CONFERENCE CENTRE OF ANTIBES JUAN-LES-PINS - FRANCE

From 1 to 6 May 2022

7th International Congress of Nematology

ICN 2022

Antibes Juan-les-Pins - France

"Crossing borders:
a world of nematode diversity and impact to discover"

Book of Abstracts

Program - Abstracts

Lists of posters

Lists of Public partners,
Sponsors & Exhibitors

Exhibition plan

IFNS
INTERNATIONAL FEDERATION OF NEMATOLOGY SOCIETIES

European Society of Nematologists

"Crossing borders: a world of nematode diversity and impact to discover"

7th International Congress of Nematology
ICN 2022
Antibes Juan-les-Pins - France

WELCOME

Dear Colleagues,

On behalf of the Organizing Committee, it is our pleasure to welcome you to Antibes Juan-les-Pins in May 2022 for the 7th International Congress of Nematology!

The International Congresses (ICN) have been hosted in cities around the world every six years since 1984. The 7th ICN was organized by the European Society of Nematology (ESN) under the auspices of the International Federation of Nematology Societies (IFNS). Antibes Juan-les-Pins, long a center of nematology, is also one of the most mythical seaside resorts of the French Riviera, dating to the 'Belle Epoque' period of the 1920s.

Delayed for two years by a global pandemic, the 7th ICN is the first to offer both on-site and virtual participation to accommodate and share this occasion with our colleagues still enduring health/ travel restrictions.

The Seventh Congress will run from May 1 to 6, 2022 with the theme of **'Crossing borders: a world of nematode diversity and impact to discover'**, a timely subject as we reconcile the global importance of agricultural production with that of environmental conservation.

Over the five days of the Congress, we will have an academic program that includes 11 plenary lectures and 31 parallel sessions covering a comprehensive variety of topics including biodiversity, biology, ecology, genetics, epidemiology, management, biocontrol, regulatory/quarantine, phylogeny and taxonomy. Poster sessions and workshops will provide further opportunities for discussing results and ideas. Significant support was available to assist students and scientists from middle and low-income countries to attend the conference.

More than a conference, the ICN 2022 will be an experience, a celebration of the best of nematology and the gathering of our global community. As you might expect when visiting France, this spirit of celebration will extend to a vibrant social program. We have scheduled a free day of activities to allow attendees to visit France and the French Riviera… unforgettable experiences.

Our sponsors are an integral part in the success of the conference, without whom none of this would be possible.

We look forward to meeting with you this week in Antibes Juan-les-Pins in 2022.

Dr. Pierre Abad, *7th ICN Chair - May 2022*
Ernesto San-Blas, *Scientific Program Chair*
Larry Duncan, *IFNS President*

"Crossing borders: a world of nematode diversity and impact to discover"

7th International Congress of Nematology
ICN 2022
Antibes Juan-les-Pins - France

COMMITTEES

THE 7th ICN ORGANIZING COMMITTEE

- IFNS President: Dr. Larry **Duncan**
- IFNS Vice President: Dr. Ernesto **San-Blas**
- IFNS Secretary: Dr. Andreas **Westphal**
- 7th ICN Chair: Dr. Pierre **Abad**

LOCAL ORGANIZING COMMITTEE

Institut Sophia Agrobiotech
INRAe, Sophia Antipolis, France

- Dr. Pierre **Abad**
- Dr. Marc **Bailly-Bechet**
- Dr. Philippe **Castagnone**
- Dr. Caroline **Caporalino**
- Dr. Etienne **Danchin**
- Dr. Janice **De Almeida-Engler**
- Dr. Daniel **Esmenjaud**
- Dr. Bruno **Favery**
- Dr. Stéphanie **Jaubert**
- Dr. Michael **Quentin**
- Dr. Laetitia **Zurletto**

Institut national de recherche pour l'agriculture, l'alimentation et l'environnement
INRAe, Sophia Antipolis, France

- Sylvie **Fajon**
- Armelle **Favery**

Biology of organisms and populations applied to Plant Protection
INRAe, Le Rheu, France

- Dr. Sylvain **Fournet**
- Dr. Eric **Grenier**
- Dr. Josselin **Montarry**

SCIENTIFIC COMMITTEE

- Dr. Ernesto **San-Blas** - *Institute of Agrifood, Animal and Environmental Sciences (ICA3), O´Higgins University, Chile*
- Prof. Ralph-Udo **Ehlers** - *e-nema GmbH, Biotechnology and biological pest management Schwentinental, Germany*
- Dr. Hans **Helder** - *Laboratory of Nematology, Wageningen University, Wageningen, The Netherlands*
- Dr. Eric **Grenier** - *Biology of organisms and populations applied to Plant Protection, INRAe, Le Rheu, France*
- Dr. Philippe **Castagnone** - *Institut Sophia Agrobiotech, INRAe, Sophia Antipolis, France*
- Dr. Catherine **Lilley** - *Centre for Plant Sciences, University of Leeds, Leeds, United Kingdom*
- Dr. Raquel **Campos-Herrera** - *Instituto de Ciencias de la Vid y del Vino, Universidad de La Rioja, Logroño, Spain*
- Dr. Sebastian **Eves-van den Akker** - *Depart. of Plant Sciences, University of Cambridge, Cambridge, United Kingdom*
- Dr. Wilfrida **Decraemer** - *Department of Biology, Ghent University, Belgium*
- Dr. Pierre **Abad** - *Institut Sophia Agrobiotech, INRAe, Sophia Antipolis, France*

7th International Congress of Nematology
ICN 2022
Antibes Juan-les-Pins - France

"Crossing borders: a world of nematode diversity and impact to discover"

COMMITTEES

SPONSORSHIP COMMITTEE

- Dr. Pierre **Abad** - *Institut Sophia Agrobiotech, INRAe, Sophia Antipolis, France*
- Dr. Caroline **Caporalino** - *Institut Sophia Agrobiotech, INRAe, Sophia Antipolis, France*
- Dr. Bruno **Favery** - *Institut Sophia Agrobiotech, INRAe, Sophia Antipolis, France*
- Dr. Sylvain **Fournet** - *Biology of organisms and populations applied to Plant Protection, INRAe, Le Rheu, France*
- Dr. Larry **Duncan** - *University of Florida, IFAS, Lake Alfred, USA*
- Dr. Johan **Deseager** - *University of Florida, IFAS, Wimauma, USA*
- Dr. Andreas **Westphal** - *University of California Riverside, Parlier, USA*
- Dr. Mieke **Daneel** - *ARC-Institute for tropical and subtropical crops, Nelspruit, South Africa*

STUDENT COMMITEE

- Dr. Stéphanie **Jaubert** - *Institut Sophia Agrobiotech, INRAe, Sophia Antipolis, France*
- Dr. Sylvain **Fournet** - *Biology of organisms and populations applied to Plant Protection, INRAe, Le Rheu, France*
- Dr. Janette **Brito** - *University of Florida, USA*

BURSARY COMMITTEE

- Larry **Duncan** - *IFNS*
- Eric **Grenier** - *ESN*
- Deliang **Peng** - *CSPN*
- Fahiem **El-Boraï** - *ESAN*
- Ralf-Udo **Ehlers** - *ESN*
- Hans **Helder** - *ESN*
- Philippe **Castagnone** - *ESN*
- Catherine **Lilley** - *ESN*
- Sebastian **Eves-van den Akker** - *ESN*
- Nathan **Garcia** - *ICN*
- Stéphanie **Jaubert** - *ICN*
- Manzoor **Soomro** - *PSN*
- María **Achinelly** - *ONTA*
- Janete **Brito** - *ONTA*
- Koon-Hui **Wang** - *SON*
- Gregory **Tylka** - *SON*
- Isgouhi **Kalosian** - *SON*
- Danny **Coyne** - *NSSA*
- Natsumi **Kanzaki** - *JSN*

ACKNOWLEDGMENT

The Organizers would like to thank **public partners, exhibitors and sponsors** for their support of the 7th International Congress of Nematology (ICN)

PUBLIC PARTNERS

Antibes Juan-les-Pins
COMMUNAUTÉ D'AGGLOMÉRATION SOPHIA ANTIPOLIS
INRAE
RÉGION SUD PROVENCE ALPES CÔTE D'AZUR
UNIVERSITÉ CÔTE D'AZUR

LEAD SPONSOR

BAYER

GOLD SPONSORS

ADAMA
BASF We create chemistry
syngenta

SILVER SPONSORS

CORTEVA agriscience
GROUPE ROULLIER

BRONZE SPONSORS

Hello NATURE
Koppert
Marrone Bio Innovations

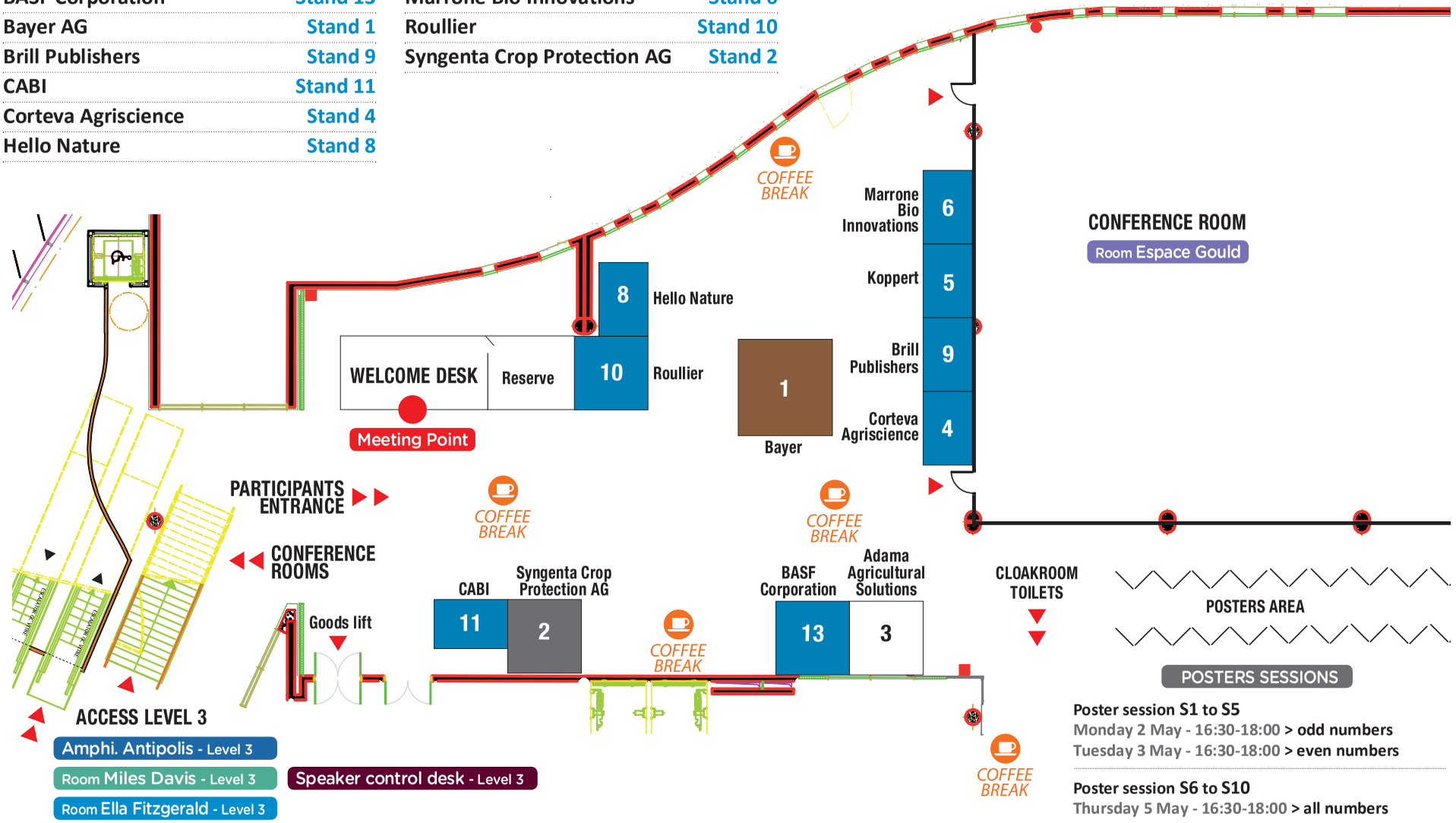
SPONSORS

TUTA SUB AEGIDE PALLAS · 1683 ·
BRILL
CABI
Limagrain from earth to life
PICO
RIJK ZWAAN
SESVANDERHAVE sugar beet seed

ESPACE GOULD, LEVEL 2 - CONFERENCE CENTRE OF ANTIBES JUAN-LES-PINS

LIST OF BOOTHS

Adama Agricultural Solutions	Stand 3	Koppert BV	Stand 5
BASF Corporation	Stand 13	Marrone Bio Innovations	Stand 6
Bayer AG	Stand 1	Roullier	Stand 10
Brill Publishers	Stand 9	Syngenta Crop Protection AG	Stand 2
CABI	Stand 11		
Corteva Agriscience	Stand 4		
Hello Nature	Stand 8		

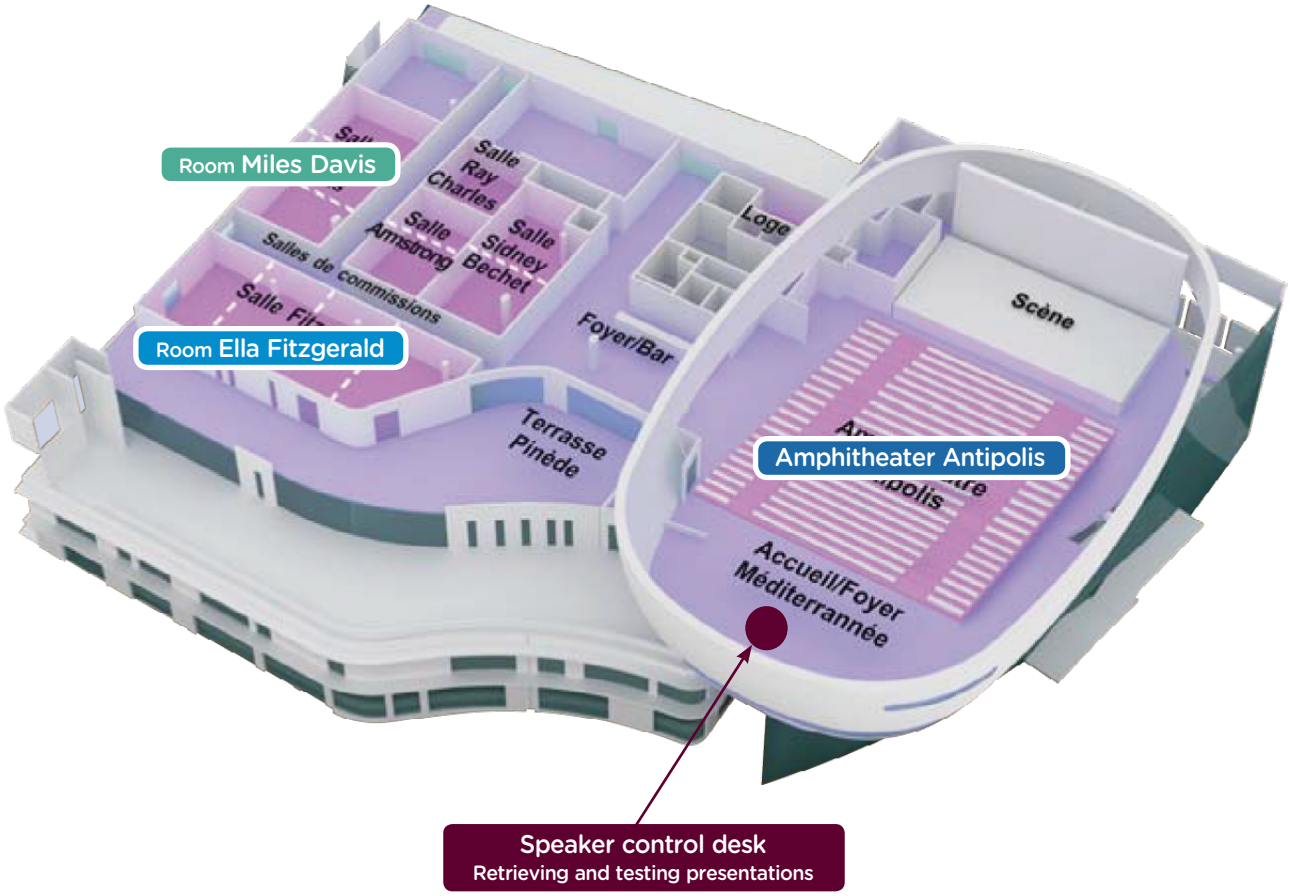


POSTERS SESSIONS

Poster session S1 to S5
Monday 2 May - 16:30-18:00 > odd numbers
Tuesday 3 May - 16:30-18:00 > even numbers

Poster session S6 to S10
Thursday 5 May - 16:30-18:00 > all numbers

FOYER MÉDITERRANÉE, LEVEL 3



TOURISM INFORMATION

The tourist office, located on the 1st level of the Congress Center, can advise you on your stay from Monday to Saturday: 9:30am to 12:30pm & 2pm to 6pm and on Sunday: from 9am to 1pm.

TABLE OF CONTENTS

Program

Abstracts Monday 2 May

Plenary Sessions

Parallel Sessions

Abstracts Tuesday 3 May

Plenary Sessions

Parallel Sessions

Abstracts Thursday 5 May

Plenary Sessions

Parallel Sessions

Abstracts Friday 6 May

Plenary Sessions

Parallel Sessions

Plenary Session

Abstracts posters

Lists of Posters

7th International Congress of Nematology
ICN 2022
Antibes Juan-les-Pins - France

PLANNING AT A GLANCE

"Crossing borders : a world of nematode diversity and impact to discover"

SUNDAY 1 MAY

Time CET*	
16:00	*Congress registration*
18:00	**WELCOME RECEPTION**
21:00	

MONDAY 2 MAY

Time CET*		
07:30	*Congress registration*	
	PLENARY SESSIONS	
08:30	Opening Ceremony	
09:00	Plenary 1	
09:30	Plenary 2	
10:00	Coffee break	
10:30	PARALLEL SESSIONS	
	Oral session 1	
	Oral session 2	
	Oral session 3	
	Oral session 4	
12:30	Lunch break	
14:00	PARALLEL SESSIONS	
	Oral session 5	
	Oral session 6	
	Oral session 7	
	Oral session 8	
16:00	Coffee break	
16:30	PARALLEL SESSIONS	Poster Session **S1** to **S5** (odd numbers)
	Workshop 1	
	Workshop 2	
	Workshop 3	
	Workshop 4	
18:00	Drinks reception	
19:00	**IFNS** Executive Meeting 1	

TUESDAY 3 MAY

Time CET*		
	PLENARY SESSIONS	
08:30	Plenary 3	
	Plenary 5	
10:00	Coffee break	
10:30	PARALLEL SESSIONS	
	Oral session 9	
	Oral session 10	
	Oral session 11	
	Oral session 12	
12:30	Lunch break	
14:00	PARALLEL SESSIONS	
	Oral session 13	
	Oral session 14	
	Oral session 15	
	Oral session 16	
16:00	Coffee break	
16:30	PARALLEL SESSIONS	Poster Session **S1** to **S5** (even numbers)
	Workshop 5	
	Workshop 6	
	Workshop 7	
	Forum 1	
18:00	**ESN** Executive Meeting 2	
19:00	**ONTA** Executive Meeting 3	

WEDNESDAY 4 MAY

Time CET*	
	Departure in front of the 'Palais des Congrès'
	• 08:30: Dive in the Cannes'islands
	• 08:45: Monaco tour
	• 09:00: Roses & Rosé tour
	• 09:15 Nice tour
10:00	CONFERENCE TOURS
18:00	

*Central European Time

THURSDAY 5 MAY

Time CET*		
	PLENARY SESSIONS	
08:30	Plenary 6	
09:00	Plenary 7	
09:30	Plenary 8	
10:00	Coffee break	
10:30	PARALLEL SESSIONS	
	Oral session 17	
	Oral session 18	
	Oral session 19	
	Oral session 20	
12:30	Lunch break	
14:00	PARALLEL SESSIONS	
	Oral session 21	
	Oral session 22	
	Oral session 23	
16:00	Coffee break	
16:30	PARALLEL SESSIONS	Poster Session **S6** to **S10** (all posters)
	Workshop 8	
	Workshop 9	
	Forum 2	
18:00		
18:15	18:15-19:30 Bus departure 19:30 Gala Evening *Villa Eilenroc*	

FRIDAY 6 MAY

Time CET*		
	PLENARY SESSIONS	
08:30	Plenary 4	
09:00	Plenary 9	
09:30	Plenary 10	
10:00	Coffee break	
10:30	PARALLEL SESSIONS	
	Oral session 24	
	Oral session 25	
	Oral session 26	
	Oral session 27	
12:30	Lunch break	
14:00	PARALLEL SESSIONS	
	Oral session 28	
	Oral session 29	
	Oral session 30	
	Oral session 31	
16:00	PLENARY SESSIONS	
16:10	Plenary 11	
16:40	Closure congress	
17:00		

Espace Gould - Level 2
Foyer Méditerranée - Level 3
Room Espace Gould - Level 2
Amphi. Antipolis - Level 3
Room Miles Davis - Level 3
Room Ella Fitzgerald - Level 3

Video conference and Q&A
Video conference only

SUNDAY 1 MAY 2022

16:00-18:00 Congress registration — Espace Gould - Level 2

18:00-21:00 Welcome reception — Foyer Méditerranée - Level 3

MONDAY 2 MAY 2022

07:30-08:30 Congress registration — Espace Gould - Level 2

Plenary Sessions

08:30-09:00 Opening Ceremony — Amphi. Antipolis - Level 3

- **Local Committee Chair** (Dr. Pierre Abad),
- **IFNS President** (Dr. Larry Duncan),
- **ESN President** (Pr. Ralf-Udo Ehlers),
- **Bayer** (Sumeja Ouyeder),
- **Université Côte d'Azur President** (Pr. Jeanick Brissewalter)

09:00-09:30 Plenary 1

- **Microgravity effect on entomopathogenic nematodes' ability to find and kill insects**
  Fatma Kaplan, David Shapiro-Ilan, Karl Cameron Schiller

09:30-10:00 Plenary 2

- **Plant Root-knot Nematode Interaction: a sophisticated dialogue**
  Pierre Abad

10:00-10:30 Coffee break - Visit of exhibition — Espace Gould - Level 2

Parallel Sessions

10:30-12:30 Oral session 1 — Amphi. Antipolis - Level 3

Plant resistance and nematode virulence

Chairs: Aska Goverse and Sebastian Kiewnick

Oral presentations

10:30-10:50
- **Multiple mechanisms of resistance to SCN encoded by soybean Rhg1**
  Andrew Bent, Shaojie Han, Adam Bayless, Katelyn Butler, John Smith, Ryan Zapotocny, Christina Fliege, Matthew Hudson, Brian Diers, Shiyan Chen, Xiaohong Wang, Derrick Grunwald

10:50-11:10
- **Epigenetic modifications in *Meloidogyne incognita* virulence to resistance gene *Mi* in tomato**
  Laetitia Perfus-Barbeoch Zurletto, Rahim Hassanaly-Goulamhoussen, Christoph Grunau, Etienne G.J. Danchin, Bruno Favery, Philippe Castagnone, Pierre Abad

11:10-11:30
- **Tackling new virulent *Globodera pallida* populations using new sources of resistance in wild potato species**
  Sebastian Kiewnick

11:30-11:45
- **The genetic basis for root-knot nematode virulence**
  David Bird

… /…

11:45-12:00 • **Proline-rich extensin-like receptor kinases mediate damage-triggered immune responses to nematode infections**
Jose L. Lozano Torres, Jaap-Jan Willig, Sonja Warmerdam, Casper C. van Schaik, Jaap Bakker, Aska Goverse, Geert Smant

12:00-12:15 • **Functional analysis of the *Ma* gene for resistance to root-knot nematodes in *Prunus***
Simon Saucet, Cyril Van Ghelder, Daniel Esmenjaud

Poster flash presentations

12:15-12:20 • **Deciphering the mechanism of ascarosides perception in plants**
Sharon Letia, Badou Mendy, Stephan H. von Reuss, Florian M.W. Grundler, M. Shamim Hasan

12:20-12:25 • **Screening of plantain and banana cultivars for resistance against *Radopholus similis* with prospects of using macropropagation plantlets for selection**
Emmanuel Olajide, Laura Cortada, Delphine Amah, Danny Coyne, Wim Bert, Rony Swennen, Yao Kolombia

12:25-12:30 • **Identification of candidate resistance genes in chickpea *(Cicer arietinum)* against *Pratylenchus thornei* using GWAS**
Sonal Channale, John Thompson, Rajeev Varshney, Mahendar Thudi, Rebecca Zwart

---

10:30-12:30 **Oral session 2** Room Espace Gould - Level 2

Soil suppressiveness and nematode control using cover crops

Chairs: Caroline Djian-Caporalino and Camille Chauvin

Oral presentations

10:30-10:50 • **Cover cropping for nematode management in nut crop production**
Andreas Westphal, Yu-Chen Wang, J. Ole Becker, Jeff Mitchell, Amelie Gaudin

10:50-11:10 • **Cover crops and litter mulching improves the soil food web structure and suppressiveness in banana fields**
Camille Chauvin, Jean-Michel Risede, Cécile Villenave, Marc Dorel

11:10-11:30 • **Evaluating sorghum as cover crop against root-knot nematodes**
Caroline Djian-Caporalino, Thierry Mateille, Marc Bailly-Bechet, Nathalie Marteu, Ariane Fazari, Pierre Bautheac, Alizée Raptopoulo, Luan Van Duong, Johannes Tavoillot, Bernard Martiny, Claire Goillon, Philippe Castagnone-Sereno

11:30-11:45 • **Suppressive effect of microbial communities extracted from the rhizospheres of tropical trees, on *M. enterolobii***
Milad Rashidifard, Hendrika Fourie, Gerhard Engelbrecht, Samad Ashrafi, Mieke Stefanie Daneel, Sarina Claassens

11:45-12:00 • **Root-knot nematode suppression and effects on soil food web by cover crop and tillage practices in a bare-ground vegetable production system**
Josiah Marquez, Timothy Coolong, Bhabesh Dutta, Abolfazl Hajihassani

12:00-12:15 • **Minimum tillage, cover crops, and dead mulch foster free-living nematodes, microbial activity and soil fertility**
Jan Henrik Schmidt, Maria R. Finckh, Matthias von Ahn, Xorla Kanfra, Holger Heuer, Johannes Hallmann

Poster flash presentations

12:15-12:20 • **Host status of different cover crops for three *Pratylenchus* species**
Betre Estifanos, Ank Theelen Peeten, Philippe Packbier, Bernd Honermeier, Johannes Hallmann

12:20-12:25 • **Using nematode community analysis to assess the resilience of agricultural soils in the Netherlands**
Anja Kombrink, Hilde Coolman, Egbert Schepel

12:25-12:30 • **Characterization and steering of the native, soil microbiome-based suppression of plant-parasitic nematodes**
Robbert van Himbeeck, Stefan Geisen, Johannes Helder

10:30-12:30 **Oral session 3** Room Miles Davis - Level 3

Ecology of free-living nematodes

Chairs: Candice Jansen van Rensburg and Thomae Kakouli-Duarte

Oral presentations

10:30-10:50 • **Mitochondrial metagenomics for evaluation of nematode diversity: paving a way into the future**
Dorota L. Porazinska, Eli Gendron, Joseph L. Sevigny, Peter Mullin, Kris Powers, Thomas O. Powers, W. Kelley Thomas

10:50-11:10 • **On the importance of species diversity in ecosystem functions and services**
Howard Ferris

11:10-11:30 • **Protected areas in southern Africa, what do we know of the nematode fauna?**
Candice Jansen van Rensburg

11:30-11:45 • **Distribution and diversity of moss-dwelling nematodes**
Bianca Kreuzinger-Janik, Nabil Majdi, Walter Traunspurger

11:45-12:00 • **Natural ecosystem diversity and functioning of nematode communities in a semi-desert ecosystem in Mexico**
Hugo H. Mejía-Madrid, Sara Sánchez-Moreno

12:00-12:15 • **Bacterial nematode grazing enhances P mobilization from phytate by *Bacillus subtilis*: what are the mechanisms involved?**
Claude Plassard, Mercedes Garcia-Sanchez, Houssein Monder, Mathilde Souche, Anne-Laure Pablo, Carlos Trives-Segura, Leyla Slamti, Julien Brillard

Poster flash presentations

12:15-12:20 • **The relations between glyphosate and soil health based on nematode trophic groups in Turkish hazelnut orchards**
Sevilhan Mennan, Gökhan Aydınlı, Ivo Brants, Xavier Belvaux, Husrev Menna

12:20-12:25 • **Soil nematode communities around gopher tortoise burrows in native and degraded ecosystems**
Rachel M. Shepherd, L.M. Uy, K.M. Gattoni, J.P. McQueen, E.M.S. Gendron, P.G. Hahn, M.A. Lashley, D.L. Porazinska

12:25-12:30 • **CRISPR and mutagenesis experiments reveal mechanisms of integration of horizontally acquired cellulases in *Pristionchus***
Ziduan Han, Ralf J. Sommer

10:30-12:30 **Oral session 4** Room Ella Fitzgerald - Level 3

EPN ecology and biology

Chairs: Raquel Campos-Herrera and Antoinette Malan

Oral presentations

10:30-10:50 • **How the spores of Isaria *fumosorosea* strain CCM8367 are spread by entomopathogenic nematodes**
Jiří Nermuť, Jana Konopická, Vladimír Půža, Rostislav Zemek

10:50-11:10 • **Machines and machine learning for applied nematology: potential and opportunity**
Denis S. Willett, Fahiem E. El-Borai, Larry W. Duncan, Camila C. Filgueiras

11:10-11:30 • **Role of *Xenorhabdus* on the metabolism of *Steinernema* infective juveniles (Nematoda: Steinernematidae)**
S. Patricia Stock, Emilie Lefoulon

11:30-11:45 • **Interactions between entomoparasitic nematodes within their host species, rose chafer grub**
Luca Eszter Balog, Oleksandr Holovachov, Júlia Katalin Török

11:45-12:00 • **Molecular effectors in immune modulation and pathogenicity by entomopathogenic nematodes**
Adler Dillman, Sophia Parks, Chau Nguyen, Shyon Nasrolahi, Dihong Lu, Raghavendran Ramaswamy, Anna Buchman, Omar Akbari, Naoki Yamanaka, Martin Boulanger

12:00-12:15 • **Key transcriptional changes in the shifting from free-living to infective stage of *Heterorhabditis bacteriophora***
Nelson Simões, Duarte Toubarro

… /…

Poster flash presentations

12:15-12:20 • **War in the darkness: the use of volatile organic compounds and entomopathogenic nematodes to control wireworms**
Diana la Forgia, François Verheggen, Andrea Chacon

12:20-12:25 • **Mass spectrometry-driven discovery of neuropeptidergic systems regulating nictation in free-living and parasitic nematodes**
Bram Cockx, Rose Boelen, Johnathan Dalzell, Liesbet Temmerman

12:25-12:30 • **Standardized surveys confirm greater EPN presence and diversity in a subtropical compared to Mediterranean citrus orchards**
Alexandros Dritsoulas, Fahiem El-Borai, Mostafa Hammam, Mahfouz Abd-Elgawad, Larry Duncan, Ioannis Giannakou

---

12:30-14:00 Lunch - Visit of exhibition — Espace Gould - Level 2

Parallel Sessions

14:00-16:00 **Oral session 5** — Amphi. Antipolis - Level 3

Plant resistance and nematode virulence (continued)

Chairs: Peter DiGennaro and David Bird

Oral presentations

14:00-14:20 • **How plants recognize nematodes: Signals and signalling**
Shahid Siddique, Florian Grundler

14:20-14:40 • **Inducing plant resistance against parasitic nematodes: Preparing for battle**
Tina Kyndt, Willem Desmedt, Anikó Meijer, Jonas De Kesel, Mohammad Reza Atighi, Richard Raj Singh, Eva Degroote

14:40-15:00 • **30 years of research on resistance against *Globodera pallida*: an overview**
Sylvain Fournet, Marie Claire Kerlan, Josselin Montarry, Eric Grenier

15:00-15:15 • **Resistance loci interactions discovery through comprehensive transcriptomics**
Peter DiGennaro, Weiming Hu, Vincent Colantonio

15:15-15:30 • **Dehydroascorbate activates induced resistance in rice against root-knot nematode *Meloidogyne graminicola***
Satish Namdeo Chavan, Jonas De Kesel, Willem Desmedt, Eva Degroote, Richard Raj Singh, Nguyen Thu Giang, Kristof Demeestere, Tim De Meyer, Tina Kyndt

15:30-15:45 • **Comparative transcriptome analysis reveals the specific activation of defense pathways against *Globodera pallida* in Gpa2 resistant potato roots**
Qi Zheng, André Bertran, Anouk Brand, Casper C. van Schaik, Stefan van de Ruitenbeek, Geert Smant, Aska Goverse, Mark G. Sterken

Poster flash presentations

15:45-15:50 • **Investigation of resistance against *Ditylenchus dipsaci* on sugar beet**
Alan Storelli, Andreas Keiser, Sebastian Kiewnick, Mario Schumann, Werner Beyer, Matthias Daub, Michael Rostás, Anne-Kathrin Mahlein

15:50-15:55 • **Prospective identification through DNA-capture technologies of a rice resistance gene to control *Meloidogyne graminicola***
Sophie Mantelin, Hue Nguyen Thi, Miles Armstrong, Hai Ho-Bich, Lizhong Xiong, Mathias Lorieux, John Jones, Ingo Hein, Stéphane Bellafiore

15:55-16:00 • **Host status of Crop plants to *Meloidogyne enterolobii* populations**
Hemanth Kumar Koniganahalli Gopal, Sebastian Kiewnick, Etienne G.J. Danchin

14:00-16:00 **Oral session 6** Room Espace Gould - Level 2

Phylogenetics/Phylogenomics: the latest updates on the Phylum Nematoda

Chairs: Sergei Subbotin and Johannes Helder

Oral presentations

14:00-14:20 • **Evolution and diversification of plant and animal parasitism within the phylum Nematoda**
Johannes Helder, Martijn Holterman, Sven Van den Elsen, Geert Smant, Stefan Geisen, Michaela Schratzberger

14:20-14:40 • **Nematode phylogenomics - how far did we progress?**
Oleksandr Holovachov, Mohammed Ahmed, Ashleigh B. Smythe, Kevin M. Kocot

14:40-15:00 • **Phylogeny and systematics of Aphelenchoididae: overview and problems**
Natsumi Kanzaki, Robin Giblin-Davis

15:00-15:15 • **An update to the phylogeny of Aphelenchoidea; Ektaphelenchinae, Seinurinae and Tylaphelenchinae, as case studies**
Majid Pedram, Farahnaz Jahanshahi Afshar

15:15-15:30 • **High-quality genome assembly of an emerging root-knot nematode species, *Meloidogyne luci***
Nik Susič, Georgios D. Koutsovoulos, Cristian Riccio, Etienne G. J. Danchin, Mark L. Blaxter, David H. Lunt, Polona Strajnar, Saša Širca, Gregor Urek, Barbara Gerič Stare Maybe

15:30-15:45 • **Phylogeography, phylogeny and DNA barcoding of the cyst nematodes**
Sergei Subbotin

Poster flash presentations

15:45-15:50 • **Genetic and phylogenetic characterization of different populations of *Meloidogyne izalcoensis*, a new species from coffee in Brazil**
Marcilene Santos, Sheila Almeida, Vanessa Mattos, Marina Carneiro, Philippe Castagnone-Sereno, Regina Carneiro

15:50-15:55 • **Phylogeography of *Ditylenchus gallaeformas***
Samara A. Oliveira, Saara DeWalt, Paula Agudelo

15:55-16:00 • **Molecular identification and phylogenetic diversity of cereal cyst nematode (*Heterodera* spp.) populations from Algeria**
Mehalaine Khawla, Imren Mustafa, Dababat Abdelfattah

14:00-16:00 **Oral session 7** Room Miles Davis - Level 3

Biodiversity of aquatic nematodes

Chairs: Federica Semprucci and Nabil Majdi

Oral presentations

14:00-14:20 • **Effects of duration and frequency of non-flow periods on stream-dwelling nematode communities**
Nabil Majdi, Miriam Colls, Linette Weiss, Vicenç Acuña, Sergi Sabater, Walter Traunspurger

14:20-14:40 • **Taxonomic and functional diversity of marine nematodes in the biomonitoring: reality or utopia?**
Federica Semprucci

14:40-15:00 • **Community structure of deep-sea nematodes: a metabarcoding approach**
Lara Macheriotou, Sofie Derycke, Ann Vanreusel

15:00-15:15 • **Resource diversity effects on cryptic marine nematode species: a multi-faceted approach**
Rodgee Mae Guden, Derycke Sofie, Tom Moens

15:15-15:30 • **Can we predict sediment quality with nematode metabarcoding?**
Janina Schenk, Nils Kleinbölting, Sebastian Höss, Marvin Brinke, Walter Traunspurger

15:30-15:45 • **Aquatic nematodes as bio-indicators of crude oil water-soluble fractions (WSFs) toxicity: a microcosm approach**
Luana Monteiro, Tom Moens, Walter Traunspurge

… /…

Poster flash presentations

15:45-15:50 • **Tobrilidae communities in western Nebraska sandhill lakes are driven by alkalinity and biotici interactions**
Kaitlin Gattoni, Abigail Borgmeier, J. Parr McQueen, Peter Mullin, Kirsten Powers, Thomas Powers, Dorota Porazinska, Eli Gendron

15:50-15:55 • **Nematode predators catalyze an increase of chloroviruses by foraging on the symbiotic hosts of zoochlorellae**
Kirsten Powers, Zeina Al-Ameeli, John Delong, Steven Thomas, James Van Etten, David Dunigan, Thomas Powers

15:55-16:00 • **Spatial distribution patterns of microbiome and free-living benthic nematodes in response to sediment ecological conditions in Sado estuary, Portugal**
Soraia Vieira, Kasia Sroczyńska, Marta Martins, Maria Costa, Joana Neves, Helena Adão, Cláudia Vicente

---

14:00-16:00 **Oral session 8** Room Ella Fitzgerald - Level 3

Management of nematodes in tropical conditions

Chairs: Biodun Claudius-Cole and Haddish Melakeberhan

Oral presentations

14:00-14:20 • **Management of potato cyst nematode and bacterial wilt by use of banana fiber paper in potatoes**
Effie Ochieng, John Kimenju, Douglas Miano, Kalpana Sharma, Charles Opperman, Tahira Pirzada, Laura Cortada-Gonzalez, Danny Coyne

14:20-14:40 • **Management of nematodes in peri-urban vegetable farms in West Africa**
Abiodun Claudius-Cole

14:40-15:00 • **Challenges and management of plant-parasitic nematodes on plantain, *Musa* spp. AAB-subgroup, in Nigeria**
Monioluwa Omolara Olaniyi

15:00-15:15 • **Adopting integrated nematode-soil health management in smallholder potato farms in the highlands of Guatemala**
Stephen Kakaire, Amilcar Sanchez, Anibal Sacbaja, Catherine Chan, Brent Sipes, Haddish Melakeberhan

15:15-15:30 • **The effects of orange and lemon juice on root-knot nematode population densities and plant growth under greenhouse**
Grace Tefu, Mieke Daneel, Driekie Fourie

15:30-15:45 • **Integrated management of *Meloidogyne incognita* and *Fusarium oxysporum* in cucumber under protected cultivation system**
Jaydeep Ashok Patil, Anil Kumar, Sewak Ram

Poster flash presentations

15:45-15:50 • **EUPHRESCO – MELORISK: Preventing *Meloidogyne graminicola* spread in European rice paddies**
Maria Lurdes Inácio, Leidy Rusinque, Filomena Nobrega, Rita Varela

15:50-15:55 • **Dominance index of soil nematodes on a coffee plantation after cost-effective bionematicide application**
Iis Nur Asyiah, Laeli Nordiana, Balawara Andika, Sugeng Winarso, Lenny Widjayanthi, Dwi Nugroho, Kurniawan Firmansyah, Ankardiansyah Pandu Pradana

15:55-16:00 • **Control of the rice root-knot nematode *Meloidogyne graminicola* using rice plants as trap crops**
Giulia Torrini, Stefano Sacchi, Leonardo Marianelli, Giuseppe Mazza, Annachiara Fumagalli, Beniamino Cavagna, Pio Federico Roversi, Mariangela Ciampitti

---

16:00-16:30 Coffee break - Visit of exhibition Espace Gould - Level 2

16:30-18:00 **Poster Session S1 to S5** **(odd numbered posters presented)** Espace Gould - Level 2

Parallel Sessions

16:30-18:00 **Workshop 1** Room Miles Davis - Level 3

Expanding indicator qualities of nematodes to identify sustainable soil health

Chairs: Haddish Melakeberhan

- **Abstract general**

16:30-18:00 **Workshop 2** Room Ella Fitzgerald - Level 3

Aphelenchoides besseyi: reemergence of a forgotten parasite

Chair: Johan Desaeger

- **Abstract general**
- **Occurrence and control of *Aphelenchoides besseyi* in China**
  Hongli Ji, Jialian Xie, Fang Yang, Xing Xu, Yunliang Peng
- **Summer crimp disease (*Aphelenchoides besseyi*): Species delimitation and feeding habits**
  Clemen De Oliveira, Johan Desaeger, Sergei Subbotin, Renato Inserra, Janete Brito, Natalia Peres
- ***Aphelenchoides besseyi:* Emerging challenge in Brazilian crops**
  Andressa Machado
- **General discussion**

16:30-18:00 **Workshop 3** Amphi. Antipolis - Level 3

Advances and challenges in CRISPR-mediated technologies in parasitic and free-living nematodes

Chairs: Sebastian Eves-van den Akker and Ralf J. Sommer

- **Abstract general**
- **CRISPR-mediated reverse genetic approaches in *P. pacificus* and potential modifications for plant parasitic nematodes**
  Ralf Sommer, Witte Hanh
- **TransPPN: The transformation of plant-parasitic nematodes consortium**
  Sebastian Eves-van den Akker, The TransPPN Consortium
- **Liposome-mediated CRISPR and RNAi in parasitic and free-living nematodes**
  Andre Pires da Silva, Sally Adams, Hongguang Shao, James B. Lok, Prachi Pathak
- **General discussion**

16:30-18:00 **Workshop 4** Room Espace Gould - Level 2

DNA barcoding of nematodes

Chairs: Thomas Powers and Dorota Porazinska

- **Abstract general**

18:00-19:00 **IFNS - Executive Meeting 1** Room Ella Fitzgerald - Level 3

18:00 **Drinks reception** Espace Gould - Level 2

TUESDAY 3 MAY 2022

Plenary Sessions

08:30-09:00 Plenary 3 — Amphi. Antipolis - Level 3

- **A model nematode outside the laboratory: *Caenorhabditis elegans* habitat and biotic interactions**
  Marie-Anne Félix

09:30-10:00 Plenary 5

- **Nematode chemosensation: implications on insect pest management**
  Ivan Hiltpold

10:00-10:30 Coffee break - Visit of exhibition — Espace Gould - Level 2

Parallel Sessions

10:30-12:30 Oral session 9 — Amphi. Antipolis - Level 3

'Omics' in nematology

Chairs: Adler Dillman and Etienne Danchin

Oral presentations

10:30-10:50
- **Revealing the 'box' code: the spatial and temporal regulation of plant-parasitic nematode pathogenicity**
  Clement Pellegrin, Helen Beasley, Badou Mendy, M.Shamim Hasan, Divykriti Chopra, Clarissa Hiltl, Huiyan Chen, Volkan Cevik, Oliver Chitambo, Florian M.W. Grundler, Shahid Siddique, Sebastian Eves-van den Akker

10:50-11:10
- **Parthenogenomics - using population genomics to understand the evolution of parthenogenetic triploid nematodes**
  Laura Villegas, Laura Pettrich, Ann-Marie Waldvogel, Philipp Schiffer

11:10-11:30
- **Evolutionary and comparative genomics of tropical root-knot nematodes**
  Georgios Koutsovoulos, Corinne Rancurel, Laetitia Zurletto, Martine Da Rocha, Djampa K. Kozlowski, Benjamin Noël, Patrick Wincker, Etienne G.J. Danchin

11:30-11:45
- ***Meloidogyne hapla* with improved assembly and genome annotation**
  Pallavi Shakya, Alison Coomer, Valerie Williamson, Shahid Siddique

11:45-12:00
- **Genome evolution of *Aphelenchoides besseyi* strains**
  Cheng-Kuo Lai, Peichen J. Chen, Yi-Chien Lee, Huei-Mien Ke, Hsin-Han Lee, Chan-Yi Ivy Lin, Isheng Jason Tsai

12:00-12:15
- **A genome-wide comparison between two Dutch *Globodera pallida* populations**
  Martijn Holterman, Mark Sterken, Joris Van Steenbrugge, Sven Van den Elsen, Anne Van Diepeningen, Geert Smant, Johannes Helder

Poster flash presentations

12:15-12:20
- **In search of a common target for control of nematode and aphid pests**
  Maria Maqsood, John Fosu-Nyarko, Michael G.K. Jones

12:20-12:25
- **Metagenomics mining improves analysis of horizontal gene transfers involved in parasitic function in plant-parasitic nematodes**
  Carole Belliardo, Adrien Deceneux, Corinne Rancurel, Marc Bailly-Bechet, Etienne G.J. Danchin

12:25-12:30
- **Development of mitometagenomics protocols for the enhancement of nematode identification and biodiversity study**
  Eli Gendron, Joseph Sevigny, Thomas Powers, W. Kelly Thomas, Dorota Porazinska

10:30-12:30 **Oral session 10** Room Espace Gould - Level 2

Advances in precision agriculture: instrumentation and nematode IPM applications

Chairs: Joseph Noling and Mieke Daneel

Oral presentations

10:30-10:50 • **Precision nematode management, a modern approach to incorporate new technologies and chemistries to control nematodes**
Pablo A. Navia Gine

10:50-11:10 • **Assessing crop impacts and nematode management options using NDVI and canopy greenness from aerial imaging**
Joseph W. Noling

11:10-11:30 • **Estimating field populations of *Heterodera schachtii* from hyperspectral signatures of sugar beet canopies**
Matthias Daub, Kai Schmidt

11:30-11:45 • **The use of infrared spectroscopy and machine learning tools for detection of *Meloidogyne* infestations**
Mayamaru Guerra, Ana Maria Casassa, Nestor Cubillan, Ernesto San-Blas

11:45-12:00 • **Multiseasonal modelling of plant-nematode interactions reveals efficient resistance deployment strategies**
Vincent Calcagno, Suzanne Touzeau, Ludovic Mailleret, Samuel Nilusmas, Caroline Dijan Caporalino, Philippe Castagnone

12:00-12:15 • **Combining traditional and precision agricultural tools to improve management of plant-parasitic nematodes in cotton production**
Zane Grabau, R. Barocco, C. Liu, D.L. Wright, I. Small

12:15-12:30 • **Models for map-based prediction of *Pratylenchus penetrans* based on soil variables**
Ann MacGuidwin, Gary Pack, Ibrahim Saeed

10:30-12:30 **Oral session 11** Room Miles Davis - Level 3

Nematode-vector relationships

Chairs: Juan Palomares-Rius and Gérard Demangeat

Oral presentations

10:30-10:50 • **Capsid determinants involved in the specific transmission of the grapevine fanleaf virus by its nematode vector, *Xiphinema index***
Gérard Demangeat

10:50-11:10 • **The importance of nematode dauer in the adaptation to its life cycle and ecological niche**
Lieke Vlaar, Mehran Rahimi, Andre Machado Bertran, Aska Goverse, Jan Kammenga, Harro Bouwmeester

11:10-11:30 • **Linking biodiversity and pathogen abundance in natural nematode populations**
Lisa van Sluijs, Jessica N. Sowa, Hala Tamim El Jarkass, Aaron Reinke, Wenjia Wu, Robbert van Himbeeck, Sem Aslan, Joost A. G. Riksen

11:30-11:45 • **Unraveling the biotic interactions between *Bursaphelenchus xylophilus, Pinus pinaster* and nematophagous fungi, *Esteya* spp.**
David Pires, Cláudia Vicente, Manuel Mota, Maria L. Inácio

11:45-12:00 • **Molecular nematode-viroid-fungus interface on *Celosia argentea* and *Solanum lycopersicum***
Pakize Gok Guler, Refik Bozbuga, Semiha Yucee

12:00-12:15 • **Diversity of fungal communities associated with Pinus pinaster infected by *Bursaphelenchus xylophilus***
Cláudia Vicente, Jorge Faria, Helena Bragança, Luís Bonifácio, Edmundo Sousa, Margarida Espada, Steve Woodward, Anna Maria Vettraino, Filomena Nóbrega, Maria L. Inácio

12:15-12:30 • **Durability of muscadine-derived resistant material to *Xiphinema index* and first detection of sexual reproduction events of the nematode under controlled conditions**
Van Chung Nguyen, Jean-Pascal Tandonnet, Samira Khallouk, Cyril Van Ghelder, Ulysse Portier, Mohamed Youssef Banora, Nathalie Ollat, Maria-Das-Dores Lafargue, Daniel Esmenjaud

… /…

10:30-12:30 **Oral session 12** Room Ella Fitzgerald - Level 3

Chemical control of nematodes

Chairs: Billy Crow and Brigitte Slaats

Oral presentations

10:30-10:50 • **NanoEngineering gone viral: plant virus-nanotechnologies for precision farming**
Nicole F. Steinmetz

10:50-11:10 • **Tymirium™: The story of a powerful new molecule**
Brigitte Slaats, Matthias Gaberthueel, Olivier Loiseleur, Anthony Flemming, Torsten Luksch

11:10-11:30 • **"Wrap and Plant": A novel concept for managing plant-parasitic nematodes with banana paper in sub-Saharan Africa**
Charles Opperman, Antoine Affokpon, Laura Cortada, Danny Coyne, Juliet Ochola, Baldwyn Torto, Richard Guenther, Steve Koenning, Reny Mathew, Tim Sit, Med Bird, Tahira Pirzada, Lokendra Pal, Eric Davis, Saad Khan

11:30-11:45 • **Effect of New non-fumigant nematicides on different trophic groups of nematodes**
David Moreira, Johan Desaeger

11:45-12:00 • **Evaluation of sequential applications of chemical and biological nematicides in vegetable production in southern Italy**
Helmut Fuersch, Joachim Meyer, Alberto Boebel, Svenja Bellof

12:00-12:15 • **The distinct profile of the inhibitory effects of fluensulfone on *Globodera pallida* hatching**
Emily Feist, James Kearn, Yogendra Gaihre, Vincent O'Connor, Lindy Holden-Dye

Poster flash presentations

12:15-12:20 • **Salibro™ (Reklemel™ Active): A novel nematicide for the control of *Meloidogyne* spp. in key annual and perennial crops in North America & Mexico**
Bo Braxton, Leonel Aviles, Sunil Tewari, Julian Mejia, Laura Smith, Tim Thoden, Yannis Stamatas

12:20-12:25 • **Discovery and characterization of bioactivated nematicides for selective control of parasitic nematodes**
Jessica Knox, Savina R. Cammalleri, Andrew R. Burns, Jack M.P. Castelli, Megan Kitner, Inga Zasada, Peter J. Roy

---

12:30-14:00 Lunch - Visit of exhibition Espace Gould - Level 2

Parallel Sessions

14:00-16:00 **Oral session 13** Amphi. Antipolis - Level 3

'Omics' in nematology (continued)

Chairs: Ralf Sommer and Georgios Koutsovoulos

Oral presentations

14:00-14:20 • **Uncovering the 'dark matter' of worm biology: a universe of signalling molecules**
Frank Schroeder

14:20-14:40 • **Juxtaposition of extreme genomic variability and stability in HYP effectors of potato cyst nematodes**
Unnati Sonawala, Helen Beasley, Sebastian Eves-van den Akker

14:40-15:00 • **Analysis of the banana root transcriptome in response to root-knot nematode infection and water deficit**
Robert Miller, Leticia Dias de Freitas, Jose Dijair Antonino, Gabriel Sergio Costa Alves, Claudia Fortes Ferreira, Mauricio Antonio Coelho Filho, Edson Perito Amorim, Roberto Coiti Togawa, Priscila Grynberg

15:00-15:15 • **miR167-ARF8, an auxin responsive couple involved in the formation of galls induced by root-knot nematodes in tomato**
Yara Noureddine, Martine Da-Rocha, Clémence Médina, Mohamed Zouine, Joffrey Mejias, Michael Quentin, Pierre Abad, Bruno Favery, Stéphanie Jaubert-Possomai

15:15-15:30 • **Genetic variation in *Globodera pallidalinked* to virulence in North-West Europe**
Mark Sterken, Joris van Steenbrugge, Dennie te Molder, Casper Van Schaik, Stefan van de Ruitenbeek, Sven Van Elsen, Geert Smant

… /…

15:30-15:45 • **Single nematode genome assemblies**
Erna King, Lewis Stevens, Christopher Laumer, Mark Blaxter

Poster flash presentations

15:45-15:50 • **Mining new nematode effectors interacting with plant transcription factors by Cr-Y2H**
Hui Xiang, Stojilković Boris, Gheysen Godelieve

15:50-15:55 • **SMART UP – Spatial Mapping of Root Transcriptomes Upon Nematode Parasitism**
Anna Pijnacker, Rik Korswagen, Aska Goverse, Geert Smant1, Jose Lozano Torres

15:55-16:00 • **The strange chromosome ends of root-knot nematodes**
Georgios D. Koutsovoulos, Evelin Despot-Slade, Martine Da Rocha, Claire Caravel, Pierre Abad, Nevenka Meštrović, Etienne G.J. Danchin

---

14:00-16:00 **Oral session 14** Room Ella Fitzgerald - Level 3

Social impact of nematode management

Chairs: Danny Coyne and Miguel Talavera

Oral presentations

14:00-14:20 • **Global 'worming': changing economic and environmental impact of plant-parasitic nematodes**
Wim Wesemael

14:20-14:40 • **Social Implications of new advances in nematode management**
Johan Desaeger

14:40-15:00 • **An industry perspective on the global social impact of nematode management - innovation and food security**
John Wiles, Tim C. Thoden

15:00-15:15 • **Microbial-based pesticides, a novel source of new tools for nematode management**
Jared P. Jenssen

15:15-15:30 • **Global impact of agricultural intensification on soil nematodes: a meta-analysis**
Jeremy Puissant, Cécile Villenave, Camille Chauvin, Eric Blanchart, Jean Trap

15:30-15:45 • **A cost-benefit and efficacy analysis of *Meloidogyne* management strategies in Mediterranean intensive horticulture**
Miguel Talavera, Luis Miranda, María Dolores Vela, Soledad Verdejo-Lucas

Poster flash presentations

15:45-15:50 • **Abundance and diversity of plant-parasitic nematodes in the rhizosphers of maize cultivars grown by commercial farmers in rural areas of South Africa**
Nancy Ntidi, Akhona Mbatyoti, Driekie Fourie, Lindokuhle Qwabe

15:50-15:55 • **Banana fibre paper: effectively delivering ultra-low nematicide dosages for more acceptable nematode management**
Danny Coyne, Laura Cortada-González, Solveig Haukeland, Baldwyn Torto, Charles Opperman

15:55-16:00 • **Best management practices for root-knot nematode *(Meloidogyne hapla)* in daylily *(Hemerocallis* spp.) production**
Amanda Howland, Emilie Cole, Kristin Poley, Marisol Quintanilla

---

14:00-16:00 **Oral session 15** Room Miles Davis - Level 3

Advances in nematode detection and identification: instrumentation and applications

Chairs: Laurent Folcher and Barbara Geric-Stare

Oral presentations

14:00-14:20 • **Non-invasive detection of plant parasitic nematodes using hyperspectral imaging**
Barbara Gerič Stare

14:20-14:40 • **European and national reference laboratories: an EU network for improved surveillance of plant-parasitic nematodes**
Maíra Grossi de Sá, Nicole Viaene, Sylvie Gamel, Nicole Damme, Françoise Munaut, Laurent Folcher

… /…

14:40-15:00 • **Use of automated image analysis techniques for species identification and classification**
Romain Thevenoux, Heloïse Villesseche, Laurent Folcher, Eric Grenier, Nicolas Parisey

15:00-15:15 • **Lab-on-chip for *Globodera pallida* detection**
Maria João Camacho, Verónica C. Martins, Eugénia Andrade, Débora C. Albuquerque, Maria L. Inácio, Manuel Mota, Paulo Freitas

15:15-15:30 • **Digitize your nematode control**
Sumeja Ouyeder and Marc Rist, Hartwig Dauck, Jose Luis Robles

15:30-15:45 • **Developing a droplet digital PCR assay for detection of the stubby root nematode *Paratrichodorus allius***
Guiping Yan, Zhuoyu Wang, Addison Plaisance

Poster flash presentations

15:45-15:50 • **A novel approach for applying machine learning for detection and phenotyping of cyst nematodes in soil extracts**
Long Chen, Martin Strauch, Matthias Daub, Marcus Jansen, Susanne Schultz-Kuhlmann, Stefan Krüssel, Hans-Georg Luigs, Dorit Merhof

15:50-15:55 • **ATR-FTIR spectroscopy and hyperspectral imaging in determining quality of formulated entomopathogenic nematodes**
Nicholas Kagimu, Antoinnette P. Malan

15:55-16:00 • **Rapid detection and quantification of plant-parasitic nematodes from large volumes of soil**
Valeria Orlando, Thomas Prior, Michael R. Surrey, Steven Bryce, Wellcome Ho, Brett J. R. Alexander

---

14:00-16:00 **Oral session 16** Room Espace Gould - Level 2

Biological control of nematodes

Chairs: Sylvia Schleker and Eric Nguema-Ona

Oral presentations

14:00-14:20 • **Biological agents to invigorate the health of established coffee trees by managing plant parasitic nematodes**
Kanan Saikai, Celestine Oduori, Wanjala Situma, Simon Njoroge, Ruth Murunde, Danny Coyne

14:20-14:40 • **Soil microbiota effect on the efficiency of root exudates to hatch cyst nematodes**
Josselin Montarry, Camille Gautier, Lisa Martinez, Sylvain Fournet, Eric Nguema-Ona, Anne-Yvonne Guillerm-Erckelboudt, Christophe Piriou, Juliette Linglin, Christophe Mougel, Lionel Lebreton

14:40-15:00 • **Activity of *Bacillus firmus* against plant-parasitic nematodes**
A. Sylvia S. Schleker, Mengmeng Huang, Aylin Bulut, Christiane Matera, Bidhya Shrestha, Volkan Cevik, Ute Schlee, Florian M. W. Grundler

15:00-15:15 • **BIODERA project: a teamwork effort to develop biocontrol solutions against plant parasitic nematodes**
Eric Nguema-Ona

15:15-15:30 • **Co-occurrence of soil microbial communities and root-knot nematode populations in strawberry farms, Egypt**
Mahfouz Abd-Elgawad , Hassan Abd-El-Khair, Mostafa M.A. Hammam, Wafaa M. A. El-Nagdi, Mahfouz M. M. Abd-Elgawad

15:30-15:45 • **Genome-wide association identifies genomic regions involved in production of nematicidal compounds in the biocontrol fungus *Clonostachys rosea***
Mudassir Iqbal, Martin Broberg, Deepak Haarith, Anders Broberg, Kathryn E. Bushley, Mikael Brandström Durling, Maria Viketoft, Dan Funck Jensen, Mukesh Dubey, Magnus Karlsson

15:45-16:00 • ***Pochonia chlamydosporia* var. mexicana response to physicochemical factors, rhizosphere colonization and egg parasitism**
María Gabriela Medina-Canales, Adriana Joselly Del Razo-Méndez, Brenda Estefany Ortiz-González, Gabriela Susana Alba-Ramírez, Rosa Helena Manzanilla-López, Alejandro Tovar-Soto

16:00-16:30 Coffee break - Visit of exhibition — Espace Gould - Level 2

16:30-18:00 **Poster Session S1 to S5** (even numbered posters presented) — Espace Gould - Level 2

Parallel Sessions

16:30-18:00 **Workshop 5** — Room Ella Fitzgerald - Level 3

Nematode-bacteria symbiosis

Chair: Patricia Stock

- Abstract general
- Take my breath away – physiological adaptations of symbiotic marine nematodes to oxic-anoxic interfaces
  Silvia Bulgheresi
- Nematode – bacterial interactions in a beetle ecosystem: ‚Boom and bust' dynamics of predatory nematode *P. pacificus*
  Ralf Sommer, Tess Renahan
- To have and to hold: on the mutualistic partnership of *Steinernema* nematodes and their *Xenorhabus* symbionts
  Patricia Stock
- General discussion

16:30-18:00 **Workshop 6** — Room Espace Gould - Level 2

Nemaplex and NINJA: Features and Uses

Chairs: Sara Sánchez-Moreno and Ron de Goede

- Abstract general
- Briefly review of the origins and evolution of Nemaplex from around 1979 to the present day
  Howard Ferris
- Demonstration of the features and navigation of the Nemaplex website…
- Demonstrations of the use of NINJA Faunal Analysis feature to evaluate the ecosystem and soil food web conditions suggested by specific nematode assemblages…

16:30-18:00 **Workshop 7** — Room Miles Davis - Level 3

Grape vines nematode management

Chairs: Daniel Esmenjaud and Cesar Bauer Gomes

- Abstract general
- Grape-associated phytoparasitic nematodes in Brazil
  Cesar Bauer Gomes
- Management approaches of plant-parasitic nematodes in grape in the changing agricultural landscape of California
  Andreas Westphal, Tom Buzo, Zin Thu Zar Maung, Gabriel Torres, Matthew Fidelibus, Andrew Walker
- Plant parasitic nematodes infesting grapevines in Portugal
  Carlos Gutiérrez-Gutiérrez, Manuel Mota
- Natural rootstock resistance to control *Xiphinema index* and RKNs in Vines.
  Daniel Esmenjaud
- Resistance durability to X. *index* in grapevine
  Van Chung Nguyen

16:30-18:00 **Forum 1** — Amphi. Antipolis - Level 3

Young nematologists Network

Chair: TBD

• Abstract general

18:00-19:00 **ESN - Executive Meeting 2** — Room Miles Davis - Level 3

18:00-19:00 **ONTA - Executive Meeting 3** — Room Ella Fitzgerald - Level 3

WEDNESDAY 4 MAY 2022

CONFERENCE TOURS

08:30 **Dive in the Cannes'islands** — Departure in front of the Congress Center

08:45 **Monaco tour** — Departure in front of the Congress Center

09:00 **Roses & Rosé tour (Agricultural tour)** — Departure in front of the Congress Center

09:15 **Nice tour** — Departure in front of the Congress Center

THURSDAY 5 MAY 2022

Plenary Sessions

08:30-09:00 Plenary 6 — Amphi. Antipolis - Level 3

- ***Caenorhabditis elegans*** **and other nematodes as biological models**
  Lindy Holden-Dye

09:00-09:30 Plenary 7

- **Using *A. suum* to study the intestine of animal/human parasitic nematodes**
  Makedonka Mitreva

09:30-10:00 Plenary 8

- **Legislation and regulatory aspects of plant nematodes**
  Manuel Mota

10:00-10:30 Coffee break - Visit of exhibition — Espace Gould - Level 2

Parallel Sessions

10:30-12:30 Oral session 17 — Amphi. Antipolis - Level 3

Integrated nematode management

Chairs: Kathy S. Lawrence and Lais Fontana

Oral presentations

10:30-10:50 • **Integrated management of cyst nematodes in agricultural important crops in China**
Deliang Peng, Ricardo Holgado, Huan Peng, Wenkun Huang, Jingwu Zheng, Shulong Chen

10:50-11:10 • **Bridging the vegetable IPM GAP in Africa: from here to sustainerty**
Danny Coyne

11:10-11:30 • **IPM options in cotton production with plant parasitic nematodes in the USA**
Kathy Lawrence

11:30-11:45 • **Can *Meloidogyne* and *Helicotylenchus* populations be manipulated using soil nutrient levels?**
Mieke Daneel, Lesego Matlala, Hendrika Fourie

11:45-12:00 • **Pathogenic variability of the population of *Pratylenchus brachyurus* in cover crops**
Lais Fontana, Bruna Orlandini Toninato, Glaucia Leticia Sete da Cruz, Ana Cristina Da Silveira, Angélica Sanchez Melo, Guilherme Tarini, Cláudia Regina Dias-Arieira

12:00-12:15 • **A complex between the plant parasitic nematode and a fungus - reevaluating *Pratylenchus capsici* disease etiology**
Sigal Braun Miyara, Patricia Bucki, Qing Xue, Svetlana Duvrinin, Ohad Avraham, Nathalia Sichov, Abraham Gamliel

Poster flash presentations

12:15-12:20 • **Synergies between climate change impacts and conservation tillage practices on agricultural soils functionality**
Paula Lillo, Mª Ángeles Hernández, Miguel Ángel Porcel, Sara Sánchez-Moreno

12:20-12:25 • **A new mode of action classification scheme for nematode control agents ("nematicides") - nematode working group of IRAC**
John Wiles, Ionit Iberkleid, Huazhang Huang, Tim C. Thoden, Ralf Nauen, Andrew Crossthwaite, Ekaterini Riga, Marc Rist, Matthias Gaberthueel, Russell Eldridge

12:25-12:30 • **Damage threshold, population dynamics and host-status of *Meloidogyne chitwoodi* on five selected crops**
Misghina Goitom Teklu, Fantahun F. Addisu, Thomas H. Been, Corrie H. Schomaker, Johnny Visser, Leendert P.G. Molendijk

10:30-12:30 **Oral session 18** Room Espace Gould - Level 2

Nematodes as bioindicators

Chairs: Sara Sanchez-Moreno and Deborah Neher

Oral presentations

10:30-10:50 • **Historical perspective on nematodes as indicators**
Deborah Neher

10:50-11:10 • **Nematode responses to oak forest dieback in Mediterranean landscapes**
Sara Sánchez-Moreno, Jorge Curiel Yuste

11:10-11:30 • **Nematodes communities used for soil health indication in Midwestern United States agricultural systems**
Carmen Ugarte

11:30-11:45 • **Characterization of the biological state of soils by studying nematofauna as bio-indicators: ELISOL methodology**
Cecile Villenave, Anne Jimenez, Manon Trambolho, Hélène Cérémonie, Camille Chauvin

11:45-12:00 • **Zinc effects on terrestrial nematodes: from field studies towards a toxicokinetic approach**
Péter Nagy, Krisztina Hrács, Lola Virág Kiss, Zoltán Sávoly, Gergely Boros, Katalin Posta

12:00-12:15 • **Regenerative agriculture and its ability to restore soil ecosystem health and functioning**
Gerhard du Preez, Driekie Fourie, Ané Loggenberg

Poster flash presentations

12:15-12:20 • **A study on the ecological impact of recycling derived fertilisers (RDFs) using nematodes as environmental bioindicators**
Anna Karpinska, Thomaé Kakouli-Duarte

12:20-12:25 • **Evaluating the environmental risks of microplastics using nematodes as bioindicators**
Sebastian Höss, Marie Theres Rauchschwalbe, Hendrik Füser, Walter Traunspurge

12:25-12:30 • **Nematodes of argan biosphere: Biodiversity and assessment of soil quality**
Amina Braimi, Amine Idhmida, Abdolhadi Ajerrar, Hinde Benjlil, Mohamed Ait hamza, Fouad Msanda, Zahra Ferji, Abdelhamid El Mousadik, Thierry Mateille, El Hassan Mayad

---

10:30-12:30 **Oral session 19** Room Miles Davis - Level 3

Future of nematology: legislation, education and training

Chairs: Wim Bert and Ricardo Holgado

Oral presentations

10:30-10:50 • **Teaching nematology–what do students need to know?**
Cynthia Gleason

10:50-11:10 • **What is next? Nematology Education from a Ghent Perspective: together we create impact**
Inge Dehennin, Laura Cortada-Gonzalez, Aäron Plovie, Danny Coyne, Solveig Haukeland, Hugues Baimey, Antoine Affokpon, Awol Seid Ebrahim, Beira Hailu Meressa, Nasamu Bawa, Christopher Okolo, Biodun Claudius-Cole, Julius Bulus, Hendrika Fourie, Antoinette P. Malan, Herbert Talwana, Shahasi Athman, Francis Onyilo, Njira Njira Pili, Shem Nchore, Pierre Abad, Matthew Back, Richard Sikora, Mieke Daneel, Raymond Collett, Elvire Line Sossa, Alfred Alumai, Dora Scott, Paul Sule Chindo (in memoriam), Wim Bert

11:10-11:30 • **Nematode legislation contributes to food sustainability by management of *Globodera rostochiensis* and *G. pallida* in Norway**
Ricardo Holgado, Christer Magnusson, Randi Knudsen

11:30-11:45 • **Nematology 101 - A collection of lectures for plant nematology as slide presentations, videos, and textbook chapters**
Jonathan Eisenback

11:45-12:00 • **Best4Soil Nematode database tool to design clever crop rotations**
Leendert Molendijk, Paulien van Asperen

… /…

12:00-12:15 • **Regulation of potato cyst nematodes in Kenya**
Solveig Haukeland, Miriam W. Mbiyu, Moses Nyongesa, George Ngundo

12:15-12:30 • **Pine wilt disease and the pinewood nematode in Portugal and Europe: it was 20 years ago today; what have we learned?**
Manuel Mota

10:30-12:30 **Oral session 20** Room Ella Fitzgerald - Level 3

Natural Products as nematicides

Chairs: Nicoletta Ntalli and Karla Medina

Oral presentations

10:30-10:50 • **Attraction of *Meloidogyne* juveniles to essential oil constituents and derivatives**
Yuji Oka

10:50-11:10 • **Nematotoxicity of vetiver extracts against root-knot nematodes by systemic defense activation in sa-pathway**
Kansiree Jindapunnapat, Buncha Chinnasri

11:10-11:30 • **Biostimulants or Bionematicides: experience with seaweed extracts**
Bruno Ngala, Nicolas Mariette, Melina Ianszen, Pauline Dewaegeneire, Marie-Christine Denis, Catherine Porte, Christophe Piriou, Emilie Robilliard, Antoine Couetil, Eric Nguema-Ona, Virginie Gobert, Amélie Beury, Anne-Claire Le Roux

11:30-11:45 • **Entomopathogenic nematodes, a powerful weapon to control foliar insect pests**
Kay Moisan, Magda Galeano, Mireya Baños, Kelly Arkoumanea, Stavroula Louka, Olga Kostenko, Roxina Soler, Jose E. Belda

11:45-12:00 • **Nematicidal plant secondary metabolites with plant and soil enhancement properties**
Nikoletta Ntalli

12:00-12:15 • **What it takes for development and launch of a successful biological nematicidal product**
Sebastian Hartmann-Wittulsky

Poster flash presentations

12:15-12:20 • **Nematicidal plants for root-knot nematode management in tomato agrosystems**
Cliven Njekete, Anne-Violette Lavoir, Antoine Bedel, Vincent Michel, Caroline Djian-Caporalino

12:20-12:25 • **Investigation on the effectiveness of some plant extractions against *Ditylenchus dipsaci* and *Meloidogyne incognita***
İbrahim Halil Elekcioğlu, Ece Börtecine Kasapoglu Uludamar, Adem Taskın

12:25-12:30 • **Effects of synthetic and phytonematicides on the reproductive potential of *Meloidogyne* species on potato plants**
Kgabo Pofu

12:30-14:00 Lunch - Visit of exhibition Espace Gould - Level 2

Parallel Sessions

14:00-16:00 **Oral session 21** Amphi. Antipolis - Level 3

Nematode-plant interactions

Chairs: Shahid Siddique and Stéphanie Jaubert

Oral presentations

14:00-14:20 • **Strigolactones enhance rice susceptibility to root-knot nematode infection by antagonizing jasmonate based defense**
Godelieve Gheysen, Zobaida Lahari, Chhana Ullah, Tina Kyndt, Jonathan Gershenzon

14:20-14:40 • **Copper microRNAs modulate the formation of giant feeding cells induced by the root-knot nematode *Meloidogyne incognita* in *Arabidopsis thaliana***
Stéphanie Jaubert, Yara Noureddine, Joffrey Méjias, Martine Da Rocha, Sébastien Thomine, Pierre Abad, Bruno Favery

… /…

14:40-15:00 • **Two sides of the same coin: *Serendipita indica* alters different development of sedentary plant-parasitic nematodes in *Arabidopsis***
Michael Opitz, Virginia Ruiz-Ferrer, Fernando Evaristo Díaz-Manzano, Carolina Escobar, Roshanak Daneshkhah, Roland Ludwig, Cindy Lorenz, Siegrid Steinkellner, Krzysztof Wieczorek

15:00-15:15 • **Molecular markers for cell damage induced by root-knot nematodes**
Janice de Almeida Engler

15:15-15:30 • **PCN hijack carbon-for-nutrient exchange between potato and arbuscular mycorrhizal fungi to enhance reproduction**
Christopher Bell, Emily Magkourilou, P. Urwin, Katie Field

15:30-15:45 • **Unravelling the mechanisms underlying cyst nematode hatching**
Lemeng Dong, Kristyna Flokova, Alessandra Guerrieri, Harro Bouwmeester

Poster flash presentations

15:45-15:50 • **Trade-offs between virulence and breaking resistance in root-knot nematodes**
Alison Coomer, Henok Zemene Yimer, Valerie Williamson, Shahid Siddique

15:50-15:55 • **Parasitic worms redirect host metabolism via NADPH oxidase-mediated ROS to promote infection**
M. Shamim Hasan, Divykriti Chopra, Christiane Matera, Oliver Chitambo, Sina Valerie-Mahlitz, Ali Naz, Slawomir Janakowski, Miroslaw Sobczak, Axel Mithöfer, Tina Kyndt, Florian Grundler, Shahid Siddique

15:55-16:00 • **Meta-analysis of wild *Arachis* transcriptome data unravels candidate genes for combined nematode and drought resistance**
Patricia M. Guimaraes, Ana Paula Zotta Mota, Roberto Coiti Togawa, Ana Claudia Guerra Araujo, Ana Cristina Miranda Brasileiro

---

14:00-16:00 **Oral session 22** Room Ella Fitzgerald - Level 3

Metabolism & Physiology of nematodes and host plants

Chairs: Florian Grundler and Vicky Hunt

Oral presentations

14:00-14:20 • **Tricky parasites: How nematodes take their vitamins from plants**
Clarissa Hiltl, Shahid Siddique, Zoran S. Radakovic, Anna Gioran, Muhammad Shahzad Anjam, Esther Riemer, Samer S. Habash, Syed Jehangir Shah, Julia Holbein, Divykriti Chopra, Miroslaw Sobczak, Daniele Bano, Sebastian Eves-van den Akker, Alexander Graf, Florian M. W. Grundler

14:20-14:40 • **Extending the survival of *Heterorhabditis bacteriophora* through phenotype selection and marker-assisted breeding**
Carlos Molina, Nanette Hope Nellas Sumaya, Giulia Godina, Rolish Singh, Carlotta Kirsch, Bart Vandenbossche, Verena Dörfler, Mike Barg, Ralf-Udo Ehlers

14:40-15:00 • **Reaction of genotypes of *Phaseolus vulgaris* L BAT-306 and Triunfo-70 to *Meloidogyne incognita* and the behavior of enzymes related to the defense of both cultivars**
Dainé Hernandez-Ochandía, Yailén Arias-Vargas, Ivonne González-Marquetti, Susana Gorrita-Ramírez, M.G. Rodriguez, I. Miranda, Belkis Peteira Delgado-Oramas, R. Holgado

15:00-15:15 • **Defining the combined stress response in wild *Arachis*: the whole is not the sum of its parts**
Ana Paula Zotta Mota, Ana Cristina Miranda Brasileiro, Bruna Vidigal, Thais Nicolini Oliveira, Andressa da Cunha Quintana Martins, Mario Alfredo Passos Saraiva, Ana Claudia Guerra de Araújo, Roberto C. Togawa, Maria Fatima Grossi-de-Sá, Patricia Messenberg Guimaraes

15:15-15:30 • **Loss of the ERGO-1 small RNA pathway in *Caenorhabditis inopinata***
Vicky Hunt, Akemi Yoshida, Ryusei Tanaka, Kazunori Murase, Taisei Kikuchi

15:30-15:45 • **A Comparative study of the development and reproduction of *Meloidogyne enterolobii* and other thermophilic *Meloidogyne* sp**
Raymond Lesley Collett, Mieke Stefanie Daneel, Mariette Marais, Hendrika Fourie

15:45-16:00 • **Steroidal alkaloids as hatching factors for the potato cyst nematode, *G. rostochiensis***
Juliet Ochola, Laura Cortada, Margaret Ng'ang'a, Ahmed Hassanali, Danny Coyne, Baldwyn Torto

14:00-16:00 **Oral session 23** Room Espace Gould - Level 2

Integrated nematode management (continued)

Chairs: Mara Da Rocha and Hendrika Fourie

Oral presentations

14:00-14:20 • **Integrated nematode control options for cereal and leguminous crops in South Africa**
Hendrika Fourie, Raymond Collett, Nancy Ntidi, Mieke Daneel

14:20-14:40 • **Reaction of *Phaseolus vulgaris* accessions from EMBRAPA core collection as to resistance to *Heterodera glycines***
Mara Rubia Da Rocha, Daniela Patricia Balduino, Cristiano Augusto Mendes Junior, Thiago Livio Pessoa Oliveira De Souza, Rosana Pereira Vianello Brondani, Paula Pereira Torga

14:40-15:00 • **Detrimental impact of soil fumigants on nematode suppressiveness**
Tristan Watson, Johan Desaeger, Tom Forge, Louise Nelson, Denise Neilsen, Gerry Neilsen

15:00-15:15 • **Effects of some spice extracts on *Melodiogyne arenaria***
Hissein Mahamat Haroun, Gökhan Aydınlı, Sevilhan Mennan

15:15-15:30 • **Biotic factors of mulch-induced soil suppressivity against *Meloidogyne incognita* may depend on soil microclimate**
Krisztina Boziné Pullai, Renáta Petrikovszki, Franciska Tóthné Bogdányi, Péter Nagy, Zoltán Mayer, Katalin Posta, Rozália Tóth, Ferenc Tóth

15:30-15:45 • **Management of *Meloidogyne paranaensis* and *M. exigua* in coffee with chemical and biological nematicides**
Andressa Cristina Zamboni Machado, Daniel Sala Faria, Santino Aleandro Silva

Poster flash presentations

15:45-15:50 • **Case studies of root-knot nematode (*Meloidogyne* spp.) control in protected vegetables in Hungary**
Anita Gódor, Franciska Tóthné Bogdányi, Ferenc Tóth

15:50-15:55 • **Ozone treatments for the management of *Meloidogyne* sp. in greenhouse tomato cultivation in southeastern Spain**
Caridad Ros Ibáñez, Mª Ángeles Hernández Colucho, Daniel Soler Cárceles, José Luis Lozano González

15:55-16:00 • **Interaction between *Fusarium* spp and root lesion nematode *Pratylenchus capsici* on pepper crops in the Arava (Israel)**
Patricia Bucki, Maoz Aizikowitz, Marcel Maymon, Marina Benichis, Ohad Abraham, Qing Xue, Abraham Gamliel, Sigal Brown Miyara

---

16:00-16:30 Coffee break - Visit of exhibition Espace Gould - Level 2

16:30-18:00 **Poster Session S6 to S10** **(all numbered posters presented)** Espace Gould - Level 2

Parallel Sessions

16:30-18:00 **Workshop 8** Amphi. Antipolis - Level 3

Nematode-fungal interactions and complex diseases

Chairs: Richard Sikora and Driekie Fourie

- **Abstract general**
- **Nematode-disease complexes: a neglected factor in improving crop health**
  Richard A. Sikora, Ann MacGuidwin, Driekie Fourie, Danny Coyne, Holger Heuer, Matthias Gaberthüel
- ***Verticillum / Pratylenchus* for potato: both do the damage, but only one gets the credit**
  Ann MacGuidwin
- **Cereal root rot - nematode interrelationships: vision for the future?**
  Driekie Fourie, Amer Dababat
- **Nematode-fungal root-rot complexity on banana: a perennial problem of unknown importance?**
  Danny Coyne

… /…

- **Metagenomics: the future to disentangle complex diseases?**
  Holger Heuer
- **Present and future management systems for complexity management**
  Matthias Gaberthueel

16:30-18:00 **Workshop 9** Room Espace Gould - Level 2

Slime time: Nematodes associated with terrestrial molluscs

Chairs: Jenna Ross and Solveig Haukeland

- **Abstract general**
- **Welcome and opening remarks**
  Jenna Ross and Solveig Haukeland
- **Survey of slug-parasitic nematodes in East and West Flanders, Belgium, and description of *Angiostoma gandavense* n. sp. (Nematoda: Angiostomatidae) from arionid slugs**
  Phougeishangbam Rolish Singh, Marjorie Amoto, Marjolein Couvreur, Wilfrida Decraemer, Wim Bert
- **Looking for nematodes in the apple snail *Pomacea canaliculata* – a recent invasive in Kenya**
  Solveig Haukeland, Fernadis Makale, Ivan Rwomushan
- **Molluscs and EPNs: Symbiotic bacteria may help with slug control**
  Jiří Nermuť, Victoria Weijler, Jakub Savula, Vanessa Bachinger, Shakeel Zahid, Jana Konopická, Jiří Borák, Martin Janouch and Vladimír Půža
- **SlugBot: Developing an autonomous monitoring and biocontrol system for slugs**
  Jenna Ross, Tom Ashford, Archita Barua, Faye McDiarmid, Ben Scott-Robinson, Andy Hall, Pat Barretto, Tom Watsham, Tom Walters, Sam Herring, Rowan Duckworth, Michael Alcock, Tim Knott, Geoff Osmond and Daniel Rowe
- **Discussion points, note of interest for Horizon Europe bid and closing remarks**
  Jenna Ross and Solveig Haukeland

16:30-18:00 **Forum 2** Room Ella Fitzgerald - Level 3

Nematology Education Forum: the current situation and the ideal way forward

Chairs: Inge Dehennin, Driekie Fourie and Wim Bert

- **Abstract general**

18:15-19:30 **Bus departure for the Gala Evening**

Gala Evening - *Villa Eilenroc*

19:15 • **Speech: Jean Leonetti**, Mayor of Antibes Juan-les-Pins

19:30 • **Aperitif and Dinner**

FRIDAY 6 MAY 2022

Plenary Sessions

08:30-09:00 Plenary 4 — Amphi. Antipolis - Level 3

- **New perspectives on nematode – bacteria interactions, their causes and consequences**
  Tom Moens

09:00-09:30 Plenary 9

- **From human to plant diseases, scientific approaches of pest control in the big data era.**
  Corentin M. Barbu

09:30-10:00 Plenary 10

- **IFNS Global capacity building programs in nematology**
  Ernesto San-Blas

10:00-10:30 Coffee break - Visit of exhibition — Espace Gould - Level 2

Parallel Sessions

10:30-12:30 Oral session 24 — Room Miles Davis - Level 3

New challenges in nematodes taxonomy and evolution

Chairs: Oleksandr Holovachov and Majid Pedram

Oral presentations

10:30-10:50
- **Constraints on nematode evolution: how to conserve a Bauplan constructed from the substrate of an ever changing genome**
  Philipp Schiffer

10:50-11:10
- **Analysis of *Meloidogyne hapla* in soil using Loop-mediated Isothermal Amplification technique**
  Zahra Omer, Maria Viketoft, Ann-Charlotte Wallenhammar, Stina Andersson

11:10-11:30
- **Nematode species: their evolution and description in theory and practice**
  Michael Hodda

11:30-11:45
- **Satellitome evolution illuminates complex species history and satellite DNA transcriptomes show coordinated expression in *Meloidogyne* nematodes**
  Evelin Despot-Slade, Saša Širca, Brankica Mravinac, Philippe Castagnone-Sereno, Miroslav Plohl, Nevenka Meštrović

11:45-12:00
- **Species diagnosis, hosts and distribution of cyst nematodes of the genus *Heterodera* (Tylenchida: Heteroderidae) with a focus on species of concern for Australia**
  Daniel C. Huston, Manda Khudhir, Mike Hodda

12:00-12:15
- **New cyst nematode species from the Tepeaca Valley, Puebla, Mexico**
  Ilia Mariana Escobar Avila, Alejandro Tovar Soto, Sergei Subbotin

Poster flash presen tations

12:15-12:20
- **Morphological and molecular characterization of several *Paratylenchus* spp. from Belgium**
  Phougeishangbam Rolish Singh, Gerrit Karssen, Catherine Malike Etongwe, Marjolein Couvreur, Wim Bert

12:20-12:25
- **An update to the identification compendium of *Aphelenchoides Fischer*, 1894 (Aphelenchoidea)**
  Samira Fadakar, Majid Pedram

12:25-12:30
- **Unravelling the race complex: A first look Into the population genetics of the stem nematode *Ditylenchus dipsaci***
  Dennie te Molder, Stefan van de Ruitenbeek, Joost Riksen, Jaap Bakker, Geert Smant, Mark Sterken

10:30-12:30 **Oral session 25** Room Espace Gould - Level 2

Metabolism & Physiology of nematodes and host plants (continued)

Chairs: John Jones and Stephan von Reuss

Oral presentations

10:30-10:50 • **Identification of a *Globodera rostochiensis* eggshell annexin and analysis of its potential role in the control of hatch**
James Price, Terry Smith, John Jones

10:50-11:10 • **Modulation of anaphase-promoting complex genes impacts root-knot nematode gall development**
Aline Kohn Carneiro, Kercia Siqueira, Danila Cabral, Nubia Eloy, Harald Keller, Paulo Ferreira, Adriana Hemerly, Janice de-Almeida Engler

11:10-11:30 • **Nematode derived modular metabolites and their functions in chemical ecology**
Stephan Von Reuss

11:30-11:45 • **Tapping into signaling interactions between nematodes and aphids**
Jessil Ann Pajar, Nicole M. van Dam

11:45-12:00 • **The effects of female pheromone exposure on lethal fighting in *Steinernema carpocapsae* males**
Maria Cassells, Christine Griffin

12:00-12:15 • **Calcium signaling events modulate host immune responses during parasitic interactions**
M. Shamim Hasan, Chhana Ullah, Sakil Mahmud, Clement Pellegrin, Sharon Letia, Mirosław Sobczak, Ute C. Vothknecht, Sebastian Eves-van den Akker, Florian M.W. Grundler

12:15-12:30 • **Silencing a new female-specific multi-gene family of *Pratylenchus penetrans* can reduce nematode propagation**
Cláudia Vicente, Jordana Branco, Margarida Espada, Manuel Mota, Paulo Vieira

---

10:30-12:30 **Oral session 26** Amphi. Antipolis - Level 3

EPN commercialization and application

Chairs: Ralf Udo Ehlers and Patricia Navarro

Oral presentations

10:30-10:50 • **Conservation biocontrol with entomopathogenic nematodes: biotic and abiotic factors driving its potential**
Raquel Campos-Herrera

10:50-11:10 • **Host searching behavior of native and introduced entomopathogenic nematodes for control of *Aegorhinus superciliosus* (Coleoptera: Curculionidae) in blueberry crops**
Patricia Navarro, Almendra Monje, Graciela Berrios, Ivonne Alvarez, Camila Herrera

11:10-11:30 • **Entomopathogenic nematodes for insect pest management: progress and prospects for commercialization in South East Asia**
Nanette Hope Sumaya, Prakaijan Nimkingrat

11:30-11:45 • **Evaluating locally isolated entomopathogenic nematodes against *Thaumatotibia leucotreta* in laboratory bioassays**
François Du Preez, Antoinette Malan, Pia Addison

11:45-12:00 • **Exosome-like vesicles are a key process of non-canonical protein secretion in *S. carpocapsae***
Duarte Toubarro, Jorge Jorge, António Marcilla, Nelson Simões

12:00-12:15 • ***Diabrotica v. virgifera* management using genetically improved strains of *Heterorhabditis bacteriophora***
Ralf-Udo Ehlers, Carlos Molina, Bart Vandenbossche, Stefan Toepfer

Poster flash presentations

12:15-12:20 • **Efficacy of species mixtures of entomopathogenic nematodes against different larval stages of cockchafers**
Bart Vandenbossche, Mike Barg, Helge Postel, Edith Ladurner, Mauro Piergiacomi, Ralf-Udo Ehlers

…/…

12:20-12:25 • **Early season use of *Heterorhabditis bacteriophora* increases strawberry yield in fields infested by the white grub *Temnorhynchus baal***
Ibrahim Shehata, Mostafa Hammam, Fahiem El- Borai, Larry Duncan, Mahfouz Abd-Elgawad

12:25-12:30 • **The undetectable killer: *Steinernema carpocapsae* avoid recognition when infecting *Drosophila suzukii* larvae**
Anna Garriga, Duarte Toubarro, Nelson Simões, Ana Morton, Fernando García-del-Pino

---

10:30-12:30 **Oral session 27** Room Ella Fitzgerald - Level 3

Next-generation nematicides

Chairs: Kurtz Benedikt and Tim Thoden

Oral presentations

10:30-10:50 • **Reklemel™ Active: a novel highly selective nematicide**
Tim Thoden, Marta Garcia, John Wiles

10:50-11:10 • **Nematicidal or nematistatic? New insights into the mode of action of fluopyram in plant-parasitic nematodes**
A. Sylvia S. Schleker, Marc Rist, Christiane Matera, Arunas Damijonaitis, Katja Twelker, Corinna Saalwaechter, Sven Geibel, Oliver Gutbrod, Svend Matthiesen, Koichi Matsuoka, Samer S. Habash, Ute Schlee,
Florian M. W. Grundler

11:10-11:30 • **NIMITZ®, efficacious, safer and selective nematode management tool: technical review**
Bruno Lovato, Pablo Navia, Morten Boisen

11:30-11:45 • **A Pipeline for the discovery of modulators of neuromuscular function with potential utility as nematicidal leads**
Sean Harrington, Jacob Pyche, Jess Knox, Aaron Au, Inga Zasada, Ken-Loon Choo, Cassandra d'Amata, Mark Lautens, Peter Roy

11:45-12:00 • **Sensitivity of *Pratylenchus vulnus* to Salibro™ nematicide *in vitro* and *in planta***
Yu-Chen Wang, Tim Thoden, Zin Thu Zar Maung, Andreas Westphal

12:00-12:15 • **Biological activity of Tymirium™ molecule as a soil- and seed-applied nematicide**
Matthias Gaberthueel, Shannon Morsello, Jon Hamill, Dale Ireland, Jeff Simmons, André Bachiega

Poster flash presentations

12:15-12:20 • **Selective control of parasitic nematodes using bioactivated nematicides**
Andrew R. Burns, Rachel Ross, Megan Kitner, Jessica Knox, Jack M. P. Castelli, Kirsten Yeung, Emily Puumala, Bruna M. Palmeira, Elizabeth M. Redman, Sagar Marwah, Jens Tiefenbach, Jamie Snider, Constance A. M. Finney, Igor Stagljar, Henry M. Krause, Sonya A. MacParland, John S. Gilleard, Leah E. Cowen, Hui Peng, Susan Meyer, Mark Lautens, Inga Zasada, Peter J. Roy

12:20-12:25 • **The effect of 1-octen-3-ol and 3-octanone on plant parasitic nematodes**
Arben Myrta, Nicola Sasanelli, Astha Gurung, Pasqua Veronico, Salim Khoja, Alberto Troccoli, Ian Baxter, Tariq M. Butt

12:25-12:30 • **Impacts of long-term SDHI nematicide use on turfgrass**
Christian Kammerer, William Crow

12:30-14:00 Lunch - Visit of exhibition Espace Gould - Level 2

Parallel Sessions

14:00-16:00 **Oral session 28** Room Ella Fitzgerald - Level 3

Interactions of nematodes with micro-organisms

Chairs: Maria Viketoft and Johan Desaeger

Oral presentations

14:00-14:20 • **Microbiota associated with phytonematodes in the rhizosphere of cultured plants**
Holger Heuer, Ahmed Elhady, Shimaa Adss, Johannes Hallmann

…/…

14:20-14:40 • **Cultivating relationships: genetics shape microbiome form and function in *C. elegans***
Buck S. Samuel

14:40-15:00 • **Can microbiomes protect phytonematodes against antagonists?**
Olivera Topalovic, Frederik Bak, Susana S. Santos, Mette H. Nicolaisen, Flemming Ekelund, Mette Vestergård

15:00-15:15 • **Microbiome analysis of malacopathogenic nematodes suggests no evidence of a single bacterial symbiont responsible for gastropod mortality**
Laura Sheehy, James Cutler, Gareth Weedall, Robbie Rae

15:15-15:30 • **Nematodes rely on microbial facilitation in extreme environments**
Jesse Jorna, Byron J. Adams

15:30-15:45 • **Nematode–microbe complexes play an essential role in apple replant disease**
Xorla Kanfra

Poster flash presentations

15:45-15:50 • **Soil-born endophytic fungi antagonize plant-parasitic root-knot nematodes in tomato**
Polina Marchenko, Tina Austerlitz, Krzysztof Wieczorek

15:50-15:55 • **External and Internal microbiomes of Antarctic dry valley nematodes are distinct, but more similar to each other than the surrounding environment**
J. Parr McQueen, Kaitlin Gattoni, Eli Gendron, Pacifica Sommers, Steven Schmidt, Dorota Porazinska

15:55-16:00 • **Characterization of nematode-bacteria associations with gastropods and their virulence towards crop pests in norhtern Mindanao, Philippines**
Veronica Tañan, Nanette Hope Sumaya, PhD

14:00-16:00 **Oral session 29** Room Miles Davis - Level 3

Trade and market access implications of plant parasitic nematodes

Chairs: Francesca De Luca and Valeria Orlando

Oral presentations

14:00-14:20 • **Nematology in support of plant health – role of EPPO**
Françoise Petter, Madeleine McMullen, Charlotte Trontin, Fabienne Grousset, Camille Picard, Muriel Suffert, Rob Tanner, Baldissera Giovani, Anne-Sophie Roy

14:20-14:40 • **Nematodes associated with damage on *Taxus media* cv. Hillii in Norway**
Christer Magnusson, Dag Ragnar Blystad, Zahra Ben Hellal

14:40-15:00 • **Movement of plant-parasitic nematodes associated with the turf industry**
Valeria Orlando, Tom Prior, Rebecca Lawson, Lesley Jutson, Margarida Correia, Paul Taylor, James Stubbs, Charlotte Howard

15:00-15:15 • **Plant-parasitic nematodes pose a serious threat to the production and nutritional quality of popular biofortified cassava**
Aminat Oyetunde, Steve Afolami, Peter Kulakow, Danny Coyne

15:15-15:30 • **Plant-parasitic nematodes and export to non-European Union countries**
Anne Sophie van Bruggen, Evelyn van Heese

15:30-15:45 • **Detection and diagnostics of tropical *Meloidogyne* spp. within the Euphresco project MeloTrop**
Saša Širca, Barbara Gerič Stare, Anne-Marie Chappe, Fabrice Ollivier, Laurent Folcher, Maria L. Inácio, Filomena Nóbrega, Leidy Rusinque, Eugénia Andrade, Carla Maleita, Luci Conceição, Isabel Abrantes, Evelyn Y.J. van Heese, Gerrit Karssen, Jasmina Bačič

Poster flash presentations

15:45-15:50 • **Negative binomial modeling of nematode count data yield more accurate mean and variance estimates**
Gideon Alake, Inga Zasada, Edzard van Santen, Peter DiGennaro

…/…

15:50-15:55 • **FAGUSTAT: Investigating Beech Leaf Disease, a threat to beech trees and forests in Europe**
Nicole Viaene, Negin Ebrahimi, Annelies Haegeman, Ondrej Douda, Anne Sophie van Bruggen, Nikica Ogris, Saša Širca, Barbara Gerič Stare, Thomas Prior, Ana Pérez Sierra, Mariana Groza, Mihaela Coman, Marie-Jo Hurley, Déborah Lanterbecq, Simon Van Kerkhove, Quentin Leroy

15:55-16:00 • **Plant-parasitic nematode: a potential threat to medicinal plants in Vietnam**
Nguyen Huu Tien, Bert Wim, Trinh Quang Pha

14:00-16:00 **Oral session 30** Room Espace Gould - Level 2

Nematode community assemblies

Chairs: Byron Adams and Eric Grenier

Oral presentations

14:00-14:20 • **Impact of cultural practices and environmental conditions on plant-parasitic nematode communities through a long-term agroecological field trial**
Nathan Garcia, Eric Grenier, Laurent Folcher

14:20-14:40 • **A novel metabarcoding strategy for studying nematode communities**
Md Maniruzzaman Sikder, Mette Vestergård, Rumakanta Sapkota, Tina Kyndt, Mogens Nicolaisen

14:40-15:00 • **Community assembly in the wake of glacial retreat: a meta-analysis**
Satyendra K. Pothula, Byron J. Adams

15:00-15:15 • **Exploring belowground biodiversity in Lincoln, Nebraska's tallgrass prairie corridor**
Abigail Borgmeier, Kaitlin Gattoni, Timothy Harris, Peter Mullin, Dorota Porazinska, Kirsten Powers, David Wedin, Thomas Powers

15:15-15:30 • **DNA barcoding individual specimens using COI and 18S in nematode community analyses: advantages of a combined approach**
Thomas Powers, Dorota Porazinska, Byron Adams, Abigail Borgmeier, Rebecca Higgins, Timothy Harris, Peter Mullin, Kirsten Powers

15:30-15:45 • **Agriculture land-use intensification causes biotic homogenization on phytonematode communities at regional scale**
Antonio Archidona Yuste, Thorsten Wiegand, Nico Eisenhauer, Carolina Cantalapiedra-Navarrete, Juan E. Palomares Rius, Pablo Castillo

Poster flash presentations

15:45-15:50 • **Nematode communities in organic and conventional rice production in Indonesia: a morphological and metabarcoding approach**
Muhammad Aksan, Prabowo Lestari, Marjolein Couvreur, Antarjo Dikin, Johannes Helder, Wim Bert

15:50-15:55 • **Association of beneficial terrestrial nematodes with glyphosate-tolerant and conventional soybean-based cropping systems**
Akhona Mbatyoti, Mieke Daneel, Antoinette Swart, Dirk De Waele, Hendrika Fourie, Rinus Knoetze

15:55-16:00 • **Research on free-living terrestrial nematodes (Order Dorylaimida) from tropical rain forest in Vietnam**
Thi Anh Duong Nguyen, Joaquin Abolafia, Reyes Pena-Santiago

14:00-16:00 **Oral session 31** Amphi. Antipolis - Level 3

Effectors in plant parasitic nematodes

Chairs: Bruno Favery and Geert Smant

Oral presentations

14:00-14:20 • **Effector gene birth in plant parasitic nematodes: neofunctionalization of a housekeeping glutathione synthetase gene**
Sebastian Eves-van den Akker, Catherine Lilley, Abbas Maqbool, Duqing Wu, Hazijah Yusup, Laura Jones, Paul Birch, Mark Banfield, Peter Urwin

… /…

14:20-14:40 • **A root-knot nematode effector targets the spliceosomal plant machinery allowing the giant cell formation**
Michaël Quentin

14:40-15:00 • **Nematode-encoded RALF peptide mimics facilitate parasitism of plants through the FERONIA receptor kinase**
Xin Zhang, Huan Peng, Hongdong Liao, Deliang Peng, Feng Yu

15:00-15:15 • **NemaWAARS: A motif to unveil mechanisms of parasitism gene regulation in the pinewood nematode as a target for disease control and plant resistance**
Margarida Espada, Cláudia Vicente, Jorge Faria, Paulo Vieira, Sebastian Eves-van den Akker, Lurdes Inácio, Manuel Mota

15:15-15:30 • **Discovery of novel stylet-secreted NLS-containing effector candidates from the soybean cyst nematode *Heterodera glycines***
Anju Verma, Rick Masonbrink, Tom Maier, Thomas J. Baum, Melissa G. Mitchum

15:30-15:45 • **The *Meloidogyne incognita* effector MiL648 contributes to nematode virulence through its interactions with OPR2 in tomato**
Ava Verhoeven, Anna Finkers-Tomczak, Pjotr Prins, Debbie Valkenburg-van Raaij, Koen Varossieau, Hein Overmars, Erik Slootweg, Mark Sterken, Aska Goverse, Geert Smant

Poster flash presentations

15:45-15:50 • **The Root-knot nematode effector Mj-NEROSS suppresses plant immunity by interfering with the ROS production in plastids**
Boris Stojilkovic, Yujin Chen, Xiang Hui, Godelieve Gheysen

15:50-15:55 • **Studying root-knot nematode *Meloidogyne javanica* MAP-1 variation and their implication in parasitism**
Anil Kumar, Patricia Bucki, Sigal Braun Miyara

15:55-16:00 • **The M. javanica effector Mj-10A08 downregulates ethylene receptors via protein-protein interactions to facilitate tomato parasitism**
Yujin Chen, Boris Stojilković, Godelieve Gheysen

Plenary Session

16:10-16:40 **Plenary 11** — Amphi. Antipolis - Level 3

• **Multi-criteria assessment in agriculture: how can agroecology help?**
Anne Mottet

16:40-17:00 **Closure congress**

• **Pierre Abad, Larry Duncan** and **Florian Grundler**

7th International Congress of Nematology
ICN 2022
Antibes Juan-les-Pins - France

"Crossing borders: a world of nematode diversity and impact to discover"

European Society of Nematologists

ABSTRACTS MONDAY 2 MAY

PLENARY SESSIONS

Microgravity effect on entomopathogenic nematodes' ability to find and kill insects.

Fatma Kaplan[1] (fkaplan@pheronym.com), David Shapiro-Ilan[2], Karl Cameron Schiller[1]

[1] *Pheronym Inc., Davis, CA, United States*; [2] *USDA-ARS, Byron, GA, United States*

Entomopathogenic nematodes (EPNs) are microscopic roundworms which are part of a healthy soil microbiota and pollinator-friendly biocontrol system for agricultural insect pest control. The nematodes kill insects with the aid of mutualistic bacteria [1]. The nematodes enter insects through natural body openings or the insect cuticle, release their symbiotic bacteria, then the symbiotic bacteria infect and kill the insects, and the nematodes feed off the bacteria. Once the insect host is consumed, a pheromone signal tells the nematodes that they are too crowded, need to leave, disperse and find a new insect to infect [2]. Dispersal is a key feature contributing to EPNs' efficacy in finding and killing insects. Dispersal is strongly regulated by environmental stress such as vibration, sub optimal temperatures, barometric pressure changes, electrical fields, and pheromones [2-5]. We hypothesize that microgravity is an environmental stress factor that changes pheromone production and affects EPN dispersal and efficacy. The microgravity experiment was conducted at the International Space Station (ISS) National Laboratory (NL) and used the EPN, *Steinernema feltiae* as the subject organism; the nematodes were exposed to microgravity between Dec 5, 2019 and Jan 7, 2020. We will present the following: 1- The ability of EPNs to forage, infect, reproduce, and emerge in microgravity, 2- analysis of whether or not EPNs that developed and emerged in space can adapt to the earth's gravity after the voyage, 3- how microgravity affects insect host physiology and 4- whether space travel affects EPN infectivity.

Keywords: Microgravity - Entomopathogenic nematodes - Efficacy - Foraging - Space travel.

References:

- [1] Stock 2005. J Invertebr Pathol. 89: 57-66.
- [2] Kaplan et al. 2012. PLoS ONE. 7: e38735.
- [3] Lewis et al. 1993. Can J Zool. 71: 765-769.
- [4] Shapiro-Ilan et al. 2012. J Invertebr Pathol. 109: 34-40.
- [5] Ilan et al. 2013. International J Parasitol. 43: 781-784.

Plant root-knot nematode interaction: a sophisticated dialogue.

Pierre Abad (pierre.abad@inra.fr)

Institut Sophia Agrobiotech, INRAe - Université Côte d'Azur - CNRS, Sophia Antipolis, France

Root-knot nematodes (RKNs), *Meloidogyne* spp., are a worldwide agronomic problem. These plant parasites trigger a long-lasting and intimate relationship with their host plants. They infect roots and establish specialized feeding structures called 'giant-cells', from which they withdraw nutrients. The nematode effector proteins secreted *in planta* are key elements in the molecular dialogue of parasitism. We developed an integrated approach to functionally characterize nematode parasitism proteins and their plant targets. These so-called susceptibility genes may in fact constitute new targets for the development of new resistance strategies. The RKN *M. incognita* is an apomictic mitotic parthenogenesis species with emergence of virulent populations bypassing plant resistance genes. Such adaptation raises questions about genome plasticity leading to genetic variation and adaptive evolution. We reasoned that epigenetic mechanisms might in part be responsible for the generation of phenotypic variants that provide material for rapid adaptation. Finally, we investigated factors affecting the control of RKN in cropping systems by combining genetic resistance and cultural practices in order to propose effective resistance deployment strategies. Thus, RKNs constitute a unique model system to study the links between variation in genome structure, mode of reproduction, and adaptation to environment and hosts in relation with parasitic success.

Keywords: Plant-parasitic nematodes - Effectors - Resistance - Virulence - Epigenetic.

ORAL SESSION 1

Plant resistance and nematode virulence

Multiple mechanisms of resistance to SCN encoded by soybean *Rhg1.*

Andrew Bent[1] (afbent@wisc.edu), Shaojie Han[2], Adam Bayless[2], Katelyn Butler[2], John Smith[2], Ryan Zapotocny[2], Christina Fliege[3], Matthew Hudson[3], Brian Diers[3], Shiyan Chen[4], Xiaohong Wang[4], Derrick Grunwald[2]

[1] *Plant Pathology, University of Wisconsin, Madison, WI, USA*; [2] *University of Wisconsin, Madison, USA*; [3] *University of Illinois at Urbana-Champaign, Urbana, USA*; [4] *USDA-ARS, Ithaca, USA*

Soybean cyst nematode (SCN) is the most yield-reducing pathogen of U.S. soybeans. We will describe new discoveries about the mechanisms of soybean resistance to SCN. Soybean breeders rely heavily on the complex *Rhg1* locus. We previously reported that one of the *Rhg1* genes encodes an atypical, cytotoxic α-SNAP protein that disrupts host cell vesicle trafficking. α-SNAP and NSF are essential eukaryotic housekeeping proteins that normally interact to sustain vesicular trafficking. We more recently learned that 100% of soybean lines with *Rhg1* carry, at an unlinked locus, the gene for an NSF protein with unusual amino acids at the predicted α-SNAP/NSF interaction site. This NSF-RAN07 exhibits better interaction with *Rhg1* α-SNAPs and partially mitigates their cytotoxicity. Separately, we learned that *Rhg1*-mediated resistance can involve depletion of wild-type α-SNAP abundance. In unpublished work, we have found that the responsible gene at another SCN resistance QTL encodes a gamma-SNAP protein. Hence for disease resistance, SCN selection pressure has apparently driven multiple types of re-wiring of soybean NSF and SNAP proteins, which in other organisms are highly conserved components of the SNARE recycling machinery. Transgenic transfer of this cyst nematode resistance can function in other plant species against other cyst nematodes. Lastly, new findings reveal that a second functional *Rhg1* protein product (the *Rhg1* amino acid permease-like protein) is upregulated at stages of the infection process that differ from the primary site of the *Rhg1* α-SNAP abundance increases, and that the *Rhg1* amino acid permease-like protein interacts with unique protein partners. The findings indicate that *Rhg1* is a resistance pyramid that encodes more than one distinct mode of action.

Epigenetic modifications in *Meloidogyne incognita* virulence to resistance gene *Mi* in tomato.

Laetitia Perfus-Barbeoch Zurletto[1] (laetitia.zurletto@inrae.fr), Rahim Hassanaly-Goulamhoussen[1], Christoph Grunau[2], Etienne G.J. Danchin[3], Bruno Favery[3], Philippe Castagnone[3], Pierre Abad[3]

[1] *Institut Sophia Agrobiotech/Interactions Plantes Nematodes (IPN), Université Côte d'Azur, Sophia Antipolis, Alpes Maritimes, France*; [2] *Ecologie et Evolution des Interactions (2EI), Université de Perpignan Via Domitia, Perpignan, Pyrénées-Orientales, France*; [3] *Institut Sophia Agrobiotech/Interactions Plantes Nematodes (IPN), INRAe, Sophia Antipolis, Alpes Maritimes, France*

Want to blame your experiments for something that doesn't seem to be (only) genetic? Look for epigenetics! To be put as a simplified definition, epigenetics is the study of biological mechanisms that will switch genes on and off, in a reversible manner. Any outside stimulus has the potential to cause epigenetic modifications. In *M. incognita*, there have been hints that epigenetic modifications may be involved in virulence to a resistance gene, most of the strongest evidence so far comes from research done on *M. incognita* isolates by isofemale line analysis (Castagnone-Sereno et al. 2006, 2014). Thanks to the application of high-throughput sequencing technologies to the characterization of epigenomes, we investigated if the exposure to the *Mi* resistant gene affects epigenetic marks. Our analysis of virulence to resistant gene *Mi*, in *M. incognita* isolates of two different geographical origins (isofemale lines morelos and kursk), revealed that the avirulent and virulent lineages differ in their epigenomes; and that the virulent lineages shared common epigenome signatures. Some of these findings point to previous identified genes, such as effectors.

Keywords: Apomixis - Plant resistance - Virulence - Epigenetics - Histone post-transcriptional modifications.

References:

- Hassanaly-Goulamhoussen et al., 2021. Front Cell Dev Biol.
- Pratx et al., 2018. BMC Genomics. 19:321
- Blanc-Mathieu et al., 2017. PLoS Genet.13(6):e1006777.
- Perfus-Barbeoch et al., 2014. Front Physiol. 5:211.
- Castagnone-Sereno and Danchin. 2014. J Evol Biol. 27(7):1323-33.
- Castagnone-Sereno. 2006. Heredity (Edinb). 96(4):282-9.

Tackling new virulent *Globodera pallida* populations using new sources of resistance in wild potato species.

Sebastian Kiewnick (sebastian.kiewnick@julius-kuehn.de)

Inst. A./Nematology, Julius Kuehn Institut, Braunschweig, Germany

Potato cyst nematodes, and in particular *Globodera pallida*, are a mayor constraint for the potato production in Europe. Until recently, the use of resistant starch potato varieties was the most efficient way to control *G. pallida* as part of national official control program in Germany. However, intensive use of these varieties with Grp1 resistance has led to the selection of a new virulence type of *G. pallida*, which was found in the Emsland region in Germany and officially reported in 2014. In addition, authorities from The Netherlands also reported the presence of *G. pallida* populations of a new virulence type from areas with intensive starch potato production. Intensive testing of available potato varieties revealed that currently no commercial variety is available to the farmers for control of these populations. Therefore, new varieties with new sources of resistance are urgently needed. Controlling populations of this new virulence type is challenging, as they seem to have a higher biological fitness resulting in higher reproduction rates on both, resistant and susceptible cultivars. Furthermore, cyst size and content seems correlated with increased virlulence. In addition, the selection for virulence is not reversible even after several reproduction cycles on susceptible cultivars such as Desiree. To identify new sources of resistance, several accessions of wild potato species were screened in pot experiments against one representative virulent population “Oberlangen”. To improve the phenotyping process, tissue culture plants were utilized to speed up the selection of resistant genotypes. From this screening process, over 50 genotypes were identified that carry resistance against “Oberlangen” as well as the reference Pa3 population “Chavornay”. As a next step, the selected genotypes are now evaluated for their suitability in breeding programs and if different modes of action are responsible for suppressing *G. pallida*. One main goal is to combine different modes of resistance to breed for a durable resistance in starch potatoes.

Keywords: G. pallida - Virulence - Resistance - Varieties.

The genetic basis for root-knot nematode virulence.

David Bird (bird@ncsu.edu)

Bioinformatics Research Center, NC State University, Raleigh, NC, USA

Here I assume a broad definition of "virulence" as "a measure of the degree of harm caused by an infective organism." For plant parasitic nematodes (PPN), virulence may be manifest as overcoming host resistance, amplification of the nematode population, or localized or systemic disruption of normal plant physiology and/or anatomy. Mechanisms underpinning such complex phenotypes will most likely be conditioned by expression of multiple nematode and plant genes functioning in regulatory and physiological networks. To address the question: "which PPN genes influence expression of what host genes" we turned to the tools of quantitative (QTL) genetics (*Genetics* 206:4175-4184, 2017). Based on the **genotype** of each of the 14,000 genes in *Meloidogyne hapla*, numerous polymorphic PPN loci were shown to influence the host transcriptome (*Medicago truncatula*). These loci capture natural variation in field isolates. Transcript levels (the **phenotype**) of each of the 21,000 Medicago genes were experimentally determined in the mapping lines (8 biological replicates). Together our genetic and genomic data directly address the question we proposed above. On-going network analyses are revealing functional pathways. For example, infection by one parent increases expression of AGAMOUS 90-fold compared to infection with the other parent. The network shows a cascade of numerous MADs box transcription factors presumably necessary to achieve this amplification. We also have "re-discovered" genes previously isolated by molecular means, including an early RNASeq experiment (*MPMI* 7:419-424, 1994). Interestingly, none of the classic effectors were revealed. We suspect that many effectors were "filtered" by our stringent *P*-values for multiple testing corrections (i.e., a technical barrier). Alternatively, it may reflect some unique biology of effectors, such as specific spatio/temporal expression patterns.

Keywords: Cross-species eQTL - Linkage mapping - Meloidogyne hapla.

Proline-rich extensin-like receptor kinases mediate damage-triggered immune responses to nematode infections.

Jose L. Lozano-Torres (jose.lozano@wur.nl), Jaap-Jan Willig, Sonja Warmerdam, Casper C. van Schaik, Jaap Bakker, Aska Goverse, Geert Smant

Laboratory of Nematology, Wageningen University, Wageningen, Netherlands

Plant-parasitic nematodes constitute a major threat to global food production. Despite causing considerable damage to host tissue during parasitism, little is known on the role of plant basal immunity in resistance to nematodes. We have recently discovered that suppression of basal immunity to nematodes involves a specific class of surface-localized receptors i.e. proline-rich, extensin-like receptor kinases (PERKs). However, the role of PERKs as mediators of damage-triggered basal immunity is not well understood. PERKs are differentially regulated upon infections with cyst and root-knot nematodes. Arabidopsis *perk*-mutants show increased susceptibility to both cyst and root-knot nematodes. By contrast overexpression of AtPERK-A results in enhanced resistance to cyst nematodes. Furthermore, Arabidopsis lines heterologously overexpressing AtPERK-A homologs also show enhanced resistance to cyst nematode infections. AtPERK-A is strongly induced in nematode feeding sites and neighbouring cells. The basal immune response mediated by PERK-A during nematode infection involves binding of the extracellular domain to polygalacturonic acid and the downstream regulation of innate immune suppressors. We, therefore, conclude that PERKs function as damage-triggered immune receptors during nematode infections and as such can be exploited as a novel source of nematode resistance.

Keywords: Proline-rich extensin-like receptor kinases - Damage-triggered immune responses - Plant-parasitic nematodes.

Functional analysis of the *Ma* gene for resistance to root-knot nematodes in *Prunus*.

Simon Saucet (simon.saucet@inrae.fr), Cyril Van Ghelder, Daniel Esmenjaud

INRAe, Université Côte d'Azur, CNRS, Institut Sophia Agrobiotech, Sophia Antipolis, France

The root-knot nematodes (RKNs) *Meloidogyne* spp. represent a global threat for annual and perennial crops causing huge crop losses worldwide. Only a few genes have been isolated for their ability to provide resistance to RKNs. Several of them encode nucleotide-binding (NB) and leucine-rich repeat (LRR)-containing receptors (NLRs). In plants, NLRs are intracellular receptors able to detect pathogen–secreted proteins known as effectors, through direct or indirect interaction. Recent studies indicate that NLR proteins function as multimeric complexes in order to activate defense responses. However, it remains unclear how the different NLR domains act precisely in the formation and activation of those complexes, as well as in the interaction with signaling partners in cells. The *Ma* gene, from the Toll/Interleukin-1 receptor/Resistance protein (TIR)-NLR (TNL) family, has been cloned from the plum *Prunus cerasifera* where it confers resistance against numerous *Meloidogyne* species such as *M. arenaria*, *M. incognita*, *M. javanica*, *M. floridensis*, *M. enterolobii* and *M. ethiopica* [1, 2]. In addition its their core TNL structure, TNLs often carry a Post-LRR/Immunoglobulin/Jelly roll (PIJ) domain C-terminally to the LRR. While this domain is mostly present as a single copy, Ma carries five of them that are encoded by five consecutive exons [3]. We are investigating how Ma functions for RKNs resistance in *Prunus* and we focus particularly on the repeated PIJ domain. The resistant allele of *Ma* (*Ma* R) was originally cloned from the *Prunus* accession P2175 which also carries a non functional susceptible allele (*Ma* S). Using the modular Golden Gate cloning method, we tested *Ma* chimeras and truncated versions for their ability to provide RKNs resistance in transgenic *Prunus* hairy roots. We identified parts in *Ma* S gene that may be responsible for its loss of function. We also initiated a functional analysis of the PIJ domain using two other TNLs; N from tobacco and RPS4 from Arabidopsis. The analysis of Ma and its duplicated PIJ domains at intra and intermolecular levels will enable us to better understand the role of this domain for immunity and, ultimately, to contribute to the development of methods controlling durably RKNs infection in crops.

Keywords: TNL - Root-knot nematode - PL domain - Hairy roots.

References:

- [1] Claverie et al. 2011. Plant Physiol 156:779-792.
- [2] Duval et al. 2018. Phytopathol 109(4): 615-622.
- [3] Van Ghelder and Esmenjaud. 2015. BMC Genomics 17, 317.

S1-PF1

Deciphering the mechanism of ascaroside perception in plants.

Sharon Letia[1] (s7shleti@uni-bonn.de), Badou Mendy[1], Stephan H. von Reuss[2], Florian M.W. Grundler[1], M. Shamim Hasan[1]

[1] *Molecular Phytomedicine (MPM), INRES, Rheinische Friedrich-Wilhelms-University of Bonn, Bonn, Germany*; [2] *Laboratory for Bioanalytical Chemistry, Institute of Chemistry, University of Neuchâtel, Neuchâtel, Switzerland*

The vital role of ascarosides in nematode development and signalling coupled with its evolutionarily conserved nature across the Nematoda phylum renders ascarosides a perfect Nematode Associated Molecular Pattern (NAMP) candidate. Ascarosides species #18 (ascr#18) is the most abundant in plant-parasitic nematodes and has been reported to induce Pattern Triggered Immunity (PTI). However, its recognition mechanism(s) remains elusive to date. Our preliminary results show that micromolar concentrations of ascr#18 induce the expression of genes associated with jasmonic acid and salicylic acid-mediated defence signalling pathways as well as activation of mitogen-activated protein kinases. Also, ascr#18-mediated plant defence response results in enhanced resistance to nematodes in the form of the priming effect. However, ascr #18 perceptions does not lead to increased ROS bursts or growth inhibitory effects in plants. Therefore, ascr#18-mediated plant responses do not reflect the complete canonical pattern of PTI. The current work aims to provide a detailed investigation identifying the plant´s molecular machinery of perceiving ascarosides from nematodes and its downstream signalling networks responsible for defence responses using the *Arabidopsis thaliana* and *Heterodera schachtii* model system.

Keywords: *Arabidopsis* - Ascarosides - PRRs - Cyst nematodes - Immune response.

S1-PF2

Screening of plantain and banana cultivars for resistance against *Radopholus similis* with prospects of using macropropagation plantlets for selection.

Emmanuel Olajide[1, 2] (olajide.o.emmanuel@gmail.com), Laura Cortada[2], Delphine Amah[1], Danny Coyne[3], Wim Bert[2], Rony Swennen[4], Yao Kolombia[1]

[1] *International Institute of Tropical Agriculture, Ibadan, Nigeria*; [2] *Nematology Research Unit, Ghent University, Ghent, Belgium*; [3] *International Institute of Tropical Agriculture, Nairobi, Kenya*; [4] *International Institute of Tropical Agriculture, Kampala, Uganda*

Radopholus similis, a migratory endoparasitic nematode species that feeds and reproduces in its host roots, is a major biotic constraint of banana and plantain (*Musa* spp.) production worldwide. In Nigeria, the wide distribution of this species has been reported as a major cause of economic concern, and its high incidence is associated with root necrosis and dead roots. Recently, *Musa* breeding programmes have strengthened the availability of improved genotypes, some of which could be resistant against nematodes. In this study, 17 improved plantain hybrids (PITA), 13 diploids, and 10 plantain landraces were evaluated for resistance to *R. similis* in two trials using a conventional screenhouse screening. We compared nematode reproduction (Pf/Pi) and the percentage of root necrosis of the tested genotypes with two well-documented susceptible and resistant reference genotypes. All the genotypes were infected by *R. similis*, with several PITAs lines being susceptible to *R. similis*. However, PITA 11, PITA 24, PITA 4, PITA 14 and PITA 17 sustained significantly lower nematode reproduction rates than the susceptible controls. A higher nematode reproduction similar to the susceptible reference genotypes was observed from the genotypes in the plantain subgroups (Bobby Tannap, Cantebalon, DH Rojo, French Reversion, Mbi Egome, Obubit Ntanga, Obino l'Ewai, Okoyo Ukom and Orishele), whereas most of the diploid lines (1297-3, 1448-1, 25291-582, 25447-57, 2829-62, 8075-7, 9128-3, Calcutta 4, SH 3142, SH 3362, Tjau Lagada) were found to sustain significantly lower nematode reproduction than the susceptible reference genotypes. The percentage of root necrosis was generally low (<15%) and no significant difference were observed in the percentage of root necrosis across all the genotypes in our study. The genotypes identified with less susceptibility should be further evaluated in the field. As a valuable alternative of conventional screening using suckers, the use of macropropagation plantlets is also forwarded and discussed to accelerate resistance selection in bananas and plantain.

Keywords: Plant host resistance - Nigeria - Germplasm - Plant-parasitic nematode.

S1-PF3

Identification of candidate resistance genes in chickpea (*Cicer arietinum*) against *Pratylenchus thornei* using GWAS.

Sonal Channale[1] (Sonal.Channale@usq.edu.au), John Thompson[1], Rajeev Varshney[2], Mahendar Thudi[3], Rebecca Zwart[1]

[1] *Centre for Crop Health, University of Southern Queensland, Toowoomba, Queensland, Australia*; [2] *Murdoch University, Perth, Australia*; [3] *Department of Agricultural Biotechnology and Molecular Biology, Dr. Rajendra Prasad Central Agricultural University, Pusa, Bihar, India*

Chickpea (*Cicer arietinum*) is a popular legume due to its nutritional and commercial value and ranks third globally among legumes for its area under cultivation. The root-lesion nematode, *Pratylenchus thornei* is the predominant plant-parasitic nematode affecting chickpea crops in Australia, causing up to 25% yield loss [1]. The life-cycle of *P. thornei* takes 45-65 days and thus multiple generations are completed throughout the growing season of chickpea, leaving behind a damaging population in the soil to attack the subsequent crop [2]. Furthermore, the broad host range of *P. thornei*, which includes many cereals and pulses, means *P. thornei* can cause extensive damage to whole farming systems. Due to the polygenic resistance of chickpea to *P. thornei* [3], we deployed a genome-wide association approach to identify genomic regions and candidate genes associated with *P. thornei* resistance in the chickpea reference set [4] containing 278 chickpea genotypes. The chickpea reference set was phenotyped for *P. thornei* multiplication in two years (2018 and 2020) followed by Genome-wide association study (GWAS) using 492,849 SNPs after filtration [5]. GWAS revealed six significant SNPs in all, distributed on chromosome 5, 6 and 7 with number of SNPs as 2, 1 and 3 respectively. SNP located on chromosome 6 and two SNPs on chromosome 7 had negative effects and were associated with lower nematode reproduction whereas SNPs on chromosome 5 and a SNP on chromosome 7 had positive effects, associated with higher nematode reproduction. Haplotype analysis revealed the presence of a haploblock for the SNP on chromosome 7. The flanking regions of SNPs were analysed and we report identification of two candidate genes on chromosome 6 and five candidate genes on chromosome 7 for *P. thornei* resistance. The candidate genes for resistance to *P. thornei* in chickpea can be explored further to develop molecular markers and accelerate the incorporation of *P. thornei* resistance into elite chickpea cultivars.

Keywords: Cicer arietinum - Chickpea - Pratylenchus thornei - Root-lesion nematode - GWAS.

References:

- [1] Reen RA et al., 2014. Crop and Pasture Science. 65(5):428-41.
- [2] Vanstone VA et al., 2008. Australasian Plant Pathology. 37(3):220-34.
- [3] Channale S et al., 2021. Scientific Reports. 11(1):1-1.
- [4] Upadhyaya HD et al., 2008. BMC plant biology. 8(1):1-2.
- [5] Varshney RK et al., 20019. Nature genetics. 51(5):857-64.

ORAL SESSION 2

Soil suppressiveness and nematode control using cover crops

Cover cropping for nematode management in nut crop production.

Andreas Westphal[1] (andreas.westphal@ucr.edu), Yu-Chen Wang[1], J. Ole Becker[2], Jeff Mitchell[3], Amelie Gaudin[4]

[1] *Department of Nematology, University of California, Riverside, Parlier, CA, United States*; [2] *Department of Nematology, University of California, Riverside, CA, United States*; [3] *Department of Plant Sciences, University of California, Davis, Parlier, CA, United States*; [4] *Department of Plant Sciences, University of California, Davis, CA, United States*

In California, almond and walnut acreages continually increase. Risks for infection by common root-knot nematodes are mitigated by rootstock choice, but both crops are susceptible to *Pratylenchus vulnus*. This California-wide pest can be suppressed by pre-plant soil fumigation with 1,3-D. Environmental and human health concerns call for alternative management options. Cover cropping can improve soil health and may be a practice for nematode management, but uncertainties about water use and requirements for nut harvest currently limit adoption. Risk for nematode population density increases by specific cover crops needs to be assessed. Some information on host status towards root-knot nematodes is available [1]. The objectives of this study were to determine (a) the host status of brassica and legume cover crops, (b) the nematode infection and reproductive potential at temperatures typical at in-orchard planting, and (c) cover crop benefits in the field. (a) In greenhouse experiments, 37 Brassica lines and 9 legumes were planted to sand/sandy loam soil in 150-ml conical pots and inoculated with one of the following: *P. vulnus*, *Meloidogyne arenaria, M. incognita*, and *Mesocriconema xenoplax*. After two months, plants were classified as susceptible or resistant based on the reproductive rates. Compared to the susceptible peach 'Nemaguard', most plant lines allowed low reproduction of *P. vulnus*, lines of *Raphanus sativus* and *Crotalaria juncea* being among the poorest hosts. Overall, susceptibility to the root-knot nematodes was higher. Many of the legumes were susceptible to *M. xenoplax.* (b) Three Brassica species and the prunus rootstock 'Krymsk86' were planted to sandy soil in 600-ml cups submerged in water bath temperature tanks set at 15, 20, or 25 °C. Two months after inoculation with *M. incognita* or *P. vulnus*, fewer *M. incognita* were extracted from plant roots grown at lower soil temperatures but numbers in *P. vulnus* were less impacted by soil temperature. (c) Two nematode-infested young almond orchards were planted to cover crop mixes immediately after nut harvest timing. Potentially up to 6 to 9 tons/ha DW biomass was produced under supplemental irrigation and mowing twice before crop termination in the following spring. Cover crops were identified with limited risk of a build-up of population densities of plant-parasitic nematodes. There is considerable potential for orchard management improvement with cover crops.

Keywords: Cover crops - Root lesion nematode - Ring nematode - Root-knot nematode.

References:

- [1] Edwards and Ploeg. 2014. J. Nematol. 46:287-295.

Cover crops and litter mulching improves the soil food web structure and suppressiveness in banana fields.

Camille Chauvin[1] (camille.chauvin@elisol-environnement.fr), Jean-Michel Risede[2], Cécile Villenave[1], Marc Dorel[2]

[1] *ELISOL environnement, Congénies, France*; [2] *CIRAD, Montpellier, France*

Suppression of plant-parasitic nematodes and decomposition of organic matter are important soil food web functions in agriculture [1]. Cover-crop identity, availability and nature of carbon substrates in soil drive the structure, dynamics, and activities of soil food webs and may alter the way they function in the ecosystem [2,3]. We assessed the impacts of planting and incorporating the litter from three fallow covers (Cz: *Crotalaria zanzibarica*, Pn: *Paspalum notatum; PnCz:* a mixed cover of both species) on nematode communities, soil's nematode infective potential and soil mineral nitrogen content in a banana field over a 12 months period. The field was naturally infested with banana plant-parasitic nematodes while *Cz* and *Pn* are non-hosts of the main banana plant-parasitic nematodes *Pratylenchus coffeae* and *Radopholus similis. C*over-crops were also selected on the basis of their difference in cellulose and nitrogen litter content.

All cover crops fostered a more structured and diversified nematode food web in a 9-month period. They also induced changes in parasitic nematode communities by reducing banana plant-parasitic nematodes but favouring other plant parasitic nematodes. Litter mulching represented an important organic resource input causing bacterivorous and fungivorous nematode populations to increase and changes in nitrogen flux while maintaining soil food web structure. N-rich litter of *Cz* and PnCz favoured bacterivorous-cp1 nematodes and high nitrogen flux in soil while *Pn* induced nitrogen immobilization.

A measure of the soil's nematode infective potential revealed a significant regulation of plant-parasitic nematodes after the 12 months of the trial. Fallow cover with litter mulching of *PnCz* induced bottom-up and top-down regulation of soil nematode communities and improved soil suppressiveness in banana fields.

Keywords: Soil food web - Nematode communities - Cover crops - Litter quality - Host status.

References:

- [1] Power, A. G. (2010). Philosophical transactions of the royal society: Biological sciences, 365 (1554), 2959-2971.
- [2] Chauvin, C. et al., (2015). Applied Soil Ecology, 96, 131-140.
- [3] Djigal, D., et al. (2012). Soil B. and Biochemistry, 48, 142-150.

Evaluating sorghum as cover crop against root-knot nematodes.

Caroline Djian-Caporalino[1] (caroline.caporalino@inrae.fr), Thierry Mateille[2], Marc Bailly-Bechet[1], Nathalie Marteu[1], Ariane Fazari[1], Pierre Bautheac[1], Alizée Raptopoulo[1], Luan Van Duong[1], Johannes Tavoillot[2], Bernard Martiny[2], Claire Goillon[3], Philippe Castagnone-Sereno[1]

[1] *UMR ISA (Institut Sophia Agrobiotech), INRAe, Université Côte d'Azur, CNRS, Sophia-Antipolis, France*; [2] *Montpellier SupAgro, Univ. Montpellier, CBGP, IRD, CIRAD, INRA, Montpellier, France*; [3] *Association Provençale R&E Légumière, APREL, Saint-Rémy-de-Provence, France*

Current restrictions on the use of chemical nematicides have led to an increase in root-knot nematode (RKN) damage in horticultural crops. The effects of two sorghums as summer cover crops, *Sorghum sudanense* sudangrass cv. 'Piper' or sudangrass hybrid [*S. bicolor x S. sudanense*] '270911', respectively with low and high dhurrin contents, were compared for their ability to suppress RKN in a vegetable production system. The use of both sorghums 'Piper' and '270911' as a green manure was found to be an effective strategy for decreasing RKN infestation in the soil, thereby protecting the subsequent planting of RKN susceptible crops (chard, lettuce or melon) [1,2]. Analytical experiments were further conducted in growth chamber and greenhouse pot experiments to investigate and compare the susceptibility of the sorghums and the factors affecting their efficacy for RKN management, in order to better explain the results obtained in the field trial. The two sorghums were poor hosts of RKN, acted as trap crops and as a biofumigant releasing hydrogen cyanide. Time of planting, time of biofumigation, and type of soil affected their efficacy. For best RKN suppression, the sorghum cover crops need to be cultivated during one month or less and biofumigated for one month prior to crop planting. The trapping effect of both sorghums in clayey soil was less efficient than in sandy or sandy-loamy soils. Combining less than 30-days of sorghum culture and 10-days soil incorporation with solarization mulch was particularly efficient in suppressing nematodes. No effect relative to the sorghum type was detectable as long as they were used appropriately. Therefore, the efficacy of sorghums clearly depends on the management strategy to be set up in the field. In particular, incorporating sorghums into the soil before the end of the RKN cycle plays a key role in the efficient control of these parasites. All these required conditions may explain why some authors found sorghums efficient to control RKN populations in vegetable cropping systems, while others have been disappointed. In a sample of 28 farms from the South of France, diversified or specialized and faced with the problem of RKN, a survey revealed that using sorghums as summer cover crops by shortening the cropping cycle is well accepted [3,4]. This study thus provides information potentially useful to breeders and farmers for the sustainable management of RKN with a cover crop in protected vegetable systems.

Keywords: Meloidogyne sp. - Sorghum vulgare var. sudanense - Sudangrass hybrids - Trap crop - Biofumigation.

References:

- [1] Djian-Caporalino et al., 2019. Biotechnologie, Agronomie, Société et Environnement (BASE) 23 (1):7-21.
- [2] Djian-Caporalino et al., 2019. Crop Protection 122:142-150.
- [3] Navarrete et al., 2016. Agronomy and Sustainable Development 36:68-7.
- [4] Goillon et al., 2016. Phytoma La défense des végétaux 698:39-44.

Suppressive effect of microbial communities extracted from the rhizospheres of tropical trees, on *M. enterolobii.*

Milad Rashidifard[1] (milad.rashidifard@nwu.ac.za), Hendrika Fourie[1], Gerhard Engelbrecht[1], Samad Ashrafi[2], Mieke Stefanie Daneel[3], Sarina Claassens[1]

[1] *Unit for Environmental sciecnces, North West University, Potchefstroom, South Africa*; [2] *Julius Kühn-Institut, Brunswick, Germany*; [3] *Tropical and Subtropical Crops, Agricultural Research Council, Mbombela, South Africa*

Plant-parasitic nematodes are one of the main biotic factors limiting agricultural production worldwide, with root-knot nematodes (*Meloidogyne* spp.) being the most damaging group. This study was conducted to evaluate the efficacy of soil microbiome associated with various subtropical fruit trees on the management of a *Meloidogyne enterolobii* population. Of 14 soil microbiomes tested for their nematode suppressiveness, nine had significantly ($P \leq 0.05$) lower numbers of eggs and J2 compared to the untreated control. The highest nematode suppression was recorded for SA12 extracted from a papaya orchard with a 38% reduction in nematode population. In addition, the presence of some bacterial (*Bacillus aryabhattai, B. funiculus* and *B. simplex*) and fungal (*Metarhizium marquandii, Acremonium* sp. and *Mortierella* sp.) isolates were correlated with the higher suppression potential in some samples. Substantial variations were observed for the diversity of bacterial and fungi isolates among the samples collected from various hosts and regions. This indicates that the nematode suppression potential of different soil microbiomes depends on the abundance and diversity of fungal and bacterial strains present in the soil. Ultimately, the study confirmed that among all variables, soil dryness, pH, Fe, Zn, organic matter, altitude, and crop genotype strongly influenced soil microbial composition.

Keywords: Biological control - Root-knot nematode - Microbial communities - M. enterolobii - Greenhouse experiment.

Root-knot nematode suppression and effects on soil food web by cover crop and tillage practices in a bare-ground vegetable production system.

Josiah Marquez[1] (josiah.marquez@uga.edu), Timothy Coolong[2], Bhabesh Dutta[1], Abolfazl Hajihassani[3]

[1] *Department of Plant Pathology, University of Georgia, Tifton, GA, United States*; [2] *Department of Horticulture, University of Georgia, Athens, GA, United States*; [3] *Department of Entomology and Nematology, University of Florida, Davie, FL, United States*

Cover crop rotation is an important cultural practice for plant-parasitic nematode (PPN) management [1]. Tropical sunn hemp (*Crotalaria juncea*) and winter rye (*Secale cereale* cv. Wrerens Abrussi) are excellent candidates for vegetable rotations in southeastern United States due to their ability to produce a large volume of biomass [3], be a poor host to root-knot nematodes (RKN; *Meloidogyne* spp.) [2,3], and suppress soilborne pathogens through allelopathy [4]. This study aimed to determine the best combination of seasonal cover cropping and tillage practices in a vegetable rotation. Trials were conducted in 2019-2021 and consisted of three vegetable crops, tomato (*Solanum lycopersicum* cv. BHN 602), squash (*Cucurbita pepo* cv. Spineless Beauty), and cabbage (*Brassica oleracea* cv. Bravo) grown in the spring, fall, and winter, respectively. Tillage treatments (shallow tillage or deep tillage after termination of crops) were main-plot treatments. Sub-plot rotation treatments consisted of two factors; type of rotation (cover crop or fallow) and season of rotation (spring and fall = sunn hemp; winter = rye; or vegetables for all seasons). Sunn hemp successfully suppressed RKN root galling and abundance in the subsequent season when rotated in the spring or fall compared to vegetable rotation ($P<0.05$), while fall planting of sunn hemp also suppressed root galling in the second subsequent season probably due to higher biomass. Rotating sunn hemp in the spring or fall improved vegetable yield. Though sunn hemp and winter rye rotation treatments did not affect fungal soilborne incidence nor abundance of other PPNs, deep tillage suppressed the incidence of *Rhizoctonia solani* in the fall,*Sclerotinia sclerotiorum* in the winter, and *Nanidorus* spp. in the fall and winter. Winter rye had no impact on any pathogens examined; however, it enhanced omnivore abundance, richness, and maturity index in the subsequent season, indicating improved soil food web structure. In contrast, sunn hemp suppressed structure index and carnivore percent in the subsequent season of the spring and fall rotation, suggesting that sunn hemp disturbs the soil food web. This study shows that sunn hemp can successfully suppress RKN in soil for the following vegetable crop independent of tillage practice. Deep tillage treatments had no impact on RKN but can suppress *R. solani, S. sclerotiorum,* and *Nanidorus* spp. in the fall and winter.

Keywords: Cover crop - Deep-tillage - Vegetables - Management - Root-knot nematode.

References:

- [1] McSorley, R. 1999. J. Nematology. 31(4S):619-623.
- [2] Wang et al., 2002. Nematropica. 32(1):35-58.
- [3] McSorley et al., 2009. Nematropica. 39(2):235-245.
- [4] Boppré et al., 2010. Nematology. 12(1):1-24.

Minimum tillage, cover crops, and dead mulch foster free-living nematodes, microbial activity and soil fertility.

Jan Henrik Schmidt[1] (jschmidt@uni-kassel.de), Maria R. Finckh[1], Matthias von Ahn[1], Xorla Kanfra[2], Holger Heuer[2], Johannes Hallmann[2]

[1] *Ecological Plant Protection, University of Kassel, Witzenhausen, Germany*; [2] *Epidemiology and Pathogen Diagnostics, Julius Kühn-Institut, Braunschweig, Germany*

Minimum tillage, organic fertilizers and permanent soil cover by living and dead plants are key components of sustainable agro-ecosystems. Under temperate conditions, soil warming is less in minimum tillage than conventional tillage. As a consequence, mineralization is often reduced and leads to nutrient deficiencies, particularly in organic farming where synthetic fertilizer are lacking. This negative effect might be compensated by fostering an active soil life that increases the level of nutrient turnover and thus fertility. We hypothesize that compost application in combination with high cover crop intensity improves the overall soil fertility as indicated by increasing levels of bacteria feeding nematodes of cp1 class. To test this hypothesis, two identical long-term organically managed field trials (Exp1, Exp2) were studied. The crop rotation included cover/catch crops whenever possible. The treatments applied since 2010/11 were plough (~25 cm) versus minimum tillage (5-10 cm) plus transferred fresh mulch to potatoes. The second factor was the application of 5 t/ (ha yr) yard waste compost versus mineral P and K fertilization matching the contents in the compost. Soil samples were taken from the top 15 cm and Corg, macro-, micronutrients, microbial respiration, and free-living nematodes up to family level analyzed. In summary, P, K, and Ntot were more than 50% higher in minimum tillage than plough tillage. The total number of free-living nematodes was 130% and 37% higher under minimum compared to plough tillage for Exp1 and Exp2, respectively. Already in 2015, the number of free-living nematodes significantly correlated with potato yields (R=0.56, $P<0.001$). In Exp2, *Cephalobidae, Rhabditidae,* and *Panagrolaimidae* (all bacterial feeders) were positively correlated with the microbial respiration and Corg (R=0.61-0.80, $P<0.02$) and their densities were 1.6–4.4-fold higher under minimum compared to plough tillage. The density of *Aphelenchidae* (fungal feeder) under plough tillage was twice as high as under minimum tillage and negatively correlated with microbial respiration and Corg (R=-0.74, $P<0.01$). Omnivorous nematodes were overall low but lowest under plough tillage with mineral fertilization. We conclude that intensive cover cropping and permanent soil cover with living and dead plants will raise the nutritional soil status of the soil and therefore overall soil fertility in organic minimum tillage systems indicated by bacterial feeding nematodes.

Keywords: Organic conservation agriculture - Long-term experiments - Plant nutrition - Bacterivorous nematodes.

S2-PF1

Host status of different cover crops for three *Pratylenchus* species.

Betre Estifanos[1] (betretadesse@yahoo.com), Ank Theelen Peeten[2], Philippe Packbier[2], Bernd Honermeier[3], Johannes Hallmann[1]

[1] *Epidemiology and Pathogen Diagnostics, Julius Kühn-Institut, Federal Research Centre for Cultivated Plants, Münster, North Rhine-Westphalia, Germany*; [2] *Joordens Zaden, Kessel, Netherlands*; [3] *Institute of Agronomy and Plant Breeding I, Justus Liebig University, Gießen, Hessen, Germany*

Root lesion nematodes belonging to the genus *Pratylenchus* are migratory obligate endo-parasites that cause substantial damage to a wide range of economically important crops. Different species often occur concomitantly. The use of cover crops that either suppress or maintain low numbers of root lesion nematodes has increased in recent years. However, data about the host status of different cover crops for mixed species of *Pratylenchus* are scarce. Greenhouse experiments were carried out to study eight cover crops for suppression of three major *Pratylenchus* species, i.e. *P. neglectus*, *P. crenatus* and *P. thornei*. The cover crops tested were 1) berseem clover (*Trifolium alexandrinum*), 2) common vetch (*Vicia sativa*), 3) phacelia (*Phacelia tanacetifolia*), 4) white mustard (*Sinapis alba*), 5) Ethiopian mustard (*Brassica carinata*), 6) Japanese/bristle oat (*Avena strigosa*), 7) oilseed radish (*Raphanus sativus*) and 8) rocket (*Eruca sativa*). Oat (*Avena sativa*) and French marigold (*Tagetes patula*) were used as suceptible and resistant controls, respectively. The average number of nematodes per root systems and nematode reproduction rate was determined 10 weeks after inoculation with 700 mixed stage nematodes of each *Pratylenchus* species. Results obtained indicated that *P. neglectus* was found to have the highest reproduction on common vetch followed by berseem clover, Ethiopian mustard, white mustard and the susceptible control oat with reproduction rate of 6.8, 5.3, 4.8, 2.6 and 2.4, respectively. Regarding *P. crenatus*, the highest reproduction was obtained on common vetch (2.7) followed by berseem clover (1.2). The final population of *P. crenatus* on all other cover crops was below the initial population density (Pf/Pi < 1). *Pratylenchus thornei* only reproduced on common vetch (3.1) and the susceptible control oat (1.3). This study demonstrated that the host status of the tested cover crops varies among the three *Pratylenchus* species. The project was partially funded by Joordans Zaaden B.v. Kessel, the Netherlands.

S2-PF2

Using nematode community analysis to assess the resilience of agricultural soils in the Netherlands.

Anja Kombrink (a.kombrink@hlbbv.nl), Hilde Coolman, Egbert Schepel

HLB BV, Wijster, Netherlands

Soil resilience is a complex concept that depends on the interaction between the soil`s biological, chemical and physical properties. Concerning its biology, the soil is a living ecosystem with a variety of organisms (biodiversity) that all play their role in the soil food web: bacteria, fungi, nematodes, earthworms and insects. A healthy ecosystem is related to and affects (favourably) chemical and physical conditions, such as organic matter, pH and soil structure. Pathogenic organisms, including nematodes, get most attention in arable soil biology because of their apparent damage to crops. However, it is evident that the total soil biology and biodiversity contribute to soil resilience and its capacity to supress diseases. The amounts and diversity of nematodes is relatively easy to measure and analysis of different trophic groups in soil samples has been linked to determining soil quality and soil resilience in various studies. As a private institute for research and consultancy in agriculture HLB BV aims to carry out applied research to specifically increase the knowledge on nematode diversity in agricultural soils in the Northern Netherlands. The obtained data will be used to develop tools that help farmers to gain insight in the resilience of their soils. A research project was started last year that allows for the analysis of a relatively high number of soil samples over three years. (Parts of) parcels with and without problems with soil-borne pathogens will be sampled to investigate differences in nematode communities. In order to limit the expected data variation between samples we will focus on clay/loamy soils and take samples from fields where potatoes will be grown and with the same preceding crop. The results of the nematode analyses will be combined with data on the chemical, physical and microbial properties of the soil and all other relevant information (e.g. fertilization, tillage) about the specific parcels. In this manner we will develop a database that allows the study of relationships between nematode diversity and other soil parameters, with a focus on disease suppressiveness.

Keywords: Nematode community - Trophic groups - Agriculture - Biodiversity.

S2-PF3

Characterization and steering of the native, soil microbiome-based suppression of plant-parasitic nematodes.

Robbert Van Himbeeck (robbert.vanhimbeeck@wur.nl), Stefan Geisen, Johannes Helder

Laboratory of Nematology, Wageningen University & Research, Wageningen, Netherlands

Root-knot and cyst nematodes are among the most harmful plant-parasitic nematodes in the world and are responsible for substantial agricultural losses each year. To control these plant-pathogens, various management strategies can be applied. Due to its adverse side effects the use of nematicides is currently highly restricted, crop rotation is effective but limited to oligophagous plant parasites, and host plant resistance is available for a few major crops only. Thus, there is an urgent need for the development of additional management tools to control plant-parasitic nematodes. Disease suppressiveness (DS) can be defined as the ability of the soil microbiome to suppress the multiplication and proliferation of pathogens. Native DS has the potential of becoming an additional biological control tool. However, despite substantial research efforts in the past to characterize DS, they have not so far resulted in robust management protocols. Most likely, this is due to the highly complex and biodiverse nature of soils. DS is likely the result of a complex of (interacting) antagonists, rather than a one-on-one nematode-antagonist relationship. Hence, it so far remains a barely manipulatable and predictable phenomenon. This project seeks to characterize the complex of biological actors that underlies DS and to develop handles to steer suppressiveness in arable soils. To achieve this, we will first identify suppressive soils and verify the microbial origin of the disease suppression in controlled greenhouse experiments. We will then use next-generation sequencing technologies to characterize both the resident (rDNA) and active (rRNA) microbial communities of these soils. The complex linkage between the microbiome and the suppressive state will be unravelled by using a “microbiome-wide association study” approach. Subsequently, we will identify agronomic variables that are associated with the suppressive state of arable soils. By combining microbiome data with the most promising agronomic variables, we want to develop handles for end-users to exploit the native suppressive potential present in their fields. Thus, this research aims to deliver management tools that can be used to maximize the endogenous ability of soils to suppress root-knot and cyst nematodes. Although this study will focus on arable fields in the Netherlands, this new tool has the potential to contribute to the global demand for environmentally sound management practices.

Keywords: Biological control - Root-knot nematodes - Cyst nematodes - Soil microbiome - Disease suppressiveness.

ORAL SESSION 3

Ecology of free-living nematodes

Mitochondrial metagenomics for evaluation of nematode diversity: paving a way into the future.

Dorota L. Porazinska[1] (dorotalp@ufl.edu), Eli Gendron[1], Joseph L. Sevigny[2], Peter Mullin[3], Kris Powers[3], Thomas O. Powers[3], W. Kelley Thomas[2]

[1] *Department of Entomology and Nematology, University of Florida, Gainesville, FL, United States*; [2] *Molecular, Cellular, and Biomed Sciences, University of New Hampshire, Durham, NH, United States*; [3] *Department of Plant Pathology, University of Nebraska, Lincoln, NE, United States*

Tremendous progress in molecular identification of nematodes has been made during the last decade. Most of the progress in high-throughput sequencing (HTS) has relied on genes encoding ribosomal subunits (i.e., 18S and 28S rRNA) in part because of highly conserved flanking regions that serve as reliable binding sites for “universal” primers [1, 2, 3]. Unfortunately, rRNA markers display insufficient taxonomic resolution to distinguish among closely related species. This lack of resolution can have important consequences for analyses of nematode diversity. Among the more promising markers for species level resolution are protein coding gene markers (PCGs) derived from the mitochondrial genome [e.g., 4, 5]. However, the nematode phylum is characterized by substantial variation within PCGs limiting their use in HTS metabarcoding.

We propose a transition from a single rRNA- or PCG-based marker identification to a shot-gun sequencing of all mitochondrial genomes simultaneously known as mitochondrial metagenomics (mtMG). We applied the mtMG approach to artificially assembled mock communities from well characterized nematode cultures as well as natural communities residing in soils of temperate broadleaf and mixed forests in the Missouri Glades, USA. We used these samples primarily for their full scope of characterization with traditional identification methods (total counts, trophic group counts, 125 randomly selected individuals morphologically identified to species, 25 randomly selected individuals confirmed molecularly using COI and 18S barcodes). We discuss the results in the context of advantages and limitations of the mtMG protocol and provide a path for accurate nematode community identification in the near future.

Keywords: Diversity - Metabarcoding - Mitochondrial metagenomics - Nematode species identification - Sequencing.

References:

- [1] Porazinska et al., 2009. Molecular Ecology Resources. 9:1439-1450.
- [2] Waeyenberge et al., 2019. Diversity. 11(4):52.
- [3] Ahmed et al., 2019 Metabarcoding and Metagenomics. 3:e36408.
- [4] Janssen et al., 2016. Scientific Reports. 6:22591.
- [5] Powers et al., 2018. J Nematol. 50(3):399-412.

On the importance of species diversity in ecosystem functions and services.

Howard Ferris (hferris@ucdavis.edu)

Department of Entomology and Nematology, University of California Davis, Davis, California, United States

Diversity is the condition of being composed of different elements. In ecological contexts, diversity may be taxonomic (species, genera, families) or functional (guilds or trophic habits). Soil and aquatic environments comprise a variety of micro-niches determined by substrate characteristics, resource distribution, microclimatic conditions, and perturbation frequency. The micro-niches provide habitat for myriad organisms of different size, physiological activity, behavior and ecosystem function. Based on physiological, behavioral or morphological attributes, nematode species may differ in adaptations to a range of available microhabitats. Measurement of diversity of whole assemblages of nematodes, irrespective of their ecosystem function, does not provide useful information on functional equivalence among taxa. The diversity and abundance of species within functional classes provides insights into the nature of ecosystem functions and services and to the health of the soil. Species with broad tolerance of edaphic conditions may function throughout a soil profile; species with narrow tolerance may be restricted in seasonal activity or to a limited soil stratum. Similarly, large-bodied organisms may be unable to access resources confined in small spaces; species with vigorous activity may agitate the resource suspension and increase its accessibility. Clearly, the magnitude of an ecosystem function is enhanced by the diversity of species in that functional class. In practice, identification of nematode species and resolution of their function present research opportunities and challenges. The aggregation of species into higher taxonomic or generalized functional groups to facilitate identification results in loss of resolution. Further, the opacity of the soil presents challenges in the measurement and interpretation of the functional diversity of soil organisms. A sample from one depth stratum may provide information on function and implied functional equivalence of organisms with similar physiological, but not necessarily behavioral, attributes. Alternatively, species diversity of the nematode assemblage determined within a core of soil representing several depth strata may provide information on functional responses of the assemblage to seasonal climatic cycles and even to sensitivity of the function to climate change. Consequently, diversity of species within functional classes is a key element of the biological component of soil health.

Keywords: Diversity - Ecosystem Function - Equivalence - Effective Species.

Protected areas in southern Africa, what do we know of the nematode fauna?

Candice Jansen van Rensburg (jvrensc@ufs.ac.za)

Department of Zoology & Entomology, University of the Free State, Bloemfontein, South Africa

South Africa has a total of 1617 protected areas that includes nature reserves and national parks, while in contrast Botswana only has 22. Most fauna from these areas are well known, however this is not the case for smaller invertebrates such as nematodes. Very few recent studies on the biodiversity of plant-parasitic and free-living nematodes from natural communities in southern Africa exist [1]. Moreover, a few scattered reports of nematodes from protected areas in southern Africa can be found in literature, with the earliest record in 1963 from the Kruger National Park [2] and the most recent in 2019 from the Telperion Nature Reserve in South Africa [3]. Over the last number of years, soil and leaf litter samples were collected from six protected areas within and outside the borders of South Africa. The South African reserves were Seekoeivlei, Willem Pretorius and Soetdoring, all from the Free State Province; Jonkershoek and De Hoop Nature Reserves both in the southern Cape, as well as the Okavango Delta in Botswana. Nematodes were extracted from samples and processed using standard techniques at the nematology laboratory in Bloemfontein. A total of 37 genera from 21 families from Seekoeivlei Nature Reserve; 38 genera from 23 families from Willem Pretorius Nature Reserve; 35 genera from 21 families from Soetdoring Nature Reserve; 32 genera from 18 families from Jonkershoek and 18 genera belonging to 12 families from the De Hoop Nature Reserve, were identified. Fourteen nematode genera belonging to 11 families were collected in the Okavango Delta. Some of the nematode families identified include: Plectidae, Cephalobidae, Criconematidae, Aphelenchoididae, Teratocephalidae, Mononchidae, Qudsianematidae, Desmodoridae and Prismatolaimidae. To date, only six protected areas have been studied, the main aim and focus will be to continue with the mapping of the biodiversity of these lesser studied areas. In doing so, new species will be described and the ecological importance of nematodes within these areas will also be determined.

Keywords: Protected area - South Africa - Biodiversity - Botswana - Free-living nematodes.

References:

- [1] Procter, 2012. J. Arid Environ. 76: 142-146.
- [2] Van der Vegte and Heyns, 1963. Koedoe 6: 134-136.
- [3] Girgan et al., 2019. Zootaxa 4561(2): 201-234.

Distribution and diversity of moss-dwelling nematodes.

Bianca Kreuzinger- Janik (bianca.kreuzinger-janik@uni-bielefeld.de), Nabil Majdi, Walter Traunspurger

Bielefeld University, Bielefeld, Germany

Moss can be found both in aquatic and terrestrial environments and grows on all kinds of substrates like soil, stones or wood. It serves as structurally complex habitat and provide shelter from unfavorable environmental conditions such as desiccation or temperature extremes and offer refuges from predators. Moss carpets form extensive interfaces between lake and forest ecosystems, functioning as a transition zone between two ecosystems (so-called ecotones). They are characterized by a particular set of microclimatic and ecological conditions that favour the establishment of nematode species from adjacent ecosystems (i.e., from the lake littoral and forest soil). Surprisingly, our knowledge of the moss-dwelling nematode community is scarce. Here we present the density, diversity, and distribution of nematodes in aquatic and terrestric moss patches along a transect from submerged mosses underwater, over the splashwater zone, to exposed mosses of terrestrial habitats along a Swedish Lake. We found a total of 74 nematode species and maximum diversity was attained 1 m away from the shoreline and at the splash-water zone. Only two species, *Plectus opisthocirculus* and *Teratocephalus terrestris*, were detected at all sites. Moss-dwelling nematodes may use a variety of food resources such as organic material, algae, bacteria and small protozoans accumulating in the moss matrix and are represented by a wide range of feeding strategies that allows exploitation of the diverse microbial resources found in this environment. Evidenced by the multiple shifts in dominance, our study clearly showed that feeding types differed between habitats. The taxonomic and functional segregation of the nematode community along the ecotone further indicated that the composition of nematode species assemblages has the potential to mirror the ecological changes of the study site, even at smallscales.

Keywords: Feeding types - Community structure - Freshwater ecology - Bryophytes - Lake.

Natural ecosystem diversity and functioning of nematode communities in a semi-desert ecosystem in Mexico.

Hugo Mejía-Madrid[1] (hugo_mejia_madrid@ciencias.unam.mx), Sara Sánchez-Moreno[2]

[1] *Departamento de Ecología y Recursos Naturales, Universidad Nacional Autónoma de México, México, Mexico*; [2] *Departamento de Medio Ambiente y Agronomía, Centro Nacional Instituto de Investigación y Tecnología Agraria y Alimentaria |, Madrid, Spain*

The aim of the present study was to detect the heterogeneity of diversity in seasonal, site, tree species, and soil properties' effects on structural and functional attributes of the nematode communities under *Prosopis laevigata* and *Cericidium praecox* within the Biosphere Reserve Valle de Tehuacán-Cuicatlán. Soils under biological crusts and neighboring exposed sites were surveyed during the dry and wet seasons of 2018. Assessment of effective diversity (D) and diversity weighted by metabolic footprint (DMFP) were calculated as they reflect taxonomic and functional diversity. Trophic group abundances and both indices showed marked seasonal variation. Soil water content and bulk density were the only physicochemical variates that showed seasonal variation whereas bulk density, total C and total N content showed significant variation between soil under biological crusts and exposed sites. DMFP seemed to have a better chance of explaining a relationship with physicochemical heterogeneity than D as shown by PCA analyses. This places DMFP as an index for measuring diversity related to ecosystem services and therefore should be used as a tool for biological integrity assessments at the soil level and a support aid in stewardship soil management policies.

Keywords: Nematodes - Metabolic footprint - Diversity - Desert - Mexico.

References:

- [1] Ferris and Tuomisto, 2015. Soil. Biol. Biochem. 85 (2015): 101-109.
- [2] Sánchez-Moreno and Ferris, 2018. Chapter 3. Plant Parasitic Nematodes in Subtropical and Tropical Agriculture, CAB International and USDA, pp. 62-86.

Bacterial nematode grazing enhances P mobilization from phytate by *Bacillus subtilis*: what are the mechanisms involved?

Claude Plassard[1] (claude.plassard@inrae.fr), Mercedes Garcia-Sanchez Garcia-Sanchez[1], Houssein Monder[1], Mathilde Souche[1], Anne-Laure Pablo[2], Carlos Trives-Segura[1], Leyla Slamti[3], Julien Brillard[4]

[1] *UMR Eco&Sols, INRAe, Montpellier, France*; [2] *UMR Eco&Sols, IRD, Montpellier, France*; [3] *Institut MICALIS, INRAe, Jouy-en-Josas, France*; [4] *UMR DGIMI, INRAe, Montpellier, France*

Objectives:
In soil, forms of organic P (Po), and particularly phytate (myo-inositol-hexakisphosphate), can constitute a very large proportion of total P. To be used by living organisms, the phosphate (Pi) group of Po molecules must be released by enzymes (phosphatases and phytases). Unlike plants, many bacterial species are capable of using phytate as the only source of P, thanks to their production of phytase. Among these species, those producing extracellular phytases such as *Bacillus subtilis* are potentially important for the mineralization of phytate. However, our previous work [1,2] showed that the presence of bacterivorous nematodes was essential for the plant to benefit from the P of phytate mineralized by *B. subtilis*. The objective of the present work was to investigate the possible mechanisms explaining the positive effect of bacterial grazing by nematodes.
Methods:
We used monoaxenic populations of *Acrobeloides*sp. fed on three strains of *B. subtilis*168 (native and two strains expressing the green fluorescent protein (GFP) under the dependence of the bacterial phytase promoter). Plant species was durum wheat. All experiments were carried out in controlled conditions, in agarose medium supplemented with 1 mM sodium phytate. In a first experiment, we located the deposit of nematodes or bacteria (*B. subtilis*168) alone. After 3 weeks of incubation, agarose was split in two parts to measure the dispersal of the organisms and the release of free Pi from phytate. In a second experiment, durum wheat (*Triticum durum*) seedlings were grown alone or inoculated with the three bacterial strains (native and GFP ones) with or without nematodes. After 1 month, fluorescence of bacteria and total plant P amounts were measured.
Results:
The first experiment shows that bacterial grazing improves the mineralization of phytate by increasing the dispersion of bacteria but also the release of Pi per unit of bacteria, suggesting that grazing could modulate the release of phytase by the bacteria. This was confirmed in the second experiment with GFP strains showing higher levels of fluorescence in presence than in absence of nematodes. In addition, nematodes increased the growth of the wheat root system and thus plant capacity to take Pi from mineralized phytate.
Conclusion:
Nematode grazing improves the mineralization of phytate by increasing (i) the dispersion of bacteria, (ii) the expression of the *B. subtilis* phytase gene and (iii) the growth of the wheat root system.

Keywords: Bacterial grazing nematodes - Acrobeloides sp. - Extracellular phytase - GFP - Phytate mineralization.

References:

- [1] Irshad et al., 2012, Plant & Soil, 358: 148-161.
- [2] Ranaorisoa et al., 2020, Soil Biol. Biochem., in press.

S3-PF1

The relations between glyphosate and soil health based on nematode trophic groups in Turkish hazelnut orchards.

Sevilhan Mennan[1] (smennan@omu.edu.tr), Gökhan Aydınlı[2], Ivo Brants[3], Xavier Belvaux[3], Husrev Mennan[1]

[1] *Plant protection dep., Ondokuz Mayıs University, Samsun, Turkey*; [2] *Animal and plant production department, Ondokuz Mayıs University, Bafra Vocational School, Bafra, Samsun, Turkey*; [3] *Bayer Agriculture BVBA, Brussel, Belgium*

Nematodes can be used as effective soil health bioindicators because they occur in any environment that provides a source of organic carbon, in every soil type, are easy to sample, occupy well classified functional groups, and nematode taxa are well classified [1, 2]. Scientific studies about the impact of glyphosate, the most widely used herbicide in the world, on soil micro-organisms have provided contrasting results to date [3]. We aimed to see the effects of glyposate on the soil nematode community and the soil health. In order to examine the long and short-term effects of glyphosate, two orchards, one in which glyphosate was continuously applied for many years (O1) and another with no historical glyphosate use (O2) were selected and 3 treatments were applied in each orchard: glyphosate applied at 3 lt/ha or 6 lt/ha, and diquat at 8 lt/ha. Experiments were started in April. 2018 by applying herbicides and taking soil samples before applications. Soil sampling was done at 2, 5, 10, 15, 20, 30, 45, 65 days after application. A second herbicide application was done July. 2018 and same soil sampling repeated. Nematodes were extracted by using Baerman funnel method and each nematode genus was classified according to feeding type. Each nematode genus identified was assigned to colonizer-persister and nematode community indices maturity index, plant parasitic index and combined MI (∑MI) were quantified and enrichment (EI) and structure indices (SI) were computed [4, 5]. A total of 33 and 34 soil nematode genera were identified in O1 and O2 respectively. Compared to gardens, all free-living and bacteria-feeding nematodes were higher in O1. After two glyphosate applications, just predators were more abundant in O1. A decrease in opportunistic nematodes reflects an overall decrease of biological productivity in the system. Glyphosate had no adverse effects on bacteria-feeding, fungal-feeding, plant-parasitic or omnivorous nematodes. When comparing nematode community indices, although the main ecosystem disturbance (MI, PPI, and ∑MI) and soil food web (EI and SI) indices did not show consistent trends across treatments or time, some of the ∑MI and SI results are noteworthy. High ∑MI and SI indicate more diverse and stable soil food webs, thus the results of this study revealed complex relationships among nematode communities and glyphosate applications.

Keywords: Glyphosate - Nematode community index - Soil health - Ecosystem disturbance.

References:

- [1] Neher, 2001. J. Nematol. 33, 161–8.
- [2] Pattison et al., 2008. Appl. Soil Ecol. 40, 155–164.
- [3] Greenslade et al., 2010. Soil Biol Biochem 42:1172–1175.
- [4] Bongers, 1990. Oecologia 83, 14–19.
- [5] Yeates, 1994. Pedobiology 38:97-101.

S3-PF2

Soil nematode communities around gopher tortoise burrows in native and degraded ecosystems.

Rachel M. Shepherd[1] (r.shepherd@ufl.edu), L .M. Uy[1], K. M. Gattoni[1], J. P. McQueen[1], E. M. S. Gendron[1], P. G. Hahn[1], M. A. Lashley[2], D. L. Porazinska[1]

[1] *Department of Entomology and Nematology, University of Florida, Gainesville, United States*; [2] *Department of Wildlife Ecology and Conservation, University of Florida, Gainesville, United States*

Ecosystem engineers change, manage, and disproportionately affect their surrounding biodiversity. Among these are the threatened gopher tortoises (*Gopherus polyphemus*) found in the savannas of the Southern Coastal Plain, USA. Known for their role in promoting aboveground diversity (e.g. plants, insects, and vertebrates) in their native habitat, gopher tortoises have also been proposed as an effective method of restoration of degraded ecosystems. Additionally, they are known to affect soils and belowground communities both directly (e.g., soil chemical change through tortoise burrowing) and indirectly (e.g., plant community change through tortoise foraging). To examine the extent to which gopher tortoises influence soil communities, we selected 15 paired sites representing two ecosystem types (native vs. degraded) from the Ordway-Swisher Biological Station, Florida in Fall 2021 and collected soil samples around 3 areas of gopher tortoise activity (apron, mound, and 15m away from the burrow). Nematodes were extracted from 100 g subsamples, counted, and identified with microscopy, and then processed for 18S rRNA (NF1-18Sr2b) metabarcoding. All samples were also evaluated for biogeochemistry.

We hypothesized that 1. Native and degraded ecosystems would be characterized by similar nematode communities, and 2. Areas of gopher activity would be characterized by distinct nematode communities. Specifically, their burrowing activity, represented by the apron samples, would contain low diversity communities which can be indicative of disturbed soil ecosystems. In contrast, communities in the mound and sites away from burrows would be highly diverse due to the presence and stabilizing effect of plants.

Contrary to our 1st hypothesis, communities in native ecosystems were more diverse than in degraded ecosystems. In agreement with our 2nd hypothesis, communities of aprons were indeed least diverse and distinct from those in mound and away from burrows. However, these patterns were variable across trophic groups with bacterial-feeding nematodes largely driving patterns, with pH explaining significant variation in degraded sites and organic matter in native sites. Our results indicate that different gopher tortoise activities influence nematode communities, and their potential role in restoration of degraded ecosystems must be further investigated by observing trends in other organisms in the ecosystem.

S3-PF3

CRISPR and mutagenesis experiments reveal mechanisms of integration of horizontally acquired cellulases in *Pristionchus*

Ziduan Han (ziduan.han@tuebingen.mpg.de), Ralf J. Sommer

Department 4, Max Planck Institute, Tuebingen, BW, Germany

Horizontal gene transfer (HGT) is the exchange of genetic material between non-related species and it has been shown to contribute to the evolution of nematodes, specifically for plant-parasitism. *Pristionchus* are microbial-feeding nematodes and have horizontally acquired cellulase genes from eukaryotic origin. We recently demonstrated that these cellulases are likely produced in the gland cells and are secreted to digest the resilient structure of bacterial biofilms increasing the fitness of the nematodes [1]. However, how these horizontally acquired genes are regulated by the recipient is still unclear. We found that the expression levels and patterns of cellulase genes are affected by environmental cues, such as starvation and bacterial foods. Moreover, as *P. pacificus* represents an example of developmental plasticity which results in two distinct life styles, a predator and a straight microbial-feeder, we have found higher cellulase expression levels associated with the microbial-feeders. Thus, cellulase genes have integrated into the complex gene regulatory networks of *P. pacificus*. Here, we are investigating the upstream regulators of the main cellulase gene (*cel-2*) in *P. pacificus.* We are conducting an EMS-induced mutagenesis screen on a fluorescent reporter line driven by the promotor of *cel-2* (*cel-2p*::RFP) to identify mutants with abnormal *cel-2* expression. Subsequently, we will ascertain the causative genes with genetic mapping and CRISPR genome-editing. With further characterization of these genes and illustration of the biological pathways involved, we will be able to demonstrate the integration of horizontally acquired genes in *P. pacificus* and provide new insights into the molecular mechanism of HGT in animal evolution.

Keywords: Pristionchus - Upstream regulator - Forward genetics - Horizontal gene transfer - Animal evolution.

References:

- [1] Han et al., 2022. Molecular Biology and Evolution. 39(2), msab370.

ORAL SESSION 4

EPN ecology and biology

How the spores of *Isaria fumosorosea* strain CCM8367 are spread by entomopathogenic nematodes.

Jiří Nermuť[1] (nermut@entu.cas.cz), Jana Konopická[2], Vladimír Půža[1], Rostislav Zemek[1]

[1] *Institute of Entomology, Biology Centre CAS, České Budějovice, Czechia*; [2] *Faculty of Agriculture, University of South Bohemia, České Budějovice, Czechia*

Entomopathogenic nematodes are almost ubiquitous soil dwelling organisms that play an important role in natural management of insect populations. Nematodes, especially plant-parasitic, are known to be able to spread and transmit plant-pathogenic viruses and fungi. *Isaria fumosorosea* (syn. *Paecilomyces fumosoroseus*) strain CCM 8367 was shown to be virulent to Colorado potato beatle (*Leptinotarsa decemlineata*) causing 92.6 % mortality of larvae. The combined application of the fungus with the entomopathogenic nematodes increased the mortality up to 98.0 %. Based on this finding there was a question if entomopathogenic nematodes are able to spread spores of this fungus on their bodies and how effective can be such transport. For this purpose we designed several types of simple agar and more complex (sand and soil) experimental arenas, olfactometers included. Our goal was to obtain the answer to the question, if and how the spreading of *I. fumosorosea* can be affected by the presence of entomopathogenic nematodes.

Keywords: *Steinernema feltiae* - *Heterorhabditis bacteriophora* - Entomopathogenic fungus - Bilogical control.

Machines and machine learning for applied nematology: potential and opportunity.

Denis S. Willett[1] (deniswillett@cornell.edu), Fahiem E. El-Borai[2], Larry W. Duncan[3], Camila C. Filgueiras[1]

[1] *Applied Chemical Ecology Technology, Cornell AgriTech, Cornell University, Geneva, NY, United States*; [2] *Gulf Coast Research and Education Center, University of Florida, Wimauma, FL, United States*; [3] *Citrus Research and Education Center, University of Florida, Lake Alfred, FL, United States*

Recent advances in technology have opened the doors to opportunities for automation and enhanced exploration in the applied sciences. Nematologists spend an inordinate amount of time visually identifying, counting, and manually separating individual nematodes in samples. Here we discuss how specific advances in instrumentation coupled with machine learning can automate much of this process with high precision and accuracy. We also discuss opportunities for applying these advanced technologies in addressing unanswered questions in nematode ecology and management.

Keywords: Machine Learning - Counting - Automation - Sorting - Technology.

Role of *Xenorhabdus* on the metabolism of *Steinernema* infective juveniles (Nematoda: Steinernematidae).

S. Patricia Stock (spstock@email.arizona.edu), Emilie Lefoulon

School of Animal and Comparative Biomedical Sciences, University of Arizona, Tucson, AZ, United States

The intimate association of *Steinernema* nematodes with their *Xenorhabdus* bacterial symbionts is an important component of their biology. *Xenorhabdus* bacteria colonize a specialized receptacle in the anterior portion of free-living, infective juvenile (IJ) stages of the nematode where they are provisioned and protected. *Xenorhabdus* symbionts are released upon invasion into a suitable insect host producing a suite of secondary metabolites and toxins which subdue the insect's immune system and produce a suitable environment for nematode maturation and reproduction [1-3]. Recent studies have demonstrated that this symbiosis is mutually beneficial and impacts the fitness of both the nematode host and bacterium [4-5]. Arguably the most vulnerable and important aspect of the dynamic lifecycle of these organisms is IJ. This free-living stage is non-feeding, which must be resilient to environmental conditions to be able to perpetuate not only the life cycle of the nematodes but that of *Xenorhabdus* symbionts for which they must provision on their stored nutrients. To investigate the impact of symbiont colonization on IJ metabolism, we performed a comparative dual RNA-seq analysis of IJs of two nematode-bacteria partnerships: *Steinernema carpocapsae-Xenorhabdus nematophila* and *Steinernema. puntauvense-Xenorhbdus bovienii.* For each association, three conditions were studied: (1) IJs reared in the insect (in vivo colonized), (2) colonized IJs reared on liver-kidney agar (in vitro colonized), and (3) IJs depleted by the bacteria reared on liver-kidney agar (in vitro aposymbiotic). Our study revealed the downregulation of numerous genes involved in metabolism pathways, such as carbohydrate, amino acid, and lipid metabolism when IJs were reared in vitro, both colonized and without the symbiont. This downregulation appears to impact the longevity pathway, with the involvement of glycogen and trehalose metabolism, as well as arginine metabolism. Additionally, a differential expression of the venom protein known to be secreted by the nematodes was observed when both *Steinernema* species were depleted of their symbiotic partners. These results suggest *Steinernema* IJs may have a mechanism to adapt their virulence in absence of their symbionts.

Keywords: *Steinernema* - *Xenorhabdus* - Symbiont - Infective-juvenile - Transcriptome.

References:

- [1] Boemare, N.E. 2002.Entomopathogenic Nematology. 2002 Jan 31:35-56.
- [2] Nielsen-LeRoux et al. 2012. Curr. Opin. Microbiol. 12(15):220–231.
- [3] Castagnola, A. and Stock, S.P. 2014. Insects, 15:139–166.
- [4] Cahpuis, E et al. Proc. Biol. Sci. 279: 2672-2680.
- [5] McMullen et al. Microbiology. 163: 510-522.

Interactions between entomoparasitic nematodes within their host species, rose chafer grub.

Luca Eszter Balog[1] (luca.eszter.balog@ttk.elte.hu), Oleksandr Holovachov[2], Júlia Katalin Török[1]

[1] *Dep. of Systematic Zoology and Ecology, Eötvös Loránd University, Budapest, Hungary*; [2] *Department of Zoology, Swedish Museum of Natural History, Stockholm, Sweden*

Various entomoparasitic nematodes found in scarab beetles (Scarabaeoidea) differ in their effect on the host. Obligate parasitic nematode species spend their whole life inside the host's body, as opposed to the facultative entomopathogenic nematodes, which have both free-living and insect-parasitic stages. In this study, rose chafer, *Cetonia aurata*'s (Coleoptera: Scarabaeoidea) grubs were collected from soil and compost in Hungary. Isolated nematodes were identified using light and scanning electron microscopy. The intensity of the nematode infection and the host's condition were also determined. The interaction between different parasitic nematodes and their effect on the host were analyzed using statistical methods. The hypothesis to be tested in this project assumes that nematode taxa with different evolutionary strategies differ in their effect on the host. The obligate parasitic and the necromenic nematodes inhabiting host's intestine never kill the host but influence the density of other parasitic nematodes present in the same individual. The gut of numerous grub specimens was infected with the Thelastomatidae nematode species, while the insect's coelom was infected mainly with nematode species of the Rhabditidae and Panagrolaimidae families. The rhabditid entomopathogenic nematodes significantly weakened the condition of the host and, in many cases, killed it, whereas the obligate parasitic nematodes had no significant effect on the host condition. The intensities of facultative entomoparasitic and obligate parasitic nematode infections correlated positively with each other, but high nematode intensity in the grub's coelom was found only in dead specimens. Better understanding of ecological interactions between parasitic nematodes and their hosts is essential for the effective development of biocontrol methods, as we found that the parasites in multispecies infections had a significant impact on the state of the host. However, many fundamental studies are yet to be done before developing new biological control tools, as our knowledge about host specificity among Rhabditidae and Panagrolaimidae species and their impact on insects is still far from complete.

Keywords: Rose chafer grub - Parasitic interactions - Necromenic nematodes - Obligate parasitic nematodes - Entomopathogenic nematodes.

Molecular effectors in immune modulation and pathogenicity by entomopathogenic nematodes.

Adler Dillman[1] (adlerd@ucr.edu), Sophia Parks[1], Chau Nguyen[1], Shyon Nasrolahi[1], Dihong Lu[1], Raghavendran Ramaswamy[2], Anna Buchman[3], Omar Akbari[3], Naoki Yamanaka[4], Martin Boulanger[2]

[1] *Department of Nematology, University of California, Riverside, CA, United States*; [2] *Biochemistry and Microbiology, University of Victoria, British Columbia, Canada*; [3] *Division of Biological Sciences, University of California, San Diego, CA, United States*; [4] *Department of Entomology, University of California, Riverside, CA, United States*

Entomopathogenic nematodes are lethal parasites of insects that utilize mutualistic bacteria in their parasitism of insects. The free-living, developmentally arrested infective juvenile (IJ) stage infects hosts. IJs release insect-pathogenic bacteria along with an arsenal of nematode proteins into hosts. The mixture of proteins released by activated IJs is toxic at low doses to a variety of insect hosts, illustrating that nematodes contribute to the killing of their hosts. It has been shown that activated *Steinernema carpocapsae* and *S. feltiae* IJs release hundreds of proteins into their hosts, but the role of most of these proteins in parasitism is unknown. We have identified several fatty acid- and retinol-binding proteins (FARs) released by activated IJs. Here we provide evidence suggesting that these FAR proteins are potent modulators of immunity, decreasing host resistance to bacterial infection. We present the results of our research aimed at understanding how FAR proteins modulate host immunity. Our data suggest that activated steinernematid IJs release a complex mixture of proteins that modulate host immunity and contribute to host death.

Keywords: Toxins - Immune modulation - *Steinernema* - Effectors - EPNs.

References:

- Lu et al. 2017. PLoS Pathogens 13(4): e1006302.
- Chang et al. 2019. PLoS Pathogens 15(5): e1007626.

Key transcriptional changes in the shifting from free-living to infective stage of *Heterorhabditis bacteriophora*.

Nelson Simões (nelson.jo.simoes@uac.pt), Duarte Toubarro

Centro de Biotecnologia dos Açores (CBA), Universidade dos Açores, Ponta Delgada, Portugal

Heterorhabditis bacteriophora is an entomopathogenic nematode largely used to control insect pests. The third juvenile of this nematode is a free-living nematode seeking a suitable host. Whenever the potential host is found the juveniles shift to an infective stage that is able to invade, to overcome defences, to reach the haemocoel and to kill the insect in less than 48 hours. In order to elucidate the mechanisms underlying such abilities we performed a comparative analysis of transcripts in both stages. Total RNA was extracted, cDNA libraries generated and sequenced by an Illumina HiSeq2000. The *de novo* assembly produced 65 517 non-redundant transcripts corresponding to 17 017 distinct genes and to 57 326 predicted proteins. BLASTX search against Swiss-prot and NCBI databases revealed that 45 560 sequences have 74%, 17% and 9% significant similarity with proteins in animal parasitic-, free-living- and plant parasitic- nematodes, respectively. A Venn diagram distribution of genes with a threshold of 10 FPKM, allowed the identification of 8 223 genes common to free-living and infective stages, 92 and 1 982 unique in the free-living and in the infective stages, respectively. Negative regulation of gene expression and nematode larval development were the GOs most represented in free-living nematodes. To go deep in the identification of genes underlying the activation of the infective stage we stimulated arrested nematodes with different stimuli. These experiments allowed the identification of GOs expressed differently. Exsheathment promoted the activation of pharyngeal development, nervous system and muscle contraction GOs. Neuronal stimulation induced the overexpression of response to external stimulus and anatomic structure morphogenesis GOs. Stimulation with insect haemolymph induced the overexpression of vesicle mediated transport and locomotion GOs. It is noteworthy that 243 genes were expressed only in juveniles stimulated with insect haemolymph, suggesting they are a nematode/insect arms race signature. These genes belong to metabolism, genetic information processing, environmental information processing and cellular processes KEGGs. The most represented GO domains were molecular biding and catalytic activity, the latter with 269 genes encoding for peptidases, 57 of them with signal P, thus suggesting they are secreted and potentially released to participate in the parasitic process.

Keywords: Entomopathogenic nematode - Infective juvenile - Transcripts - Enriched KEEG - Enriched GO.

S4-PF1

War in the darkness: the use of volatile organic compounds and entomopathogenic nematodes to control wireworms.

Diana la Forgia[1] (diana.laforgia@doct.uliege.be), François Verheggen[1], Andrea Chacon[2]

[1] *Chemical and Behavioural Ecology, University of Liege, Gembloux Agro-BioTe, Gembloux, Belgium*; [2] *Functional and Evolutionary Entomology, Gembloux Agro-Bio Tech - TERRA (ULiège), Gembloux, Belgium*

Plant roots release in the soil Volatiles Organic Compounds (VOCs) that guide belowground phytophagous insects, such as wireworms (Coleoptera: Elateridae). Wireworms are generalist insect pests that can be found in many crops where they are responsible for high economical losses, killing the plants at early stages. Attract-and-kill strategies have great potential because wireworms are attracted by plant odors released from the rhizosphere and, later on, entomopathogenic nematodes (EPNs) have shown a great utility to control the larvae of various soil insect pests. In order to develop such an attract-and-kill strategy, we archeived different aims: (1) selection of entomopathogenic nematode species that readily infect and kill wireworms, combined with attractive VOCs (2) development of a blend of VOCs that could attract wireworms and induce feeding on a lure with EPNs (3) comparison of conventional screening to the attract-and-kill system, showing increased efficiency (4) wireworm monitoring for a possible effect on metabolism following EPN infection in conventional screening compared to the attract-and-kill feeding. By increasing the efficiency of attract-and-kill, we aim to further improve this promising alternative to pesticides.

Keywords: Attract-and-kill - Alginate beads - Entomopathogenic nematode - Feeding attractants - Volatile Organic Compounds.

S4-PF2

Mass spectrometry-driven discovery of neuropeptidergic systems regulating nictation in free-living and parasitic nematodes.

Bram Cockx[1] (bram.cockx@kuleuven.be), Rose Boelen[1], Johnathan Dalzell[2], Liesbet Temmerman[1]

[1] *Lab of Molecular and Functional Neurobiology, KULeuven, Leuven, Belgium*; [2] *Queens University Belfast, Belfast, United Kingdom*

As in many nematodes, behavioral changes are regulated by neuropeptidergic signaling (e.g. *Caenorhabditis elegans*) [1]. Emerging evidence suggests that behavioral changes associated with host finding - like observed in entomopathogenic nematodes (EPNs) - may be regulated by neuropeptides [2]. Hence, peptidomic discovery in EPNs, like *Steinernema* species, provide a fundament upon which differential and functional work can build. The peptidomic similarities between the well-known *C. elegans* peptidome and predicted *S. carpocapsae* peptidome are profound, indicating evolutionary conserved behavioral regulations. This, combined with the extensive peptidomic knowledge of our lab, gives us a broad fundament for peptidomic research in other nematodes. We aim to understand the neuropeptidergic signaling system underlying nictation – the evolutionary conserved behavior for foraging in nematodes – in *S. carpocapsae* and *C.* elegans.
An in-house method based on acidified methanol was used to extract endogenous neuropeptides of *Steinernema carpocapsae*. Neuropeptide identification was done by state-of-the-art sub-Dalton high accuracy tandem mass spectrometry coupled online to an ultra-high performance liquid chromatograph (UHPLC-MS/MS). Currently, we can detect 30% (139) of the predicted peptidome. Selected neuropeptide precursors have been tested for nictation deficits using a micro-dirt system. Up to now, we found at least one *C. elegans* neuropeptidergic signaling system that is involved in nictation modulation. Results of all tested systems in *C. elegans* and *S. carpocapsae* will be presented at the conference. *Steinernema* spp. are used as a potent and environment-friendly alternative for chemical pesticides to combat pest insects. For now, EPN only have a limited applicability and there is a need for host-range and ecological niche expansion, while controlling host specificity. Knowledge on neuropeptidergic regulation of host-finding strategies will help us understand how EPNs regulate their behavior, hence, contribute to improving EPN applicability and host specificity.

Keywords: Host-finding - Mass spectrometry - Neuropeptidomics - Nictation - Pest control.

References:

- [1] Schoofs et al, 2019, Frontier in Endocrinilogy, 10, Art.No. ARTN 64.
- [2] Dalzell et al. 2017, PLOS Pathogens, 13(3): e1006185.

S4-PF3

Standardized surveys confirm greater EPN presence and diversity in a subtropical compared to Mediterranean citrus orchards.

Alexandros Dritsoulas[1] (alexdritsi@gmail.com), Fahiem El-Borai Kora[2], Mostafa Hammam M. A.[3], Mahfouz Abd-Elgawad M. M.[3], Larry Duncan W.[4], Ioannis Giannakou[1]

[1] *Laboratory of Agriculture Zoology and Entomology, Agricultural University of Athens, Athens, Greece*; [2] *Gulf Coast Research and Education Center, University of Florida, Wimauma, FL, USA*; [3] *Department of Phytopathology, National Research Center El-Tahrir St., Dokki, Giza, Egypt*; [4] *Department of Entomology and Nematology/ CREC, University of Florida, Lake Alfred, Florida, USA*

Characterizing entomopathogenic nematode (EPN) biogeography requires fine-scale taxonomic resolution, because closely related EPN species can exhibit divergent phenotypes for properties such as habitat adaptation and host specificity. We used high throughput sequencing (HTS) in surveys to evaluate the suitability of conservation biological control (CBC) to exploit endemic EPN in citrus orchards in Greece, Egypt and Florida. We hypothesize that species diversity and abundance are related directly to the potential for naturally occurring EPN to regulate pests. We further speculate that EPN are especially abundant in Florida orchards based on numerous reports that varied in methodology. Here we characterize EPN communities in a humid-subtropical (Florida central ridge) and 5 Mediterranean (Epirus, Argos, and Chania in Greece, and Nile delta and reclaimed desert in Egypt) ecoregions, using the same sampling, extraction, and detection methods for each. We designed improved ITS2 rDNA primers to discriminate EPN species and we characterized the soil texture of Greek orchards and five properties of the Egyptian soils. Six species were detected in Egypt, with *H. indica, H. bacteriophora,* and *H. taysearae* found in both ecoregions, and *S. scapterisci, Steinernema* sp. and *S. glaseri* also in the Nile delta. *Steinernema feltiae, S. carpocapae* and *H. bacteriophora* were detected in Greek orchards and *S. glaseri, S. scapterisci, H. indica* and *H. zealandica* occurred in a temporal survey (96 samples) in a single Florida orchard. Species richness (S) and diversity (Shannon H') in Florida far exceeded that in Mediterranean ecoregions with Greece having the lowest values. Both ecological statistics in the Florida survey were congruent with estimates from a previous survey of 30 central ridge orchards that utilized qPCR rather than HTS. EPN were detected in 53% (reclaimed desert), 28% (Nile delta), 62% (Chania), 10% (Argos), 40% (Epirus) and 100% (Florida) of samples. Stepwise regression revealed that S and H' were inversely related ($P<0.01$) to soil pH in the Egyptian orchards. In Greece, *H. bacteriophora* occurrence and species richness increased with soil sand content. These surveys support previous reports that EPN in Florida citrus orchards are especially amenable to CBC tactics. They are a basis for additional study of citrus habitats to determine whether ecoregions are reliable predictors of EPN biogeography and potential biocontrol significance.

Keywords: High throughput sequencing - Entomopathogenic nematodes - Community structure - Species detection - Species diversity.

ORAL SESSION 5

Plant resistance and nematode virulence (continued)

How plants recognize nematodes: Signals and signalling.

Shahid Siddique[1] (ssiddique@ucdavis.edu), Florian Grundler[2]

[1] *UC Davis, Davis, CA, United States*; [2] *University of Bonn, Bonn, Germany*

The initial sensing of infection is mediated by germline encoded pattern recognition receptors (PRRs) in both plants and animals. Plant PRRs are primarily membrane-localized receptor-like kinases or receptor-like proteins that recognize highly conserved microbial- or damage-associated molecular patterns (MAMPs, DAMPs) leading to the activation of PRR-triggered immunity (PTI). BAK1 has been shown to act as a co-receptor for various PRRs, which typically detect proteinaceous ligands. There are well characterized examples of PAMPs and PRRs for bacterial, fungal and insect pathogens in plants. However, the details of PTI responses during plant–nematode interactions have remained relatively unexplored. Our recent work showed that nematode invasion of root is capable of inducing both PAMP and DAMP responses in a manner dependent on the common immune co-receptor BAK1. Using co-immunoprecipitation and mass spectrometry analyses, we identified several receptor-like kinases that dynamically associated with BAK1 upon infection. In my presentation, I will discuss the relevance of various signaling pathways in nematode-induced PTI.

Keywords: PTI - Defence - Virulence - DAMP - PRR.

Inducing plant resistance against parasitic nematodes: Preparing for battle.

Tina Kyndt (tina.kyndt@ugent.be), Willem Desmedt, Anikó Meijer, Jonas De Kesel, Mohammad Reza Atighi, Richard Raj Singh, Eva Degroote

Biotechnology, Ghent University, Ghent, Belgium

The use of induced resistance (IR) to control pests and diseases is increasingly investigated as a more sustainable alternative to conventional pesticides. We have recently discovered different molecules that induce resistance by systemic activation of the plant's innate immune response against parasitic nematodes. Reductions in gall formation and/or nematode reproduction of typically around 30-60% can be attained by IR, and broad-spectrum activity against multiple pathogens/pests has been observed in different host plants such as rice, tomato and sugar beet.

Although the specific target(s) of different IR-inducing compounds varies, typical hallmarks of the activated immune response involve the direct or primed accumulation of H2O2, phenylpropanoid biosynthesis, DNA hypomethylation and/or jasmonate accumulation in treated plants. Novel IR-establishing compounds can now be easily screened using our recently identified rice PTI marker genes, for which gene expression profiling on fully grown plants or in rice cell suspension cultures provides a versatile tool to predict IR induction in rice. The IR-induction by some of these compounds and upon root-knot nematode infection is potentially transmittable to the next generation of plants, and the possible epigenetic basis for this phenomenon is currently being investigated in our lab.

Keywords: Induced resistance - Priming - Epigenetics - Plant defence.

30 years of research on resistance against *Globodera pallida*: an overview.

Sylvain Fournet[1] (sylvain.fournet@inrae.fr), Marie-Claire Kerlan[2], Josselin Montarry[1], Eric Grenier[1]

[1] *IGEPP 1349 INRAe-Agrocampus Ouest-Université Rennes1, INRAe, Le Rheu, France*; [2] *IGEPP 1349 INRAe-Agrocampus Ouest-Université Rennes1, INRAe, Ploudaniel, France*

For 30 years INRA has been involved in the identification of genetic resistance to major potato pathogens such as the potato late blight *Phytophthora infestans* and the potato cyst nematode *Globodera pallida*. This long-time research plan is based on the association of skills in genetics and pathology with the use of both worldwide collections of wild *Solanum* species and genetic resources and of *G. pallida* populations coming from South America and Europe. This makes possible the understanding of genetic and dynamic processes involved in resistance breakdown in order to predict the effectiveness of resistance over time and space, but also the selection of innovative genetic material to be used against virulent or newly introduced populations.
In this talk we will present results about the adaptation of *G. pallida* to resistant potato cultivars, about life-history traits associated with the selection for virulence and finally about the efficiency of new original genetic construction to control virulent *G. pallida* lines or populations from Europe and worldwide.

Keywords: *Globodera* - Resistance - Virulence.

Resistance loci interactions discovery through comprehensive transcriptomics.

Peter DiGennaro (pdigennaro@ufl.edu), Weiming Hu, Vincent Colantonio

Entomology and Nematology, University of Florida, Gainesville, FL, United States

The general withdrawal of broad-spectrum fumigants has highlighted the need to develop alternative strategies to control plant parasitic nematode damage in agricultural settings. Plant resistance is a potent source of nematode control and while, in some cases, extensively deployed, the molecular aspects of plant resistance have been largely unexplored. Recent work on soybean resistance to cyst nematode has unveiled non-canonical (R-gene) based molecular underpinnings. We believe similar types of non-R-gene mediated resistance may explain other forms of incompatible plant parasitic nematode interactions. One such case is the identity and induced pathways of the *N* gene in pepper which confers resistance to a range of root-knot nematode species. Here, we combined the available genetic map and physical map data to estimate the location of the *N* gene in cultivated pepper. Detailed annotation of this 200kb region of the pepper genome revealed a putative glutamate carboxypeptidase. This extracellular protease may have role in similar pathways to those induced during soybean resistance to cyst nematode. To explore the possibility of this region of the pepper genome containing a novel nematode resistance pathway, we performed comprehensive transcriptomic analyses. Using the resistant pepper cultivar Charleston Belle and its susceptible parent, Keystone Resistance Giant, we harvested whole roots for RNA resequencing at 1, 4, and 7 days post inoculation with root-knot nematode *M. incognita.* Comparative transcriptomics between timepoints, pepper genotypes and uninfected tissues included over 400 million reads and revealed insights into the molecular underpinnings of compatible and incompatible pepper-nematode interactions. In close proximity are a number of other known resistance loci, including *Me*, conferring resistance to *M. enterolobii*. Our data also reveals how nematode transcriptomes are modulated based on differences in hosts and potential resistance.

Keywords: Resistance - Virulence - Root-knot - Pepper - RNA-seq.

Dehydroascorbate activates induced resistance in rice against root-knot nematode *Meloidogyne graminicola.*

Satish Namdeo Chavan[1, 2] (satish.namdeochavan@ugent.be), Jonas De Kesel[1], Willem Desmedt[1], Eva Degroote[1], Richard Raj Singh[1], Nguyen Thu Giang[1], Kristof Demeestere[3], Tim De Meyer[4], Tina Kyndt[1]

[1] *Department of Biotechnology, Faculty of Bioscience Engineering, Ghent University, Ghent, Belgium*; [2] *ICAR-Indian Institute of Rice Research, Hyderabad, India*; [3] *Department of Green Chemistry and Technology, Faculty of Bioscience Engineering, Ghent University, Belgium*; [4] *Department of Data Analysis and Mathematical Modelling, Ghent University, Ghent, Belgium*

Rice is one of the most important food crops globally. The root-knot nematode *Meloidogyne graminicola* is one of the most devastating rice pests and causes crop loss in all types of rice ecosystems [1]. Induced resistance is a promising approach for the management of pests and diseases in view of environment-friendly control strategies. Induced resistance prepares the plants for future attacks from pests and diseases, is often long-lasting, and can even be inherited to its offspring. Exogenous foliar application of dehydroascorbate (DHA), the reversibly oxidized form of ascorbic acid (vitamin C), was found to activate systemic induced resistance in rice against the root-knot nematode *M. graminicola*. Detailed transcriptome analysis done on roots of rice plants showed an early and robust transcriptional response upon foliar DHA treatment, with induction of several genes related to plant stress responses, immunity, hormones, antioxidant activity, and secondary metabolism. Through the quantitative and qualitative evaluation of hydrogen peroxide (H_2O_2) levels, we confirmed the appearance of a reactive oxygen species (ROS) burst (a primary feature of induced resistance) in rice upon DHA treatment. The role of H_2O_2 in DHA-induced resistance was further confirmed using chemical ROS inhibitors or scavengers. Hormone measurements in DHA-treated rice plants showed a significant systemic accumulation of the defence hormone salicylic acid (SA). The role of the SA pathway in DHA-based IR was confirmed by nematode infection experiments using an SA-signaling deficient *WRKY45*-RNAi line and qRT-PCR on SA marker genes. We are currently investigating if DHA is able to induce transgenerational acquired resistance (TAR) in rice and will try to unravel the mechanisms underlying DHA-TAR, with a first focus on global DNA methylation. Our results collectively revealed that DHA is a potent inducer of plant defence pathways in rice and can be used as a novel control strategy against nematode infection in rice.

Keywords: Dehydroascorbate - Induced resistance - Rice - *Meloidogyne graminicola*.

References:

- [1] Mantelin et al., 2017. Mol. Plant Pathol. 18(1): 3-15.

Comparative transcriptome analysis reveals the specific activation of defense pathways against *Globodera pallida* in Gpa2 resistant potato roots.

Qi Zheng (qi.zheng@wur.nl), André Bertran, Anouk Brand, Casper C. van Schaik, Stefan van de Ruitenbeek, Geert Smant, Aska Goverse, Mark G. Sterken

Laboratory of Nematology, Wageningen University, Wageningen, Netherlands

Cyst nematodes are considered a dominant threat to yield for a wide range of major food crops. Current control strategies mainly depend on crop rotation and using resistant cultivars. Various crops exhibit single dominant resistance (*R*) genes able to activate effective host-specific resistance to certain cyst nematode species and/or populations. An example is the potato *R* gene *Gpa2*, which confers resistance against the potato cyst nematode (PCN) *Globodera pallida* population D383. Activation of Gpa2 results in a delayed resistance response which is characterized by a layer of necrotic cells formed around the developing nematode feeding structure. However, knowledge about the Gpa2-induced defense pathways is still lacking. Here, we uncover the transcriptional changes and gene-expression network induced upon *Gpa2* activation in potato roots infected with *G. pallida*. To this end, resistant potato genotype SH was infected with the Gpa2 avirulent population D383 and virulent population Rookmaker. Infected root segments were harvested at 3 and 6 dpi for RNA sequencing. Comparative transcriptomics revealed a total of 1,743 differentially expressed genes (DEGs) upon nematode infection, of which 559 DEGs were specifically regulated in response to D383 infection. D383-specific DEGs associated with Gpa2-mediated defence mainly involve calcium binding activity, salicylic acid biosynthesis, and systemic acquired resistance. These data reveal that cyst nematode resistance in potato roots depends on conserved downstream signalling pathways involved in plant immunity, which are also required for *R* genes-mediated resistance against various other pathogens with different lifestyles.

Keywords: *Globodera pallida* - Gpa2 - Defense response - Transcriptome - NB-LRR.

S5-PF1

Investigation of resistance against *Ditylenchus dipsaci* on sugar beet.

Alan Storelli[1] (alan.storelli@bfh.ch), Andreas Keiser[1], Sebastian Kiewnick[2], Matthias Daub[2], Mario Schumann[3], Werner Beyer[3], Michael Rostás[4], Anne-Kathrin Mahlein[5]

[1] *School of Agricultural, Forest and Food Sciences HAFL, Bern University of Applied Sciences, Zollikofen, Switzerland*; [2] *Institute for Plant Protection in Field Crops and Grassland, Julius Kuehn Institute, Braunschweig, Germany*; [3] *KWS SAAT SE & Co. KGaA, Einbeck, Germany*; [4] *Departement for Crop Sciences, Georg-August-Universistät Göttingen, Göttingen, Germany*; [5] *Institute for Sugar Beet Research IfZ, Göttingen, Germany*

Ditylenchus dipsaci has emerged as an economically threatening pest in the European sugar beet (*Beta vulgaris* L.) production. As no effective controls are available to reduce *D. dipsaci* populations in the field, the development of resistant cultivars, therefore, is a major challenge to maintain the sustainability of sugar production in the affected regions. The Ph.D. project aimed to investigate the resistance of sugar beet genotypes against *D. dipsaci*. Before investigating the interaction between sugar beet and the nematode, the development of an *in vivo* test system was required. It aimed to replace above-ground *D. dipsaci* inoculation with a soil inoculation more closely related to field conditions. The most suitable inoculation time point, inoculum level, and positioning on sugar beets, as well as rearing process on carrots, were determined.

To find potentially resistant sugar beet restricting reproduction and penetration of *D. dipsaci*, *in vivo* bioassays were carried out with 15 prebreeding populations and 79 breeding lines. It could be demonstrated that none of the genotypes showed complete resistance towards *D. dipsaci*. However, a high variation of the penetration rate by *D. dipsaci* was observed among the genotypes. They also responded differently to the fresh biomass reduction caused by the nematode combined with soil-borne pathogens.

Finally, virulence and pathogenicity of four *D. dipsaci* populations were investigated under *in vivo* conditions. No difference was found in *D. dipsaci* penetration rate into sugar beet seedlings. However, Seeland (CH) population showed a significantly higher reproduction on sugar beets than the others populations, validating observations obtained in microplot experiments.

Keywords: Breeding line - Nematode population - Pre-breeding population - Resistance breeding - Test system.

S5-PF2

Prospective identification through DNA-capture technologies of a rice resistance gene to control *Meloidogyne graminicola.*

Sophie Mantelin[1] (sophie.mantelin@hutton.ac.uk), Hue Nguyen Thi[2], Miles Armstrong[3], Hai Ho-Bich[4], Lizhong Xiong[5], Mathias Lorieux[6], John Jones[1], Ingo Hein[1], Stéphane Bellafiore[7]

[1] *Cell & Molecular Sciences, The James Hutton Institute, Dundee, United Kingdom*; [2] *IRD - LMI RICE, University of Science and Technology of Hanoi, Hanoi, Vietnam*; [3] *Division of Plant Sciences, University of Dundee, Dundee, United Kingdom*; [4] *IOIT/University of Science and Technology of Hanoi, VAST and UMMISCO, IRD, Hanoi, Vietnam*; [5] *National Key Laboratory of Crop Genetic Improvement, Huazhong Agicultural University, Wuhan, Hubei, China*; [6] *UMR DIADE, IRD, Montpellier, France*; [7] *UMR IPME, IRD, Montpellier, France*

An emerging pest in Europe, the root-knot nematode *Meloidogyne graminicola,* is already considered a major threat to rice agriculture worldwide, particularly in Asia where changes in agricultural practices in response to environmental and socioeconomic conditions have led to a dramatic increase in the nematode disease pressure. Breeding resistant rice varieties that could be used as a component of an effective integrated pest management strategy is essential to mitigate the spread of the disease and to promote sustainable control. Resistance to *M. graminicola* was identified in the Chinese rice cultivar Zhonghua11 (Zh11) that demonstrates the hallmarks of a typical nucleotide-binding domain leucine-rich repeat (NB-LRR)-containing type of disease resistance gene: dominant resistance, as determined by genetic characterisation of crosses with susceptible rice IR64 and Nipponbare, accompanied by a hypersensitive reaction that occurs as soon as two days after penetration and impairs the development of the nematode feeding site. Therefore, a targeted resistance gene-enrichment sequencing (RenSeq) approach was developed in rice to accelerate the mapping of this new resistance. Combined with bulk-segregant analysis of F2 progenies, RenSeq identified two resistance loci, including a major resistance island at the bottom of the rice chromosome 11. RenSeq-derived single nucleotide polymorphisms (SNPs) within the target regions were converted into allele-specific PCR-based KASP markers to screen the recombinants and further defined the position of the resistance, possibly leading to the identification of candidate resistance genes. In addition, a complementary targeted-enrichment approach in rice has been undertaken that is based on generic-mapping enrichment sequencing (GenSeq), which relies on single/low-copy number genes. The prospective results should strengthen our confidence in the resistance mapping, possibly help refining the current 3.19-Mb interval that carries the resistance locus and extend the pool of genetic markers linked to the resistance gene. Fine mapping of the gene and development of molecular markers will provide new tools for introgressing the Zh11 resistance in rice cultivars favoured in Asia.

Keywords: *Meloidogyne graminicola* - Rice - Resistance - Targeted resistance gene-enrichment sequencing (RenSeq) - NB-LRR.

S5-PF3

Host status of Crop plants to *Meloidogyne enterolobii* populations.

Hemanth Kumar Koniganahalli Gopal[1] (hemanth.gopal@julius-Kuehn.de), Sebastian Kiewnick[1], Etienne G J Danchin[2]

[1] *Institute for Plant Protection in Field Crops and Grassland, Julius Kühn Institute, Braunschweig, Lower-Saxony, Germany*; [2] *Institut Sophia-Agrobiotech, INRAe: French National Institute for Agriculture, Food and Environment, Paris, France*

Meloidogyne enterolobii is a polyphagous and highly pathogenic root-knot nematode and considered as economically very important due to its ability to develop and reproduce on host plants carrying resistance to other other major root-knot nematodes. To mitigate its further spread and develop control options, it is critical to understand the host range of *M.enterolobii* and its potential to adapt to new hosts. In the project "AEGONE" funded by the German Science Foundation (DFG project No 431627824) and ANR (FR) 20 crop species previously classified as non-, minor- or major-hosts were challenged under greenhouse conditions with 10 *M. enterolobii* populations (collected from different geographic areas and hosts) reared as single egg mass lines. Based on the determined reproduction factor (RF), crops were categorized as good-host (RF is ≥1), poor-host (RF between 0 and 1) or non-host (RF=0). Ten out of 20 species (cucumber, eggplant, pepper, tobacco, tomato, beans, melon, sugar beet, yellow mustard, and potato) showed RF values ≥ 1 for all 10 populations. Contrary to recent studies, maize, sunflower and soybean were good-hosts for only for 2, 2 and 8 populations, respectively. Although several reports suggested roses as minor-host, no reproduction was observed for all populations tested. Furthermore, cotton was a non-host for two populations. In addition, fodder radish and phacelia both showed RF values ≥1 for 7 populations, whereas the RF values for yellow mustard were > 1 for all 10 populations and consequently identified as a good-host.

Based on the results it was demonstrated that several populations of *M. enterolobii* are capable of reproducing on crops previously reported as non-host. Therefore, further studies are underway to investigate the potential of *M. enterolobii* to adapt to initially poor hosts, the related costs of fitness and to determine variations on the genome level with differences in host compatibilities.

ORAL SESSION 6

Phylogenetics/Phylogenomics: the latest updates on the Phylum Nematoda

Evolution and diversification of plant and animal parasitism within the phylum Nematoda.

Johannes Helder[1] (Hans.Helder@wur.nl), Martijn Holterman[1], Sven Van den Elsen[1], Geert Smant[1], Stefan Geisen[1], Michaela Schratzberger[2]

[1] *Laboratory of Nematology, Wageningen University, Wageningen, The Netherlands*; [2] *Fisheries and Aquaculture Science, Centre for Environment, Lowestoft, United Kingdom*

Parasitism is a popular life-style among members of the phylum Nematoda. Around 46% of the described nematode species uses either a plant or an animal as a primary food source. Animal parasites are almost equally distributed over terrestrial and marine habitats (both harbour » 4,000 species), whereas plant-parasitic nematodes (about 4,100 species) are almost exclusively present in soil ecosystems.

Objective:

Our objective was to use molecular phylogenetics to decipher patterns of evolution and diversification among plant and animal parasitic nematodes.

Method:

An alignment comprising over 5,000 nearly full-length SSU rDNA sequences (each appr 1,700 bp) with a fairly good coverage of all extant nematode families allowed us to pinpoint patterns with regard to the appearance of animal and plant parasitism.

Results and conclusions:

Whereas plant parasitism arose in Clades 1, 2, 10 and 12, vertebrate-parasitic nematodes evolved and diverged exclusively in Clades 2, 8, 9, 10. So only two clades, Clades 2 and 10, harbour both plant and animal parasites. Within Clade 2, vertebrate parasitic orders Trichinellida and Dioctophymatida are positioned sister to the predacious nematode-dominated order Mononchida that together diverged from the common ancestor of all Dorylaimia. Clade 10 is dominated by fungivores and facultative plant parasites, and within this clade the animal-parasitic family Strongyloididae arose. Insights will be given in the diversification and distribution of increasing levels of intimacy with regard to parasite-host interaction; transition from ecto- to endoparasitism, and the transitions towards feeding sites with increasing levels of complexity and dependency (from the parasite point of view).

Keywords: Plant parasitism - Animal parasitism - Habitat transitions - SSU rDNA.

Nematode phylogenomics – how far did we progress?

Oleksandr Holovachov[1] (oleksandr.holovachov@nrm.se), Mohammed Ahmed[1], Ashleigh B. Smythe[2], Kevin M. Kocot[3]

[1] *Zoology, Swedish Museum of Natural History, Stockholm, Sweden*; [2] *Virginia Military Institute, Lexington, United States*; [3] *The University of Alabama, Tuscaloosa, United States*

The first phylogenomic analysis of the phylum Nematoda that was published in 2004 used EST data from 30 different nematode species. Since then, the amount of publicly available transcriptomic and genomic datasets has grown to nearly 230 species. However, the quality of available data is variable, ranging from small EST datasets to complete genomes. Genomes and transcriptomes with contamination from non-target organisms is another problem. Here we summarise publicly available genomic data suitable for phylogenetic studies and highlight needed improvements to data generation and analysis. The newly generated nematode phylogenomic tree [1] is unsurprisingly similar to the phylogenies based solely on 18S rDNA, not only in general topology, but in sampling bias towards economically important and model species. So far, genomic sequencing has been largely focused on selected animal parasitic (e.g. *Trichinella*), plant parasitic (*Meloidogyne*) and model rhabditid (*Caenorhabditis* and *Pristionchus*) species. Thus, the quality of genome assemblies and annotations for these groups are generally high. On the other hand, neglected clades remain underrepresented and undervalued, partly because many of the available genomic datasets for these species are of lower quality. These neglected taxa are not limited to free-living nematodes – huge diversity of parasitic species are also not sequenced. Nonetheless, there are interesting evolutionary and biological questions that can be addressed by studying non-model organisms, among them the origin of parasitism in aquatic environments, evolution of ectoparasitism, evolution of parthenogenesis and hermaphroditism, adaptation to life in extreme environments, cryobiosis, anhydrobiosis. Generation of high quality genomes and transcriptomes from understudied groups of nematodes will become more and more important to improve understanding of the origin of nematodes, disentangle the evolutionary relationships among numerous animal and plant parasitic lineages, understand adaptations that allowed nematodes to inhabit almost every possible habitat on Earth, and develop a robust and comprehensive phylogenetic framework for comparative studies.

Keywords: Genome - Transcriptome - Phylogeny - Phylum - Evolution.

References:

- [1] Ahmed et al., 2022. Front. Ecol. Evol. 9: 769565. doi.org/10.3389/fevo.2021.769565

Phylogeny and systematics of Aphelenchoididae: overview and problems.

Natsumi Kanzaki[1] (nkanzaki@ffpri.affrc.go.jp), Robin Giblin-Davis[2]

[1] *Kansai Research Center, Forestry and Forest Products Res. Inst., Kyoto, Japan*; [2] *Ft. Lauderdale REC, University of Florida, Davie, United States*

The family Aphelenchoididae is a group of highly divergent nematodes hypothesized to have evolved from soil-dwelling fungal feeders. The family is currently separated into five molecularly inferred phylogenetic groupings which generally correspond to the traditional classification system. Clade 1 is equivalent to the subfamily Tylaphelenchinae which is found in soil and dead wood, and often associated with termites. Clade 2 is equivalent to Aphelenchoidinae and is further separated into two subclades (2a and 2b). These subclades contain highly specialized and morphologically convergent fig-associated groups that arose independently within each subclade. Clade 4 is equivalent to the Parasitaphelenchinae, with many of its members being fungal feeders that are phoretically associated with various groups of beetles (bark beetles, ambrosia beetles, weevils, longhorn beetles, nitidulid beetles, etc.). Contrastingly, clade 3 consists of five predatory and insect parasitic subfamilies, Acugutturinae, Anomyctinae, Ektaphelenchinae, Entaphelenchinae, and Seinurinae. The phylogenetic relationships among these subfamilies are unclear because of the shortage of molecular sequences for taxonomically characterized members from the Acugutturinae, Entaphelenchinae, Seinurinae and paraphyly in the Ektaphelenchnae. Currently, the taxonomic system (systematics) of clades 1 and 4 have been updated and morphological and biological characters of the species are generally congruent with their inferred molecular phylogenetic relationships. Unfortunately, the characterization of clade 2 species suffers from too many old and unclear descriptions without any associated molecular data. The subclades and species of clade 2 require re-isolation and recharacterization based on accepted molecular barcodes and re-observed morphological traits using type-cultured materials. The majority of clade 3 species are unculturable or fastidious because of their predatory and/or insect parasitic life histories. Thus, clade 3 representatives must be re-isolated alive (and cultured, if possible) and re-observed to morphologically and phylogenetically modernize the characterization and systematics of its members.

Keywords: Aphelenchoididae - Systematics - Paraphyly - Parasite - Predator.

An update to the phylogeny of Aphelenchoidea; Ektaphelenchinae, Seinurinae and Tylaphelenchinae, as case studies.

Majid Pedram[1] (majid.pedram@modares.ac.ir), Farahnaz Jahanshahi Afshar[2]

[1] *Faculty of Agriculture, Tarbiat Modares University, Tehran, Iran*; [2] *Iranian Research Institute of Plant Protection, AREEO, Tehran, Iran*

Molecular data and corresponding phylogenetic analyses are greatly helping to resolve the taxonomy of Aphelenchoidea. The conserved gross morphology of several aphelench genera is the first problem for their taxonomy, and absense of molecular data for most early taxa is the second. The subfamily Ektaphelenchinae includes predatory or insect related forms, at first delimited by having non-functional rectum and anus. The increasing molecular data show the three genera *Ektaphelenchus*, *Ektaphelenchoides* and *Deviburs aphelenchus* are not monophyletic, while, *Cryptaphelenchus* is monophyletic. The increasing molecular data for *Seinura* yielded new observations on its phylogeny. Until recently a non-monophyletic nature was assumed for *Seinura*, while we showed its currently sequenced species form a maximally supported clade. Tylaphelenchinae, the third studied subfamily, was established by two genera *Tylaphelenchus* and *Pseudaphelenchus*, and later updated by including *Albiziaphelenchus* and *Basilaphelenchus* [1]. The latter genus has a conserved general morphology with *Aphelenchoides*, still inside the clade of the subfamily Tylaphelenchinae, corroborating the usefulness of reverse taxonomy for studying of tylaphelenchs, and highlighting a need to sequence two other genera *Tylaphelenchus* and *Albiziaphelenchus*. The updated phylogenies of three aforementioned subfamilies using maximal number of species are presented and discussed in this study.

Keywords: *Albiziaphelenchus*, Aphelenchoides - *Basilaphelenchus, Deviburs aphelenchus* - *Ektaphelenchus*, Ektaphelenchoides, phylogeny - Reverse taxonomy, *Seinura*, taxonomy.

References:

- [1] Pedram et al., 2018. Nematology. 20: 567-582.

High-quality genome assembly of an emerging root-knot nematode species, *Meloidogyne luci.*

Nik Susič[1] (nik.susic@kis.si), Georgios D. Koutsovoulos[2], Cristian Riccio[3], Etienne G. J. Danchin[2], Mark L. Blaxter[4], David H. Lunt[5], Polona Strajnar[1], Saša Širca[1], Gregor Urek[1], Barbara Gerič Stare[1]

[1] *Plant Protection Department, Agricultural Institute of Slovenia, Ljubljana, Slovenia*; [2] *Institut Sophia Agrobiotech, Sophia-Antipolis, France*; [3] *Wellcome Sanger Institute, Hinxton, United Kingdom*; [4] *Institute of Evolutionary Biology, Edinburgh, Scotland, United Kingdom*; [5] *University of Hull, Kingston-upon-Hull, United Kingdom*

Root-knot nematode (RKN) *Meloidogyne luci* can parasitize a wide range of crop plants and was recently reported from Argentina, Bolivia, Brazil, Chile, Ecuador, Greece, Guatemala, Iran, Italy, Portugal, Slovenia and Turkey. *M. luci* is one of the tropical species belonging to *Meloidogyne* Clade I and reproduces asexually by mitotic parthenogenesis. Several genomes of Clade I tropical *Meloidogyne* spp. have been sequenced and have revealed themselves to be complex allopolyploids with heterozygous duplicated genome regions and abundant transposable elements. Previous genome assemblies largely relied on short-read next-generation sequencing which limited the contiguity of the assemblies. We used long-read Pacific Biosciences Sequel and short-read Illumina HiSeqX sequencing data to produce a high-quality *M. luci* genome assembly. The *M. luci* population was isolated from tomato plants grown commercially in Šmartno, Slovenia. Long-read data were assembled with HGAP4 pipeline and polished using trimmed Illumina data. The 209.2 Mb *M. luci* genome assembly consists of 327 contigs with a minimum contig length of 10,147 bp and N50 of 1,711,905 bp. The genome is estimated to be triploid (AAB). The assembly is currently the most contiguous RKN assembly publicly available with estimated coding space coverage of 95.2% based on Core Eukaryotic Genes Mapping Approach (CEGMA). The polished assembly was 88% complete based on the eukaryote set (n=303) of Benchmarking Universal Single-Copy Orthologs (BUSCO). The assembly of *M. luci* can now be used to determine the correct phylogenetic position of the clade, identification of genetic changes related to the origins of virulence, and in the study of evolutionary history of this organism.

Keywords: Genome - *Meloidogyne luci* - Root-knot nematode.

Phylogeography, phylogeny and DNA barcoding of the cyst nematodes.

Sergei Subbotin (sergei.a.subbotin@gmal.com)

Plant Pest Diagnostic Center, CDFA, Sacramento, CA, USA

Cyst nematodes cause significant economic losses on different crops around the world. In our study we provide comprehensive phylogenetic analyses of ITS rRNA and mtDNA gene sequences of cyst nematodes from the *Schachtii, Goettingiana, Humuli* groups of the genus *Heterodera* and the genera *Globodera, Punctodera* and *Cactodera* using Bayesian inference, maximum likelihood and statistical parsimony. Several hundreds of new COI, cytb and ITS rRNA gene sequences were obtained from more than 50 species collected from different countries and continents. Based on the results of phylogeographical analysis and age estimation of clades with a molecular clock approach, it is hypothesised that cyst nematodes originated and diversified from several centres of speciation located in: Irano-Anatolian region, Western Cape in South Africa, Andean region of South America, Sierra Madre of Mexico and several others and then dispersed across the world from these regions. Analysis of phylogenetic relationships of populations of potato cyst nematode species and representatives of the *Schachtii* group of the *Heterodera* the revealed incongruence in topology between networks inferred from mtDNA or ITS rRNA genes, which might be an indication of possible recombination and selective introgression events through gene flow between previously isolated populations. This puts some limitations on the use of the mtDNA marker as universal DNA barcoding identifier for cyst nematodes

Keywords: mtDNA - *Heterodera* - *Globodera*.

S6-PF1

Genetic and phylogenetic characterization of different populations of *Meloidogyne izalcoensis*, a new species from coffee in Brazil.

Marcilene Santos[1] (lenebio@gmail.com), Sheila Almeida[1], Vanessa Mattos[1], Marina Carneiro[1], Philippe Castagnone-Sereno[2], Regina Carneiro[1]

[1] *Nematology, Embrapa Recurs Genéticos e Biotecnologia, Brasília, Df, Brazil*; [2] *Nematology, INRAe, Université Côte d'Azur, CNRS, ISA, Sophia Antipolis, France*

A new species *Meloidogyne izalcoensis* was detected on coffee in Minas Gerais state, Brazil in 2019 [1]. This species was first described in El Salvador by Carneiro *et al.* (2005) [2] and recently detected in Africa. This species causes small galls with external egg masses and severe destruction on coffee roots, showing different root symptoms when compared with other *Meloidogyne* spp. from coffee. Although already detected in two continents, there are no reports about the genetic diversity for this species. DNA-based tools and the characterization of different genetic regions have been extensively employed to evaluate the genetic variability and phylogenic relationships of *Meloidogyne* spp. related to coffee. The objectives of this study were to assess the genetic variability of five *M. izalcoensis* populations from different geographic areas in the world (El Salvador, Kenya, Tanzania, Vietnam and Brazil) and their phylogenetic relationships inferred from distinct regions of ribosomal DNA (rDNA), mitochondrial gene cytochrome *c* oxidase II (COII) and nuclear protein coding gene HSP90. All populations were identified by esterase phenotype and SCAR-specific markers. Based on RAPD and AFLP markers, a low intraspecific variability was detected among *M. izalcoensis* populations from Africa (Kenya and Tanzania), Vietnam and Brazil, with 91% bootstrap, except for the population from El Salvador that showed a few genetic differences compared to the other populations. Phylogenetically, all populations of *M. izalcoensis* from different locations (El Salvador, Kenya, Tanzania, Vietnam and Brazil) grouped with 90% and 69% bootstrap for COII and HSP90 regions, respectively, indicating that these markers are highly conserved for the species. In addition, both markers allowed the separation between *M. izalcoensis* populations and other important coffee *Meloidogyne* species, including *M. exigua*, *M. paranaensis*, *M. incognita*, *M. arabacida*and *M. lopezi*. However, in the analyses obtained from the D2D3 regions of the 28S and intergenic ITS1-5.8S-ITS2 gene from the rDNA, *M. izalcoensis* populations did not form cohesive clusters, demonstrating a phylogenetic limitation of these markers.

Keywords: Root-knot nematode - Coffea arabica - Phylogeny - RAPD/AFLP - COII.

References:

- [1] Stefanelo et al. 2019. Tropical Plant Pathology 44:209–212.
- [2] Carneiro et al. 2005. Nematology 7: 819-832.

S6-PF2

Phylogeography of *Ditylenchus gallaeformans.*

Samara A. Oliveira[1] (solivei@g.clemson.edu), Saara DeWalt[2], Paula Agudelo[1]

[1] *Nematology, Plant and Environmental Sci, Clemson University, Clemson, South Carolina, United States*; [2] *Biological Sciences, Clemson University, Clemson, South Carolina, United States*

Ditylenchus gallaeformans is a migratory ectoparasite nematode that induces galls in the stem, leaves and flowers of several Melastomataceae species, including invasive species from *Miconia* and *Clidemia* genera. This nematode has been considered as a possible biological control agent against these invasive plant species. However, the culture of this nematode in artificial conditions is challenging, and has not been established yet. For this reason, it is important to focus studies on the biology and geographic ditribution of *D. gallaeformans* to better understand the nematode's relation with the hosts and environment. A more comprehensive understanding of the phylogeography of *D. gallaeformans* may provide insight into the coevolution between nematode and host, nematode phylogeny and genetic differences among the populations, which may have influence on host preference, virulence and pathogenesis. *D. gallaeformans* populations used in this study were collected from Costa Rica, Dominica and Trinidad. DNA was extracted from individual nematodes and the COI region was amplified and sequenced. Phylogenetic analysis was performed using maximum-likelihood (ML) and Bayesian inference (BI), and haplotype networks were constructed for each population according to geographical location and host species. Phylogenetic reconstruction using the COI sequences yielded trees with similar topologies and revealed four major clades by location. Haplotype networks for COI were congruent with the phylogenetic reconstructions, showing segregation of the mitochondrial haplotypes according to locality and not by host species. The results show us that our *Ditylenchus gallaeformans* samples are monophyletic, and that the *Ditylenchus* group is paraphyletic. The haplotype networks revealed that geographic location is the trait responsible for the population structure observed, and the host species have no influence on the haplotypes.

Keywords: Phylogeography - Biological control - Invasive plants - Haplotype.

S6-PF3

Molecular identification and phylogenetic diversity of cereal cyst nematode (*Heterodera* spp.) populations from Algeria.

Mehalaine Khawla[1] (khemayna@hotmail.com), Imren mustafa[2], Dababat Abdelfattah[3]

[1] *Zoology, Ensa (High school of agronomy), Algiers, Algeria*; [2] *Bolu Abant Izzet Baysal University, Bolu, Turkey*; [3] *Cymmit, Bolu, Turkey*

Cereal cyst nematodes (CCNs), members of the genus of *Heterodera*, are the most damaging plant–parasitic nematodes on wheat, causing severe global economic losses. In 2018, a survey was conducted to investigate parasitic nematodes on wheat in several wheat-growing regions of Algeria. The study revealed that 41.6% of the investigated wheat fields were infested with cereal cyst nematodes. Forty-eight CCN populations from twenty-two locations were obtained and morphologically identified. To verify the morphological identification, the internal transcribed spacer (ITS) of rDNA of the nematodes was sequenced and analyzed using BLASTn searches. Consequently, all populations were classified into *Heterodera avenae*, *H. hordecalis*, *H. carotae*, and *H. cruciferae*. *Heterodera carotae* and *H. cruciferae* were reported for the first time from Algeria and detected from two and three locations, respectively. *Heterodera carotae* and *H. cruciferae* are closely related to the populations from Italy and the Netherlands, respectively. *Heterodera avenae* and *H. hordecalis* were found in four and ten localities, respectively. The Algerian population of *H. avenae* showed high similarity with that of the Spanish population, whereas the representatives of the *H. hordecalis* population were highly similar to those of an Israeli population. Due to the variation among the Algerian populations of *H. avenae* and *H. hordecalis*, they can be assumed to be multi-introduced populations. *Heterodera carotae* and *H. cruciferae* have formed a well-supported cluster with the corresponding populations.

Keywords: Cereal cyst nematodes - *Heterodera* spp. - species identification.

ORAL SESSION 7

Biodiversity of aquatic nematodes

Effects of duration and frequency of non-flow periods on stream-dwelling nematode communities.

Nabil Majdi[1] (nabil.majdi@uni-bielefeld.de), Miriam Colls[2], Linette Weiss[1], Vicenç Acuña[3], Sergi Sabater[3], Walter Traunspurger[1]

[1] *Animal Ecology, Bielefeld University, Bielefeld, Germany*; [2] *Catalan Institute of Water Research, Girona, Spain*; [3] *GRECO, ICRA, University of Girona, Girona, Spain*

Streams may experience non-flow periods during which their resident fauna may be altered. The frequency and duration of those non-flow periods is expected to increase with climate change, especially in Mediterranean areas. The meiofauna (microscopic benthic invertebrates comprising nematodes) are intermediaries in food webs and thereby deemed as important actors of stream ecosystems, however the effects of the non-flow period on their taxonomic and functional diversity is barely known. Here, we monitored the hydrological, physical and chemical variables of 22 streams spread across 5 basins of the north-eastern Iberian Peninsula over a period of 250 days, and we sampled the sediment-dwelling meiofauna. We aimed to define how much the temporal components of non-flow periods (i.e. duration and frequency) as well as other environmental variables could affect the structure of meiofaunal communities. Overall, meiofauna abundance was among the highest ever reported for a streambed, nematodes representing >50% of individuals counted. We identified a total of 113 different nematode species, from which 55 were specific to intermittent streams, whereas only 5 species were specific to permanent streams. The nematode community in intermittent streams was characterized by a smaller proportion of bacterivores and a higher proportion of omnivorous and fungivorous species, thus resembling more the functional structure encountered in forest floors. A variance partitioning analysis showed that the duration and frequency of non-flow had a strong structuring effect on nematode assemblages, superior to the effect of sediment granulometry and nutrient availability. Interestingly, most meiofaunal groups as well as nematode species correlated positively with the frequency of non-flow periods. However, prolonged periods of continuous flow, as well as prolonged periods of non-flow, decreased abundances of most taxa; with the exception of tardigrades, which thrived-well under prolonged periods of non-flow. Our results thus suggest that increased duration of flow or non-flow phases could have a rather deleterious effect on the diversity of streambed fauna. In contrast, frequent alternance of flow and non-flow seem to favor the settlement of a luxuriant nematofauna at the crossroads between aquatic and terrestrial ecosystems.

Keywords: Community Ecology - Functional Diversity - Intermittent Streams - Disturbance - Climate Change.

Taxonomic and functional diversity of marine nematodes in biomonitoring: reality or utopia?

Federica Semprucci (federica.semprucci@uniurb.it)

Dipartimento di Scienze Biomolecolari (DiSB), University of Urbino, Urbino, Italy

Research over the past fifty years illustrates the role of free-living nematodes in the marine ecosystem functioning. Their response to natural and anthropogenic environmental changes is important for seafloors worldwide, but when scientists and policy makers refer to biodiversity in the benthic domain, they take in consideration only angiosperms, macro-algae or macrofauna. Accordingly, the use of nematodes as bioindicators is still limited, despite their potential advantages in biomonitoring programs. This presentation deals with several study cases focused on the taxonomical and functional diversity of free-living nematodes from coastal to transitional habitats of a wide geographical span. The results reveal that taxonomical community structure documents perfectly the changes after anthropogenic disturbance, but Shannon-diversity and maturity indices are the most suitable available tools when the ecological quality has to be evaluated. These insights suggest that nematodes could be efficiently used as descriptors also according to the European directives (WFD, 2000/60/EC and MSFD 2008/56/EC).

Keywords: Nematodes - Marine sediments - Human impacts - Ecological quality assessment.

Community structure of deep-sea nematodes: a metabarcoding approach.

Lara Macheriotou[1] (lara.macheriotou@ugent.be), Sofie Derycke[2], Ann Vanreusel[1]

[1] *Marine Biology Research Group, Ghent University, Ghent, Belgium*; [2] *Aquatic Environment and Quality, Institute for Agricultural and Fisheries Research (ILVO), Oostende, Belgium*

Free-living marine nematodes dominate the meiofaunal component of deep-sea sediments across the globe (Vanreusel et al., 2010). The combined application of morphology- and DNA-based methodologies for the identification of nematodes over the years has revealed notable species richness in this phylum (Hauquier et al., 2018), the ubiquity and intraspecific morphological variability of specific genera (Miljutin & Miljutina, 2016; Vanreusel et al., 2010), as well instances of regionally-restricted taxa and low genetic divergence between geographically disparate areas (Bik et al., 2012). This talk will describe the findings of our exploration into the phylogenetic- and occurrence-based community structure of deep-sea nematodes using metabarcoding in two regions of contemporary and imminent anthropogenic exploitation: 1) the polymetallic nodule-rich abyssal plains of the Clarion-Clipperton Fracture Zone (CCFZ); an area targeted for industrial-scale deep-sea mining operations, and 2) the Mozambique Channel (MC); currently being explored for further hydrocarbon extraction. Ribosomal (18S) Amplicon Sequence Variants (ASVs) were predominantly phylogenetically clustered and aggregated (i.e., exhibit higher relatedness and co-occurrences than expected at random) in both the CCFZ and MC, suggesting that the environment is selecting for ecologically similar taxa and a suppressed role of competitive interspecific interactions. Most ASVs were highly localized, usually occurring in just a single replicate. The dominant genera *Acantholaimus*, *Desmoscolex* and *Halalaimus* were more often randomly structured, especially in the MC Pockmark fauna, an indication of the influence of neutral dynamics in this particular environment. Our results are discussed in the context of community ecology within the framework of the "Theory of ecological communities" (Vellend, 2010).

Keywords: Deep sea - Community structure - Metabarcoding - Polymetallic nodules - Pockmark.

References:

- Bik, H. M., Sung, W., De Ley, P., Baldwin, J. G., Sharma, J., Rocha-Olivares, A., & Thomas, W. K. (2012). Metagenetic community analysis of microbial eukaryotes illuminates biogeographic patterns in deep-sea and shallow water sediments. Molecular Ecology, 21(5), 1048–1059. https://doi.org/10.1111/j.1365-294X.2011.05297.x
- Hauquier, F., Macheriotou, L., Bezerra, T. N., Egho, G., Martínez Arbizu, P., & Vanreusel, A. (2018). Geographic distribution of free-living marine nematodes in the Clarion-Clipperton Zone: implications for future deep-sea mining scenarios. Biogeosciences Discussions, December, 1–29. https://doi.org/10.5194/bg-2018-492
- Miljutin, D. M., & Miljutina, M. A. (2016). Intraspecific variability of morphological characters in the species-rich deep-sea genus Acantholaimus Allgén, 1933 (Nematoda: Chromadoridae). Nematology, 18(4), 455–473. https://doi.org/10.1163/15685411-00002970
- Vanreusel, A., Fonseca, G., Danovaro, R., Da Silva, M. C., Esteves, A. M., Ferrero, T., Gad, G., Galtsova, V., Gambi, C., Da Fonsêca Genevois, V., Ingels, J., Ingole, B., Lampadariou, N., Merckx, B., Miljutin, D., Miljutina, M., Muthumbi, A., Netto, S., Portnova, D., … Galeron, J. (2010). The contribution of deep-sea macrohabitat heterogeneity to global nematode diversity. Marine Ecology, 31(1), 6.
- Vellend, M. (2010). Conceptual synthesis in community ecology. The Quarterly Review of Biology, 85(2), 183–206. https://doi.org/10.1017/CBO9781107415324.004

Resource diversity effects on cryptic marine nematode species: a multi-faceted approach.

Rodgee Mae Guden[1] (rodgeemae.guden@ugent.be), Derycke Sofie[2], Moens Tom[1]

[1] *Marine Biology Lab, Ghent University, Ghent, Belgium*; [2] *Aquatic Environment and Quality, Institute for Agricultural and Fisheries, Oostende, Belgium*

Resource partitioning is central to our understanding of the dynamics of species composition and coexistence in biological communities. Based on the principle of competitive exclusion, species occupying the same ecological niche cannot coexist for a long time due to strong interspecific competition for resources. The approach to reduce competition is to diversify niches, for instance, through resource partitioning. In addition, there can be tradeoffs between competitive and dispersal ability. Nematodes are one of the most abundant and species-rich taxa in marine environments. They can perceive many attractive and repellent chemical cues in the environment, among others emanating from their food, which can be important in their foraging, reproduction and survival strategies. Cryptic species, i.e., morphologically (nearly) identical but genetically different species, is prominent among nematodes which may have significant implications on biodiversity estimates and ecosystem functioning. Here, we demonstrate the effects of resource diversity on food preference of nematodes using taxis (i.e. a directed movement) to food assays, on population development, and on interspecific interactions of four cryptic bacterivore nematode species (Pm I-IV) of *Litoditis marina* which consistently co-occur in the field. Three resource (bacteria) diversity levels (low, medium, high) were used as treatments for the experiments. Results revealed differences in taxis-to-food between the cryptic species and between different levels of resource diversity: with the exception of Pm I, all cryptic species were significantly more attracted towards medium-diversity food than to higher- and lower-diversity food treatments. Higher population growth was also observed at medium and high food diversity treatments for each cryptic species, but their relative abundances changed in the presence of other species. Pm III clearly suffered from the interspecific interactions while these had a positive effect on Pm II. These results suggest that resource diversity has differential effects on taxis-to-food and on nematode population development and can alter the interspecific interactions among the cryptic species of *L. marina*, indicating that competitive equilibria between species are likely very context dependent.

Keywords: Cryptic species - Coexistence - Resource partitioning - Resource diversity - Nematodes.

Can we predict sediment quality with nematode metabarcoding?

Janina Schenk[1] (janina.schenk@uni-bielefeld.de), Nils Kleinbölting[2], Sebastian Höss[3], Marvin Brinke[4], Walter Traunspurger[1]

[1] *Animal Ecology, Bielefeld University, Bielefeld, Germany*; [2] *Center for Biotechnology, Bielefeld University, Bielefeld, Germany*; [3] *Ecossa, Starnberg, Germany*; [4] *Federal Institute of Hydrology, Koblenz, Germany*

Bioindication is a widely applied approach to evaluate ecological conditions, and several indices have been designed for this purpose. To assess the impact of pollution, especially polluted sediments, a pollution-sensitive index based on nematodes, one of the most abundant and species-rich groups of metazoa, was developed. The NemaSPEAR[%] index in its original form relies on the morphological inspection of nematode species. The present study evaluated a NemaSPEAR[%] index based on metabarcoding of nematode communities and tested the potential of fragments from the 28S rDNA, 18S rDNA. At the genus level, NemaSPEAR[%] values based on morphological data strongly correlated with those based on molecular values for both the 28S rDNA and the 18S rDNA gene fragments (R2= 0.86 and R2=0.74, respectively). Within the dominant genera (> 3%) identified by morphology, 68% were detected by at least one of these three molecular markers. At the species level, however, concordance was less pronounced, as there were several deviations of the molecular from the morphological data. These differences could mostly be attributed to shortcomings in the reference database used in the molecularly based assignments. The application of a morphologically based NemaSPEAR[%] at the genus-level was previously validated. Our pilot study shows that a molecularly based, genus-level NemaSPEAR[%] can be successfully applied to evaluate sediment quality.

Keywords: Bioindication - Metabarcoding - NemaSPEAR[%].

References:

- Höss, S., Claus, E., Von der Ohe, P C, Brinke, M., Güde, H., Heininger, P., Traunspurger, W., 2011. Nematode species at risk--a metric to assess pollution in soft sediments of freshwaters. Environ. Int. 37(5), 940–949. https://doi.org/10.1016/j.envint.2011.03.013
- Höss, S., Heininger, P., Claus, E., Möhlenkamp, C., Brinke, M., Traunspurger, W., 2017. Validating the NemaSPEAR[%]-index for assessing sediment quality regarding chemical-induced effects on benthic communities in rivers. Ecol. Indic. 73, 52–60. https://doi.org/10.1016/j.ecolind.2016.09.022

Aquatic nematodes as bio-indicators of crude oil water-soluble fractions (WSFs) toxicity: a microcosm approach.

Luana Monteiro[1] (luana.dacostamonteiro@ugent.be), Tom Moens[2], Walter Traunspurger[3]

[1] *Nematology + Marine Biology Research Units, Ghent University, Ghent, Belgium*; [2] *Marine Biology Research Unit, Ghent University, Ghent, Belgium*; [3] *Animal Ecology Department, Bielefeld University, Bielefeld, Germany*

The water-soluble fractions (WSFs) of oils contain highly toxic compounds, despite their low persistence in aquatic environments. Their effects may be instantaneous or delayed, provoking immediate mortality or sublethal effects, for instance on growth and reproduction. We investigated the effects of crude oil WSFs on both marine and freshwater (FW) nematode communities, in microcosm experiments lasting up to 15 weeks. Both experiments were performed simultaneously. Nematoda was the most abundant group, comprising ca. 90% of the meiofauna in both marine and freshwater sediments. Significant impacts on total nematode abundance, diversity and species composition only became apparent after 15 weeks, indicating that delayed effects are far more pronounced than instantaneous effects. In the short-term, significant oil WSF effects occurred in marine but not in freshwater microcosms: After one week, oil WSFs reduced the number of deposit- and epistrate feeders. In freshwater microcosms, significant effects on nematode feeding types were only detected by differences in the index of trophic diversity, but not by the multivariate comparison of feeding-type composition. Overall, sensitivity was species-specific in both marine and freshwater microcosms, with sometimes opposing responses between even congeneric species. Our results showed that oil WSFs can yield strong effects on both marine and freshwater nematode assemblages, and demonstrate the need to assess WSF effects on communities at the species level and over time periods well exceeding the residence time of WSF compounds in the environment.

Keywords: Oil pollution - Nematoda - Benthic communities - Direct toxicity - Experiments.

S7-PF1

Tobrilidae communities in western Nebraska sandhill lakes are driven by alkalinity and biotici interactions.

Kaitlin Gattoni[1] (kgattoni@ufl.edu), Abigail Borgmeier[2], J. Parr McQueen[1], Peter Mullin[2], Kirsten Powers[2], Thomas Powers[2], Dorota Porazinska[1], Eli Gendron[1]

[1] *Department of Entomology and Nematology, University of Florida, Gainesville, FL, United States*; [2] *Department of Plant Pathology, University of Nebraska, Lincoln, NE, United States*

Nematodes from the family Tobrilidae are commonly found in freshwater sediments and deep soil samples yet little is known about their lifestyle and ecology. It is expected that they are predators or omnivores of small microbial eukaryotes, but there is no clear evidence. Additionally, they are found to be active under a wide range of abiotic conditions including ecosystems with non-neutral pH and unique biogeochemistry. Despite this, Tobrilidae are not commonly reported in extreme aquatic environments, indicating abiotic conditions may limit the distribution of these nematodes. To determine how both biotic and abiotic factors influence the ecology of Tobrilidae, we examined four lakes in the Western Nebraska Sandhills spanning a unique alkalinity gradient (Gimlet pH 8.1, Island pH 8.7, Border pH 9.9, and Bean pH 10.1) driven by high concentrations of potassium and sodium ions. In October of 2020, we dredged 4 replicate sediment samples from each lake, extracted nematodes from 100g subsamples, counted, and identified them with microscopy, and then processed for 18S rRNA (NF1-18Sr2b) metabarcoding. In addition, we described the microbial communities (16S/18S rDNA metabarcoding) and biogeochemistry of all samples. We hypothesized that: 1) distribution of Tobrilidae would be most influenced by abiotic factors (i.e., alkalinity) and 2) biotic factors (i.e., microbial diversity) would be less important. We used variance partitioning and general linear modelling to test these hypotheses. We found that Tobrilidae were the dominant nematode family comprising 67-100% of the total nematode community within each lake. In the most alkaline lakes (Border and Bean) *Epitobrilus* sp. was the most abundant, whereas *Tobrilus* sp. were more abundant in the more neutral lakes (Island and Gimlet). Although alkalinity was indicated as more important (29%) than biotic (15%) factors, most of the variance explaining Tobrilidae distribution and composition was shared between both (33%) factors. This could indicate feedback relationships within this ecosystem where Tobrilidae, alkalinity, and the microbial communities affect each other simultaneously. Future experimental work with these nematodes in a microcosm assay will allow for a more thorough understanding of these potential relationships.

S7-PF2

Nematode predators catalyze an increase of chloroviruses by foraging on the symbiotic hosts of zoochlorellae.

Kirsten Powers[1] (kpowers1@unl.edu), Zeina Al-Ameeli[2], John Delong[3], Steven Thomas[4], James Van Etten[2], David Dunigan[2], Thomas Powers[1]

[1] *Dept. of Plant Pathology, University of Nebraska, Lincoln, NE, United States*; [2] *Nebraska Center for Virology, University of Nebraska, Lincoln, NE, United States*; [3] *School of Biological Sciences, University of Nebraska, Lincoln, NE, United States*; [4] *School of Natural Resources, University of Nebraska, Lincoln, NE, United States*

In surveys of viruses in the freshwater lakes of Nebraska (USA), chlorovirus populations cyclically increase up to 104-fold [1]. Because a virus must contact its host to replicate, what accounts for such population growth when physical barriers preclude contact? Chloroviruses are large DNA viruses that infect and replicate in certain types of green algae, zoochlorellae. Zoochlorellae, however, live in a mutually beneficial endosymbiotic relationship within the body of a freshwater or marine invertebrate or protozoan protected from chlorovirus contact.
Preliminary findings indicate that nematode predators catalyze virus population growth by breaking down physical barriers between viruses and their endosymbiont hosts through whole consumption or messy feeding [2, 3]. Under laboratory conditions, *Chlorella variablis* Syngen 2-3-infecting chloroviruses attach to the exterior of *Paramecium bursaria*, a common zoochlorellae-bearing freshwater protozoan. We tested whether the nematodes, *Mononchus sp*. and *Pristionchus* sp., foraging on *P. bursaria*, can disrupt the mutualism and pass endosymbiontic zoochlorellae through their guts, exposing them to chloroviruses. We observed zoochlorellae in the nematode gut 30 s after ingestion of paramecium by *Mononchus*. No interaction was detected with *Pristionchus*. Virus density time series over 3 days demonstrated that nematode predation by *Mononchus* on paramecia increased densities of chloroviruses by about 102-fold. This catalytic role for predaceous nematodes in natural systems is novel but consistent with similar observations using different *P. bursaria* predators, increasing our understanding of predator prey dynamics in protozoan food webs.

Keywords: *Mononchus* - Symbiosis - Paramecium - Giant virus.

References:

- Quispe et al., 2016. Arch Virol. 161:1839-1847.
- Delong et al., 2016. Proc Natl Acad Sci U S A. 113:13780-13784.
- DeLong et al., 2018. Microb Ecol. 75:847-853.

S7-PF3

Spatial distribution patterns of microbiome and free-living benthic nematodes in response to sediment ecological conditions in Sado estuary, Portugal.

Soraia Vieira[1] (svvieira@uevora.pt), Kasia Sroczyńska[1], Marta Martins[2], Maria Costa[2], Joana Neves[2], Helena Adão[1], Cláudia Vicente[3]

[1] *MARE-UÉ - Marine and Environmental Sciences Centre, University of Évora, University of Évora, Évora, Portugal*; [2] *MARE – Marine and Environmental Sciences Centre, NOVA School of Science and Technology (FCT NOVA), Lisbon, Portugal*; [3] *MED – Mediterranean Institute for Agriculture, Environment and Development, University of Évora, Évora, Portugal*

Sediment microbiome has an essential role in regulating ecosystem functions, not only regulating primary productivity and nutrient cycling but also shaping trophic interactions with higher trophic levels [1]. While the importance of microbiome in terrestrial soil systems is being widely recognized, its role in marine aquatic environments remains much less studied [2]. Microbiome can be highly affected by bottom up (abiotic factors) and top down (predation by meiofauna) effects [3]. Understanding the interaction effect between abiotic and biotic factors on microbiome communities will be an essential step for future predictions of ecosystem stability. To address this knowledge gap we studied spatial distribution patterns of microbiome communities and nematode assemblages in highly heterogenous Sado Estuary, SW Portugal. The samples were taken from three contrasting sites with varying sediment characteristics and human impact degrees. From each site, three replicate samples for sediment characterization (total organic matter, granulometry, total C and N, chlorophyll a and phaeopigments, contaminants: heavy metals and metalloids, organochlorine pesticides, PAH and PCBs), microbiome (*sensu lato*) and nematode community were taken. Total DNA from sediment was extracted using DNeasy Power Soil kit® (MOBIO, Qiagen) and processed for Illumina MiSeq platform sequencing targeting the V3 and V4 region of 16S rRNA gene. Sediment characterization indicated heterogeneity between sites with distinct levels of contamination, which resulted in contrasting microbial communities. All sites showed a high α-biodiversity with predominance of Proteobacteria phylum, particularly Woeseiaceae, Desulfobacteraceae and Desulfobulbaceae families. Beside this heterogeneity in microbiome community, β-diversity of microbiome communities was demonstrated to be very high, greatly discriminating among all three sites. Instead, nematode assemblages did not yield clear distributional patterns suggesting that their response is rather driven by the within site specific factors, acting at the smaller spatial scales. Studying the relations between sediment ecological conditions and microbiome and meiobenthic communities greatly advance our understanding on benthic ecosystem functioning.

Keywords: Sediment microbiome - Benthic nematodes - Metagenomics - spatial distribution - Interactions.

References:

- [1] Thakur, M. P., & Geisen, S. (2019). Trophic regulations of the soil microbiome. Trends in Microbiology. 27(9):771-780.
- [2] Avó, A. P., Daniell, T. J., Neilson, R., Oliveira, S., Branco, J., & Adão, H. (2017). DNA barcoding and morphological identification of benthic nematodes assemblages of estuarine intertidal sediments: advances in molecular tools for biodiversity assessment. Frontiers in Marine Science. 4: 66.
- [3] Zhang, Q., Goberna, M., Liu, Y., Cui, M., Yang, H., Sun, Q. et al. (2018). Competition and habitat filtering jointly explain phylogenetic structure of soil bacterial communities across elevational gradients. Environmental Microbiology. 20(7): 2386-2396.

ORAL SESSION 8

Nematode management in tropical conditions

Management of potato cyst nematode and bacterial wilt by use of banana fiber paper in potatoes.

Effie Ochieng[1] (lilefy@gmail.com), John Kimenju[1], Douglas Miano[1], Kalpana Sharma[2], Charles Opperman[3], Tahira Pirzada[4], Laura Cortada-Gonzalez[5], Danny Coyne[6]

[1] *University of Nairobi, Nairobi, Kenya*; [2] *International Potato Center, Nairobi, Kenya*; [3] *Department of Chemical and Biomolecular Engineering, North Carolina University, Raleigh, United States*; [4] *Department of Entomology and Plant pathology, North Carolina University, Raleigh, United States*; [5] *Nematology Research Unit, Ghent University, Ghent, Belgium*; [6] *Nematology Unit, International Institute for Tropical Agriculture, Nairobi, Kenya*

Potato cyst nematodes (PCN) and *Ralstonia solanacearum* (bacterial wilt- BW) are both quarantine organisms that have been found in Kenya causing significant yield losses to potato farmers across the country. Where the two pathogens co-exist, the presence of nematodes increases chances of opportunistic infections by BW, which penetrates through root wounds caused during nematode invasion, leading to increased crop losses[1]. Our study aimed to determine if the combination of organic soil amendments (OSA) with nematicides is an effective management strategy to minimize nematode damage and hence, reduce BW infestation of potato in field conditions. Field trials were conducted in Kenya in farms that presented a history of severe BW and PCN infestation. The experimental approach consisted of a 3 ×5 factorial design with 2 different types of OSA: i) Plantmate®- 1000 Kg/ha, ii) Neemgold®- 1000 Kg/ha, and iii) no OSA- negative control, which were individually combined with 4 nematicidal treatments: i) banana fiber paper (BFP) impregnated with abamectin (Wrap & Plant technology), deployed by wrapping potato seeds at planting stage, ii) Velum® (0.625 l/ha), iii) Tervigo®(8 l/ha), iv) Real Trichoderma®(*Trichoderma asperellum*; 0.2 l/ha), and v) no nematicide-negative control. Each of the 15 treatments was replicated three times, and trials were repeated twice. At the end of the experiment cyst counts per g of soil were reduced ($p< 0.05$) in plots with BFP, and more so when it was used with any of the two OSA. Plantmate® and Neemgold® tubers harvested in the BFP + Plantmate® and BFP + Neemgold® tested negative for BW, whereas tubers in Tervigo®+Plantmate® and control+ Plantmate® had visual symptoms of BW and tested positive for BW. Velum®+ Neemgold® ($p < 0.05$) had the highest average yield of potato (11.03 Kg/plot), BFP+Plantmate® seconded at (10.75 Kg/plot), whereas control+control (farmers' practice) had the lowest yields at (6.1 Kg/plot). Our results indicate that the use of OSA in combination with Velum® can have a positive effect on reducing inoculum levels of PCN and BW in the soil and increase potato yields. Very positive results were achieved with the BFP, which represents an environmentally friendly method for combating PCN and BW that severely affect potatoes threatening food security of small-holder farmers in Kenya.

Keywords: Potato cyst nematode - Bacterial wilt - Banana fiber paper - Nematicides - Organic soil ammendment.

References:

- [1] Back et al., 2002. Plant Pathology.51(6)683-697.

Management of nematodes in peri-urban vegetable farms in West Africa.

Abiodun Claudius-Cole (bi_cole@yahoo.com)

Dept. of Crop Protection and Env. Biology, University of Ibadan, Ibadan, Oyo, Nigeria

West African cities are getting larger and more cosmopolitan, and urban farming enterprises are increasingly being established close to cities to meet the vegetable needs and tastes of its residents. These systems use intense production practices and are prone to rather quick build-up of nematode populations. The knowledge deficit about nematode problems and the tendency to use any available pesticide as the first approach for any symptom requires that attention be given to available management options in these peri-urban systems. Health conscious consumers in the region seeking higher quantities of vegetables versus the chiefly starch based diets are also becoming aware of pesticide residues on local and exotic' vegetables produced in these systems and their dangers. Among the several management options, not all are practical or available for farmers in West Africa. An integrated approach remains the more sustainable option as each separate method has its shortcoming in the system. The presentation discusses each approach used in the system and growers perceptions on the success and application in the peri-urban system.

Keywords: Amaranthus - Cabbage - Carrots - Integrated management - Pesticide residue.

References:

- Adedayo, V. 2014. Journal of Agriculture and Sustainability, 5(2): 171-182.
- Affokpon, A. et al., 2014. Acta Horticulturae, (1021), pp.409-419.
- Maconachie, R. 2008. Urban Agriculture magazine 20: 22-24.

Challenges and management of plant-parasitic nematodes on plantain, *Musa* spp. AAB-subgroup, in Nigeria.

Monioluwa Omolara Olaniyi (momolaraolaniyi@yahoo.com)

Department of Pure and Applied Sciences, National Open University of Nigeria, Abuja, FCT, Nigeria

Plant-parasitic nematodes pose a serious constraint to plantain production in Nigeria, which until now has not been identified. The black Sigatoka disease had earlier been considered the most important threat to plantain in Nigeria reducing yield by 33 % in the first crop. However, nematode-induced yield losses in Nigeria are greater in the first crop cycle. The impact of plant-parasitic nematodes on plantain production was evaluated in multifactorial experiments and field surveys in Southern Nigeria over 12 years. Results revealed a range of 46-54 % yield reduction due to species mixture of plant-parasitic nematodes. Reaction of plantain was dependent on genotype and environmental factors including soil management and fertility status. Additionally, an absolute yield loss due to toppling over of plants bearing immature bunches may be up to 33 %. In diagnostic surveys, eleven plant-parasitic nematode species were observed on plantain in Southern Nigeria whereas farmers were largely unaware of plant-parasitic nematode problems. *Pratylenchus coffeae, Radopholus similis* and *Helicotylenchus multicinctus* were identified as the key species of plant-parasitic nematodes on plantain in Nigeria with *P. coffeae* considered the most damaging, followed by *R. similis* while *H. multicinctus* was ubiquitous. However, the predominance of each species was region specific with *R. similis* of significance in South-Eastern and South-Southern Nigeria while *P. coffeae* was predominant and more damaging in the South-West. The need for concerted efforts by breeding programmes to develop plantain hybrids with acceptable traits and resistance to the key nematode species in Nigeria cannot be overemphasised. Although crop failure associated with plant parasitic nematode damage often results in plantation abandonment, farmers associated plantation decline with superstitious beliefs. In a bid to explore cost-effective interventions, organic mulches and extracts of medicinal plants were evaluated in the management of plant-parasitic nematodes on plantain in Nigeria. Organic mulches enhanced plantation productivity in the first crop, while yield declined in subsequent years. Of the medicinal plants assayed, *Acalypha wilkesiana* leaf extract showed promise in nematode control *in vitro* and was further explored *in vivo* with resultant reduction in plantation decline. This is an attractive option for resource-poor farmers that produce the crop in Nigeria.

Keywords: Key nematodes - Yield loss - Plantation decline - Organic mulch - Plant extracts.

References:

- [1] Olaniyi, 2011. Plant Parasitic Nematode Constraint to Plantain Production in Nigeria. LAP Lambert Publishing. ISBN 978-3-8454-2312-8. 240pp.
- [2] Oso et al., 2016. International Journal of Agriculture and Environmental Research 2(3): 549-557.
- [3] Oso et al., 2014. Asian Journal of Agricultural Extension, Economics & Sociology 3(6): 630-637.
- [4] Oso et al., 2014. Nigerian Journal of Nematology 2: 63-76.

Adopting integrated nematode-soil health management in smallholder potato farms in the highlands of Guatemala.

Stephen Kakaire[1] (kakaire1@msu.edu), Amilcar Sanchez[2], Anibal Sacbaja[2], Catherine Chan[3], Brent Sipes[3], Haddish Melakeberhan[1]

[1] *Agricultural Nematology Laboratory, Department of Horticulture, Michigan State University, East Lansing, MI, United States*; [2] *University of San Carlos, Guatemala City, Guatemala*; [3] *Univerisity of Hawaii at Manoa, Honolulu, HI, United States*

Food and nutritional insecurities, plant-parasitic nematodes (PPN), including the potato cyst nematode (PCN; *Globodera* spp.), and poor soil health are among the intertwined challenges that the USAID's Horticulture Innovation Laboratory has been focusing on in smallholder farming systems in the highlands of Guatemala. To address these challenges, an interdisciplinary team of nematologists, social scientists, agronomist, and soil scientist initiated experiments in 2017 and 2018 in the Mollisols of Huehuetenango and the Andisols of Xela, by amending soils either with or without bio-mix and 0, 318, or 454 kg of composted chicken manure at eight locations. The bio-mix (BioCopia) consisted of Guatemalan isolates of *Purpureum* and *Bacillus* applied at 1.8 kg/m2 to suppress PPN. Nematodes were extracted from 100 cm3 of soil, fixed in double TAF solution at USAC and enumerated at MSU. Over the growing seasons, nematode abundance averaged about 300 to 600 individuals/100 cm3 of soil. Herbivores, predators, and omnivores tended to increase with time in Andisols plots more so than in Mollisols plots. PCN population density was very high before planting in both years with PPN accounting for 20–40% of the nematode fauna in both regions. At mid-season in both years, striking visual differences in plant growth between compost and non-compost amended treatments existed in both regions. PCN population density trended similar to potato yield, with both greater in Andisols than in Mollisols. Soil amendments increased yield and PCN population densities in both soil groups. Soil pH in the Andisols averaged 5.5 and 5.0 in Mollisols. Whereas K was above recommended levels in both soils, K levels were greater in Andisols than in Mollisols. Potato yield, soil pH and % OM were positively (P = 0.05) correlated in Andisols. Percent OM and C:N ratio were significantly higher in Mollisols than in Andisols, suggesting nutritional imbalances between the soil groups. The Andisols are likely to benefit more from soil amendments than the Mollisols, which are higher in organic matter. Based on the Soil Food Web (SFW) model [1], both soil groups have a need for biological activity to release nutrients. The SFW and biophysiochemical data suggest that neither soil group has sustainable agroecosystem conditions. The growers are being encouraged to consider soil health in their production system.

Keywords: Biological control - Potato cyst nematode - Soil food web - Soil type.

References:

- [1] Ferris et al., 2001, Applied Soil Ecology, 18:13–29.

The effects of orange and lemon juice on root-knot nematode populations densities and plant growth under greenhouse conditions.

Grace Tefu[1] (grace@arc.agric.za), Mieke Daneel[2], Driekie Fourie[1]

[1] *Integrated Pest Management, North West University, Potchefstroom, North West, South Africa*; [2] *Crop Protection, ARC-Tropical and Subtropical Crops, Nelspruit, Mpumalanga, South Africa*

Earlier research showed that citrus-based amendments (e.g. as organic material or fruit extracts such as juice or powders) have potential to reduce *Meloidogyne* spp. densities and increase plant growth. This was evident especially for glasshouse trials, while results from field trials were less promising. For example, addition of orange juice to potted tomato plants consistently showed a plant growth promoting effect, although a reduction in *Meloidogyne incognita* densities was inconsistent. Conversely, application of lemon juice and powder extract to potted tomato plants resulted in superior reduction of *M. incognita* densities (92%), while results from field trials were inconsistent. To further investigate the effect of such amendments, two glasshouse trials were conducted during two consecutive seasons in Venda (University Venda, South Africa) using tomato seedlings (cv. Monica) that were inoculated with ±3000 eggs and second-stage juveniles of *M. incognita.* Five concentrations (200, 400, 600, 800 and 1000 L/ha) of lemon and orange juice represented the treatments, as well as the synthetic nematicide cadusafos (upper control) and an untreated control. Each treatment was replicated six times for each trial. The different concentrations of both crop juices resulted in a significantly (P≤0.05) higher plant growth effect of tomato plants compared to the untreated control and the cadusafos (both seasons) treatments. Plant growth was hence positively correlated with the lemon juice concentration; lower for low concentrations and higher for high concentrations. Addition of the individual juices also substantially reduced *M. incognita* densities in tomato roots, *viz*. for lemon juice between 98-100% and for orange juice between 57-100%; except for the lowest orange juice concentration that had no effect on reducing nematode densities in the second season. Reduction of *M. incognita* root densities by lemon and orange juice treatments compared well with the registered nematicide cadusafos (96-100%). It is evident from this study that juice from both lemon and orange fruits can be used by developing producers since they can obtain it from produce that cannot be marketed due to poor quality. It will, however, be helpful to determine the active molecules in the amendments to optimise/enhance their nematicidal effect.

Keywords: Citrus-based amendments - *Meloidogyne* - Tomato - Plant growth.

Integrated management of *Meloidogyne incognita* and *Fusarium oxysporum* in cucumber under a protected cultivation system.

Jaydeep Ashok Patil (rajhau99@gmail.com), Anil Kumar, Sewak Ram

Department of Nematology, CCS Haryana Agricultural University, Hisar, Haryana, India

Cucumber (*Cucumis sativus*) is attacked by root knot nematode under protected cultivation systems. Continuous availability of host, moisture and aeration in polyhouses favours the growth of nematodes and soil borne pathogens. Root-knot nematode, *Meloidogyne incognita* and *Fusarium oxysporum* f. sp. *cucumerinum* form a disease complex and sometimes complete failure of a crop. A study was conducted under polyhouse conditions in pots to determine the potential of various combinations of organic amendments, fumigant and biocontrol agent as soil application and seed treatment viz., neem cake, neem leaves, neem oil, formalin and *Purpureocillium lilacinum* against *Meloidogyne incognita* and *F. oxysporum* f. sp. *cucumerinum* on cucumber. Sixteen treatments combinations along with checks with carbofuran @ 0.1 g/ kg soil and Bavistin @ 2 g/ l water as well as untreated check (inoculated and uninoculated) were also maintained. Potted soil was treated with various combinations of organic amendments and biocontrol agents. Waiting periods were given whenever required. All the treatments significantly improved the plant growth parameters and reduced nematode reproduction as compared to untreated, inoculated control. Among the various treatments, maximum shoot length was observed in fumigation with formalin @ 30 % i.e. 5 ml/ kg soil + seed treatment with *P. lilacinus* @ 20 gm/ kg seed (160.6 cm), followed by soil treatment with neem cake + seed treatment with *P. lilacinus* @ 20 gm/ kg seed (159.9 cm) as compared with untreated inoculated check (84.7 cm). The minimum number of galls per plant was observed in formalin @ 30 %, i.e. 5 ml/ kg soil+ seed treatment with *P. lilacinus* @ 20 gm/ kg seed (24), followed by formalin @ 30 % i.e. 5 ml/ kg soil+ seed treatment with neem oil @ 20% v/w (38).

Keywords: Root-knot nematode - Fungus - Bio-control agent - Plant growth parameters - Protected cultivation.

S8-PF1

EUPHRESCO – MELORISK: Preventing *Meloidogyne graminicola* spread in European rice paddies.

Maria Lurdes Inácio[1] (lurdes.inacio@iniav.pt), Leidy Rusinque[1], Filomena Nobrega[1], Rita Varela[1], Ines gabl[2], Nicole Viaene[3], Ani Becheva[4], Mirela Borisova[4], Giulia Torrini[5], Agostino Strangi[5], Evelyn van Heese[6], Gerrit Karsen[6], Barbara Geric Stare[7], Tim Warbroek[6], Daniel Apolônio[6], Cristiano Bellé[8], Giovanna Curto[9], Francesca De Luca[10], Alberto Troccoli[10], Elena Fanelli[10], Stefano Sacchi[11], Antonio Evidente[12], Alessio Cimmino[12], Marco Masi[12], Marta Santos[13], Marieke Beltman[13], Salvador Noges[14], Xavier Serrat[14], Sandra Mace[14], Refik Bozbuga[15], Dilek Dinçer[16], Hulya Demirbas Pehlivan[16], Isabel Abrantes[17], Luci Conceição[17], Carla Maleita[17]

[1] *Instituto Nacional de Investigação Agrária e Veterinária, INIAV, Oeiras, Portugal*; [2] *Austrian Agency for Health and Food Safety, Graz, Austria*; [3] *Flanders Research Institute for Agriculture, Fisheries and Food Research, Brussels, Belgium*; [4] *Bulgarian Food Safety Agency, Sofia, Bulgaria*; [5] *Council for Agricultural Research and Economics, Rome, Italy*; [6] *Netherlands Food and Consumer Products Safety Authority, Utrecht, Netherlands*; [7] *Ministry of Agriculture Forestry and Food, Ljubljana, Slovenia*; [8] *Instituto PHYTUS, Itaara, Rio grande do Sul, Brazil*; [9] *Emilia Romagna Plant Protection Service, Bologna, Italy*; [10] *National Research Council, Rome, Italy*; [11] *Plant Protection Service Lombradia, Milan, Italy*; [12] *University of Naples Federico II, Naples, Italy*; [13] *Clear Detections, Wageningen, Netherlands*; [14] *University of Barcelona, Barcelona, Spain*; [15] *Eskisehir Osmangazi University, Odunpazarı, Turkey*; [16] *Biological Control Research Institute, Adana, Turkey*; [17] *University of Coimbra, Coimbra, Portugal*

Rice (*Oryza sativa* L.) is the most important staple food being the primary source of energy for over half of the world's population. The total area growing rice within Europe is about 450,000 ha and according to the European Commission, the region produces ≈70% of the total rice consumed domestically, with an average of 3.1 million tons/year. Plant-parasitic nematodes (PPN) represent an important constraint to agricultural production worldwide. Among the PPN, root-knot nematodes *Meloidogyne*spp., and particularly *M. graminicola*(*Mg*), are serious pests of rice, accounting for up to 70% of yield losses. Although *Mg*is considered a pest of tropical rice production, it was recently reported in Italy and included in the European and Mediterranean Plant Protection Organization (EPPO) Alert List. Projections by the Intergovernmental Panel for Climate Change (IPCC) indicate that there will be an increase in mean annual temperature and rainfall that may result in an increasing rate of *Mg*'s infection, development and reproduction, causing shifts in their abundance and geographic distribution. Such effects may have a detrimental impact on cereal production, mainly rice in temperate regions. Since its detection in 2016, in the Piedmont region, it has further spread to other Italian regions, such as Lombardy, which poses a risk to the whole region. This project aims 1) to investigate the occurrence and distribution of *Mg* in the partnering countries 2) to validate the identification methods (morphology, biochemical and molecular), 3) to sequence (NGS) the genome of Italian and other *Mg* isolates; and 4) to evaluate host suitability of various cultivars. Therefore, the expected outcomes are: Distribution maps of *Mg* in Europe; Knowledge to support risk analysis of spread with trade, especially in Mediterranean countries; Accessibility of isolates for morphological, biochemical and molecular studies; A network of research and quarantine nematologists to harmonize and validate molecular protocols to support *Mg* diagnosis; Availability of robust sequence data through Q-bank and Genbank; Better understanding of *Mg* epidemiology through accurate characterisation; Sustainable management strategies.

Keywords: Surveillance - Diagnosis - Sequencing - Networking.

References:

- Fanelli, E. et al., 2017. European Journal of Plant Pathology 149:467-476.
- Mantelin, S., et al., 2017. Molecular Plant Pathology 18(1): 3-15.
- Petitot, A.S., et al., 2016. Molecular Plant Pathology 17(6): 860-74.
- Soriano, I.R., et al., 1999. Nematology 1:395-398.
- Zhant, L.P., et al., 2018. Journal Integ. Agriculture 17:621-630.

S8-PF2

Dominance index of soil nematodes on a coffee plantation after cost-effective bionematicide application.

Iis Nur Asyiah[1] (iisnaza.fkip@unej.ac.id), Laeli Nordiana[1], Balawara Andika[1], Sugeng Winarso[2], Lenny Widjayanthi[3], Dwi Nugroho[4], Kurniawan Firmansyah[5], Ankardiansyah Pandu Pradana[6]

[1] *Biology Education, University of Jember, Jember, East Java, Indonesia*; [2] *Soil Science, University of Jember, Jember, East Java, Indonesia*; [3] *Agricultural Extension, University of Jember, Jember, East Java, Indonesia*; [4] *Indonesian Coffee and Cocoa Research Institute, Jember, East Java, Indonesia*; [5] *Tiga Kreasi Bersama Limited Partnership, Bondowoso, East Java, Indonesia*; [6] *Plant Protection, University of Jember, Jember, East Java, Indonesia*

Some areas of smallholder coffee plantations in Indonesia are located on acid dry land and have steep slopes. This can lead to soil degradation and lower coffee productivity. One of the efforts to overcome this problem is biofertilizer. In previous research, we have succeeded in finding a cost-effective bionematicide formula with an active ingredient of 3 rhizobacteria species and 1 endophytic bacterial species [1]. This formula has proven to be effective in controlling some plant parasitic nematodes, namely *Meloidogyne incognita*, *Pratylenchus coffeae, Radopholus similis,* and *Globodera rostochiensis* [1, 2]. This research aims to evaluate the effect of bionematicides on the diversity of soil nematodes in smallholder coffee plantations. The research was conducted at the arabica coffee plantation in the mountainous area of Ijen, Bondowoso Regency, East Java, Indonesia. The treatment in this research followed a randomized block design pattern with 7 treatments and 5 replications. Each replication consisted of 8, five-years-old coffee plants. The treatments used per plant included control (T1), 5 L of organic matter (T2), 60 mL of bionematicide (T3), 30 mL of bionematicide + 5 L of organic matter (T4), 60 mL of bionematicide + 5 L of organic matter (T5), 90 mL of bionematicide + 5 L of organic matter (T6), and 120 mL of bionematicide + 5 L of organic matter (T7). Five orders (Rhabditida, Dorylaimida, Enoplida, Mononchida, and Tylenchida) and 18 genera of nematodes were found. The application of biofertilizers plays a role in increasing the diversity and population of soil nematodes, especially bacterivore, predatory, omnivore and fungivore nematodes, as well as reducing the population of parasitic nematodes in coffee field. Treatment T7 had a significant effect in reducing the parasitic nematode population by 74.61% and increasing the free-living nematode population by 268.11% compared to T1. The dominance value of free-living nematodes in T7 treatment was the highest compared to other treatments, namely bacterivores (16.62), omnivores (6.32), predators (7.62) and fungivores (4.81). In addition, parasitic nematodes in T7 treatment also showed the lowest dominance value (0.93) compared to other treatments.

Keywords: Arabica - Bacterial-feeder - Diversity - frequency - Fungal-feeder.

References:

- [1] Asyiah et al., 2020. Biodiversitas. 21 (10): 4702-4708.
- [2] Asyiah et al., 2021. Biodiversitas. 22 (6): 3256-3264.

S8-PF3

Control of the rice root-knot nematode *Meloidogyne graminicola* using rice plants as trap crops.

Giulia Torrini[1] (giulia.torrini@crea.gov.it), Stefano Sacchi[2], Leonardo Marianelli[1], Giuseppe Mazza[1], Annachiara Fumagalli[3], Beniamino Cavagna[4], Pio Federico Roversi[1], Mariangela Ciampitti[4]

[1] *CREA - Plant protection and Certification, Florence, Italy*; [2] *Phytopathological Laboratory, Fondazione Minoprio, Vertemate Con Minoprio, Como, Italy*; [3] *Phytosanitary Service, ERSAF Lombardy, Milan, Italy*; [4] *DG Agricoltura Servizio Fitosanitario Regionale, Lombardy Region, Milan, Italy*

Meloidogyne graminicola is one of the most harmful organisms in the rice cultivation throughout the world. In 2016 it was detected for the first time in mainland Europe (Northern Italy) and subsequently added to the EPPO Alert List [1, 2].
Italy is the main rice growing country in Europe, so the Italian NPPO quickly issued phytosanitary measures to eradicate this pest. The rice field submersion is the technique usually carried out, but in the Lombardy region (where this pest has been detected in 2018) this practice is not applicable due to the sandy soil structure. For this reason, some field trials using rice plants as trap crops, were conducted to identify new control strategies against this pest. This agronomic technique stimulates juveniles to hatch and invade the roots of rice plants and, before the nematodes complete their life cycle, the crop is destroyed thus reducing the nematode population in the soil. In 2019, field trials were performed in a Lombard rice field that was highly infested with *M. graminicola*. In this area, 15 plots (5 m x 5 m) were set up, five plots for three treatments: (i) Uncultivated; (ii) Rice sown and left to grow until the end of the trials; (iii) Three separate cycles of rice production; where plants were sown and destructed at the second leaf stage each time. At the end of the three cycles, the number of eggs and juveniles in the soil was compared with the data recorded before the experiment and throughout treatments. In addition, all the plots were weeded and subsequently sown with the same amount of rice at the same time. At the second leaf stage of the plants, the severity of root galling was assessed (120 plants for treatment). Moreover, the evaluation of plant growth and density of the seedlings were evaluated.
The results showed that in the plots of the three seeding-weeding cycles, the density of the nematodes was statistically lower than the other two treatments, as well as the root gall index. Furthermore, the density of the seedlings and their epigeal growth was statistically higher than the other two treatments. In conclusion, the use of trap crop technique for control of this pest gave good results in a relatively short time and thus it could be used as new phytosanitary measure.

Keywords: Europe - Phytosanitary measures - Phytoparasitic nematode - Rice crop - Root gall index.

References:

- [1] EPPO, 2016. Reporting service (2016/211).
- [2] Fanelli et al., 2017. European Journal of Plant Pathology. 149: 467-476.

WORKSHOP 1

Expanding indicator qualities of nematodes to identify sustainable soil health

Expanding indicator qualities of nematodes to identify sustainable soil health.

Haddish Melakeberhan[1] (melakebe@msu.edu), Alemayehu Habteweld[2]

[1] *Agricultural Nematology Laboratory, Michigan State University, East Lansing, MI, US*; [2] *Entomology and Nematology, University of Florida, Gainesville, FL, United States*

Soil health fragility from intensive land use practices and the value of nematodes as a bioindicator of changes in soil ecosystems as influenced by production and nematode management practices are well established. Healthy soil requires balanced biological, physiochemical, nutritional, structural, and water holding components. Despite the advances in basic and applied aspects of soil health components and nematode management, achieving and sustaining healthy soil remains challenging. In part, this is due to lack of simultaneous integration of the different components of soil health and the desired ecosystem services of improved soil structure, physiochemistry, water holding capacity and nutrient cycling, suppressing harmful while increasing beneficial organisms, and crop yield that they generate. Aligning the desired ecosystem services is challenging. For example, the biological component, which drives the soil food web (SFW) and nutrient cycling, includes both harmful and beneficial nematodes as central players in the same environment. Under these circumstances, sorting out ecosystem services with conflicting trajectories is difficult, as is aligning all of the desirable ecosystem services. This workshop will provide an overview of the soil health components and the role of the SFW therein. This will be followed by two interactive modules that use nematodes as indicators to demonstrate how to separate effects of agricultural practices on the desired ecosystem services, leading to sustainable and steady-state of soil health conditions simultaneously. The first module will use changes in population dynamics of harmful nematodes and the second module beneficial ones as indicators. Attendants are expected to bring computers and they will be provided with data used for the demonstration. The broad impact will be improved use of nematodes as an indicator to translate basic and complex biophysiochemical information into integrated practical application, leading to improved long-term environmental, economic and quality of life needs.

Keywords: Decision-making - Ecosystem services - Nutrient cycling.

WORKSHOP 2

Aphelenchoides besseyi: reemergence of a forgotten parasite

Aphelenchoides besseyi: reemergence of a forgotten parasite.

Johan Desaeger (jad@ufl.edu)

University of Florida

The genus *Aphelenchoides* contains many ubiquitous fungal feeding nematode species, as well as nematodes species that are parasites of plants. The plant-parasitic species often have a very wide host range, occurring as ectoparasites or endoparasites of the buds, leaves and growing points of many economically important crops. *Aphelenchoides besseyi* is the most important plant-parasite in this group. This species has a global distribution, mostly in rice-growing regions of the world, but also in strawberry, ornamental, and soybean crops. This workshop will provide an overview of the importance of *Aphelenchoides besseyi* in different geographic regions (US, Brazil and China) by experts from these three regions, and focused on the complexity, species differentiation, and feeding preferences of this cryptic species. The regional overviews will be followed by a group discussion on the reemergence of foliar nematodes in relation to changing climate, and agricultural and nematode management practices.

Occurrence and Control of *Aphelenchoides besseyi* in China.

Hongli Ji (hongli.ji@jihongli.com), Jialian Xie, Fang Yang, Xing Xu, Yunliang Peng

MOA Key Laboratory of Integrative Pest Management on Crops in Southwest China, Institute of Plant Protection of Sichuan Academy of Agricultural Sciences, Chengdu, Sichuan Province, P.R. China

The white tip disease of rice in China was first reported in the 1940s and records of the disease were subsequently found in all of the country where *japonica* rice was planted. Tolerance of the nematode was found in many of the *indica* varieties of rice and the disease was almost neglected in more southern and warmer parts of China where *indica* rice was planted. The loss of the disease was limited due to the limited range of the individual rice varieties and the prevention of the disease by quarantine measures. However, the late 1980s and early 1990s have witnessed the outbreak of the disease with the symptom of “smaller panicles” in the eastern part of China. Treatment of the seeds with nematicides has resulted in successful control of the disease. Nevertheless, severe losses also result from the summer-dwarf symptoms caused by the nematode on strawberry in China since late 1980s, although attracting less attention recently due to the changes in the conditions of strawberry production. The nematode has become one of the quarantine problems for the export of seeds of hybrid rice. Larger numbers of nematodes and common symptoms is observed on the sub-species hybrids. New initiatives to study and control of the disease have resulted from CARS projects and some advances in understanding and controlling the disease have been made.

Keywords: White tip disease - Control - Hybrid rice - Smaller panicles.

Summer crimp disease (*Aphelenchoides besseyi*): Species delimitation and feeding habits.

Clemen Oliveira[1] (clemendeoliveira@ufl.edu), Johan Desaeger[1], Sergei Subbotin[2], Renato Inserra[3], Janete Brito[3], Natalia Peres[4]

[1] *Entomology and Nematology Department, University of Florida, Wimauma, Florida, United States*; [2] *California Department of Food and Agriculture, Sacramento, Florida, United States*; [3] *Florida Department of Agriculture and Consumer Services, Gainesville, Florida, United States*; [4] *Plant pathology Department, University of Florida, Wimauma, Florida, United States*

Summer crimp disease is caused by *Aphelenchoides besseyi*, a damaging foliar nematode in Florida strawberry. A cooperative study in the southeastern United States aimed to delimit the species within the *A. besseyi* complex using an integrated taxonomic approach. This study disclosed several cryptic species: *A. besseyi sensu stricto*; *A. oryzae*; and *A. pseudobesseyi*. A population collected from North Carolina strawberry transplants, which is the type locality for *A. besseyi*, fits the taxonomic descriptions reported by Christie and Allen on *A. besseyi* being the only representative of this species. A Louisiana rice population fit the descriptions of *A. oryzae* of both Yokoo and Fortuner. Three populations from Florida ornamental plants (*Dryopteris erythrosora*, *Echinacea* sp., and *Farfugium japonicum*) differed from those of the two above-mentioned species and were described as *A. pseudobesseyi*. The feeding habits of the Florida *A. besseyi sensu stricto* on higher plants demonstrated that this nematode infects strawberry and gerbera daisy, but not soybean and alfalfa. Furthermore, *A. besseyi* presented a fungal selectivity as its reproduction was significantly greater ($P<0.05$) on strawberry pathogenic isolates of *B. cinerea*, *C. gloesporioides,* and *P. clarispora* than on the non-pathogenic isolates of *F. oxysporum*, and *M. fructicola*. *Aphelenchoides pseudogoodeyi,* which is mainly a mycetophagous nematode, was used as a check. This nematode did not infect any crop and no feeding selectivity was observed on fungi that produced mycelium in our study. However, localized inoculation to the soybean leaves resulted in nematode penetration but infection did not expand overtime. Our research confirms the difficulty in the morphological separation of species previously considered as '*A. besseyi*' detected on rice, ornamental plants, and strawberry in the southeastern United States. This morphological variability made separation of *A. besseyi* from *A. pseudobesseyi* and *A. oryzae* unreliable without the examination of numerous specimens and molecular analyses. The feeding selectivity of *A. besseyi* toward strawberry pathogens, strawberry, and gerbera daisy plants was evident compared to *A. pseudogoodeyi*. These results have implications for integrated nematode management decisions for farmers, scientists, and global regulatory agencies.

Keywords: *A. besseyi* - Species delimitation - Foliar nematodes - Feeding habits - Summer crimp disease.

Aphelenchoides besseyi: Emerging challenge in Brazilian crops

Andressa Machado

No abstract submitted.

WORKSHOP 3

Advances and challenges in CRISPR-mediated technologies in parasitic and free-living nematodes

Advances and challenges in CRISPR-mediated technologies in parasitic and free-living nematodes.

xxxxxxxx (xxxxx)

xxxxxxx

No abstract submitted.

CRISPR-mediated reverse genetic approaches in *P. pacificus* and potential modifications for plant parasitic nematodes.

Ralf J. Sommer (ralf.sommer@tuebingen.mpg.de), Witte Hanh

Department for Integrative Evolutionary Biology, Max Planck Institute for Biology Tübingen, Tübingen, Germany

The CRISPR technology has revolutionized reverse genetics and has a major impact on biomedical research. In *C. elegans*, various CRISPR-associated approaches have been established that have further increased the methodological toolkit of this organism. In the applied sciences, plant and animal parasitic nematodes might profit substantially from the development of related tools. We study the free-living nematode *Pristionchus pacificus* and have intensive experience with transferring methodologies originally developed in *C. elegans* to *P. pacificus*. *P. pacificus* is a model system for integrative evolutionary biology with a sophisticated toolkit for functional analysis. It has a 4-day generation time (20° C) and is a self-fertilizing hermaphrodite similar to *C. elegans*. Recent studies in this species focus on the regulation of predatory vs bacterial feeing as a result of mouth-form developmental plasticity.
In this talk, we will provide an overview on CRISPR applications in *C. elegans* and *P. pacificus*. In particular, we will discuss successful troubleshooting and potential modifications of standard protocols that might make it more amenable, also to parasitic nematode species.

Keywords: Nematode - Phenotypic plasticity - Evolution - Reverse genetics.

TransPPN: The Transformation of Plant-Parasitic Nematodes Consortium.

Sebastian Eves-van den Akker[1] (se389@cam.ac.uk), The TransPPN Consortium[2]

[1] *University of Cambridge, Cambridge, Cambridgeshire, United Kingdom*; [2] *Open, Global, United Nations*

The ability to incorporate foreign DNA or to make targeted modifications in the genome of an organism has been achieved in multiple systems, and has revolutionised their fields of study.
Several independent attempts have been made to develop a transformation method for plant-parasitic nematodes over the last decade/s, all so far unsuccessful. Partly because of the lack of benchmark progress, there has been little public dissemination as to what has, and crucially, what has not worked. Recent technological and scientific advances, both within plant-parasitic nematology and further afield, impact at multiple key stages: now is the right time to revisit transformation.
In 2016, the transformation of plant-parasitic nematodes consortium (TransPPN) was established to address this constraint. TransPPN is an open, informal, and global consortium consisting of members from 12 institutes, across 7 countries. The aim is to consolidate previously unshared experiences, provide a forum to discuss ideas and techniques, foster collaborations, and maintain momentum.
We have made a concerted effort over the last few years to assemble the necessary seed-corn funding, expertise, ideas, and preliminary data to demonstrate that genetic modification of plant-parasitic nematodes is not an insurmountable problem. This talk will summarise some of the activities of the consortium, and the recent progress (failures and successes) towards genetic modification of plant-parasitic nematodes.

Keywords: Plant-parasitic nematodes - Transformation - Consortium - CRISPR.

Liposome-mediated CRISPR/Cas9 and RNAi in free-living and parasitic nematodes.

Andre Pires da Silva[1] (andre.pires@warwick.ac.uk), Sally Adams[1], Hongguang Shao[2], James B. Lok[2], Prachi Pathak[1]

[1] *School of Life Sciences, University of Warwick, Coventry, United Kingdom*; [2] *Department of Pathobiology, School of Veterinary Medicine, University of Pennsylvania, Pennsylvania, Philadelphia, United States*

Manipulation of gene expression and mutagenesis are important methods to functionally characterize genes. Although gene-silencing by RNA interference (RNAi) and genome-editing by CRISPR/Cas9 are relatively straightforward techniques to implement in model nematodes such as *Caenorhabditis elegans*, they are not easily transferable to other nematodes. For RNAi, soaking nematodes in a solution with dsRNA or small RNAs may fail because nematodes lack mechanisms that transfer the exogenous RNAs from the gut to the germline [1]. Delivery of the RNAi or RNA/Cas9 by microinjection may be difficult because of poor accessibility to gonads by microinjection [2]. To circumvent these problems, we use the transfection agent lipofectamine, which are vesicles with a lipid bilayer that can be loaded with cargo (e.g., nucleic acids and/or proteins) [3, 4]. When using lipofectamine in combination with microinjection, it is possible to generate mutants or RNAi phenotypes in the injected animal as well in a subsequent generation. We also show that the underdeveloped gonads of larval stages are amenable for transfection after injection in the body cavity. As proof-of-concept we generated CRISPR/Cas9 mutants of *Strongyloides stercoralis* by injecting the free-living infective stage. Thus, this technique may open the possibility of studying gene function in parasitic nematodes, for which the adults are inaccessible for microinjection because they are inside of the host.

Keywords: Transfection - Parasitic nematodes - CRISPR - RNAi - Gonad.

References:

- [1] Nuez, I., and Félix, M.A. (2012). PLoS One 7, e29811.
- [2] McCaig, C.M., Lin, X., Farrell, M., Rehain-Bell, K., and Shakes, D.C. (2017). Dev Biol 430, 362-373.
- [3] Felgner, P.L., Gadek, T.R., Holm, M., Roman, R., Chan, H.W., Wenz, M., Northrop, J.P., Ringold, G.M., and Danielsen, M. (1987). PNAS 84, 7413-7417.
- [4] Adams, S., Pathak, P., Shao, H., Lok, J.B., and Pires-daSilva, A. (2019). Sci Rep 9, 483.

WORKSHOP 4

DNA barcoding of nematodes

DNA barcoding of nematodes.

Thomas Powers[1] (tpowers1@unl.edu), Dorota Porazinska[2]

[1] *Department of Plant Pathology, University of Nebraska - Lincoln, Lincoln, NE, United States;* [2] *Department of Entomology and Nematology, University of Florida, Gainesville, FL, United State*

In this workshop we address one main question: "How to ensure that nematode biodiversity is accurately represented in global assessment of diversity? The problem: Technological advances in nucleotide sequencing are increasingly outstripping our ability to store, retrieve, and analyze taxonomic data. On projects that focus on nematode diagnosis or assessment of nematode diversity, it is common to encounter reference database deficiencies that prohibit an accurate description of the taxonomic composition of nematode communities. As stated by Waeyenberge et al., (2019) "The need of a larger, high quality, taxon-specific, reference database cannot be overstated". For DNA barcoding process perspective, we need to:

1. Evaluate the current status of taxonomic data repositories for nematodes.
2. Identify deficiencies or blockages that compromises our nematode databasing workflow.
3. Determine what functionality we want for our databases.
4. Evaluate collaborative strategies to address these inefficiencies.

We recognize that while nematodes are among the most abundant and diverse multicellular animals on the planet, the community of Nematologists is small compared to other taxon-oriented societies. We suggest that a collaborative effort is necessary to move DNA barcoding into the future. A major goal of this workshop would be for interested parties to contribute to a white paper that would be the basis of a proposal for grant funding.

7th International Congress of Nematology
ICN 2022
Antibes Juan-les-Pins - France

"Crossing borders: a world of nematode diversity and impact to discover"

European Society of Nematologists

ABSTRACTS TUESDAY 3 MAY

PLENARY SESSIONS

A model nematode outside the laboratory: *Caenorhabditis elegans* habitat and biotic interactions.

Marie-Anne Felix (felix@bio.ens.psl.eu)

Institute of Biology of ENS, CNRS, Paris, France

Caenorhabditis elegans is widely used as a laboratory model organism. Most laboratory studies are performed in a single reference strain in a standard laboratory environment, feeding the animals with *Escherichia coli.* We try to widen this horizon and provide an ecological and evolutionary context for *C. elegans*.
Natural populations of *C. elegans* and its relatives can be found in rotten vegetal material such as rotting stems, flowers and fruits. These food sources are ephemeral, forcing a boom-and-bust lifestyle on *C. elegans*: after population proliferation, young larvae in large populations develop into diapausing dauer larvae. These dauers can be found associated with isopods and terrestrial molluscs. We surveyed *Caenorhabditis* populations in an orchard and a wood over several years, so as to probe *C. elegans* metapopulation structure and outcrossing rate. With other colleagues, we also discovered many new species in the *Caenorhabditis* genus and thus provide an evolutionary framework for genomic and phenotypic studies. My laboratory also studies a widely distributed terrestrial rhabditid nematode species called *Oscheius tipulae*.
Natural pathogens provide strong and changing selection pressures and are thus relevant to study defense systems and their potentially rapid evolution. Several natural pathogens of *C. elegans* were isolated, including the first viruses that infect *C. elegans* or *C. briggsae*. A genome-wide association study of Orsay RNA virus load after infection of a worldwide set of *C. elegans* isolates indicates one major locus segregating in the species. We found that this major locus corresponds to a widespread deletion inactivating the homolog of vertebrate RIG-I viral sensors, thus allowing viral replication. In *C. briggsae*, we found a specificity of interaction of the Santeuil and Le Blanc viruses to different *C. briggsae* wild isolates.
I will also report on other organisms associated with *Caenorhabditis* nematodes such as microsporidia, fungi, oomycetes and bacteria. Bacteria in particular are found to have a wide range of context-dependent association types with *Caenorhabditis*, from highly deleterious to beneficial.

Keywords: *Caenorhabditis elegans* - Biotic interactions.

Nematode chemosensation: implications on insect pest management.

Ivan Hiltpold (ivan.hiltpold@agroscope.admin.ch)

Agroscope Chagins, Nyon, Switzerland

There is an increasing number of evidences suggesting that entomopathogenic nematodes are able to use volatile cues to perceive and assess their environment. Either to recognize their potential insect host, to evaluate host health status, or to locate insect damaged root systems, entomopathogenic nematodes have evolved the ability to respond to diverse volatile organic compounds to successfully and efficiently achieve their life cycle. Our recent understanding of these interactions and the identification of several of these environmental cues allows us to manipulate agroecosystems to enhance biological control potential of entomopathogenic nematodes. Focusing on plant emitted cues, we will discuss how to exploit these finely tuned signals to manage major insect pests threatening crop yield and food security in various systems. We will also explore some ideas on how to use this knowledge in the (close) future of digital farming.

ORAL SESSION 9

'Omics' in nematology

Revealing the "box" code: the spatial and temporal regulation of plant-parasitic nematode pathogenicity.

Clement Pellegrin[1] (cp678@cam.ac.uk), Helen Beasley[1], Badou Mendy[2], M.Shamim Hasan[3], Divykriti Chopra[2], Clarissa Hiltl[2], Huiyan Chen[2], Volkan Cevik[2], Oliver Chitambo[2], Florian M.W. Grundler[2], Shahid Siddique[3], Sebastian Eves-van den Akker[1]

[1] *Department of Plant sciences, University of Cambridge, Cambridge, United Kingdom*; [2] *Molecular Phytomedicine, University of Bonn, INRES, Bonn, Germany*; [3] *Department of Entomology and Nematology, University of California, Davis, California, United States*

Plant-parasitic nematodes are an important group of plant pathogens that threaten current and future food security. Among them, the cyst nematodes parasitize some of the most important crop species. To cause disease, cyst nematodes inject effector proteins into the plant. Nematode effectors are produced primarily in either the dorsal or sub-ventral glands. Hundreds of dorsal gland effectors are unified by a six base-pair non-coding DNA motif, termed the "DOG box". In a recent effort, a transcription factor that can recognize the DOG-box was identified: a candidate "DOG box reader" that could explain the spatial regulation of the dorsal gland effectors. However, subsets of these effectors are also expressed at different stages of the lifecycle, implying the existence of additional temporal regulators. In order to identify the genetic signatures and corresponding readers of gene expression in different gland cell and at different times of infection, we used Pacbio DNA-seq and Illumina RNA-seq to reconstruct the genome and lifestage-specific transcriptome of *Heterodera schachtii* infecting the model plant *Arabidopsis thaliana*. We conducted a trans-kingdom transcriptome-wide analysis of temporal expression to identify candidate regulators that will be functionally validated by RNAi interference. This time-resolved, multi-kingdom, transcriptomic analysis provides an in depth view of host:pathogen transcriptional interaction during infection. Temporal clustering of these expression data has revealed key landmarks of the infection cycle that were cross-referenced with effector annotations to categorize subsets of the effector repertoire into pseudo-functional 'waves'. We have identified host and parasite transcription factors that are tightly co/anti-regulated with these 'waves', and therefore represent attractive candidate temporal regulators of parasitism.

Keywords: Transcriptomic - Cyst - Nematode - Transcription factor - Pathogenicity.

Parthenogenomics - using population genomics to understand the evolution of parthenogenetic triploid nematodes.

Laura Villegas (lvilleg1@uni-koeln.de), Laura Pettrich, Ann-Marie Waldvogel, Philipp Schiffer

Department for Biology, University of Cologne, Cologne, NRW, Germany

Asexual reproduction is believed to lead to the accumulation of deleterious mutations (Muller's ratchet), and reduced heterozygosity due to the absence of recombination. *Panagrolaimid* nematodes display different modes of reproduction. Sexual reproduction through distinct males and females, asexual reproduction through parthenogenesis found in the genus *Panagrolaimus,* and hermaphroditism, found in *Propanagrolaimus.* Here, we compared genomic features of free-living nematode populations with different modes of reproduction isolated from geographically distant regions to study genomic diversity and genome-wide differentiation.
We firstly, estimated genome-wide spontaneous mutation rates per genome for a polyploid parthenogenetic *Panagrolaimus* strain and a diploid hermaphroditic *Propanagrolaimus* species via mutation-accumulation-lines. Secondly, we calculated population genomic parameters including nucleotide diversity and fixation index (Fst) between populations of asexually and sexually reproducing nematodes. Thirdly, we used phylogenetic network methods on sexually and asexually reproducing *Panagrolaimus* strains to understand evolutionary relationships between them. The estimated mutation rate was slightly lower for the asexual strain, as expected for taxa with this reproductive mode. Natural polyploid asexual strains revealed higher nucleotide diversity. Despite their common ancestor, a gene network revealed a high level of genetic differentiation among asexual strains. The elevated heterozygosity found in the triploid parthenogens could be explained by the third genome copy. Given their tendentially lower mutation rates it can be hypothesized that this is part of the mechanism to evade Muller's ratchet. In conclusion, our findings in parthenogenetic triploid nematode populations seem to challenge common expectations of evolution under asexuality.

Keywords: Parthenogenesis - Sexual reproduction - Mutation accumulation - Population genomics.

Evolutionary and comparative genomics of tropical root-knot nematodes.

Georgios Koutsovoulos[1] (georgios.koutsovoulos@inra.fr), Corinne Rancurel[2], Laetitia Zurletto[1], Martine Da Rocha[2], Djampa K. Kozlowski[1], Benjamin Noël[3], Patrick Wincker[3], Etienne G.J. Danchin[1]

[1] *IPN, ISA, INRAe, CNRS, Université Côte d'Azur, Sophia Antipolis, France*; [2] *PlantBIOs, ISA, INRAe, CNRS, Université Côte d'Azur, Sophia Antipolis, France*; [3] *Genoscope, Institut de Biologie François-Jacob, CEA, Université Paris-Saclay, Évry, France*

Root-knot nematodes (RKN; genus *Meloidogyne*) are plant parasitic species that cause huge economic loss in the agricultural industry and affect the prosperity of communities in developing countries. Interestingly, in this genus, the most detrimental species to agriculture are deemed to have abandoned meiosis and consequently sexual reproduction. The level of this parasitic and ecological success seems surprising given the obligatory clonal reproduction. Indeed, fully asexual reproduction in animals is extremely rare and the few clonal species are considered evolutionary dead ends with very limited adaptive potential. In order to advance our understanding of the factors involved with their astonishing parasitic success, we have sequenced three root-knot nematode species (*M. incognita*, *M. javanica*, and *M. arenaria*) with Oxford Nanopore Technologies (ONT). The ONT long reads allowed more congruent genome assemblies that were previously unfeasible due to the peculiar evolutionary history of these genomes (polyploid hybrids) [1]. Here, we are going to present the latest genomic resources for RKN that enable more robust and powerful analyses. In more detail, the genomic structure in these species consists of duplicated and divergent homologous regions (collinear blocks) that have been clearly separated during the assembly. Highly-contiguous genome assemblies give us the opportunity to survey gene neighborhoods and define stable segments that persist through evolutionary time as well as genes and gene sets that are free to accumulate changes or disappear. Therefore, we find longer stretches of these blocks that enable us to perform more accurate phylogenomic analyses in order to unravel past evolutionary events. These analyses allow us to chart the genome structure dynamics that occur in *Meloidogyne* delivering a comprehensive analysis of the association between structural gene clusters and function.

Keywords: Comparative genomics - Root-knot nematodes - Phylogenomics.

References:

- [1] Blanc-Mathieu et al., 2017, PLOS Genetics, 13(6): e1006777.

Meloidogyne hapla with improved assembly and genome annotation.

Pallavi Shakya[1] (pshakya@ucdavis.edu), Alison Coomer[2], Valerie Williamson[2], Shahid Siddique[3]

[1] *Siddique Lab, Plant Pathology Department, University of California Davis, Davis, California, United States*; [2] *Siddique Lab, Plant Pathology Department, University of California Davis, Davis, United States*; [3] *Siddique Lab, Entomology Department, University of California Davis, Davis, United States*

Root-Knot Nematodes (RKNs) of the genus *Meloidogyne* represent a major class of plant-parasitic nematodes. RKNs are sedentary endoparasites that spend most of their life cycle feeding from permanent feeding sites in a plant's vascular root system. Despite their tendency to reproduce asexually, the increase in adaptability and pathogenicity of RKNs on new hosts and new agricultural environments has remained a mystery to date. With the ability to parasitize almost every vascular plant, RKNs alter plant development, immunity, and physiology through a plethora of effector proteins secreted by their gland cells. Currently, there is a significant gap in available RKN genetic models for well-annotated reference genomes. With the help of these genomes we can characterize candidate genes responsible for virulence and adaptability of these nematodes. A genome sequence draft is available for *Meloidogyne hapla*, which reproduces by facultative meiotic parthenogenesis, i.e., they reproduce sexually in the presence of males. Due to their mode of reproduction, outcrossing and selfing is possible in *M. hapla*, which facilitates the production of nearly homozygous lines between virulent and avirulent strains, thus allowing the segregation of virulence genes. Previously, the whole genome shotgun approach was used to sequence the genome of *M. hapla*. With a genome size of 54Mbp*, M. hapla* encodes 14,420 genes among which, 544 genes have been newly predicted and 3751 protein-coding genes have been recently updated [1,2]. My research project aims to build on the available nearly homozygous lines of *M. hapla* to construct a more contiguous reference genome utilizing PacBio's HiFi and HiC sequencing technology for chromosome level assembly. I also aim to improve the genome annotation by sequencing the full-length transcriptome of mixed life stages in *M. hapla* via IsoSeq technology. These long read sequencing methods will yield an output of a reference genome with an improved assembly. This result will facilitate the further hypotheses building and investigation of biological questions related to the parasitizing capability of Meloidogyne species and the characterization of novel virulence genes.

Keywords: Root Knot Nematodes - Genomics - Parasitsm - Sequencing - Annotation.

References:

- [1] Guo, Yuelong, David McK Bird, and Dahlia M Nielsen. 2014. Worm 3 (May): e29158. https://doi.org/10.4161/worm.29158
- [2] Opperman, C. H., D. M. Bird, V. M. Williamson, D. S. Rokhsar, M. Burke, J. Cohn, J. Cromer, et al. 2008. Proceedings of the National Academy of Sciences 105 (39): 14802–7. https://doi.org/10.1073/pnas.0805946105

Genome evolution of *Aphelechoides besseyi* strains.

Cheng-Kuo Lai[1] (lihowfun@gmail.com), Peichen J. Chen[2], Yi-Chien Lee[1], Huei-Mien Ke[1], Hsin-Han Lee[1], Chan-Yi Ivy Lin[1], Isheng Jason Tsai[1]

[1] *Biodiversity Research Center, Academia Sinica, Taipei, Taiwan*; [2] *Department of Plant Pathology, National Chung Hsing University, Taichung, Taiwan*

Aphelenchoides besseyi is a plant parasitic nematode belonging to Aphelenchoidae, Clade 10b of Nematoda. It can infect the aboveground parts of over 200 species of plants in 35 genera and is problematic in many tropical and subtropical rice-growing areas. Different isolates of *A. besseyi* exhibited different pathogenicity to plants. It was probably due to the adaption of nematodes to different hosts and environments. However, the knowledge about genomic evolution of the parasites in this clade is still obscure. In this study, we used comparative genomics to understand what genetic differences were accumulated in the genome and what mechanism caused the genetic differences among these species. We sequenced four *A. besseyi* strains, two isolated from bird's-nest ferns and the other two from rice, and two other *Aphelenchoides* sp. The *A. besseyi* genomes were assembled using Pacbio and nanopore technologies into 44.7-50.7 Mb. The 6 Mb inter-host strains difference are mainly due to the expansion of transposable elements at intergenic region of rice isolates. Copy number variations of glycoside hydrolase (GH) families were identified between different strains of *A. besseyi*. In addition, when compared with five other plant parasites, GH45 was only found in Aphelenchoidae including our isolates. Among these nematode species, the isolates from fern is the only plant-parasitic nematode with coexistence of GH5 and GH45. We suggest that the different copy number of GH families among *A. besseyi* strains may be a result of different infectivity to plants. The environment or host may play an important role in the retention of GH families in nematode genomes during evolution, and it might be an ongoing differentiation within this foliar nematode species.

Keywords: Comparative genomic - Synteny - Glycoside hydrolase - *Aphelenchoides besseyi* - Plant-parasitic nematodes.

A genome-wide comparison between two Dutch *Globodera pallida* populations.

Martijn Holterman[1] (martijn.holterman@wur.nl), Mark Sterken[1], Joris Van Steenbrugge[1], Sven Van den Elsen[1], Anne Van Diepeningen[2], Geert Smant[1], Johannes Helder[1]

[1] *Laboratory of Nematology, Wageningen University and Research, Wageningen, Netherlands*; [2] *Biointeractions and Plant Health, Wageningen University and Research, Wageningen, Netherlands*

Potato cyst nematodes belong to the most serious yield-limiting biotic factors in potato production worldwide. Potato cyst nematode is a single common name that refers to two related *Globodera* species, *Globodera rostochiensis* and *G. pallida*. Due to their limited mobility and their long life cycle – typically one generation per year followed by a diapause of several months – host plant resistances have been a remarkably effective control measure for decades. However, recently a number of *G. pallida* populations have been found in Germany and The Netherlands that are no longer stopped by the current arsenal of resistance genes. Mapping the genomic variation present in virulent and avirulent populations can help to gain more insight into genetic factors underlying these differences in virulence. In 2014, a genome of the *G. pallida* population Lindley was published (Cotton *et al*., 2014), but the number of scaffolds (» 6,800) makes it hard to use it as a reference for pinpointing virulence characteristics. We used PacBio sequencing to generate a new reference genome for the old (= barely exposed to modern resistance genes) Dutch *G. pallida* population D383 (Pa2). This resulted in an assembly of 113 Mb consisting of 163 scaffolds with an N50 of 2.9 Mb. HiSeq sequencing was used to polish the assembly and determine the genetic variation present in the population. This variation was compared with the variation present in another Dutch population, Rookmaker (Pa3), which was sequenced using HiSeq and mapped on the new reference genome (≈ 250x coverage). The variation within and between these populations was compared as well as the distribution of this variation across the genome.

Keywords: *Globodera pallida* - Genome - Potato - Cyst nematode - Populations.

References:

- Cotton et al., 2014. Genome biology 15:R43.

S9-PF1

In search of a common target for control of nematode and aphid pests.

Maria Maqsood (maria.maqsood@hotmail.com), John Fosu-Nyarko, Michael G.K Jones

State Agricultural Biotechnology Centre, Murdoch University, Perth, Western Australia, Australia

Plant parasitic nematodes (PPNs) and aphids cause substantial yield losses of crops globally. Diversity in their habitats and evolutionary adaptations have led to a remarkable resilience in these pests. Both nematodes and aphids are biotrophic pests, and thus require an intimate association with their host plants to access nutrients. To initiate and maintain parasitism, both groups of pests secrete 'effectors' (proteins or small molecules) through their stylets: these are capable of altering host cell structure and function, evade defences, and enable feeding and development. Sedentary endoparasitic nematodes secrete effectors from oesophageal gland cells through their stylet into plant cells, and convert root cells into specialised feeding sites. During probing and feeding aphids secrete watery and gelling saliva, both of which also contain effectors. To understand whether PPNs and aphids employ similar effectors or mechanisms for parasitising plants, we have used comparative bioinformatics to identify putative effectors from genomic and transcriptomic data and have characterised candidate proteins *in silico* by predicting common structures, domains and possible localisations in host cells. So far, fourteen common proteins with similar structures, or with different structures but similar predicted functions, have been identified in both PPNs (*Meloidogyne incognita* and *Heterodera schachtii*) and aphids (*Myzus persicae*), suggesting common mechanisms by which these pests interact with plant hosts.
These putative common effectors have been characterised in these pests using RNA interference: this insight may provide an opportunity to develop a single common control strategy to silence activity of common essential genes required by both groups of pests for successful parasitism.

Keywords: RNAi - Aphid effectors - Nematode effectors - Pest Control - Gene silencing.

S9-PF2

Metagenomics mining improves analysis of horizontal gene transfers involved in parasitic function in plant-parasitic nematodes.

Carole Belliardo[1] (Carole.Belliardo@inrae.fr), Adrien Deceneux[1], Corinne Rancurel[2], Marc Bailly-Bechet[3], Etienne G.J. Danchin[1]

[1] *INRAe, Institut Sophia Agrobiotech, Sophia Antipolis, France*; [2] *CNRS, Institut Sophia Agrobiotech, Sophia Antipolis, France*; [3] *Université Cote d'Azur, Sophia Antipolis, France*

Plant-parasitic nematodes (PPN) are responsible for 11% losses on edible crops worldwide each year [1]. To reduce their damage, we need to progress in understanding these plant pests' evolution. These organisms have acquired fungal and bacterial genes involved in essential parasitic functions [2,3]. For example, some of these genes encode enzymes that are involved in the degradation of the plant cell wall. Furthermore, transcriptomic, biochemical and proteomic data support the functional 'domestication' of microbial genes in these plant pathogens [4]. Although several central questions remain, like how are HGT distributed in the host genomes or what are the donor organisms? We think our current understanding is currently limited by the lack of representation in public databases of the diversity of genes present in the soil, the natural environments of PPN. Our objective is thus to mine soil metagenomes to obtain a better representation of the pool of genes present in this environment [5].
A preliminary study highlighted the benefits of this approach to detect HGT. Indeed, 392 new candidate HGT were detected in the *M. incognita* genome by mining 3,601 soil metagenomes. However, the accuracy of the taxonomic annotation in public metagenomes was too low to gain information on the possible donors. Furthermore, we realized that eukaryotic species were underrepresented in these metagenomic datasets, probably because the current bioinformatics pipelines have been optimized for prokaryotes. Therefore, to gain a better insight on the identity of the possible donors we applied stringent quality checks and sought to improve the taxonomic annotation. To better account for HGT of eukaryotic origin, we also detected and sorted contigs of eukaryotic origin in metagenome assemblies to treat them separately with a dedicated pipeline for gene prediction. Finally, most of the available data generated with short reads methods are far from providing complete microbial genome reconstructions. To improve the completeness of these genome assemblies and provide a more realistic and accurate representation of the pool of genes present in the actual environment of PPN, we sequenced the metagenome of five soils in fields infected by *M. incognita* using long-read technology. The predicted proteins from the latest PPN genome assemblies will be searched against the pool of annotated proteins detected in the soil to further information on the extent and origin of HGT in these species.

Keywords: Root-knot nematodes - Metagenomic - Horizontal gene transferts - Parasitisme - Evolution.

References:

- [1] Savary, S. et al., 2019, Nat. Ecol. Evol. 3, 430–439.
- [2] Danchin, EG. et al.,2010, Proc Natl Acad Sci Usa 107, 17651–17656.
- [3] Haegeman A. et al.,2011 Mol Plant Microb Interact 24, 879-887.
- [4] Jaubert, S., et al., 2002, FEBS Lett. 522, 109–112.
- [5] Handelsman, J., 2004,Microbiol Mol Biol Rev., 68: 669–685.

S9-PF3

Development of mitometagenomics protocols for the enhancement of nematode identification and biodiversity study.

Eli Gendron[1] (egendron@ufl.edu), Joseph Sevigny[2], Thomas Powers[3], W Kelly Thomas[2], Dorota Porazinska[1]

[1] *Department of Entomology and Nematology, University of Florida, Gainesville, FL, United States*; [2] *Hubbard Center for Genome Studies, University of New Hampshire, Durham, NH, United States*; [3] *Department of Plant Pathology, University of Nebraska, Lincoln, NE, United States*

The use of DNA metabarcoding techniques have greatly increased our understanding of biodiversity on the planet. 18S and 28S ribosomal DNA (rRNA) gene markers have been most commonly used to identify and classify members of the phylum Nematoda, but these markers provide limited taxonomic resolution [1]. In recent years, the use of the mitochondrial DNA (mtDNA) *cox1* gene has increasingly been applied for the identification and classification of nematode species [2]. However, due to the hyper-variability in the priming regions of the *cox1* locus, targeted metabarcoding of *cox1* in the use biodiversity studies is cost prohibitive [3]. In contrast, mitochondrial metagenomic shotgun sequencing is able to overcome this limitation [4,5]. To test an approach for the rapid identification of complex nematode communities without the need for targeted sequencing, we used a series of 8 nematode mock community types consisting of differing abundances of 26 morphologically and molecularly confirmed nematode species representing different feeding traits, life stages, and phylogenetic relationships to examine the rate and accuracy of species and gene recovery. Utilizing our newly developed *de novo* metagenome assembly pipeline and reference database, mitochondrial sequences were first assembled and then classified at the species level at >98% sequence identity. Of the 26 expected species used to construct the mock communities we recovered 24 species. Full mitochondrial genomes were recovered for 24% of our expected community, suggesting all genes do not assemble with equal efficiency. Predictably, *cox1* was the best gene for species recovery, followed by *cox2* and *cytb.* In contrast *nad4l, nad2,* and *nad6* were highly limited in their use. However, together the recovery of additional genes beyond *cox1* greatly reinforced species identification. We also evaluated the rate of false positive errors, and found zero errors when including a query sequence coverage cutoff of 80%. We conclude that mitochondrial metagenomics is highly effective in the recovery of the nematode biodiversity without the need for the costly and time consuming development of primers, but efficacy is gene and species dependent.

Keywords: Diversity - Metabarcoding - Mitochondrial metagenomics - Nematode species identification - Sequencing.

References:

- [1] Waeyenberge et al. 2019. Diversity. 11(4).1-22.
- [2] Powers et al. 2018. Journal of Nematology. 50(3).399-412.
- [3] Humphreys and Elling 2015. Gene. 560(2):173-183.
- [4] Tang et al. 2014. Nucleic Acids Research. 42(22)1-13.
- [5] Sevigny et al. 2021. Ecological Indicators 121:106973.

ORAL SESSION 10

Advances in precision agriculture: instrumentation and nematode IPM applications

Precision nematode management, a modern approach to incorporate new technologies and chemistries to control nematodes.

Pablo A. Navia Gine (pablo.navia@adama.com)

Product Development, ADAMA U.S., Wellington, FL, United States

Monitoring soil population densities of nematodes provides the quantitative framework needed to make variable rate nematicide application recommendations. In fabrication of the prescription map needed for nematicide application, one of the biggest challenges to address is how to obtain a representative soil sample for population estimation. The objective of the Precision Nematode Management approach is to create a nematode zones map based on a prediction of nematode soil densities from representative samples. With this purpose, a new algorithm is being developed to identify appropriate sampling points. The sampling point will be generated based on the zone-based soil sampling system. In zone-based strategies, the goal is to collect samples within each zone. Management zones can be identified using three or more data layers: a. Soil information systems (soil texture, soil compaction, moisture availability and root zone dept) that are collected using electromagnetic sensors and geophysical probes b. Stress analysis from the previous season (plant stress, vegetation indexes) collected with drone or satellite imaging c. nematode samples from soil sampling points. After accepting user input, and sampling results, nematode population levels, zones of high, medium and low nematode pressure are automatically created and georeferenced and expressed in a shape file map that can be exported to a sprayer or shank to perform variable rate applications (VRA). The final step for the precision nematode management approach is to select nematicides that can be utilized within the approach, such as with fluensulfone, a new chemistry from ADAMA that have enough flexibility that can be applied with standard equipment for a Pre-Plant Incorporated (PPI) spray or chemigation for VRA. For the case study presented, aerial imaging was generated to assess plant stress from the previous season using a DJI matrice 100 equipped with a ZENMUSE X5 camera. An orthomosaic map was generated using the dronedeploy platform and images were analyzed using the ADAMA EagleEye powered by Agremo on-line platform. Then SIS mapping system from Trimble Inc. was used to make soil maps and after cross-referencing with zone-based soil samples, nematode management zones maps were generated. The nematode management zones maps were used to prescribe a nematicide (NIMITZ) to apply variable rates in different fields with the use of a PPI broadcast application.

Keywords: Variable Rates - Precision agriculture - Nematode management - Precision nematode management.

Assessing crop impacts and nematode management options using NDVI and canopy greenness from aerial imaging.

Joseph Noling (jnoling@ufl.edu)

Entomology / Nematology Department, University of Florida, IFAS, CREC, Lake Alfred, FL, United States

Since 2010, fifteen field studies were conducted in Florida with the objectives to 1) compare commercial harvest yields with relative strawberry yields determined from end of season counts of plants within four size categories, or with NDVI (Normalized Difference Vegetation Index) using a plant reflectance optical sensor measuring green canopy cover; and or with independent assessments of canopy greenness using ArcGIS analysis of aerial images; and 2) to relate differences in plant sizing, NDVI, and canopy greenness to the performance of different chemical soil treatments. Strawberry plots were commercially harvested every 3 days from November to March of every season and total yield tabulated. Plant numbers in four plant size categories were also systematically enumerated and recorded at 12 to 15 m intervals in monitored fields at season's end of each year. Plant size categories, measured as average canopy diameter, were dead (0), small (<20 cm), medium (>20 and < 30 cm) and large (>30 cm). Using plant sizes, fumigant treatment evaluations based on relative yield were determined in commercial fields with recurring histories of sting nematode problems. Hyperspectral reflectance plant imaging technology was used to relate differences in strawberry crop yield, relative yield (based on plant sizing) to within row, green vegetative cover using separate estimates of greenness and canopy cover. Besides NDVI, canopy greenness per plant row was also determined using ArcGIS analysis of aerial images. Strawberry yields from commercial harvest of large plots were always well correlated with NDVI, canopy greenness, and relative yield values determined from plants of different sizes within plots and fields displaying Sting nematode stunting. Overall, changes in strawberry crop productivity due to sting nematode and chemical soil treatment were effectively determined within experimental plots, and on a farm and industry-wide basis, from post-harvest assessments of counts of different plant sizes, NDVI, and canopy greenness measurement. This presentation will show how digital imaging and greenness analysis can be used in lieu of NDVI to provide a quantitative measure of strawberry yield and to provide growers guidance on suitable nematode management options.

Keywords: Remote sensing - Precision agriculture - Crop loss assessment - Strawberry.

Estimating field populations of *Heterodera schachtii* from hyperspectral signatures of sugar beet canopies.

Matthias Daub[1] (matthias.daub@julius-kuehn.de), Kai Schmidt[2]

[1] *Julius Kuehn-Institute, Elsdorf, Germany*; [2] *Nemaplot, Bonn, Germany*

The beet cyst nematode (BCN) *Heterodera schachtii* is widespread throughout the core production areas in central Europe where it frequently reaches damage relevant population densities in sugar beets. Today nematode tolerant sugar beet cultivars largely have replaced standard cultivars in areas with severe nematode infestation. Nevertheless, the combination of high BCN population densities, high temperatures and drought conditions have yield impacts even on these tolerant cultivars. In the three year study presented here we monitored physiological leaf symptoms of different sugar beet cultivars using hyperspectral reflection sensors. By applying a unique field method, Pi densities of BCN were preset by cultivation of resistant and susceptible catch crops prior to the sugar beet cultivation resulting in Pi ranges between nearly zero and >5000 eggs and juveniles per 100 ml of soil. Three different sugar beet cultivars (tolerant, susceptible, resistant) were grown as a strip trial overlaying the completed range of Pi densities. These population densities of *H. schachtii* were monitored by soil samples taken at the beginning and at the end of season using a centrifugal flotation method. Time series of hyperspectral measurements were taken in fortnightly intervals throughout the seasons by hand held sensors connected to different spectrometers (Agrospec, ASD FieldSpec 4).The wavelength reflection data have been fitted to a specific designed model and were analysed by multivariate methods in a second step. We found sufficient correlations (r= 0.74-0.84) in both nematode sensitive cultivars between population densities assessed by the remote sensing and ground truth data. Those correlations were found in all years, but only in short time windows during the seasons which furthermore varied with each year. The three years of sensor information revealed or confirmed multiple interactions between BCN population densities, cultivar, year and vegetation period, while the nematode effect is mainly hidden behind the more dominant factors. Having identified the basics and statistical tools of such a complex procedure, we see the perspective of future approaches to detect spatial distributions of BCN in the field and to identify significant loci before the onset of visible symptoms.

Keywords: Hyperspectral signatures - BCN - Population densities - Field trials.

The use of infrared spectroscopy and machine learning tools for detection of *Meloidogyne* infestations.

Mayamaru Guerra[1] (mguerra@utb.edu.co), Ana Maria Casassa[2], Nestor Cubillan[3], Ernesto San Blas[4]

[1] *Universidad Tecnológica de Bolivar, Cartagena, Colombia*; [2] *Universidad del Zulia, Maracaibo, Venezuela*; [3] *Universidad del Atlantico, Barranquilla, Colombia*; [4] *Independent Researcher, Santiago, Chile*

Plant parasitic nematodes are generally soil-borne pathogens which attack plants and cause economic losses in many crops. The infested plants show non-specific symptoms or many times could be asymptomatic. Therefore, diagnosis must proceed by taking soil and root tissue samples. Here we show that a combination of different infrared spectra analysis and machine learning algorithms can be used to detect plant parasitic nematode infestations before symptoms become visible, using leaves instead of roots and soil as samples. In this work, we found that tomato and guava plants infested with *Meloidogyne enterorlobii* produced different spectral patterns compared to uninfested plants. Using partial spectra from 1450 to 900 cm-1 as fingerprint zone, principal component analyses indicated that after 5 (tomatoes) or 8 weeks (guava), plants with no visible symptoms of infestations were positively diagnosed. To improve the early detection response, we used machine learning modelling. Support vector machine (SVM) was used to obtain more robust, accurate and self-adaptively models. The SVM model contained 34 support vectors, 17 for each level. The overall performance of the model was > 97 % and the total accuracy was significantly higher, demonstrating the absence of chance prediction. The best prediction of infestation was obtained at the 2nd and 4th weeks for tomatoes and guavas, respectively. Hence, the diagnostic time was reduced by half. The combined application of these techniques reduces the processing time from field to laboratory and shows enormous advantages by avoiding root and soil sampling.

Keywords: Artificial intelligence - *Meloidogyne enterolobii* - FTIR-ATR - Support Vector Machine - Genetic Algorithms - Plant parasitic nematodes.

Multiseasonal modelling of plant-nematode interactions reveals efficient resistance deployment strategies.

Vincent Calcagno (vincent.calcagno@inrae.fr), Suzanne Touzeau, Ludovic Mailleret, Samuel Nilusmas, Caroline Dijan Caporalino, Philippe Castagnone

Institut Sophia Agrobiotech, Sophia Antipolis, France

Root-knot nematodes (RKNs) are soil-borne polyphagous pests with major impact on crop yield worlwide. Resistant crops efficiently control avirulent RKNs, but favour the emergence of virulent forms. Virulence being associated with fitness costs, susceptible crops counter-select virulent RKNs. In this study we identify optimal and efficient rotation strategies between susceptible and resistant crops to control RKNs and maximize crop yield.

- We developed an epidemiological model describing the within-season dynamics of avirulent and virulent RKNs on susceptible or resistant plant root-systems, and their between-season survival. The model was fitted to experimental data and used to predict yield-maximizing rotation strategies, with special attention to the impact of epidemic and genetic parameters.
- Crop rotations were found to be efficient under realistic parameter ranges. They were characterised by low ratios of resistant plants, and were robust to parameter uncertainty. Rotations provide significant gain over resistant-only strategies, especially under intermediate fittness costs and severe epidemic contexts.
- Switching from the current general deployment of resistant crops to custom rotation strategies could not only maintain or increase crop yield, but also preserve the few and valuable R-genes available to us.

Combining traditional and precision agricultural tools to improve management of plant-parasitic nematodes in cotton production.

Zane Grabau[1] (zgrabau@ufl.edu), R. Barocco[2], C. Liu[1], D. L. Wright[2], I. Small[2]

[1] *Entomology and Nematology Dept., University of Florida, Gainesville, FL, United States*; [2] *North Florida Research and Education Center, University of Florida, Quincy, FL, United States*

Plant-parasitic nematodes are a major pest of cotton (*Gossypium hirsutum*) worldwide and in the United States. High quality and timely evaluation of nematode management solutions—such as nematicides and resistant cultivars—are a key piece in management of these pests. Traditional methods of assessing nematode populations and damage (soil nematode counts, visual ratings) are labor-intensive, making it difficult to employ them to get accurate assessments on large scales. Precision agriculture tools such as remote sensing imaging facilitated by small Unmanned Aerial Systems (sUAS) may be one means of improving precision and speed of evaluating nematode management solutions in row crops such as cotton. These tools are commonly used for other pests on row crops in the United States, but nematode-specific evaluation in cotton is needed. The objective of this research was to assess remote sensing as a tool for evaluating nematode management solutions in cotton with validation by on-the-ground assessments including agronomic measurements and nematode populations. The platform for this research was a series of small plot field trials conducted at three locations in Florida over two years. Treatments used for proof of concept were known nematicide treatments available in Florida cotton production including 1,3-dichloropropene fumigation, in-furrow aldicarb treatment, and no treatment. Remote sensing and on-the-ground data was collected regularly during the growing season and are being used to evaluate these methods.

Models for map-based prediction of *Pratylenchus penetrans* based on soil variables.

Ann MacGuidwin[1] (aem@plantpath.wisc.edu), Gary Pack[2], Ibrahim Saeed[1]

[1] *Plant Pathology, University of Wisconsin, Madison, WI, United States*; [2] *Biostatistics and Medical Informatics, University of Wisconsin, Madison, WI, United States*

Pratylenchus penetrans is a common and serious problem in sandy vegetable fields. Most management practices for *P. penetrans* have a high economic and environmental cost. Farmers are interested in adjusting application rates of products to match the level of need. This is possible if they have a spatial representation of nematode population density at the field scale. An appealing alternative to intense sampling for nematodes is to map nematode distributions using a proxy for actual counts, such as yield or soil factors related to nematode ecology. A field study showed that electrical conductivity maps had some explanatory value for nematode counts but were insufficient for variable rate fumigation. We then expanded our study to 322 sampling locations in eight fields. Models were developed for three dependent variables: spring counts of *P. penetrans* from 100 cc soil, root fragments in that volume, and the total count from both habitats. We collected data on 34 physical and chemical soil variables, combinations of variables in two-way interactions, and computed variables such as spatial filters (> 1200 variables total) from each sample. We first determined which variables were useful for predicting the negative binomial distribution of the nematode count data. Different variables were selected for the three models and all showed a reasonable fit to the nematode count data, based on model deviance. We then used the soil data to develop a global logistic model that predicted if nematode densities at locations of unknown status exceed a designated threshold value. The unknown locations were a subset of the original data points that were set aside and not used for model development. Twelve models to predict the three dependent variables at each of four density thresholds were selected. The models had reasonable predictive power, showing ROC values greater than 0.84. Our next step was to develop an ordinal logistic model that used soil factors to generate maps predicting the spatial distribution of multiple nematode density classes. For example, the model to predict low, medium, and high categories of total nematode counts included seven soil variables, 15 two-way interactions, and three computed variables. Variables with p-values < 0.05 were % sand, Zn, S x Zn, and altitude x Fe. Across all fields, the low “do not treat” class showed 75% accuracy. This study demonstrates valid approaches to predicting the spatial distribution of *P. penetrans* using soil data.

Keywords: Spatial analysis - Root lesion - Logistic regression - Edaphic - Population density.

ORAL SESSION 11

Nematode-vector relationships

Capsid determinants involved in the specific transmission of the grapevine fanleaf virus by its nematode vector, *Xiphinema index.*

Gérard Demangeat (gerard.demangeat@inrae.fr)

SVQV UMR-A 1131, INRAe, Université de Strasbourg, Colmar, France

Grapevine fanleaf virus (GFLV) and arabis mosaic virus (ArMV) are two major causal agents of grapevine degeneration disease that occurs in vineyards worldwide [1,2]. These two closely related nepoviruses are specifically transmitted by the two distinct ectoparasitic nematodes *Xiphinema index* and *X. diversicaudatum* respectively following a semi-persistent mode [2]. These transmission features suggest a highly selective molecular recognition mechanism between virus and nematodes. To identify structural domain(s) and residues within the GFLV capsid protein (CP) that are responsible for this specificity, we hypothesized that candidate residues are likely exposed at the external surface of virions, different between GFLV and ArMV isolates but highly conserved among GFLV isolates. Based on GFLV and ArMV atomic structures determined by 3 structural approaches, 5 putative candidate regions (R), termed R1 to R5 ranging from 4 to 11 residues were identified. These 5 putative GFLV domains were substituted by their ArMV counterparts in the GFLV CP [1,2]. Chimeric viruses that led to a systemic infection were evaluated for their transmissibility by *X. index*. Transmission tests of these chimeric viruses revealed that R2 and R4/R5 are two viral determinants involved in the transmission specificity [3]. Moreover, the characterization of a weakly transmitted GFLV variant, revealed the importance of a single CP mutation in Gly297Asp, belonging to R5 region in the transmission process. The comparisons of GFLV and ArMV three-dimensional structures associated to functional approaches allow us to map the viral determinants along a viral pocket-like structure named ligand binding pocket (LBP) as the putative binding site that will be recognized by a specific nematode ligand [1,3]. To further characterize this LBP in the transmission process, nanobodies, small peptides derived from heavy-chain-only antibodies found in camelids and directed against GFLV, were produced. Four GFLV specific nanobodies map with the LBP and will be used in competitive virus/nematodes interactions tests. Our results will be discussed in regards to the transmission specificity of GFLV and ArMV by their respective nematode vectors.

Keywords: Transmission - Nepovirus - *Xiphinema* - 3D structure - Viral determinant.

References:

- [1] Schmitt-Keichinger et al., (2017). In Grapevine viruses: molecular biology, diagnostics and management, Meng, B.; Martelli, G. P.; Golino, D. A.; Fuchs, M., Eds. Springer: Cham, Switzerland. pp 83-107.
- [2] Andret-Link et al., (2017). In Grapevine viruses: molecular biology, diagnostics and management, Meng, B.; Martelli, G. P.; Golino, D. A.; Fuchs, M., Eds. Springer: Cham, Switzerland. pp 505-529.
- [3] Belval et al., (2019). Viruses. 11 (12), 1146.

The importance of nematode dauer in the adaptation to its life cycle and ecological niche.

Lieke Vlaar[1] (l.e.vlaar@uva.nl), Mehran Rahimi[1], Andre Machado Bertran[2], Aska Goverse[2], Jan Kammenga[2], Harro Bouwmeester[1]

[1] *Plant Hormone Biology, University of Amsterdam, Amsterdam, Netherlands*; [2] *Laboratory of Nematology, Wageningen University, Wageningen, Netherlands*

Nematodes need to adjust their development, according to their obligate, facultative or non-parasitic life style. A crucial mechanism to achieve this is dauer, which has been studied mostly in the non-parasitic model nematode *Caenorhabditis elegans*. In *C. elegans*, dauer is a facultative dispersal stage only entered if environmental conditions are unfavourable, such as under crowding, lack of food or suboptimal temperatures. The worm can stay in this stage up to 5 months or until these conditions improve. Dauer worms are resistant against many types of stress and do not grow or feed.

The dauer stage in obligate parasites shows much resemblance to that of *C. elegans*, but it is non-facultative. It is required for the parasite to survive until a suitable host is within reach and molecular cues from the host can induce exit from the dauer stage. Since dauer release by necessity results in parasitism, it is remarkable that it has been studied only sparsely, in contrast to the extensively studied dauer entry. Moreover, parasitic dauer has until now only been acknowledged in animal parasites, whereas the economically highly relevant plant parasitic nematodes have been ignored in this light. For example, eggs of cyst nematodes of the *Heteroderidae* family can enter a dauer-like stage which can last up to 20 years, in order to prevent hatching until a suitable host plant grows nearby. Mechanisms regulating these processes are unknown.

We are unravelling these mechanisms in the plant parasitic cyst nematodes using *C. elegans* as model and other (parasitic) nematodes for comparison. Hereto we have developed dauer exit assays - based on automated image analysis and machine learning - that allow testing of the effects of compounds and conditions on dauer release in the model *C. elegans,* as well as in the plant-parasitic cyst nematodes. In addition, we do comparative analyses of dauer and dauer-like stages across the phylum *Nematoda* and study the importance of dauer in adaptation to a specific ecological niche. A better insight in these processes should lead to a better understanding of the evolution of the plant-cyst nematode interaction and will possibly also provide the basis for new control methods.

Keywords: Dauer - Parasite - Bioassay - Cyst nematode - *C. elegans*.

References:

- Crook, 2014. Int. J. Parasitol. 44, 1–8.
- Cassada & Russell, 1975. Dev. Biol. 46, 326–342.
- Perry & Clarke, 1981. Parasitology 83, 435–449.

Linking biodiversity and pathogen abundance in natural nematode populations.

Lisa van Sluijs[1] (lisa.vansluijs@wur.nl), Jessica N. Sowa[2], Hala Tamim El Jarkass[3], Aaron Reinke[3], Wenjia Wu[4], Robbert van Himbeeck[1], Sem Aslan[1], Joost A. G. Riksen[1]

[1] *Laboratory of Nematology, Wageningen University and Research, Wageningen, Netherlands*; [2] *Department of Biology, West Chester University of Pennsylvania, West Chester, United States*; [3] *Department of Molecular Genetics, University of Toronto, Toronto, Canada*; [4] *Key Laboratory of Vegetation Restoration and Management of Degraded Ecosystems, South China Botanical Garden, Chinese Academy of Sciences, Guangzhou, China*

Natural nematode populations harbor a broad range of pathogens that infect them, such as oomycetes, microsporidia and viruses. These pathogens shape nematode communities and may prove beneficial in situations where parasitic nematodes are controlled by their natural enemies and explain phenomena of suppressive soils. In macroscopic species, differences in pathogen susceptibility of hosts contribute to the 'dilution effect' where higher host biodiversity contributes to lower pathogen prevalence, but this effect is unidentified for microscopic species. Here, we investigated the effect of nematode biodiversity on the prevalence of nematode-infecting pathogens. Therefore, nematode populations were collected from decaying plant substrates in habitats where the model nematode species *Caenorhabditis elegans* was previously identified. Using (wild) *C. elegans* as a target species, we could use fluorescent reporter strains to physically identify bacterial, microsporidian and viral pathogens. To compare pathogen prevalence with nematode biodiversity, nematodes were counted and the number of species present was identified. Current work involves amplicon sequencing of the DNA that was collected from the wild populations to identify additional pathogens and characterize nematode species present. For nematode community characterization, nearly the complete eukaryotic SSU is amplified using PCR and sequenced by Nanopore sequencing. Moreover, many of the obtained (bacterivorous) nematodes and their pathogens have been maintained in the lab since field collection. Future work involves using this nematode-pathogen library for multispecies infection experiments comparing and testing correlations found in the field.

Keywords: Field communities - Pathogens - Biodiversity - *Caenorhabditis elegans* - Nanopore sequencing.

Unraveling the biotic interactions between *Bursaphelenchus xylophilus*, *Pinus pinaster* and nematophagous fungi, *Esteya* spp.

David Pires[1, 2] (david.pires@iniav.pt), Cláudia Vicente[1, 2], Manuel Mota[2], Maria L. Inácio[1, 3]

[1] *Laboratory of Nematology, Instituto Nacional de Investigação Agrária e Veterinária (INIAV,I.P.), Oeiras, Portugal*; [2] *Laboratory of Nematology, MED/University of Évora, Évora, Portugal*; [3] *GREEN-IT Bioresources for Sustainability, ITQB NOVA, Oeiras, Portugal*

The pinewood nematode (PWN), *Bursaphelenchus xylophilus*, is the causal agent of pine wilt disease (PWD), a serious threat to pine forests worldwide, especially in Asia and Europe. Due to its economic relevance and global dissemination, an enormous effort is devoted to study the virulence of *B. xylophilus* and the epidemiology of PWD. Understandably, the non-specificity of synthetic chemicals and lack of environmental-friendly options to control this parasite has led to an increasing focus on antagonists capable of suppressing the PWN. *Esteya* spp. are nematophagous fungi and promising biocontrol agents against the PWN. There are currently two described species: *E. vermicola* and *E. floridanum*. In a quest to explore their potential applications in maritime pine, *Pinus pinaster*, the main and most affected species in Portuguese forests, we take a look at plant host, nematode and antagonistic fungi interactions. To this end, various interaction assays were considered: specifically, fungus-plant, fungus-nematode and fungus-fungus. These preliminary results will help us select the most promising *Esteya* spp. for biocontrol strategies in this plant model and allow us to devise new ways to manage PWD in the foreseeable future.

Funding: this work is funded by the Portuguese Foundation for Science and Technology (Fundação para a Ciência e a Tecnologia – FCT), under the project "PineEnemy – Exploring the NEmatode-MYcobiota interactions in Pine Wilt Disease" (PTDC/ASP-PLA/28724/2017), and FEDER (Fundo Europeu de Desenvolvimento Regional) (LISBOA-01-0145-FEDER-028724), through Lisbon and Alentejo Regional Operational Programmes. David Pires is also supported by the FCT PhD Fellowship 2021.08030.BD.

Keywords: Biological control - Biological interactions - Maritime pine - Nematophagous fungi - Pinewood nematode.

References:

- Mamiya, Y. (1983). Pathology of the pine wilt disease caused by Bursaphelenchus xylophilus. Annual Review of Phytopathology, 21: 201–220.
- Mota, M., Braasch, H., Bravo, M.A., Penas, A.C., Burgermeister, W., Metge, K. & Sousa, E. (1999). First report of Bursaphelenchus xylophilus in Portugal and in Europe. Nematology, 1: 727–734.
- Vicente, C., Espada, M., Vieira, P. & Mota, M. (2012). Pine wilt disease: a threat to European forestry. European Journal of Plant Pathology, 133: 89–99.
- Inácio, M.L., Nóbrega, F., Vieira, P., Bonifácio, L., Naves, P., Sousa, E. & Mota, M. (2014). First detection of Bursaphelenchus xylophilus associated with Pinus nigra in Portugal and in Europe. Forest Pathology doi:10.1111/efp.12162.
- Pires, D., Vicente, C.S.L., Inácio, M.L. & Mota, M. (2022). The potential of Esteya spp. for the biocontrol of the pinewood nematode, Bursaphelenchus xylophilus. Microorganisms, 10(1): 168.

Molecular nematode-viroid-fungus interface on *Celosia argentea* and *Solanum lycopersicum*.

Pakize Gok Guler[1] (pakize.gokguler@tarimorman.gov.tr), Refik Bozbuga[2], Semiha Yuceer[1]

[1] *Biological Control Research Institute, Adana, Turkey*; [2] *Faculty of Agriculture, Plant Protection Department, Eskisehir Osmangazi University, Eskisehir, Turkey*

Many pathogens including nematodes, viroids and fungi may found together infecting, the same plant. Root-knot nematodes (*Meloidogyne* spp*)* are important plant parasitic nematodes infect many plants including ornamental plant species. *Meloidogyne chitwoodi* is one of the most damaging nematode species on ornamental plants. A viroid, *Potato spindle tuber viroid* (PSTVd) causes damage to several plants. The soil-inhabiting fungus *Fusarium proliferatum* also severely damages plants and may cause death. However, the effect of three (nematode, viroid and fungus) pathogens on *Celosia argentea* and *Solanum lycopersicum* plants has not been fully understood. Therefore, this study was aimed to determine the nematode-viroid-fungus effects on an important and widely used ornamental plant, *Celosia argentea,* and *Solanum lycopersicum* using molecular and screening assay. Gene expression of pathogen related genes were also investigated following the infection by the pathogens (nematode, viroid, fungus) at 1, 3, 7, 14 days post infection using qRT-PCR. RNA extraction and screening assays were performed following mechanical inoculation, and specific primers were used for the detection of the viroid. Both molecular and screening assays were achieved following the infection on different days post-infection. The replication of viroid RNA in infected plants, infection and severity of nematode, and fungus infection were determined. Gene expressions were determined to be at altered levels. Morphological data also showed that some detrimental effects and decreased growth parameters, including plant height, fresh weight and shoot width were observed in pathogen-inoculated compared to non-infected plants. In conclusion, advanced genetic studies are needed to explain effects of *M. chitwoodi* in combination with of viroids (PSTVd) and fungus *(F. proliferatum)* on different hosts.

Keywords: *Melodiogyne chitwoodi* - Potato spindle tuber viroid - *Fusarium proliferatum* - *Celosia argentea* - Gene expression.

Diversity of fungal communities associated with *Pinus pinaster* infected by *Bursaphelenchus xylophilus.*

Cláudia Vicente[1, 2] (claudia.vicente@iniav.pt), Jorge Faria[2], Helena Bragança[2], Luís Bonifácio[2], Edmundo Sousa[2], Margarida Espada[1], Steve Woodward[3], Anna Maria Vettraino[4], Filomena Nóbrega[2], Maria L. Inácio[2]

[1] *MED, Mediterranean Institute for Agriculture, Environment and Development, Institute for Advanced Studies and Research, Évora University, Évora, Portugal*; [2] *INIAV, I.P., National Institute for Agrarian and Veterinarian Research, Oeiras, Portugal*; [3] *School of Biological Sciences, University of Aberdeen, Aberdeen, United Kingdom*; [4] *DIBAF, University of Tuscia, Viterbo, Italy*

Pine wilt disease (PWD) is one of the most threatening diseases to conifer forests worldwide [1,2] and results from the complex interaction between the pinewood nematode (PWN) *Bursaphelenchus xylophilus*, its causal agent, the insect-vector from the *Monochamus* genus, and the host tree *Pinus* spp. [3,4]. In the later stages of PWD, the PWN undertakes an obligatory mycetophagous phase, feeding on fungi that colonize the declining trees [3]. Limited studies available indicate that fungi dominating the dead pines, in particular blue-stain fungi from Ophiostomatales order, may influence the population and development of PWN carried by the insect vector [5]. Hence, our aim is to identify and characterise the culturable fungal community putatively associated with the PWN life cycle using a polyphasic approach on *Pinus pinaster*. A total of 242 fungal isolates were obtained from symptomatic and asymptomatic *P. pinaster* collected in three PWD-affected areas, in Seia, Tróia and Lezírias, from which representative isolates of each species were characterized on the basis of morphology and selected molecular markers (the internal transcribed spacer region, ITS; β-tubulin, BT; elongation factor 1-α, TEF; and calmodulin, CAL) for phylogenetic inference. Our data shows that fungal communities of symptomatic *P. pinaster* are less biodiverse and dominated by Ophiostomatales in contrast with the communities of asymptomatic *P. pinaster* trees. The most abundant ophiostomatoid fungal species were *Ophiostoma ips*, followed by *Leptographium* and *Graphilbum* sp. Exploring the diversity of multispecies interactions in PWD provides a valuable insight into the successful invasion and adaptation of PWN.
This work was conducted in the national project PineEnemy: Exploring the NEmatode-MYcobiota interactions in Pine Wilt Disease (LISBOA-01-0145-FEDER-028724) which focus on the characterization of the structure and dynamics of the nematode-fungi interactions, and to which extend it can be targeted to disrupt the disease cycle.

Keywords: *Bursaphelenchus xylophilus* - Fungi - Ophiostomatales - *Pinus pinaster* - Pine Wilt Disease.

References:

- [1] Vicente C, Espada M et al. (2012) Eur J Plant Pathol 133: 89-99.
- [2] Mota and Vieira (2008) Springer Netherlands. DOI: 10.1007/978-1-4020-8455-3.
- [3] Futai (2013) Annu Rev Phytopathol 51:61-83.
- [4] Inácio et al. (2015) For Path 45: 235-238.
- [5] Vicente, CSL.; Soares, M; Faria, JMS; Ramos, AP; Inácio, ML (2021) JoF: 7, 780. DOI: 10.3390/jof7090780.

Durability of muscadine-derived resistant material to *Xiphinema index* and first detection of sexual reproduction events of the nematode under controlled conditions.

Van Chung Nguyen[1] (nchung.ppri@gmail.com), Jean-Pascal Tandonnet[2], Samira Khallouk[3], Cyril Van Ghelder[1], Ulysse Portier[1], Mohamed Youssef Banora[4], Nathalie Ollat[2], Maria-Das-Dores Lafargue[2], Daniel Esmenjaud[1]

[1] *INRAE, Universite Cote d'Azur, CNRS, ISA, Sophia Antipolis, France*; [2] *INRAE, UMR EGFV, Villenave d'Ornon, France*; [3] *ONSSA, Rabat, Morocco*; [4] *Ain Shams University, Cairo, Egypt*

Because of the ban of nematicides, control alternatives are urgently needed against plant-parasitic nematodes affecting major crops and breeding for plant varieties carrying natural resistance (R) is promising. Nevertheless, in perennial crops, the long plant-nematode interaction increases the risk for R breaking and ensuring R durability is a real challenge. In grapevine, the dagger nematode *Xiphinema index* has a high economical impact by transmitting *Grapevine fanleaf virus* (GFLV) and resistant rootstocks to this vector are being selected, using *Muscadinia rotundifolia* in particular as an R source, to arrest or delay GFLV transmission. In order to optimize *in fine* this strategy, the R durability has been studied under controlled conditions in F1 and BC1 muscadine-derived resistant material previously obtained from either hardwood-cutting or *in vitro* propagation. After inoculation with a mix, in equal proportions, of four lines representative of the *X. index* diversity, the nematode multiplication has been monitored yearly in plants between 3 and 6 years. The nematode reproduction factor increased over years in resistant material and in reference susceptible material of *in vitro* origin while it remained low and stable in resistant material obtained from cuttings. Thus the *in vitro*-culture steps preceding acclimatization into soil may have induced an improved host suitability, presumably through a modified root architecture. As the four lines retained had either microsatellite specific alleles or combinations of alleles, putative hybridization events between each other were identifiable. In each combination of accessions and propagation types, the multilocus genotype of a sample of nematode individuals was analyzed yearly from plants aged 3 to 6 years. Hybrid individuals between different lines were detected for the first time under controlled conditions, albeit their occurrence was low but increasing over years. Nevertheless, in our experimental conditions, selection pressure exerted by resistant plants on the nematode did not appear to have a quick drastic effect on sexual reproduction. Differences in aggressiveness between mixed lines were also observed, even though none of them was eliminated or conversely became highly predominant over years.

Keywords: *Xiphinema index* - Durability - Hybridization - *In vitro* - Hardwood-cutting.

ORAL SESSION 12

Chemical control of nematodes

NanoEngineering gone viral: plant virus-nanotechnologies for precision farming.

Nicole F. Steinmetz (nsteinmetz@ucsd.edu)

NanoEngineering, UC San Diego, La Jolla, CA, USA

Nanoscale engineering is leading to new materials and approaches for use in medicine and environmental applications. Viruses are playing a special role in these developments because they can function as prefabricated nanoparticles naturally evolved to deliver cargos to cells and tissues. Our research is focused on *plant* viruses and virus-based nanoparticles thereof. We have developed a library of plant virus-based nanoparticles and through structure-function studies we are beginning to understand how to tailor these materials appropriately for applications in medicine and agriculture. A particularly exciting avenue pursued is the repurposing of plant viruses for precision farming to maintain plant health. We employ principles of nanomedicine to formulate plant virus-based drug carriers [ACS Nano 2017, p. 4719] and to target pesticides to nematodes residing deep in the soil. Based on the zwitterionic nature of the protein-based nanoparticle, the plant virus-based formulation outperforms free pesticide and contemporary nanoparticles in soil mobility experiments [Nature Nanotechnology 2019, p. 712]. Based on our established protocols, the plant virus-based nanotechnology can be engineered for the delivery of small molecule drugs as well as protein or gene therapies. The engineering design space is impeccable; and we are just beginning to understand the potential of disarmed (non-infectious) plant virus-based nanoparticles for applications targeting plant health and the environment.

Keywords: Nanotechnology - Drug carrier - Soil mobility - Plant virus - Precision farming.

References:

- Steinmetz et al, Nature Nanotechnology 2019, p. 712.
- Steinmetz et al, ACS Nano 2017, p. 4719.

Tymirium™ – The Story of a Powerful New Molecule.

Brigitte Slaats[1] (brigitte.slaats@syngenta.com), Matthias Gaberthueel[2], Olivier Loiseleur[3], Anthony Flemming[4], Torsten Luksch[5]

[1] *Marketing, Syngenta Crop Protection AG, Basel, Baselstadt, Switzerland*; [2] *Crop Protection Research and Development, Syngenta Crop Protection AG, Basel, Baselstadt, Switzerland*; [3] *Chemical Research, Syngenta Crop Protection AG, Stein, Aargau, Switzerland*; [4] *Crop Protection Research, Syngenta UK Ltd, Bracknell, Warfield, United Kingdom*; [5] *Chemical Research, Syngenta Crop Protection AG, Stein, Aargau, Switzerland*

Syngenta was the first agrochemical company world-wide to develop and launch a seed treatment nematicide containing the active ingredient abamectin, in US cotton production in 2006. Since then there has been a shift in nematode management practices from in-furrow application to use of seed-applied solutions for a variety of field crops such as corn, cotton and soybean. As an agrochemical company, Syngenta Crop Protection AG has been committed to pioneer the discovery of a next-generation seed treatment nematicide, as a follow-up to the current commercial product Avicta™ (AI: Abamectin). Research and development set the ambitious goal to discover a molecule that would deliver a highly efficacious nematicide, with a novel mode of action and an improved human and environmental safety profile, for a selective and reliable control of a broad range of plant-parasitic nematodes. The conventional drench-applied centric nematicide screening techniques used across the industry were not pursued. Instead the logic was reversed, and seed treatment application tests were placed at the front end of the selection screen. From an initial series of a few thousand compounds belonging to various chemical classes and tested in the screening process for their potential nematicidal activity, a promising chemistry class was identified. With the help of the Mode of Action (MOA) Identification Platform at the Research site in Jealott`s Hill, Inhibition of Complex II was confirmed as the MOA and key amino acids in the protein, which are important for the effect were identified. Encouraged by these findings, Research chemistry expanded efforts in this area, designing and synthesizing several new subclasses of the original lead. Accurate custom design was made possible by the establishment of a three-dimensional model of the nematode Complex II through computational chemistry. Seedcare development improved and expanded testing capabilities, such as a micro-plot testing platform, against a number of target nematode pests to secure the delivery of high quality field data to be able to accurately judge the potential of the field compounds. A small multinational, multidisciplinary science team worked in a focused, efficient way, innovating at scale to exploit Syngenta expertise around the world. The outcome of this research work is the Tymirium™ molecule and its outstanding potential against a broad spectrum of plant-parasitic nematodes.

Keywords: TYMIRIUM (TM) nematicide/fungicide molecule - Chemical nematicide - Plant-parasitic nematodes - Complex II - Seed treatment.

"Wrap and Plant", a novel concept for managing plant-parasitic nematodes with banana paper in sub-Saharan Africa.

Charles Opperman[1] (warthog@ncsu.edu), Antoine Affokpon[2], Laura Cortada[3], Danny Coyne[3], Juliet Ochola[4], Baldwyn Torto[5], Richard Guenther[1], Steve Koenning[1], Reny Mathew[1], Tim Sit[1], Med Bird[6], Tahira Pirzada[7], Lokendra Pal[6], Eric Davis[1], Saad Khan[7]

[1] *Department of Entomology and Plant Pathology, North Carolina State University, Raleigh, United States*; [2] *Laboratoire d'Ecologie Microbienne/ Nematology Research Group, Université d'Abomey-Calavi, Abomey-Calavi, Benin*; [3] *International Institute of Tropical Agriculture, Nairobi, Kenya*; [4] *Chemistry Department, Kenyatta University/ICIPE, Nairobi, Kenya*; [5] *Behavioural and Chemical Ecology Unit, International Centre of Insect Physiology and Ecology, Nairobi, Kenya*; [6] *Department of Forest Biomaterials, North Carolina State University, Raleigh, United States*; [7] *Department of Chemical & Biomolecular Engineering, North Carolina State University, Raleigh, United States*

We developed an innovative, cost effective "Wrap and Plant" (W&P) methodology for crop protection from plant-parasitic nematodes. W&P technology is an innovative concept based on banana fibre paper, as a mechanism to deliver micro-doses of the nematicide abamectin to the root zone – presenting an economically viable option for smallholder growers. Importantly, incorporation of active ingredients into a lignocellulose matrix, such as banana paper, enables effective distribution of crop protection agents without interfering in smallholder farming practices. Our multi-year replicated field trial results demonstrate the efficacy and affordability of W&P technology for management of nematodes on yam and white potato crops. Yam tuber yield and quality were consistently and substantially greater than conventional farmers' practices across all on-farm sites each year, and farmers indicated that W&P treatment resulted in larger, longer, and cleaner tubers. Further, food quality and preparation of yam flour revealed a strong preference for the W&P-treated yams. Application of W&P technology resulted in significant reductions in final nematode populations compared to farmers' practice. We observed significant reductions in final yam nematode (*Scutellonema bradys*) populations in tuber peels. Not only was this largely responsible for the high tuber quality, but also indicates substantially reduced risks of post-harvest tuber damage and loss due to this nematode. Field trials were performed in white potato-growing regions in Kenya and Uganda, many conducted in fields infested with potato cyst nematode (PCN), the most important nematode pest of potato in Kenya and worldwide. Potato yield was observed to be 3-fold greater across all on-farm sites when treated with W&P compared to conventional practices, and W&P outperformed all other treatments by a substantial margin. Potato yields were consistently fourfold higher for PCN (26.41 T/ha) and RKN field trials (20.24 T/ha) than yields observed under farmers' practices (5.67 T/ha). W&P technology also provided the highest profitability for farmers, compared with other treatments. This novel and innovative technique provides a simple and effective option for nematode management in yam and potatoes, in addition to numerous other crops. We will discuss potential mechanisms for the efficacy of W&P for nematode management.

Keywords: Banana paper - Yam - Potato - Abamectin - Smallholder.

Effect of New Non-fumigant Nematicides on Different Trophic Groups of Nematodes.

David Moreira (daanmoca94@gmail.com), Johan Desaeger

Vegetable and Fruit Nematology, University of Florida, Wimauma, United States

Plant-parasitic nematodes cause more than $100 billion dollars per year in damage to crops (>14% global crop production). Non-parasitic, free-living nematodes, that co-exist with plant-parasitic nematodes in soil, are critical components of the soil food web and play important roles in nutrient cycling and making inorganic chemicals available to plants. Traditionally, especially in Florida, nematode management has relied on the use of soil fumigants and broad-spectrum insecticide/nematicides, all of which are detrimental to human health and the environment. Recently, new selective contact nematicides have emerged that offer safer alternatives, but little is known about their specific activity against different types of nematodes. Our main objective was evaluating the intrinsic activity of the new and older nematicides in a series of in-vitro assays. Different feeding groups of nematodes (bacterivore, fungivore, entomopathogenic and plant-parasitic) were exposed to varying concentrations of three new non-fumigant nematicides (Nimitz®, Salibro™ and Velum® Prime), a biological nematicide (Majestene®) and a carbamate nematicide (Vydate® *L)* and were evaluated for their effect on mortality, motility, ability to feed and ability to move through the soil. Exposure to the different nematicides was conducted using 48-well tissue culture plates, at five different time points (24h, 48h, 72h, 96h, and 168h). Living and dead nematodes were determined by adding 1 N NaOH to each well and counting the nematodes that were able to react (alive) and the nematodes that were not able to respond (dead) to 1 N NaOH. Results indicate an overall greater effect of all the nematicides on the mortality of plant-parasitic nematodes (*Meloidogyne javanica* and *Belonolaimus longicaudatus)* in comparison to free-living nematodes (*Cephalobus* sp., *Aphelenchus* sp. and *Steinernema* sp.), confirming the greater selectivity of these products. At low concentrations (1–1.25ppm, representative of soil water concentration following field applications) the overall effect on mortality of Velum® on all nematode taxa was greater than that of Nimitz® and Salibro™. Nematodes that responded to 1 N NaOH were often sluggish and lethargic; although they are technically not dead, their infectivity is likely affected. In addition to the in-vitro assays, we are currently also evaluating these products in greenhouse and field trials.

Keywords: Trophic group - Nematicide - Free-living nematodes - Plant-parasitic nematodes - Integrated management.

References:

- Tugel et al., 2000. Soil Biology Primer. 30-33.
- Bernard et al., 2017. Nematology - Concepts, Diagnosis and Control: InTech. 124-125.
- Rich et al., 2004. Nematicides: Past and present uses: CABI Publishing. 1180-1182
- Lahm et al., 2017. Bioorganic and Medicinal Chemistry Letters. 1572-1575.
- Chen et al., 2000. Journal of Nematology. 117-121.

Evaluation of sequential applications of chemical and biological nematicides in vegetable production in southern Italy.

Helmut Fuersch[1] (helmut.fuersch@bayer.com), Joachim Meyer[1], Alberto Boebel[2], Svenja Bellof[1]

[1] *Bayer AG, Monheim, NRW, Germany*; [2] *Bayer Crop Science S.r.l., Milano, Italy*

Vegetable production is an important agricultural business in many Mediterranean countries. To manage the threat of plant parasitic nematodes can be a major concern especially under protected conditions. In the future this issue is likely to become even more challenging when broad-spectrum fumigants are not available anymore. As alternatives there are some chemical products in the market as well as products, which are based of natural occurring bacteria, fungi and plant extracts ("biologics"). The aim of our investigations was to demonstrate that a combination of chemistry and biologics in an application program offers a value for the grower.

At four commercial greenhouse sites, representing key production areas for fruiting vegetables in Southern Italy, trials were started in 2017. Different sequential applications of nematicides were compared in their performance on parameters such as plant growth, damage caused by root knot nematodes (*Meloidogyne* spp.), population dynamics of the nematodes and yield.

The main goal of these multiyear experiments was to evaluate the performance of fluopyram (Velum Prime©), either in a sequence with commercial standards or in a sequence with a biologic product based on the fungus *Purpureocillium lilacinum* (BioAct Prime©).

Tomatoes were cultivated at each location as a long cycle crop once per year in alternation with cucumber or melon (short cycle crop). At all locations this cropping sequence was performed for the years 2017 and 2018; at two sites the trials continued at least till end of 2019.

It could be shown that two applications of Velum Prime© in alternation with commercial market standards resulted in a 36 % yield gain, while the addition of repeated applications of BioAct Prime© could increase the yield up to 48 % (average of all trial sites). All additional assessments like plant stand, root gall ratings and nematode population counting corresponded positively with the observed yield gain.

Results from this trial series demonstrate the beneficial effects combining biologics and chemicals for an integrated nematode control program. Investigations will continue to measure long-term effects on nematode population dynamics.

Keywords: Root-knot nematodes - Vegetables - Multiyear trials - Integrated nematode control.

The distinct profile of the inhibitory effects of fluensulfone on *Globodera pallida* hatching.

Emily Feist (ef1g12@soton.ac.uk), James Kearn, Yogendra Gaihre, Vincent O'Connor, Lindy Holden-Dye

Biological Sciences, University of Southampton, Southampton, United Kingdom

Fluensulfone (FLS) is a novel nematicide with a distinct mode of action against plant parasitic nematodes. It has previously been shown to have irreversible nematicidal effects on the model organism *C. elegans* (Kearn et al. 2014); however, it is most potent and efficacious against plant parasitic nematodes, including *Meloidogyne* spp. (Oka et al. 2009; Oka et al. 2012) and the potato cyst nematode, *Globodera pallida* (Kearn et al. 2017). Here, we utilize *in vitro* hatching assays to investigate fluensulfone's ability to inhibit *Globodera pallida* hatching, relative to the efficacy of other distinct classes of nematicides.

At low concentrations of FLS (1µM), full inhibition of hatching is achieved. This effect was reversible at concentrations ≤5µM (1.46 ppm), partially reversible at concentrations ≤50µM (14.6 ppm) and irreversible at the maximum concentration tested of 500µM (146 ppm). Treatment with 500µM FLS results in the loss of encysted juvenile structure giving rise to a granulated appearance consistent with necrosis, suggesting a nematicidal effect. At low concentrations, although inhibition of hatching is reversible, more than 50% of the juveniles that hatch from cysts pre-treated with FLS have reduced motility. Intriguingly, hatching initiated by root diffusate is arrested when egg populations are subsequently exposed to FLS.

Fluensulfone is a potent inhibitor of hatching and impacts the viability of the J2s emerging from the cysts. This activity, and the previously described impaired motility and metabolism of hatched juveniles, show that fluensulfone's distinct mode of action among existing nematicides intersects at two pivotal steps of the parasitic life cycle. Investigating the effect of FLS on hatching will improve understanding of its novel crop protecting action and inform on its mechanism of action.

Keywords: Abamectin - Aldicarb - Fluopyram - Mode of action - Plant Parasitic Nematode.

References:

- Kearn et al., 2014. Pesticide Biochemistry and Physiology. 109: 44-57.
- Oka et al., 2009. Pest Management Science. 65: 1082-1089.
- Oka et al., 2012. Pest Management Science. 68: 268-275.
- Kearn et al., 2017. Pesticide Biochemistry and Physiology. 142: 83-90.

S12-PF1

Salibro™ (Reklemel™ active): A novel nematicide for the control of *Meloidogyne* spp. in key annual and perennial crops in North America & Mexico.

Bo Braxton[1] (bo.braxton@corteva.com), Leonel Aviles[2], Sunil Tewari[3], Julian Mejia[4], Laura Smith[5], Tim Thoden[6], Yannis Stamatas[7]

[1] *Corteva Agriscience, Indianapolis, United States*; [2] *Corteva Agriscience, Guadalajara, Mexico*; [3] *Corteva Agriscience, Fresco, United States*; [4] *C, Bogota, Columbia*; [5] *Corteva Agriscience, Toronto, Canada*; [6] *Corteva Agriscience, München, Germany*; [7] *Corteva Agriscience, Halandri (athens), Greece*

Salibro™ (which contains Reklemel™ active) is a new, highly effective and selective nematicide from Corteva Agriscience™ for the control of plant-parasitic nematodes. Specificity for nematodes coupled with absence of activity against the target sites of commercial nematicides suggests that Salibro™ has a novel mode of action. It is the first member from the novel chemical class of sulfonamide nematicides. Salibro™ has been extensively tested in field trials in North America & Mexico over the past years. In those trials, Salibro™ has proven extremely effective against a range of important plant-parasitic nematode species. This paper will focus on the fit of Salibro™ in nematode management programs in key annual and perennial crops and report single and across trials data on root protection, crop safety & yield.

S12-PF2

Discovery and characterization of bioactivated nematicides for selective control of parasitic nematodes.

Jessica Knox[1, 2] (jess.knox@utoronto.ca), Savina R. Cammalleri[1, 2, 3], Andrew R. Burns[1, 2], Jack M.P. Castelli[1, 2], Megan Kitner[4], Inga Zasada[4], Peter J. Roy[1, 2, 3]

[1] *Department of Molecular Genetics, University of Toronto, Toronto, Canada*; [2] *Terrence Donnelly Centre for Cellular and Biomolecular Research, University of Toronto, Toronto, Canada*; [3] *Department of Biochemistry, University of Toronto, Toronto, Canada*; [4] *Horticultural Crops Research Laboratory, USDA-ARS, Corvallis, United States*

Plant-parasitic nematodes (PPNs) pose an enormous threat to global food security, and their management is critical in optimizing crop yields. Chemical nematicides are an important tool in combatting PPN infestation. While effective, many commercial nematicides are facing increasing restrictions and bans due to poor phylum-selectivity, jeopardizing our ability to control PPNs. Our objective is to identify and characterize novel small molecules with nematode-selective toxicity that hold potential as safe and effective agricultural nematicides.
To identify nematode-selective small molecules, we performed a screen in the model nematode *Caenorhabditis elegans* for molecular scaffolds that are metabolically bioactivated into a toxic product by cytochrome P450s (CYPs) within the worm. These molecules act as pro-nematicides and offer potential for nematode-selective activity due to the phylogenetic diversity of CYP enzymes. We identified numerous bioactivated molecular scaffolds featuring a disubstituted oxadiazole core structure. These hit scaffolds include the commercial nematicide Tioxazafen and a novel compound we have named Cyproside.
Cyproside is broadly active *in vitro* across all nematode species tested and does not disrupt viability in the non-target systems examined to date. We tracked the metabolism of Cyproside using HPLC and found it to be robustly metabolized by *C. elegans* and diverse PPN species including *Meloidogyne hapla, Ditylenchus dipsaci,* and *Pratylenchus penetrans.* Cyproside is not metabolized in the non-target systems examined to date. Using chemical CYP inhibitors, we have found that Cyproside kills PPNs in a CYP-dependent manner, suggesting that CYP-catalyzed bioactivation is required for activity in PPNs. We conclude that the nematode-selective activity of Cyproside is likely achieved via nematode-specific metabolic conversion into a lethal product. Currently, we are further characterizing the Cyproside metabolites produced in *C. elegans* and PPN species using LC-MS/MS to better understand the metabolic bioactivation and potential mechanism-of-action of this novel selective nematicide.

Keywords: *Caenorhabditis elegans* - Small molecule nematicides - Chemical screening - Drug metabolism.

ORAL SESSION 13

'Omics' in nematology (continued)

Uncovering the "dark matter" of worm biology: a universe of signaling molecules.

Frank Schroeder (fs31@cornell.edu)

Chemistry and Chemical Biology, BTI/Cornell University, Ithaca, United States

How can we complement genomics and proteomics of animal model organisms such as *C. elegans* or Drosophila with a comprehensive structural and functional annotation of the corresponding metabolomes? Growing evidence suggests that small molecules of largely undetermined structure play important roles in the biology of microorganisms, animals, and their mutual interactions, affecting key physiological pathways that regulate lifespan, development, and metabolism, with estimates for the number of metabolites ranging from 10,000 to several 100,000 in a single species.
Our goal is to develop a systematic approach for linking small molecule metabolites directly with genotypes and probable biological functions. In this lecture, I will present several novel types of small molecule signals we recently identified in the nematode *C. elegans*. We found that, using simple building blocks from conserved primary metabolism and a strategy of combinatorial assembly, *C. elegans* and other nematode species create complex molecular architectures to regulate almost every aspect of their life history. The resulting signaling molecules can be active at femtomolar concentrations, changing behavior, development, or lifespan by modulating conserved insulin or nuclear hormone receptor signaling. Additionally, recent studies revealed that nematode-derived small molecules serve diverse functions in interactions with plants and microorganisms. The discovery of new types of modular, primary metabolism-derived signaling molecules in *C. elegans* provides a strong incentive for a comprehensive re-analysis of metabolism in higher animals, including humans.

Keywords: Metabolomics - Signaling - Mass spectrometry - Chemical ecology - Second messengers.

Juxtaposition of extreme genomic variability and stability in HYP effectors of potato cyst nematodes.

Unnati Sonawala (us275@cam.ac.uk), Helen Beasley, Sebastian Eves-van den Akker

University of Cambridge, Cambridge, United Kingdom

Plant-parasitic nematodes have evolved large repertoires of "effector" proteins to manipulate their host. Like many effectors, HYP effectors are secreted into the host and are necessary for infection. Since their discovery in 2014[1], a non-exhaustive set of over 70 unique HYP genes have been cloned. HYPs can be categorised into three subfamilies, each consists of a 'hyper-variable' domain characterised by variable number, organisation, and subfamily-specific repeats. The hyper-variable domain is flanked by 410 and 94 nucleotides that have remained 95% identical for ~30 million years of evolution. Moreover, no two individual nematodes tested have the same genetic complement of HYPs. To the best of our knowledge, no known genetic mechanism can account for both the hyper-variable domain organisations and gene number variation spanning an order of magnitude among sisters of the same population, while at the same time maintain the conserved flanking domain sequence and structure. The objective of this study is to understand how it is possible for the genome of an animal to permit such variability in a single domain of a gene family, while maintaining the stability of the genome in general, and HYPs in particular. We used long-read PacBio and Nanopore direct RNA and DNA sequencing to address the variability and genomic organisation of HYP effectors. We identified individual reads that contain entire HYP genes, complete with adjacent sequence. We mapped these reads to highly-contiguous genome assemblies of multiple *G. pallida* and *G. rostochiensis* populations/pathotypes. Strikingly, we found that all HYP-containing reads can be assigned to two loci. Reads containing subfamilies HYP2 and HYP3 map to a single ~40kb locus, where HYP2s vary in copy number (but not sequence) from 2 to 3 per haplotype depending on the *G. pallida* population. Importantly, those reads containing members of the largest and most sequence diverse HYP subfamily (HYP1) map to a different ~2kb locus. We conclude that the dominant majority of all variation observed to date is allelic: in the case of HYP1, this means that there is a single locus containing at least 50 hyper-diverse alleles, but probably many more. We hypothesise that: i) there must be some as yet unknown genome biology underlying HYP loci to allow such juxtaposition of genomic variability and stability, and ii) HYPs must carry out an important and unusual biological function to necessitate such unusual characteristics.

Keywords: Cyst nematode - Effector - Genomics - Parasitism.

References:

- [1] Akker, S.E. den, Lilley, C.J., Jones, J.T., Urwin, P.E., 2014. PLOS Pathogens 10, e1004391.

Analysis of the banana root transcriptome in response to root-knot nematode infection and water deficit.

Robert Miller[1] (robertmiller@unb.br), Leticia Dias de Freitas[2], Jose Dijair Antonino[3], Gabriel Sergio Costa Alves[1], Claudia Fortes Ferreira[4], Mauricio Antonio Coelho Filho[4], Edson Perito Amorim[4], Roberto Coiti Togawa[5], Priscila Grynberg[5]

[1] *Biologia Celular, Universidade de Brasilia, Brasilia, Brazil*; [2] *Fitopatologia, Universidade de Brasilia, Brasilia, Brazil*; [3] *Departamento de Agronomia-Entomologia, Universidade Federal Rural de Pernambuco, Recife, Brazil*; [4] *Embrapa Cassava and Tropical Fruits, Cruz Das Almas, Brazil*; [5] *Embrapa Recursos Genéticos e Biotecnologia, Brasilia, Brazil*

Banana (*Musa* spp.) is one of the world's most important crops, contributing to global food security. In contrast to wild species, many commercial varieties are sterile, with limited genetic variation resulting in cultivars frequently susceptible to pests and diseases. Endoparasitic root-knot nematodes (RKN) (*Meloidogyne*spp) cause considerable losses in banana, with *Meloidogyne incognita* a predominant species in the commercially important Cavendish subgroup. Water deficit is also an important limiting factor for the cultivation of *Musa* spp., resulting in losses of up to 60% in affected areas in susceptible cultivars. Accessing the genomic information of improved genotypes can further our understanding of biotic and abiotic stress tolerance mechanisms, facilitating the development of genetic improvement measures. The aim of this study was to identify and characterize genetic components of resistance responses to *M. incognita,* tolerance responses to drought stress, and combinatory biotic and abiotic stress responses in the Embrapa *Musa acuminata* genotype 75, an improved material tolerant to drought stress. Root RNA samples were extracted from 15-day old plants, following inoculation (DAI) with nematode J2 juveniles, following drought stress, after simultaneous cross-stresses, and in non-challenged controls. CDNA libraries were paired-end sequenced using lllumina NovaSeq 6000 S4 technology. A total of 34,317 transcripts were mapped against the *M. acuminata* ssp. *malaccensis*var. Pahang reference genome (version 4). A total of 573 differentially expressed genes (DEGs) were identified in nematode-infected libraries, in relation to non-inoculated controls. Enrichment analysis revealed over-represented GO terms related to catalytic activity, oxidoreductase activity, carbohydrate binding, and nucleic acid binding. A total of 2834 DEGs potentially involved in drought stress were identified, with over-represented GO terms comprising response to stimulus, catalytic activity and nucleic acid binding. A total of 1683 cross stress-responsive DEGs were also identified, within enriched GO categories of response to stimulus, catalytic activity, oxidoreductase activity and nucleic acid binding. Advances in understanding of the plant responses to multiple biotic, abiotic and cross-stresses will benefit the development of broad-spectrum stress-tolerant improved *Musa* genotypes.

Keywords: Banana - Root-knot nematode - Drought - Transcriptome.

miR167-ARF8, an auxin responsive couple involved in the formation of galls induced by root-knot nematodes in tomato.

Yara Noureddine[1] (yara.noureddine@inrae.fr), Martine Da-Rocha[1], Clémence Médina[1], Mohamed Zouine[2], Joffrey Mejias[1], Michael Quentin[1], Pierre Abad[1], Bruno Favery[1], Stéphanie Jaubert-Possomai[1]

[1] *INRAe, Université Côte d'Azur, CNRS, ISA, Sophia Antipolis, France*; [2] *INRAe, INP-ENSAT, Castanet-Tolosan, France*

Root-knot nematodes, genus *Meloidogyne*, induce the dedifferentiation of root vascular cells into giant and multinucleated feeding cells. The formation of these feeding cells is the result of an extensive reprogramming of gene expression in targeted root cells as shown by analyses of transcriptomes from galls or giant cells from various plant species. MicroRNAs are small (20-22nt) non coding RNAs that regulate gene expression at the post transcriptional level. Small RNAs and messenger RNAs (mRNAs) from tomato (*Solanum lycopersicum*) galls and uninfected roots at 7 and/or 14 days post infection (dpi) were sequenced. *De novo* prediction of miRNAs in tomato genome was performed by using three algorithms (MirCat, Mirdeep Plant and Shortstack). A statistical analysis of the microRNAs expressed in galls and uninfected roots identified 174 miRNAs that are differentially expressed in galls. mRNA targeted by microRNAs in tomato galls were identified by a specific sequencing of cleaved mRNAs by using the degradome approach. Integrative analyses combining the sequencing data from small RNAs, degradome and transcriptome identified 12 microRNAs-mRNAs that are robust candidate to be involved in the tomato response to the nematode. Moreover, the role of miR167 and its targets, the auxin response factors 8, in the development of feeding cells was established by functional analysis.

Keywords: Root-knot nematodes - Galls - MicroRNAs - Tomato - Epigenetic.

Genetic variation in *Globodera pallidalinked* to virulence in North-West Europe.

Mark Sterken (mark.sterken@wur.nl), Joris van Steenbrugge, Dennie te Molder, Casper Van Schaik, Stefan van de Ruitenbeek, Sven Van Elsen, Geert Smant

Laboratory of Nematology, Wageningen University and Research, Wageningen, Netherlands

The cyst nematode *Globodera pallida* is an important pest in potato. Originating from South America, the species has been introduced in Europe approximately one and a half centuries ago. After its introduction the nematode has spread over the continent and is currently found in almost all potato producing countries. Furthermore, various control measures, including resistance genes, have been applied to keep populations from causing yield losses. We suspect that these control measures have shaped the genetic variation within and between European populations. Here, we present genetic links between virulence arisen by artificial and agronomical selection of Dutch *G. pallida* populations.

We conducted container tests characterizing the virulence of 15 recently collected *G. pallida* populations on a total of 28 potato cultivars. We determined that these new populations were virulent and that (remaining) potato resistance could be characterized by two main clusters. Selection of two of these populations on the potato cultivar Seresta resulted in an increased virulence. The genetic diversity of the selected and the other field populations was determined based on variant calling using a newly constructed *G. pallida* reference genome. We thus identified a region that was consistently selected for and associated with increased virulence on Seresta.

Our findings fit within the context of earlier observations from France and Germany, where it was found that *G. pallida* populations could be (artificially) selected for virulence. Given the concurrent rise of virulence in these populations within The Netherlands and the broader North-West of Europe is suggestive that this potential for virulence is wide-spread within the European populations.

Keywords: *Globodera pallida* - Genetic variation - Potato cyst nematode - Virulence.

Single nematode genome assemblies.

Erna King[1] (ek5@sanger.ac.uk), Lewis Stevens[1], Christopher Laumer[2], Mark Blaxter[1]

[1] *Tree of Life, Wellcome Sanger Institute, Hinxton, Cambridge, United Kingdom*; [2] *Natural History Museum, London, United Kingdom*

Genome sequencing for marine and estuarine nematode species has been challenging due to low individual input, together with difficulties isolating and identifying sufficient abundances from the field or sustaining them in culture. A novel low input technique overcomes these challenges through optimised long-range library amplification, permitting PacBio sequencing and genome assembly for single nematode specimens. Draft genomes ranging from 180 to 650 Mb in size have thus been assembled for 28 underrepresented free-living marine species with high contiguity (N50 range 22.5 to 700 kb). Transcriptome annotation is also possible as full transcript length cDNA libraries are prepared for these same specimens.
Species were sequenced in the Anoplostomidae, Enoplidae, Oncholaimidae and Tripyloididae families within the Enoplia. Within the Chromadoria, the paraphyletic Monhysterida and Araeolaimida were represented by five (3 within Linhomoeidae, 1 Sphaerolaimidae, 1 Xyalidae) and three species (1 within Axonolaimidae and 2 within Comesomatidae) respectively. Additional Chromadoria families presented include the Desmodoridae, Microlaimidae, Monhysterida and Xyalidae. These genomes were analysed together with a selection of published nematode genomes and transcriptomes to construct a phylogeny for the phylum Nematoda based on conserved protein coding genes. These results support previously published Nematoda phylogenies.

Keywords: Genome - Low-input - Phylogeny - Marine.

S13-PF1

Mining new nematode effectors interacting with plant transcription factors by Cr-Y2H.

Hui Xiang (Hui.Xiang@Ugent.be), Stojilković Boris, Gheysen Godelieve

Department of Biotechnology, Ghent university, Ghent, Belgium

Meloidogyne incognita is one of the most economically important species of plant pathogens and it causes serious loss of crop production worldwide. During different infection stages, *M. incognita* secretes hundreds of specific proteins, called effectors, into the host. In recent years, the functions of plant-parasitic nematodes (PPN) effectors have been found and illuminated. For example, they suppress the host immunity system, alter the cell physiology, and form special feeding cells called giant cells (GCs). The mechanism of giant cells formation is not entirely clear. RNA-seq has shown a significantly different gene expression in infected roots and giant cells compared to control roots. We hypothesize that this could be caused by interaction between effectors and plant transcription factors influencing downstream gene expression level. However, only a few effectors from PPN have been confirmed to interact with host transcription factors. Most of the interactions have been found by the Yeast-2-Hybrid (Y2H), the traditional method, but not enough to provide broad-scale protein-protein interaction mapping. Here, we use a massively multiplexed yeast two-hybrid method, CrY2H-seq [1]. In this system, protein interactions activate a Cre recombinase that will fuse the coding sequences of the two interacting partners and next-generation sequencing can then identify these interactions *en masse.* Compared to the traditional Y2H, CrY2H-seq is able to be applied to comprehensively screen in an «all-by-all» fashion a host transcription factor library and a *M. incognita* effector library. By bioinformatic analysis, we screened for and then successfully cloned 82 different effectors from *M. incognita.* All these effectors have higher expression levels in J2 to J4, have a predicted signal peptide, no transmembrane domain, and the WOLF PSORT predicts them to be localized in the nucleus, the same subcellular localization with transcription factors [2]. We also obtained a bait library that contains over 1800 transcription factors from *Arabidopsis thaliana* [1]. We have made the yeast bait and prey library and aim to screen now for interactions between nematode effectors and plant transcription factors.

Keywords: Root-knot nematode - Effector - CrY2H-seq.

References:

- [1] Trigg, Shelly A., et al.2017. Nature methods. 14.8 : 819-825.
- [2] Grynberg, Priscila, et al.2020. Genes. 11.11 : 1347.

S13-PF2

SMART UP – Spatial Mapping of Root Transcriptomes Upon Nematode Parasitism.

Anna Pijnacker[1] (anna1.pijnacker@wur.nl), Rik Korswagen[2], Aska Goverse[1], Geert Smant[1], Jose Lozano Torres[1]

[1] *Laboratory of Nematology, Wageningen University and Research, Wageningen, The Netherlands*; [2] *Royal Netherlands Academy of Arts and Sciences and University, Hubrecht Institute, Utrecht, The Netherlands*

Nematodes belong to the most damaging plant-pathogens worldwide and hence threaten global food security. Root-knot and cyst nematode species are the most damaging of the parasitic worms and both induce a permanent feeding structure inside the plant root. Currently, highly specific resistance genes are the most effective and sustainable option to combat nematode infections. However, this resistance is easily overcome by virulent nematode populations. Thus, there is an urgent need for novel, broad-spectrum nematode resistance to secure food production. Our lab has previously found that the cell wall integrity (CWI) receptor PERK13 alters host susceptibility to cyst and root-knot nematode infections. This project aims to study the molecular mechanisms underlying CWI during nematode-induced feeding site initiation and expansion. CWI-receptors continuously monitor the functional integrity of the plant cell by perceiving cell wall modifications and regulating both developmental and immune responses. We hypothesize that CWI-receptors are key regulators of nematode feeding site formation. Therefore, the spatiotemporal changes triggered by CWI-receptors for the formation of root-knot and cyst nematode feeding structures will be studied in *Arabidopsis thaliana* and tomato plants using state-of-the-art spatial transcriptomics i.e., RNA tomography, advanced microscopic and spectroscopic approaches. Ultimately, activation of basal immune responses upon cell wall damage recognition by CWI-receptors may result in broad-spectrum resistance against nematodes.

Keywords: RNA tomography - Microscopy - Broad-Spectrum Resistance - Sedentary Nematodes - Cell Wall Integrity.

S13-PF3

The strange chromosome ends of root-knot nematodes.

Georgios D. Koutsovoulos[1] (etienne.danchin@inrae.fr), Evelin Despot-Slade[2], Martine Da Rocha[1], Claire Caravel[1], Pierre Abad[1], Nevenka Meštrović[2], Etienne G.J. Danchin[1]

[1] *Institut Sophia Agrobiotech, INRAe, Université Côte d'Azur, CNRS, Sophia Antipolis, Sophia Antipolis, France*; [2] *Ruđer Bošković Institute, Zagreb, Croatia*

Advances in genome sequencing technologies have allowed progressing in the assembly of various nematode genomes several of which now reach or approach chromosome-level resolution. This allows studying genome and chromosome structures at unprecedented scales, including chromosome ends.
Using a combination of ONT long reads, Illumina short reads and Hi-C chromatin contact information we have improved genome completeness and contiguity for the root-knot nematodes *Meloidogyne incognita*, *M. javanica* and *M. arenaria*. For instance, previous versions of these genomes were assembled in >10,000 fragments with N50 values of dozens of kilobases. New genome assemblies now yield hundreds of fragments with N50 values of several megabases. We investigated whether telomere motif repeats could be found at the end of some contigs, thus representing chromosome ends. To our surprise, the canonical (TTAGGC)n telomere motif arrays found at the ends of *C. elegans* and other nematode chromosomes were not found at contig ends in these root-knot nematode genomes. Further search for any 6 to 12-mers repeated at the ends of contigs failed to identify any clear repeated motif.
Because no telomeric repeat could be identified, we investigated whether the telomerase enzyme, responsible for the addition of telomere repeats at chromosome ends was present in these genomes. Consistent with the absence of telomere repeat neither the telomerase enzyme nor telomerase reverse transcriptase domain were present. A search on 63 nematode proteomes showed the telomerase domain was otherwise widely conserved across the nematode tree of life. Besides the genus *Meloidogyne* in which the telomerase was completely absent (despite 10 species searched) this domain was also not found in *Pristionchus* and *Strongyloides* species. Interestingly, the telomerase domain was found in *Pratylenchus*, suggesting a loss specific to the *Meloidogyne* genus among Tylenchomorpha.
A more comprehensive search for enriched degenerated motifs at the extremities of contigs, allowed identifying a ca. 300bp complex motif repeated in arrays at ca. 50 contig ends. This motif is conserved and repeated at contig ends in at least all clade I root-knot nematodes surveyed so far.
We designed probes and performed fluorescent in situ hybridization in *M. incognita* cells and confirmed this complex motif was present at one end of most chromosomes. These findings suggest unusual and complex chromosome ends in the root-knot nematodes.

ORAL SESSION 14

Social impact of nematode management

Global 'worming': changing economic and environmental impact of plant-parasitic nematodes.

Wim Wesemael (wim.wesemael@ilvo.vlaanderen.be)

ILVO, Merelbeke, Belgium

The economic importance of plant-parasitic nematodes has generated considerable research effort directed primarily at understanding host-parasite relationships and determining methods to control these pests. With the restriction or banning of many chemical control options on which farmers and growers have traditionally relied, it is imperative to identify and utilise new control and management strategies. Increased trade and changing climate allows nematode problems to travel the world with increasing impact. Outbreaks of thermophilic species in temperate zones is a major concern in Europe. 'New crops' facilitate new nematode problems that might affect traditional crops but they can also create rotational options to manage plant-parasitic nematodes. Greening agriculture and precision farming are aspects that will alter nematode management. Research is needed on novel control and management options and it is essential that they are effectively passed on to end users. The change in research funding in many countries from direct government support to short-term grant-aided research has focused the need to obtain research funding from a variety of agencies. The impact of the above mentioned aspects on plant-parasitic nematode problems and management options will be discussed and illustrated with examples.

Keywords: Plant-parasitic nematodes - Greening - Climate change - Precision farming.

Social Implications of New Advances in Nematode Management.

Johan Desaeger (jad@ufl.edu)

Gulf Coast Research and Education Center, Department ofEntomology and Nematology, University of Florida, Wimauma, Florida, USA

Plant-parasitic nematodes significantly impact our lives. Nematode parasitism affects all crops, and although difficult to measure, estimated global crop losses are in excess of 10%. The enormous amount of crop damage and yield loss that plant-parasitic nematodes can cause was not known until the first trials with nematicides in the 1920s. Soil fumigants and broad-spectrum insecticides/nematicides became the cornerstone of nematode management in the decades thereafter. During the last decades, both the agricultural and economic and sociological environment has undergone major changes. Globalization, industrialization, emergence of a knowledge economy, growth of the organic food market, and changes in the regulatory environment have dramatically changed the way that pesticides and nematicides are being perceived and used. The phasing out of old broad-spectrum nematicides, and the emergence of new more selective and safer nematicides, combined with the increasing focus on soil health and sustainability, offers new opportunities, but at the same time forces growers to change often decades-old practices. These developments, combined with a growing awareness of the importance of nematodes in agriculture, and the expectation of more crop damage attributed to nematodes in the future (due to agricultural intensification, soil degradation, and warmer climate), will have major social implications for the world's rapidly changing food markets. It is difficult to predict the impact of these new developments, but the hope is that new advances in nematode management will improve food safety, as well as the livelihood and well-being of farmers, and strengthen the perception of farmers as good environmental stewards. Nematode management has acquired a new urgency in recent years, and with the expected transition from broad-spectrum nematicides to safer and more specific products, the need for more integrated nematode management programs will increase. This will require a more thorough understanding of nematode biology, and increased need for nematology research, education and extension. Only if such support is ensured, will new advances in nematode management be adopted by growers, sustainability increased, and the impact of plant-parasitic nematodes on crop production lessened. A case study of nematode management in Florida strawberries will be discussed.

Keywords: Social impact - Plant-parasitic nematodes - Nematicides - IPM - Florida.

An industry perspective on the global social impact of nematode management – innovation and food security.

John A Wiles[1] (john.wiles@corteva.com), Tim C Thoden[2]

[1] *Corteva Agriscience, Fulbourn, Cambridge, United Kingdom*; [2] *Corteva Agriscience, Munich, Germany*

Nematode management in crop production is as important today as it ever was in addressing threats to global food security. The ubiquitous nature of plant-parasitic nematodes creates serious challenges for all types of farmers, whether they are growing crops for subsistence on a small plot of land, or farming over very large areas. Global trends of population growth, increased demand for quality food, increased monocultures and climate change drive some of the conditions that can increase unseen problems with soil pests such as plant-parasitic nematodes, and affect soil health and productivity. Additionally, even though education is improving, awareness of what plant-parasitic nematodes are, which species are present and what level of impact they can have on yield may often go unnoticed or unidentified in many parts of the world. This knowledge gap widens yet further when it comes to understanding the role and function of the beneficial soil nematodes.
These challenges and societal needs are certainly being recognised by researchers, agronomists and larger research organisations. There is an abundance of research teams searching for innovations in nematode management that will bring greater prosperity to growers across the world and which aim to meet modern societal demands. The toolbox available for integrated nematode management is constantly changing, especially from a chemical perspective, and adapting as regulatory requirements continue to evolve and become more stringent. At the same time there are intense efforts to find new innovative selective solutions, within plant breeding, seed applied technology, novel modern chemistry and biologicals. The way land is managed for sustainable production and soil health is also being considered at farm-scale levels. Today, analysis of “big data” through digital systems, and integrated approaches, which also encompasses cultural and crop rotational considerations, are included in the equation. This paper provides an overview of the perspectives of Corteva Agriscience™ in how the Agricultural industry can contribute to global innovation for integrated nematode management.

Keywords: Nematode management - Soil health - Plant-parasitic nematode - Food security - Social impact.

Microbial-based pesticides, a novel source of new tools for nematode management.

Jared P. Jensen (jjensen@agbiome.com)

AgBiome, Research Triangle Park, NC, United States

With trillions of microbes spread across a diverse array of environments, there exists a near-infinite potential for producing novel biologically active molecules. At AgBiome, our mission is to partner with the microbial world for human benefit and discover unique microbes with activity against major agricultural pests, including plant diseases, insects, weeds and nematodes. Microbially-based pesticides are becoming an increasingly important tool in pest management due to their low environmental impact, potential for delivering multiple modes of action, ability to positively impact plant growth, and the important role they play in mitigating pesticide resistance. AgBiome has successfully demonstrated our ability to commercialize highly-efficacious microbial-based pesticides with our first fungicide, Howler™, with near-future products including novel fungicides, insecticides, and nematicides. Through grants with the Bill and Melinda Gates Foundation, we are developing more socially acceptable products with a global reach, and impacting agriculture across a spectrum of crops and ecologies.

Global impact of agricultural intensification on soil nematodes: a meta-analysis.

Jeremy Puissant[1] (jeremy.puissant@sfr.fr), Cécile Villenave[2], Camille Chauvin[2], Eric Blanchart[1], Jean Trap[1]

[1] *IRD, Montpellier, France*; [2] *ELISOL environnement, Congénies, France*

Nematodes are by far the most abundant invertebrates on Earth. Being involved in controlling soil microbial populations, carbon flows and nutrient cycling, these microscopic worms contribute to key ecosystem services such as primary production and pest regulation. Agriculture is one of the primary drivers of terrestrial biodiversity loss and may have a profound impact on soil nematode communities and the ecosystem services they support. By reducing local plant diversity, disturbing the soil surface by plowing, decreasing carbon stocks and altering nutrient cycling through fertilization, agricultural intensification may drastically change the composition, taxonomic richness and size of nematode populations. Despite this, we still lack a comprehensive picture of the global impact of agricultural intensification on soil nematodes with most research being perform at local scales and examining only one agricultural driver (*e.g.* fertilisation, plowing, pesticide) at a time. Here, we conducted a worldwide meta-analysis on the impact of agricultural practices on soil nematodes. Main agricultural practices under study are (i) inorganic and organic fertilization, (ii) tillage, (iii) pesticide, (iv) vegetal diversity including rotation, cover crop or presence of legumes. We collected data from published studies investigating the effects of agricultural practices on soil nematode community. To do so, we conducted a literature search, last updated in October 2019, using the Web of Science (Thomson Reuters) search engine. Out of 4962, 102 articles (1034 observations) met our criteria which consist of presenting means and variances on abundances, trophic groups and/or nematofaunal indices. We evaluated to what extent agriculture alters population size, functions and ecological network of the soil nematode community.

Keywords: Biodiversity - Agricultural practices - Tillage - Fertilisation - Nematofaunal indices.

A cost-benefit and efficacy analysis of *Meloidogyne* management strategies in Mediterranean intensive horticulture.

Miguel Talavera[1] (miguelf.talavera@juntadeandalucia.es), Luis Miranda[2], Maria Dolores Vela[3], Soledad Verdejo-Lucas[4]

[1] *Sustainable Plant Protection, IFAPA Alameda del Obispo, Cordoba, Spain*; [2] *Sustaibable Plant Protection, IFAPA Las Torres, Sevilla, Spain*; [3] *Sustaibable Plant Protection, IFAPA Rancho de la Merced, Jerez de la Frontera, Cadiz, Spain*; [4] *Sustaibable Plant Protection, IFAPA LA Mojonera, La Mojonera, Almeria, Spain*

Losses caused by phytoparasitic nematodes in crops depend directly on their soil densities at the start of the crop, so reducing their populations before planting is the main objective of nematological control. Efficacies in reducing *Meloidogyne* spp. soil populations by such different RKN-IPM strategies, as agrochemicals, botanicals, soil solarization, biofumigation, soil steaming or soilless cultivation, were calculated based on multiple field trials carried out during 15 consecutive years. *Meloidogyne* soil densities were reduced by 78 to 87% after fumigation with 1.3-dichloropropene:chloropicrin or dimethyl-disulfide. Other chemical nematicides such as fluopyram, oxamyl, dazomet, fosthiazate, fenamiphos, azadirachtin, ethoprophos, abamectin and metam-sodium showed efficacies ranging from 51 to 64%, while garlic extract, ozone or hydrogen peroxide reduced *Meloidogyne* populations by 41 to 46% and chloropicrin alone and furfural showed less efficacy of 40%. The combination of solarization with organic manure (biosolarization) reduced soil nematode populations by 73%, an efficiency slightly lower than soil fumigation, but similar to that of other agrochemicals. An economical cost-benefit study, including social and environmental externalities, of several RKN-strategies (soil disinfestation methods, resistant cultivars and grafting, soilless cultivation) in Mediterranean intensive horticulture was performed and a comparison of them is presented.

Keywords: Cost-Benefit - Meloidogyne - Nematicides - Soil disinfestation.

S14-PF1

Abundance and diversity of plant-parasitic nematodes in the rhizospheres of maize cultivars grown by commercial farmers in rural areas of South Africa.

Nancy Ntidi[1] (ntidin@arc.agric.za), Akhona Mbatyoti[2], Driekie Fourie[3], Lindokuhle Qwabe[3]

[1] *Plant Protection-Nematology Unit, Agricultural Research Council-Grain Crop, Potchefstroom, North West province, South Africa*; [2] *Plant Protection-Nematology Unit, Agricultural Research Council-Tropical &, Nelspruit, Mpumalanga province, South Africa*; [3] *Integrated Pest Management, North-West University, Potchefstroom, North West province, South Africa*

Maize (*Zea mays* L.) is the most important grain crop produced in South Africa under diverse environments and practice systems. The omnipresence of plant-parasitic nematodes in local agricultural soils, however, poses a threat to the sustainable production of maize, soybean and other rotation crops. The aim of this research was to determine the nematode assemblages associated with maize sampled from 15 farms situated in the North West and Free State provinces of South Africa. Rhizosphere, including root and soil, samples were obtained during two sampling intervals throughout the 2018/19 growing season. Eggs and second-stage juveniles (J2) of *Meloidogyne* and *Rotylenchulus* were extracted from 50g roots using an adapted NaOCl method. Individuals of other nematode pest genera were extracted and identified from 5g roots using the sugar-flotation method, while both plant-parasitic and non-parasitic nematodes were extracted from rhizosphere samples using the decanting and sieving, followed by the sugar-flotation methods. Prominence values (PV) were calculated for each nematode species identified from both soil and root (5g and 50g) samples. Two sedentary, endoparasitic plant-parasitic nematode genera were identified from 50-g root samples, *viz*. *Meloidogyne* (PVs ranging from 699 to 72 891) followed by *Rotylenchulu*s (PVs ranging from 33 to 10 635). Plant-parasitic nematode genera, species and/or families identified from the soil and root samples obtained from 15 farms were Belonolaimidae, Bacterivores, Fungivores, Criconemidae, Dorylaimidae, Longidoridae, *Rotylenchulus, Helicotylenchus, Scutellonema, Pratylenchus* and *Meloidogyne.* The present study gave new insights regarding the host status of maize cultivars to *Meloidogyne* and *Rotylenchulus* spp. Furthermore, population densities of some nematode pest genera were exceptionally high in soil samples, indicating that practicing monoculture can lead to a build-up in population densities of damaging nematodes. Results obtained from this study will enable nematologists to make informed decisions regarding cultivar use to combat nematode problems in maize-based cropping systems. This study is on-going with a followup survey commencing during January 2020.

Keywords: Frequency of occurrence - *Meloidogyne* spp. - Prominence values - Rural - Survey.

References:

- BACK, M.A., HAYDOCK, P.P.J & JENKINSON, P., 2002. Disease complexes involving plant parasitic nematode and soilborne pathogens. Plant Pathology, 51: 683 - 697.
- BEKKER, S., FOURIE, H., RASHIDI, M., DANEEL, M., SHOKOOHI, E. AND NEL, A., 2016). Discriminating between the eggs of two egg-mass-producing genera using morphometric and molecular results. Nematology, 18: 1 119 - 1 123.
- MC DONALD, A.H., DE WAELE, D. & FOURIE, H., 2017. Nematode pests of maize and other grain crops. In: Fourie, H, Spaulls, V.W., Jones, R.K., Daneel, M.S. and De Waele, D. (Eds.) Nematology in South Africa: A view from the 21st Century. Springer Publishing, Germany, pp. 183-199. DOI:10.1007/978-3-319-44210-5.
- FOURIE, H., STEENKAMP, S., MC DONALD, A.H. & DE WAELE, D., 2017. Nematode pests of leguminous and oilseed crops. In: Fourie, H, Spaulls, V.W., Jones, R.K., Daneel, M.S. and De Waele, D. (Eds.) Nematology in South Africa: A view from the 21st Century. Springer Publishing, Germany, pp. 201-230. DOI:10.1007/978-3-319-44210-5.
- NTIDI, K.N., FOURIE, H., MC DONALD, A.H., DE WAELE, D. & MIENIE, C.M.S., 2012. Plant-parasitic nematodes associated with weeds in developing agriculture in South Africa. Nematology, 14(7): 875 - 887.

S14-PF2

Banan fibre paper: effectively delivering ultra-low nematicide dosages for more acceptable nematode management.

Danny Coyne[1, 2] (d.coyne@cgiar.org), Laura Cortada-Gonzalez[2], Solveig Haukeland[3], Baldwyn Torto[3], Charles Opperman[4]

[1] *International Institute of Tropical Agriculture, Nairobi, Kenya*; [2] *Department of Biology, Ghent University, Campus Ledeganck, Nematology Research Unit, Ghent, Belgium*; [3] *International Centre of Insect Physiology and Ecology, Nairobi, Kenya*; [4] *Department of Entomology and Plant Pathology, North Carolina State University, Raleigh, North Carolina*

A lignocellulose fiber paper matrix made from banana fiber provides a novel ability to effectively manage nematode pests of various crops. Impregnated with ultra-low dosages of synthetic nematicides the paper enables the delivery of concentrations that effectively reduce nematode densities in the target rhizosphere zone with much reduced application rates. The special characteristics of the lignin and cellulose composition of banana fiber slowly release the impregnated nematicides over time, enabling a persistent protection in the root zone of the growing plant. This biodegradable matrix creates an effective delivery system for reducing the impact of chemical nematicides on the environment, improving the efficacy of some compounds and improving crop production. Additionally, the properties of the paper that enable effective adsorption of pesticides, similarly adsorbs host-pest semio-chemical signals, interfering with nematode hatching and host location. The paper matrix can also provide for the delivery of biologically based nematicides. Results from potato, sweet potato and cassava field trials and farmer demonstrations in Africa are presented.

S14-PF3

Best management practices for Root-knot nematode (*Meloidogyne hapla*) in daylily (*Hemerocallis* spp.) production.

Amanda Howland (howlan21@msu.edu), Emilie Cole, Kristin Poley, Marisol Quintanilla

Michigan State University, East Lansing, MI, United States

Bare-root production of *Hemerocallis* spp., daylilies, is of major economic importance among Michigan's ornamental industry. Production of clean nursery plant material can be a challenge for bare-root ornamental crops grown under field conditions due to the occurrence of plant-parasitic nematodes. One of the major plant-parasitic nematodes affecting daylily production resulting in 20% yield loss in Michigan is the northern root-knot nematode, *Meloidogyne hapla*. Therefore, a three-year field trial was established from 2018-2020 in a commercial nursery in Michigan, US, with the objective to establish effective management strategies by examining alternatives to fumigation. Twelve treatments, with six replications each, were established in a non-fumigated field. The treatments implemented were an untreated control, three compost blends, four bio-nematicides, two chemical nematicides, a chemical nematicide root dip, and soil steaming. First-year daylily plants (cv. 'Going Bananas') were treated with their respective treatments and planted in the field. At the same time of planting, initial soil samples were collected in each plot to serve as a baseline for nematode populations. Each year, additional soil samples were taken in the spring, mid-season, and fall in each plot. Root samples from each respective plot were taken mid-season and stained to determine the number of root-knot nematodes in each root system; plant height measurements were also taken at mid-season each year. The top treatments for controlling root-knot nematodes were two chemical nematicides: TerraClean 5.0 (AI: hydrogen peroxide) and Majestene 304 (AI: *Chromobacterium subtsugae*), and the chemical nematicide Indemnify (AI: fluopyram) as a preplant root dip. Indemnify also had the greatest plant growth and yields. This field trail provides effective *M. hapla* management methods that reduce the application of fumigants and prevent significant yield losses to increase profitability for ornamentals.

Keywords: *Meloidogyne* spp. - Daylilies - Management - Bare-root production.

ORAL SESSION 15

Advances in nematode detection and identification: instrumentation and applications

Non-invasive detection of plant parasitic nematodes using hyperspectral imaging.

Barbara Gerič Stare (barbara.geric@kis.si)

Plant Protection Depertment, Agricultural Institute of Slovenia, Ljubljana, Slovenia

Traditional visual detection of nematode infestations is based on characteristic symptoms of the plants. Nematode detection is further exacerbated by their naturally heterogeneous distribution, making a systematic approach challenging. Plants have to be checked individually, making this a time-consuming and costly endeavour. On the other hand, remote sensing (RS) methods can provide accurate detection of nematode infested plants over large areas and overcome several shortcoming of visual detection by eliminating the need for individual check of every plant and uprooting of the plant. Research of remote sensing of nematode infestations began in the 1960s, and since then a large amount of information has been gathered using various sensors, platforms, and analysis methods. By utilizing spectral information outside the visible spectrum of light, pre-symptomatic detection also becomes possible. Additionally, RS for plant protection enables non-invasive detection, without affecting the plant in any way (e.g. uprooting) where plant's response to diverse stressors can be measured. Nematodes are a good fit for remote sensing applications and precision agriculture, since they have a clustered spatial distribution, low mobility, and characteristic changes in spectral signatures of the plant canopy. Hyperspectral imaging is a type of RS where data of the light spectrum in over one hundred contiguous spectral bands with a constant width of up to 10 nm is acquired, providing detailed spectral resolution of the plants' images. We developed a methodology for non-invasive detection of *Meloidogyne incognita* infestation of tomato plants utilizing hyperspectral imaging [1]. Both root-knot nematodes (biotic stress) and water deficiency (abiotic stress) led to similar drought symptoms in the plant canopy. Pre-symptomatic detection as well as distinguishing nematode infested plants from plants in drought stress was possible by combining partial least squares discriminant analysis (PLS-DA) and support vector machine classification (PLS-SVM) [1,2]. PLS-SVM classification achieved up to 100 % accuracy differentiating between well-watered and water-deficient plants, and between 90 and 100 % when identifying nematode-infested plants. Shortwave infrared spectral regions associated with the O-H and C-H stretches were most relevant for the identification of nematode infested plants and severity of infestation. Such non-invasive detection of RKN could be very valuable for producers.

Keywords: Remote sensing - Hyperspectral imaging - Non-invasive detection - Root-knot nematodes - Drought stress.

References:

- [1] Susič et al., 2018, Sensor Actuat B-Chem. 273: 842-852.
- [2] Žibrat et al., 2019. MethodsX 6: 399-408.

European and national reference laboratories: an EU network for improved surveillance of plant-parasitic nematodes.

Maíra Grossi de Sá[1] (maira.grossidesa@anses.fr), Nicole Viaene[2], Sylvie Gamel[1], Nicole Damme[2], Françoise Munaut[3], Laurent Folcher[1]

[1] *Nematology Unit / EURL for plant parasitic nematodes, ANSES - Plant Health Laboratory, Le Rheu, France*; [2] *Plant Sciences Unit, ILVO-Flanders Research Institute for Agriculture, Fisheries and Food, Merelbeke, Belgium*; [3] *Unit G1 –Plant Health, European Commission, DG Health and Food Safety – Directorate G – Crisis manageme, Brussels, Belgium*

In 2019, the EU Commission (EC) designated the first EU Reference Laboratories (EURLs) in Plant Health. These EURLs were created to assist the EC and all National Reference Laboratories (NLRs) of the Member States (MSs) in implementing the Plant Health EU Regulation 2016/2031. The EURLs' mandate includes contributing to the improvement and harmonisation of the diagnostic methods used for official controls in the EU territory. Some plant-parasitic nematodes (PPNs) challenge the sustainable production of food worldwide. The EURL for PPNs is a consortium between ANSES (France) and ILVO (Belgium) and was established for improving accurate detection of regulated and emerging PPNs threatening EU plant health. In the EU surveillance framework of quarantine PPNs, the establishment of a network between the EURL and NRLs is of prime importance in the context of rapid and efficient interventions at EU borders and in the territory. In addition, surveillance data give a meaningful picture of PPN distribution, enabling the early detection of potential threats, assessing the impact of those already present and identifying the introduction pathways of emerging PPNs. However, PPN surveillance can only be achieved through accurate detection methods. Therefore, the EURL can operate as an EU detection tool since it enhances the application of detection methods by the NRLs. This is achieved by providing high standard diagnostic practices, including improving and giving guidance on analytical methods and organising inter-laboratory tests (IT). ITs were organised to assess the robustness of the EU network and its competence for diagnostic testing and official controls. ITs also revealed potential analytical issues leading to implementation of corrective actions and training sessions. Thus, training sessions on reference methods for diagnostic quarantine PPNs were provided by the EURL to enhance NRL performances. Research on reliable diagnostic approaches enabled the publication of EURL protocols for the detection of *Globodera* and *Meloidogyne* quarantine PPNs. Protocols and other information are published on the EURL website, an information source for NRLs. These approaches support the implementation of new acts by the EC, harmonising the application of control programmes and diagnostic methods across the MSs. When requested, the EURL also assists the EU by providing expertise, research and information on technical innovations, increasing detection and maximising PPN surveillance.

Keywords: EU laboratories network - Plant-parasitic nematodes - Detection methods - Surveillance - Plant health.

References:

- Council Directive 2017/625/EC of 15 March 2017 on official controls and other official activities performed to ensure the application of food and feed law, rules on animal health and welfare, plant health and plant protection products. (2017) Official Journal of the European Union L 95/1, pp.1-142.
- Council Directive 2016/2031/EC of 26 October 2016 on protective measures against pests of plants. (2016) Official Journal of the European Union L 260/8, pp.1-101.

Use of automated image analysis techniques for species identification and classification.

Romain Thevenoux[1] (romain.thevenoux@inra.fr), Heloïse Villesseche[2], Laurent Folcher[2], Eric Grenier[1], Nicolas Parisey[1]

[1] *UMR IGEPP, INRAe, Le Rheu, France*; [2] *Plant Health Laboratory, Nematology Unit, ANSES, Le Rheu, France*

Identification of taxonomy at a specific level is time consuming and reliant upon expert nematologists. Hence, automated species identification is a novel and accurate way to circumvent some of these limits. The analyse of data from images has recently become easier due to developments in computational technology. The cyst nematodes belonging to the genus *Globodera* includes species of major economic importance due to damage caused in potato fields. However, their identification remain complex. Strong suspicions of cryptic species within *G. pallida* exist based on recent investigations. The creation of tools capable of exploring the morphological variability within this complex on one hand and distinguishing species already described on the other hand would be an undeniable asset. Thanks to novel methods of automatized digitalisation, it is now possible to quickly study more morphological data. The automatization and standardisation of morphometric studies will i) simplify the identification of known species and ii) provide evidence about the evolution and differentiation of species complexes. In this study, we developed new metrics, relevant to automatized digitalisation. We choose 3 populations belonging to the well-known European *G. rostochiensis* and *G. pallida* species, the close species *G. mexicana* and the cryptic species suspected in Chile. For each, 30 individuals were photographed and used to build an automated method, using up-to-date image processing to predict metrics. the first step was to predict simple metrics, such as body length, and detect those anatomical parts. A second step predicts more accurate metrics in the head or tail of infective juveniles. We showed that by using such image analysis we are able to accurately detect anatomical parts, predict simple metrics and distinguish *G. pallida* from *G. rostochiensis*. We are also able to accurately predict metrics for 30% of novel infective juveniles and then assign them to one of these species with a confidence of 95%. We also showed that thanks to these new metrics and the high number of infective juveniles that were analysed, a morphological distinction can be observed between *G. pallida* and the Chilean populations which are suspected to belong to a cryptic species. Nonetheless, we are not yet able to distinguish these populations from *G. mexicana*. These data represent a new contribution for improved species identification and description within the genus *Globodera.*

Keywords: Cyst nematodes - Skeletization - Phenotypic variability - Morphological analysis - Mathematical morphology.

Lab-on-chip for *Globodera pallida* detection.

Maria João Camacho[1, 2] (mjoao.camacho@iniav.pt), Verónica C. Martins[3], Eugénia Andrade[2, 4], Débora C. Albuquerque[3, 5], Maria L. Inácio[2, 4], Manuel Mota[1,] Paulo Freitas[3,6]

[1] *Mediterranean Institute for Agriculture, Environment and Development (MED), University of Évora, Évora, Portugal*; [2] *Instituto Nacional de Investigação Agrária e Veterinária (INIAV, I.P.), Oeiras, Portugal*; [3] *INESC Microsistemas e Nanotecnologias, Rua Alves Redol, Lisbon, Portugal*; [4] *GREEN-IT Bioresources for Sustainability, ITQB NOVA, Oeiras, Portugal*; [5] *IST- Instituto Superior Técnico, University of Lisbon, Lisbon, Portugal*; [6] *INL- International Iberian Nanotechnology Laboratory, Braga, Portugal*

The potato cyst nematode (PCN) *Globodera pallida*, has acquired significant importance throughout Europe due to its widespread detection and negative effects on potato production. Thus, rapid and reliable diagnosis of PCN is critical during the surveillance programs and for the implementation of control measures. Despite the high reliability of molecular diagnostics (e.g conventional PCR, rt-PCR and LAMP methods), they require trained technicians, expensive equipment and laboratory facilities, are time-consuming and not suitable for on-site analysis. The development of innovative technologies to overcome current limitations in early detection is needed. Biosensors, in combination with microfluidics based biochemical analysis, can quickly detect the presence of specific nucleotide sequences with high sensitivity and convert the presence of biological compounds into an easy-to-read electrical signal. In this work, a specific probe and PCR primers (to amplify the internal transcribed spacer region (ITS) of *G. pallida* ribosomal DNA) were designed to be used in magnetic field biosensors which reliably discriminate *G. pallida* from other cyst nematodes. Magnetic nanoparticles are used as labelling agents of asymmetric PCR products through biotin-streptavidin interaction. Upon target hybridization to sensor immobilized oligo probes, the fringe field created by the magnetic nanoparticles produces a variation in the sensor's electrical resistance. The detection signal will correspond to the concentrations of target molecules present in the sample. These results show the specificity of the probe and suitability of the magnetic biosensor to detect *G. pallida* PCR target product, demonstrating its potential as a bioanalytical device for point-of-use applications.

Financial support was provided by National Funds through FCT—Foundation for Science and Technology under the Ph.D. fellowships BD 138724/2018, the project UIDB/05183/2020 (MED–Mediterranean Institute for Agriculture, Environment and Development, Institute for Advanced Studies and Research, University of Évora) and the project UID/05367/2020 (PROGRAMATICO – Research Unit INESC MN, Microsystems and Nanotechnologies).

Keywords: Biosensor - Digital agriculture - Potato cyst nematodes.

References:

- Camacho MJ, Inácio ML, Mota M, Andrade E. 2021. Pathogens 744:10(6).
- Camacho MJ, Andrade E, Mota M, Nóbrega F, Vicente C, Rusinque L, Inácio ML. 2020. Frontiers in Plant Science 11:606178.
- Martins V, Cardoso F, Germano J, Cardoso S, Sousa L, Piedade M, Freitas P, Fonseca LP. 2009. Biosensors & Bioelectronics 24: 2690–2695.

Digitize your nematode control.

Sumeja Ouyeder[1] (sumeja.ouyeder@bayer.com), Hartwig Dauck[2], Jose Luis Robles[3]

[1] *Global Strategic Portfolio Management, Bayer - Crop Science division, Monheim, Germany*; [2] *Segment Management, Bayer - Crop Science division, Monheim, Germany*; [3] *Crop Management, Bayer - Crop Science division, Basel, Switzerland*

Nematool, Bayer's digital solution for nematode control in open-air crops and greenhouses, is easy to use, provides instant alerts, and functions fully automatically. This functionality is supported by means of a mobile app, a Nematool probe and BioAct® Prime.
Nematode model: Based on the soil temperature at a depth of 20 cm and degree-day accumulation, Nematool gives you automatic alerts on the current generation and the percentage applicable for the appearance of next-generation eggs. Nematool provides a solid recommendation for the correct application of BioAct® and therefore for obtaining the best result of the product application.
Solarization model: Nematool indicates the quality of solarization based on the accumulation of lethal temperatures in the soil, and sends alerts when an excellent temperature for solarization is reached.

Keywords: Digital tool - Biological control of nematodes - App - I-Phone - Sensor.

Developing a droplet digital PCR assay for detection of the stubby root nematode *Paratrichodorus allius.*

Guiping Yan (guiping.yan@ndsu.edu), Zhuoyu Wang, Addison Plaisance

Department of Plant Pathology, North Dakota State University, Fargo, ND, United States

Stubby root nematodes (*Paratrichodorus* and *Trichodorus*) are migratory ectoparasites that feed on roots of many crops. These nematodes are particularly important to the potato industry as they transmit *Tobacco rattle virus* (TRV) causing corky ringspot disease of potato. *Paratrichodorus allius* is the most prevalent vector of TRV in potato fields from multiple states in the USA. Thus, it is important to have a sensitive and reliable method for detecting this nematode from infested fields. A droplet digital PCR (ddPCR) assay was developed for *P. allius*. Droplet digital PCR is a novel nucleic acid-based technique that enables absolute quantification of DNA targets without the need to construct a calibration curve, and is a robust and powerful platform for quantitative pathogen detection. Species-specific primers published for *P. allius* were selected and their specificity was confirmed with six isolates of stubby root nematode species and 28 isolates of other plant-parasitic nematodes. Optimum ddPCR conditions were established by performing annealing temperature gradient tests. Analysis of amplification efficiency (E) was conducted by creating a regression line between log values of copy numbers of linearized *P. allius* plasmid DNA determined by ddPCR and predicted values of serial dilutions of the templates. The relationship between the measurements of ddPCR assay and expected values showed good linearity (R2 = 0.9345) with E = 108%. The sensitivity and reliability of ddPCR for detecting *P. allius* are being assessed. Further studies are warranted to evaluate and validate the ddPCR technology for accurate and sensitive diagnosis of plant-parasitic nematodes from infested fields.

Keywords: Stubby root nematode - ddPCR detection - Specificity - Amplification efficiency - Sensitivity.

References:

- [1] Huang et al., 2018. Plant Disease 102: 2101-2111.
- [2] Taylor et al., 2017. Scientific Reports 7: 2409. DOI:10.1038/s41598-017-02217-x.
- [3] Zhao et al., 2016. PloS One 11(7): e0159004. doi.10.1371/journal.pone.0159004.

S15-PF1

A novel approach for applying machine learning for detection and phenotyping of cyst nematodes in soil extracts.

Long Chen[1] (matthias.daub@julius-kuehn.de), Martin Strauch[1], Matthias Daub[2], Marcus Jansen[3], Susanne Schultz-Kuhlmann[4], Stefan Krüssel[4], Hans-Georg Luigs[5], Dorit Merhof[1]

[1] *Imaging and Computer Vision, RWTH Aachen University, Aachen, Germany*; [2] *Institute for Plant Protection in Field Crops and Grassland, Julius Kuehn Institute, Elsdorf, Germany*; [3] *LemnaTec GmbH, Aachen, Germany*; [4] *Pflanzenschutzamt, Sachgebiet Zoologie, Landwirtschaftskammer Niedersachsen, Hannover, Germany*; [5] *Imaging and Computer Vision, LemnaTec GmbH, Aachen, Germany*

Cyst nematodes comprise a considerable number of 110 valid species and many of them are important pests of cultivated plants. Individuals survive inside the female body which is transformed into a cyst at the end of the female life cycle. Determination of population densities for damage prediction models usually require first the extraction of cysts and subsequently the release of eggs and juveniles from cysts which is time demanding and susceptible to subjective errors. The PheNeSens project is aiming at the development of two independent systems for the automated detection, counting, and phenotyping of cysts on a macroscopic level and cysts contents (eggs and juveniles) on a microscopic level. As an initial approach we propose an instance segmentation method that will serve as the basis of automatic quantitative detection of cysts and a framework for detecting vermiform objects in microscopic images that is based on convolutional neural networks (CNNs). We consider light microscopic images of cluttered object collections as they occur in realistic extracts of soil samples. For the detection of cysts we introduce an algorithm, LMBI (Local Maximum of Boundary Intensity) to propose instance segmentation candidates of cysts. In a second step, a SVM classifier separates the nematode cysts among the candidates from soil particles. On a data set of soil extract images, the LMBI detector achieves near-optimal recall with a limited number of candidate segmentations, and the combined detector/classifier achieves recall and precision of about 70 %. The pipeline only requires simple dot annotations and moderately sized training data, which enables quick annotating and training in laboratory applications. For the detection of juveniles we annotate nematodes with curves along the body. The trained model predicts worm «skeletons» and body endpoints. The endpoints serve to untangle the skeletons from which segmentation masks are reconstructed by estimating the body width at each location along the skeleton. With light-weight backbone networks, we achieve 75.85% precision, 73.02% recall on a potato cyst nematode data set and 84.20% precision, 85.63% recall on a public *C. elegans* data set. As a future perspective we will integrate automated detection of body dimension, shape and colour or other biological characteristics (viability, parasitation) to detect specific phenological features of nematodes on a population level.

Keywords: Cyst nematodes - Phenotyping - Detection - Machine learning.

S15-PF2

ATR-FTIR spectroscopy and hyperspectral imaging in determining quality of formulated entomopathogenic nematodes.

Nicholas Kagimu[1,2,3] (nalwogaf@gmail.com), Antoinnette P. Malan[1]

[1] *Dep. Conservation Ecology and Entomology, University of Stellenbosch, Stellenbosch, Western Cape Province, South Africa*;
[2] *Department of Zoology and Entomology, University of Pretoria, Hatfield Pretoria, Gauteng, South Africa*; [3] *FABI, University of Pretoria, Hatfield Pretoria, Gauteng, South Africa*

Beneficial nematodes known as entomopathogenic nematodes (EPNs) are used as biological control agents to fight a variety of insect pests that severely damage crops. However, they often fail to measure up to other control methods such as inorganic chemical pesticides due to quality deterioration. For nematode quality, virulence remains the most important standard for measurement, which is often determined through using either one-on-one, or sand-well, bioassays, which are costly in terms of laboratory consumables and time. Such determination calls for the use of quick, non-destructive and effective quality control techniques, which could include the application of attenuated total reflectance (ATR), in conjunction with Fourier transform infrared spectroscopy (FTIR) and hyperspectral imaging (HSI) tools, which have been proven to have a wide application in other fields of research [1,2,3,4,5]. In this study, the potential for the quality control of formulated *Steinernema jeffreyense* and *S. yirgalemense* in diatomaceous earth (DE), and the characterisation of different species using ATR-FTIR and HSI have been investigated. Results report, for the first time, the use of ATR-FTIR spectral analysis in detecting chemometric changes in the formulated EPN product and changes occurring over time, during storage. The changes are mainly for reasons of nematode survival, in response to environmental stresses. HSI was able to differentiate between variables, in terms of differences in nematode densities, in the formulated sample. For EPN characterisation, the study reports close similarities among the species, as detected by the ATR-FTIR.

Keywords: Virulence - Heterorhabditis - Trehalose - Short-wave infrared - Visible near-infrared.

References:

- [1] Bouyanfif et al., 2018. Vib. Spectrosc. 96:74–82.
- [2] Bouyanfif et al., 2017. Analyst. 142:4727–4736.
- [3] San-Blas et al., 2011. Vib. Spectrosc. 57:220–228.
- [4] Su and Su, 2018. Compr. Rev. Food Sci. Food Saf. 17:104–122.
- [5] San-Blas et al., 2012. Spectrochim. Acta Part A Mol. Biomol. Spectrosc. 93:58–62.

S15-PF3

Rapid detection and quantification of plant-parasitic nematodes from large volumes of soil.

Valeria Orlando[1] (Valeria.Orlando@fera.co.uk), Thomas Prior[2], Michael R. Surrey[3], Steven Bryce[2], Wellcome Ho[3], Brett J. R. Alexander[3]

[1] *Crop & Environment Sciences, Harper Adams University, Newport, United Kingdom*; [2] *Fera Science Ltd., York, United Kingdom*; [3] *Plant Health and Environment Laboratory, Biosecurity New Zealand - Tiakitanga Pūtaiao Aotearoa, Ministry for Primary Indu, Auckland, New Zealand*

The aim of this project, funded by New Zealand Ministry for Primary Industries, was to develop a novel method using existing molecular techniques, to enable rapid detection and quantification of nematodes from large volumes of soil. This approach could have important application for completing delimiting surveys and taking timely actions if pathogenic or quaranitine soil-borne nematodes were to be found. Employing this innovative system, the process from nematode extraction from soil to identification of target species takes about two days. The study comprised the assessment and validation of an optimal chemical solution, including relative specific gravity (sp.gr.), for the extraction of various nematode sizes from differing soil substrates using flocculation. This technique is used in other areas of biology and was initially adapted for the extraction of nematodes by Caveness & Jensen (1955); the method allows extraction of motile, immotile and even dead nematodes from soil and sediments. The compounds assessed were, sucrose, NaCl, MgSO4 and colloidal silica at sp.gr. of 1.12 – 1.22. The flocculant compounds were also assessed for any potential effect on DNA integrity. From these selected compounds, a colloidal silica solution at sp.gr. 1.15 performed closest to the controls for all soil types, with nematodes remaining alive even after extended contact with the solution. Sucrose, although the most cost-effective option, gave the poorest recovery. Detection of plant-parasitic nematodes by visual inspection requires practitioners skilled in morphological identification and requires a significant timeframe, especially when present on a background of other nematodes. Targeted detection of DNA in extracts from bulk soil samples provides a rapid route to screening survey samples. We therefore validated a method for isolation and purification of DNA from the samples suitable for analysis using PCR methods. To assess the performance of the method, 5kg of soil were spiked with known quantities of *Meloidogyne hapla* nematodes. Nematodes were extracted from samples using the flocculation method and DNA was isolated from the bulked nematodes and purified. *Meloidogyne hapla* DNA was detected using a species-specific real-time PCR assay. For this species, the measured detection (mean CT value) showed a linear relationship with (log) quantity of nematodes over the spiking range 25-2500. The limit of detection for the overall method is <25 target nematodes per 5 kg soil.

Keywords: Plant-parasitic nematodes - Soil extraction - Identification - Flocculation - Real-time PCR.

References:

- [1] Caveness and Jensen, 1955. Proc Helminthol Soc Wash. 22: 87-89.

ORAL SESSION 16

Biological control of nematodes

Biological agents to invigorate the health of established coffee trees by managing plant parasitic nematodes.

Kanan Saikai[1] (K.Saikai@cgiar.org), Celestine Oduori[2], Wanjala Situma[3], Simon Njoroge[4], Ruth Murunde[4], Danny Coyne[2]

[1] *NemAfrica, International Institute of Tropical Agriculture (IITA), Nairobi, Kenya*; [2] *International Institute of Tropical Agriculture (IITA), Nairobi, Kenya*; [3] *International Centre for Insect Physiology and Ecology (ICIPE), Nairobi, Kenya*; [4] *The Real IPM CO. (Kenya) LTD., Nairobi, Kenya*

Plant parasitic nematodes are the threat to the coffee production in Sub-Saharan Africa; however, limited nematode management options are available in the region, especially after the nematode infestation is established in coffee fields. From the environmental perspective, there is a strong need for a sustainable nematode management method that is not harmful to non-target nematodes. To achieve this, two biological agents, *Trichoderma asperellum* and *Purpureocillium lilacinus,* were evaluated for their effects on plant parasitic nematodes and the nematode community in soil. Seven coffee fields with different ages of two coffee varieties in Nairobi, Kenya were treated with either 20L of *T. asperellum* solution (2 x 107 cfu /L), 20 L of *P. lilacinus* solution (2 x 107 cfu /L), or 20 L water with four replications, consisting of five trees for each replication, at each field. Population densities of plant parasitic nematodes in soil and roots were monitored every two months and every six months, respectively, for two years. All the fields were heavily infested by *Meloidogyne hapla,* which was identified morphologically and molecularly, for the first time on coffee in Kenya. Fungal colonization was first confirmed in soil 6 months after and in roots one year after the first treatment application. Significant reduction of *M. hapla* in roots was observed for *P. lilacinus* after the fungal root colonization ($P<0.05$) but not for *T. asperellum.* All the fields demonstrated the characteristics of resource-limited soil conditions and no improvement by the bioagents based on the functional footprint. Greater number of fungivores were found for *P. lilacinus,* especially *Aphelenchoides* and *Aphelenchus* ($P<0.05$). Although not always significant at all time points, more Dorylaimid nematodes were detected for *P. lilacinus* as well. Larger value of sigma maturity index for both bioagents and enrichment index for *P. lilacinus* was observed from more recent sampling ($P<0.1$). *P. lilacinus* demonstrated the potential to be used as a sustainable nematode management option for coffee without harming soil health conditions.

Keywords: Coffee - Root-knot nematode - Biological control - Nematode community.

Soil microbiota effect on the efficiency of root exudates to hatch cyst nematodes.

Josselin Montarry[1] (josselin.montarry@inrae.fr), Camille Gautier[1, 2], Lisa Martinez[1], Sylvain Fournet[1], Eric Nguema-Ona[2], Anne-Yvonne Guillerm-Erckelboudt[1], Christophe Piriou[1], Juliette Linglin[1], Christophe Mougel[1], Lionel Lebreton[1]

[1] *IGEPP, INRAe, Le Rheu, France*; [2] *Agro Innovation International, Groupe Roullier, Saint-Malo, France*

Plant-parasitic nematodes are among the most harmful pests of cultivated crops causing important economic losses. For cyst nematodes, the hatching of juveniles is stimulated by root exudates released by the host plant into the rhizosphere. The removal of chemical nematicides requires development of alternative approaches to protect crops against nematodes. Root exudates would constitute an effective and innovative biocontrol method that could be used in the absence of the host plant to induce a “suicide hatching” of nematode juveniles and to control cyst nematodes. To anticipate the effectiveness of root exudates in different environments, many parameters, such as the microbiotic composition of soil, must be considered. Thus, the aim of this study was to investigate the effect of soil microbiota on the ability of root exudates to induce cyst nematodes hatching. Four different agricultural soils were selected based on their physicochemical and biological compositions. To disentangle the microbiota effect from the global effect, a sterile soil matrix was used and inoculated with the microbiota of the different agricultural soils. Each soil was artificially inoculated with cysts of *Globodera pallida* (which attack potato) and root exudates (two doses: dose 1 > dose 2) were applied by exogenous applications. After 45 days, larvae remaining in cysts were counted in order to determine the efficiency of root exudates to stimulate cyst hatching according to the different soils. The results showed that i) in agricultural soils, significant differences of hatching were observed between soils but the hatching rate remained high with no dose effect and ii) in recolonized soils, a strong dose effect was highlighted with dose 2 inducing a lower hatch of nematodes than dose 1. This result was probably due to carbon consumer microorganisms. This study provides key elements for the development of root exudates as new biocontrol strategy: abiotic and biotic factors do not jeopardize the efficiency of suicide hatching when root exudates are highly available for both microorganisms and cyst nematodes.

Activity of *Bacillus firmus* against plant-parasitic nematodes.

A. Sylvia S. Schleker (sylvia.schleker@uni-bonn.de), Mengmeng Huang, Aylin Bulut, Christiane Matera, Bidhya Shrestha, Volkan Cevik, Ute Schlee, Florian M. W. Grundler

Molecular Phytomedicine, University of Bonn, Bonn, Germany

Plant parasitic nematodes cause severe yield and quality losses in agricultural crop production worldwide. Utilizing antagonistic microbes like the Gram-positive rhizobacterium *Bacillus firmus I-1582* which is promoted as biological nematode control agent is one strategy to combat nematode parasitism. Although *B. firmus* is a known nematode antagonist in general, details about its interaction with nematodes and plants are rare. We therefore investigated the impact of *B. firmus I-1582* and its secreted molecules on *Heterodera schachtii* infective juveniles (J2s) and on plant-nematode interaction.

Our studies indicate a multiple mode of action profile. When treating nematodes directly with cell-free bacterial supernatant we found a specific fraction of the supernatant to be highly lethal for the J2s in a temperature-dependent manner. The toxicity can partially be addressed to proteinaceous molecules. Genome sequencing and gene expression analysis highlights post-translationally modified molecules to potentially be the active agents.

In interaction with the plant, we demonstrate that *B. firmus I-1582* is attracted by *Arabidopsis thaliana* root exudates, colonizes the root and develops there in a strictly pH-dependent manner. Root colonization by *B. firmus I-1582* significantly protects the plant from infection with *H. schachtii*. Effects can also be observed in the next generation: Juveniles hatching from cysts that developed on *B. firmus* I-1582 colonized plants are highly impaired in invading and infecting *B. firmus*-free *A. thaliana* roots as well as in development and reproduction.

Keywords: Biological control - *Bacillus firmus* - Cyst nematode - Parasitism - Mode of action.

BIODERA project: a teamwork effort to develop biocontrol solutions against plant parasitic nematodes.

Eric Nguema-Ona (Eric.NguemaOna@roullier.com)

Laboratoire de Nutrition Végétale, Pôle Stress Biotiques, Centre Mondial de l'Innovation Roullier, Saint-Malo, France

Plant-parasitic nematodes (PPNs) are microscopic worms mainly found in agricultural soils. PPNs are responsible for important damage to a large number of crops (including potato, sugar-beet, legumes, cereals or soybean). Globally, they are responsible for the reduction of the yield by 11% worldwide, a loss averaging millions of tons every year, which can be translated into hundreds of billions of dollars in losses. PPNs encompass different groups of nematode having different lifestyles. Among them some of the most damaging to plants include both the sedentary, root-knot and cyst nematodes. The ban of nematicides and fumigants for both human health and environmental concerns, renders the management of agricultural lands difficult for holders and technical sector advisers, and thus impact/reduce crop production as well as profitability of farming business. A number of natural products or biological control solutions, have been proposed to control nematode attacks, and have been tested experimentally. However, most of them strive to show efficacy in the field, as well as their ways to the market. The aim of the BIODERA project is to develop natural, environmentally friendly, nematode biocontrol solutions. The solutions tested focused on i) plant-based bioactives ability to control juvenile mobility or ii) their ability to induce egg hatching of plant parasitic nematodes. The project gathers technical sector researchers, basic and applied researchers from public and private institutions, as well as partners from agro-supply industries. It offers the possibility to tackle this challenge by taking into account from the initiation, recent scientific findings, plant-derived bioactives of interest against PPNs, together with agronomical, industrial and economical constraints that may hamper the effective delivery of such solutions in the field. Lessons learned from this ongoing project helped address some research issues related to the efficacy of nematode biocontrol solutions developed within the project (illustrated by several posters and oral presentations within the 7th ICN). They also point toward the need to question and revisit current land and pest management practices.

Keywords: Knot nematodes - Cyst nematodes - Biocontrol - Crop - Diseases.

Co-occurrence of soil microbial communities and root-knot nematode populations in strawberry farms, Egypt.

Mahfouz Abd-Elgawad[1] (mahfouzian2000@yahoo.com), Hassan Abd-El-Khair[2], Mostafa M.A. Hammam[2], Wafaa M. A. El-Nagdi[2], Mahfouz M. M. Abd-Elgawad[2]

[1]*Plant Pathology Dept, National Research Centre, Giza, Egypt;* [2] *Plant Pathology Department, National Research Centre, El-Behooth St., Dokki, Giza, Egypt.*

Root-knot nematodes (*Meloidogyne* spp., RKN) can infect and cause real yield losses of strawberry in Egypt [1]. Their control using unsafe nematicides has boosted interest in benign alternative methods to minimize these losses. Specific disease suppressive soils can safely contribute to the management of root-knot nematodes. These soils usually have high population densities of specific microbial species leading to enhanced levels of the nematode control. This study examined such microbial presence with RKN in twelve strawberry farms in Egypt. The population density of root-knot nematodes, *i.e*. second-stage juveniles (J2) in soil as well as J2, galls, egg-masses, females and eggs in roots of strawberry plants in six farms at El-Ismailia governorate were less than those recorded in six farms at El-Beheira governorate. The bacterial counts differed in the strawberry rhizosphere. The total microbial count of spore-forming bacteria was high after two months, whereas the aerobic bacteria count was high four months after sowing. Differences of the fungal counts were recorded in the plant rhizospheres among the farms. These included *Aspergillus niger*, *Aspergillus terreus*, *Aspergillus* spp., *Penicillium citrinum*, *Penicillium chrysogenum*, *Penicillium* spp., *Trichoderma* spp., *Fusarium solani*, *Rhizoctonia solani*, *Rhizopus* spp. and other common fungi. The study discusses the possible role of the encountered soil microbial communities and other factors that can cooperatively manage root-knot nematodes or root-rot fungi in strawberry plants [2].

Keywords: Strawberry - Biological control - Nematodes - Root-rot fungi - Suppressive soil.

References:

- [1] Abd-Elgawad, 2019. Bull NRC. 43: 1-13.
- [2] Topalović et al., 2020. Front Microbiol. 11: 1-15.

Genome-wide association identifies genomic regions involved in production of nematicidal compounds in the biocontrol fungus *Clonostachys rosea*.

Mudassir Iqbal[1] (mudassir.iqbal@slu.se), Martin Broberg[1], Deepak Haarith[2], Anders Broberg[3], Kathryn E. Bushley[4], Mikael Brandström Durling[1], Maria Viketoft[5], Dan Funck Jensen[1], Mukesh Dubey[1], Magnus Karlsson[1]

[1] *Forest Mycology and Plant Pathology, Swedish University of Agricultural Sciences, Uppsala, Sweden*; [2] *Department of Plant Pathology, University of Minnesota, Saint Paul, United States*; [3] *Department of Molecular Sciences, Swedish University of Agricultural Sciences, Uppsala, Sweden*; [4] *Department of Plant and Microbial Biology, University of Minnesota, Saint Paul, United States*; [5] *Department of Ecology, Swedish University of Agricultural Sciences, Uppsala, Sweden*

Biological control is a promising approach to reduce plant diseases caused by nematodes to ensure high productivity in future agricultural production. As large-scale genomic sequencing becomes economically feasible, the impact of single nucleotide polymorphisms (SNPs) on biocontrol-associated phenotypes can be easily studied across entire genomes of fungal populations. In this study, we made use of 53 whole-genome re-sequenced *Clonostachys rosea* strains to perform a genome-wide association (GWA) study for *in vitro* antagonism against plant-parasitic nematodes. Potato dextrose broth culture filtrates from *C. rosea* was inoculated with the root-lesion nematode *Pratylenchus penetrans* and nematode mortality was determined after 24 h of incubation. *In vitro* antagonism assay against *P. penetrans* displayed a significant variation among *C. rosea* strains and suggests that GWA of the trait is possible. An empirical Bayesian multiple hypothesis testing approach identified a total of 279 SNP markers significantly (local false sign rate ≤ 10-10) associated with the trait. Two non-ribosomal peptide synthetase genes (*nps4* and *nps5*) were present in the genomic regions associated with the nematicidal activity. Gene deletion strains of *nps4* and *nps5* genes were generated and showed increased growth and conidiation rates compared to the wild type. Culture filtrates from *C. rosea* Δ*nps4* and Δ*nps5* strains exhibited reduced nematicidal activity and immobilised nematodes to a significantly ($P \leq 0.05$) lower number compared to the wild type after 24 h of incubation. Furthermore, Δ*nps4* and Δ*nps5* strains showed reduced biocontrol efficacy in a naturally nematode infested soil in a pot experiment and failed to reduce the populations of nematodes in soil or in roots of wheat as efficiently as the wild type strain. Taken together, we show that NPS4 and NPS5 are biocontrol factors in *C. rosea*, presumably by producing a hitherto unknown non-ribosomal peptide compound with nematicidal properties.

Keywords: Biological control - GWAS - Plant growth - Nematode - Non-ribosomal peptide synthetases.

Pochonia chlamydosporia var. mexicana response to physicochemical factors, rhizosphere colonization and egg parasitism.

María Gabriela Medina-Canales[1] (gabymedina28@gmail.com), Adriana Joselly Del Razo-Méndez[1], Brenda Estefany Ortiz-González[1], Gabriela Susana Alba-Ramírez[1], Rosa Helena Manzanilla-López[2], Alejandro Tovar-Soto[1]

[1] *Escuela Nacional de Ciencias Biológicas, Instituto Politécnico Nacional de México, Mexico, Mexico*; [2] *CEPROBI, Instituto Politécnico Nacional de México, Yautepec, Mexico*

Pochonia chlamydosporia var. chlamydosporia has been extensively investigated as a fungal egg parasite of plant-parasitic nematodes and also as a beneficial endophyte [1]. Other varieties of the fungus, such as *P. chlamydosporia* var. mexicana, have been little investigated in relation to the most suitable conditions for development, response to soil physicochemical factors, rhizosphere colonization and potential as biocontrol agents of Meloidogyne spp. Recent studies of selected strains of *P. chlamydosporia* var. mexicana (Pcp21, Pcp2) and *P. chlamydosporia* var. *chlamydosporia* (Anarsu, Anarma 1b, Pc10 and Anarma 1c) have included: i) strain tolerance to high temperature (30°C-34°C), pH (4.5-8.5) and osmotic stress (NaCl 0.25M-1.5M); ii) fungal strain colonization of tomato and corn roots inoculated with conidia or chlamydospores; and iii) serine protease VCP1 activity [2] and how to increase egg parasitism. Preliminary results show that strains are thermostable, tolerating acidic and alkaline pH. *Pochonia chlamydosporia* var. mexicana Pcp21, *P. chlamydosporia* var. *chlamydosporia* Anarsu and Pc10 are osmotolerant and Anarma 1b, Anarma 1c and Pcp2 are moderate halophytes. Studies show that at 5, 10, 15 and 20 days post-inoculation, *P. chlamydosporia* var. mexicana chlamydospores, unlike conidia, are more efficient in rhizosphere colonization [3]. The Mexican strain Anarma 1c showed maximum VCP1 activity at 10 days but a second peak occurred at 25 days in chitosan supplemented rice medium. The percentage of M. arenaria egg parasitism also increased in chitosan supplemented medium. These preliminary results support the need for studies to assess Mexican strains as potential biological control agents for use by local farmers.

Keywords: *Pochonia chlamydosporia* var. chlamydosporia - Osmotic stress - Nematophagous fungi - VCP1 - *Meloidogyne*.

References:

- [1] Manzanilla-López et al., 2013. J. Nematol. 45(1): 1-7.
- [2] Esteves et al., 2009. Mycol Res. 113(8): 867-76.
- [3] Maciá-Vicente et al., 2009. Ann. Appl. Biol. 155: 391-401.

WORKSHOP 5

Nematode-bacteria symbiosis

Nematode-bacteria symbiosis

S. Patricia Stock (spstock@csuchico.edu)

College of Agriculture, California State University Chico, Chico, CA, USA

Associations between nematodes and bacteria range from fortuitous to obligate and from beneficial to pathogenic. The ubiquity and diversity of nematode-bacterium symbioses make them an excellent model to understand the key questions in symbiosis. Through this workshop speakers will discuss bacteria interactions with nematodes with lifestyles that span from free living to necromenic and parasitism/pathogenesis.

Take my breath away – physiological adaptations of symbiotic marine nematodes to oxic-anoxic interfaces.

Silvia Bulgheresi (silvia.bulgheresi@univie.ac.at)

University of Vienna, Vienna, Austria

Eukaryotes may experience oxygen deprivation under both physiological and pathological conditions. Because oxygen shortage leads to a reduction in cellular energy production, all eukaryotes studied so far conserve energy by suppressing their metabolism. However, the molecular physiology of animals that do not appear to suppress their metabolism is poorly known. One such animal is *Laxus oneistus,* a marine nematode invariably coated by its sulfur-oxidizing symbiont *Candidatus* Thiosymbion oneisti. This symbiotic nematode does not enter suspended animation even upon 6-day-long incubations in anoxic seawater. Here, transcriptomics and proteomics showed that, whether in anoxia or not, *L. oneistus* mostly engages in ubiquitination, energy generation, oxidative stress response, immune response, development, and translation. Importantly, ubiquitination genes appeared upregulated when the nematode was subjected to anoxic sulfidic conditions, together with genes involved in autophagy, detoxification and ribosome biogenesis. We hypothesize that degradation pathways were induced to recycle damaged cellular components (mitochondria) and misfolded proteins into nutrients. Remarkably, when *L. oneistus* was subjected to anoxic sulfidic conditions, lectin and mucin genes were also upregulated, potentially to promote the attachment of its thiotrophic symbiont. Furthermore, the nematode appeared to survive oxygen deprivation by using an alternative electron carrier (rhodoquinone) and acceptor (fumarate), to rewire the electron transfer chain. On the other hand, under hypoxia, genes involved in costly processes (e.g., amino acid biosynthesis, development, feeding, mating) were upregulated, together with the worm's Toll-like innate immunity pathway and several immune effectors (e.g., Bacterial Permeability Increasing proteins, fungicides).
In conclusion, we hypothesize that, in anoxic sulfidic sand, *L. oneistus* upregulates degradation processes, rewires oxidative phosphorylation and reinforces its coat of bacterial sulfur-oxidizers. In upper sand layers, instead, it appears to produce broad-range antimicrobials and to exploit oxygen for biosynthesis and development.

Keywords: Symbiosis - Anoxia - Immunity - Ubiquitination - Toll pathway.

Nematode – bacterial interactions in a beetle ecosystem: ‚Boom and bust' dynamics of predatory nematode *P. pacificus*

Ralf J. Sommer (ralf.sommer@tuebingen.mpg.de), Tess Renahan

Max-Planck Institute for Biology Tübingen, Tübingen, Germany

The nematode *Pristionchus pacificus* is a genetic and evolutionary model to elucidate the ubiquitous insect-nematode-bacteria complex. *P. pacificus* has a facultative affiliation with scarab beetles and relies on the bacteria that decompose the beetle carcasses as a food source. Though, when population densities increase, resources can become scarce; *P. pacificus* can respond by inducing either the dauer stage or the predatory mouth form, the latter allowing for feeding on other nematodes. However, given that scarab beetles spend most of their lives (as eggs, grubs, and pupae) below ground, little is known about the dynamics and succession of the beetle-nematode-microbe interaction. We initiated field studies to understand the succession of the nematode in association with its microbiome in the beetle ecosystem. The dynastid beetle *Oryctes borbonicus* from La Réunion Island serves as an exceptional model system due to its nematode infestation rate of more than 90% at one location, which provides ample material for experimental manipulation. Specifically, we caught and killed beetles, placed their carcasses back in the soil, and analyzed the decaying beetle over 12 weeks in two-week intervals. We found that the worm exhibits a boom and bust strategy on the carcass with an unexpected dynamic pattern that is not induced by specific bacterial species, though the presence of the nematode influenced the bacterial composition on the beetle carcass. Additionally, the microbiome of the nematode derives largely from the decaying beetle and not from the surrounding soil. We will present our current understanding of the *Pristionchus*-beetle association and will also describe our recent analysis of the nematode association with beetle grubs in the soil. These studies help towards a comprehensive understanding of the true ecology of a nematode model system.

Keywords: Necromenic nematode - Pristionchus pacificus - iIntegrative evolutionary biology - Beetles - Oryctes borbonicus.

To have and to hold: on the mutualistic partnership of *Steinernema* nematodes and their *Xenorhabus* symbionts.

S. Patricia Stock[1, 2] (spstock@csuchico.edu)

[1] *School of Animal and Comparative Biomedical Sciences, Univesrity of Arizona, Tucson, AZ, USA*; [2] *College of Agriculture, California State University Chico, Chico, CA, USA*

Microbial symbionts play key roles in the survival, nutrition, reproduction and defense of their eukaryote hosts. They can also directly or indirectly manipulate their behavior to enhance their own fitness. One fascinating example of host utilization and manipulation is represented by *Xenorhabdus* bacteria, which interact with two eukaryotic hosts: *Steinernema* nematodes, and insects [usually soil-dwelling, immature stages], to complete their life cycle. *Steinernema-Xenorhabdus*-insect partnerships are extremely diverse and represent a model system in ecology and evolution to investigate symbioses between invertebrates and microbes. Insect virulence and reproductive fitness of the nematode-bacterium partnership is tightly associated and dependent to each other and also utilizing its resources to complete their life cycle [1]. Indeed, a growing body of evidence shows *Xenorhabdus* symbionts play an important role not only on the survival of the nematodes in the soil environment, but also on their development and reproduction in the insect host [2]. Disruption of the symbiotic partnership can have detrimental effects on the fitness of both partners [3, 4]. In this presentation I will summarize recent findings on the symbiotic relationship between *Steinernema* and *Xenorhabdus*, focusing on the contributions of the bacterial symbionts to nematode fitness and in their ability to successfully access and utilize an insect host.

Keywords: Symbionts - *Steinernema* - *Xenorhabdus* - Mutualism - Fitness.

References:

- [1] Stock. 2019. Current opinion in insect science, 32: 22-27.
- [2] McMullen et al. 2017. Microbiology, 163: 510-522.
- [3] McMullen et al. 2017.BMC evolutionary biology, 17:1-14.
- [4] Lefoulon et al. 2022. Frontiers in physiology, In Press.

WORKSHOP 6

Nemaplex and NINJA: Features and Uses

Nemaplex and NINJA: Features and Uses.

Sara Sánchez-Moreno[1] (sarasm@inia.es), Ron de Goede[2]

[1] *Department of Environment and Agronomy, INIA-CSIC, Madrid, Spain;* [2] *Wageningen University & Research, Wageningen, The Netherlands*

In this 90-minute practical workshop, we will explore the Nemaplex and NINJA on-line tools to analyze and comprehend different ecological attributes of soil nematode communities. Attendants interested in soil food web ecology will have the opportunity to learn through play and perform hands-on practice with the different features provided by Nemaplex and NINJA and work with their own datasets if desired. Participants will require a laptop computer with internet access in order to complete the hands-on exercises (see 3.b). If desired, participant may bring their own data to complete their own analyses (data should be formatted according to the instructions at https://shiny.wur.nl/ninja).
PhD students and early-career scientist are especially welcome!

Briefly review of the origins and evolution of Nemaplex (http://Nemaplex.ucdavis.edu) **from around 1979 to the present day.**

Howard Ferris

No abstract submitted.

.....

Demonstration of the features and navigation of the Nemaplex website, including how to retrieve information on the characteristics, taxonomy, and ecology of plant-parasitic and free-living nematodes, exploring databases, field and laboratory methodologies, techniques, and teaching resources.

.....

Demonstrations of the use of NINJA Faunal Analysis feature to evaluate the ecosystem and soil food web conditions suggested by specific nematode assemblages. Participants will be split in two groups that will by various means (a. NemaNINJA card game; b. NINJA online internet tool) gain experience on how to analyze and interpret ecological data for monitoring, management and policy guidance. User-feedback and suggestions on the design and future development of Nemaplex and NINJA will be welcome.

WORKSHOP 7

Grape vines nematode management

Grape vines nematode management.

Daniel Esmenjaud[1] (daniel.esmenjaud@inrae.fr), Cesar Bauer Gomes[2]

[1] *INRAe, Sophia Antipolis, France*; [2] *Embrapa Clima Temperado, Pelotas, Brazil*

Nematodes affecting grapevines are very diverse and this crop suffers from major nematode pests belonging to both the Tylenchida and Dorylaimida. In the context of the worldwide challenges that viticulture has to face with the removal of chemical nematicides and the global warming, the workshop will consider diverse current aspects of nematode management in grapevines. It will give an overview of the nematode problems affecting viticulture in major producing countries from North and South America and Europe. The workshop will also deal with more specific topics focused on resistance to dagger and root-knot nematodes.

Grape-associated phytoparasitic nematodes in Brazil.

Cesar Bauer Gomes (cesar.gomes@embrapa.br)

Embrapa Clima Temperado, Pelotas, Brazil

No abstract submitted.

Management approaches of plant-parasitic nematodes in grape in the changing agricultural landscape of California.

Andreas Westphal[1] (andreas.westphal@ucr.edu), Tom Buzo[1], Zin Thu Zar Maung[1], Gabriel Torres[2], Matthew Fidelibus[3], Andrew Walker[4]

[1] *Department of Nematology, University of California, Riverside, Parlier, CA, United States*; [2] *UCCE, Tulare, CA, United States*; [3] *Department of Viticulture and Enology, University of California, Davis, Parlier, CA, United States*; [4] *Department of Viticulture and Enology, University of California, Davis, Davis, CA, United States*

California is a globally significant producer of raisin, table, and wine grapes. These commodities are becoming increasingly specialized, with unique scion varieties and production practices. In all commodities, production efficiency remains key to success, and protection from plant-parasitic nematodes is important for ensuring adequate yield and quality. Vineyards are notorious for concomitant infestations with multiple species of plant-parasitic nematodes. Often *Meloidogyne* spp., *Mesocriconema xenoplax*, *Paratylenchus hamatus*, *Tylenchulus semipenetrans* and *Xiphinema* spp. occur in various combinations. In the deep-rooting soils and extended growing conditions of California, population densities can be high and distributed at least in the upper 1.5 m of soil. Management options need to target suppression in this large volume of soil. The objectives of this study were to (a) identify grape rootstocks with resistance to multiple plant-parasitic nematodes, and (b) test new chemistry for pre-plant soil applications and post-plant application use. Four commercial rootstocks and two experimental rootstocks were planted to microplots with sandy soil. Plants were inoculated with population mixes of root-knot nematodes and ring nematodes from three different vineyards. In the second year, rootstocks were budded to a common scion. In the first year after planting, root-knot nematode population densities were low with the exception of the susceptible own-rooted 'Chardonnay'. No root-knot nematodes were found on 1017A or GRN1 rootstocks. GRN1 was also free of ring nematode. In the second year, only GRN1 was virtually free of root-knot and ring nematodes. Graft compatibility varied among the rootstocks. Despite the beneficial nematode characteristics, GRN1 grafted poorly under field-grafting conditions. In other efforts, bench grafting was found to overcome such challenges. Chemical suppression of nematode communities of various species was challenging. In preplant plus postplant application patterns, chemically intensive strategies offered some plant growth improvements, but yield improvements were limited during the 5-year study. In 2-3-year studies in >30-year-old vineyards, limited yield improvements followed postplant applications. In times of declining availability of preplant tools, intensifying genetic protection approaches to nematode management offers promise, but may require improved propagation techniques to make effective use of the most resistant stocks.

Keywords: Chemical nematode suppression - Grafting - Nematode-resistant rootstocks.

Plant parasitic nematodes infesting grapevines in Portugal.

Carlos Gutiérrez-Gutiérrez (carlosg@uevora.pt), Manuel Mota

NemaLab, MED-Universidade de Évora, Évora, Portugal

Grapevine serve as host to a large variety of plant-parasitic nematodes, the most common of which include *Criconemoides* spp. (ring nematodes), *Meloidogyne* spp. (root-knot nematodes), *Paratylenchus* (pin nematodes), *Pratylenchus* spp. (root-lesion nematodes), *Xiphinema* spp. (dagger nematodes) and same species of spiral nematodes (*Helicotylenchus* spp.) and stubby-root nematodes (*Paratrichodorus* spp) [1]. Among these, *Criconemoides* spp., *Meloidogyne* spp., *Pratylenchus* spp., and *Xiphinema* spp. are the top four genera ranked at the top in the list of major damaging plant-parasitic nematodes for grapevine worldwide [1]. However, pin nematodes are polyphagous plant ectoparasites with at least seven species that can cause damages to plants by their direct feeding on root cells [2]. Members of the genus *Paratylenchus* have not been studied in detail in Portugal [3]. Thus, updated information on the present biodiversity including molecular data, occurrence and distribution of pin nematodes in Portuguese vineyards is lacking. Sequences of ribosomal RNA (rRNA)-genes are a powerful molecular diagnostic tool for nematode identification. Nowadays, an integrative taxonomic approach based on morphological and molecular data is a strategy widely used for an accurate nematode identification [4, 5]. From 2019 to 2021, a survey to determinate the occurrence, distribution and biodiversity of *Paratylenchus* species infesting grapevine in Portugal, was carried out in main grape-growing areas of this country. Nematodes were identified using an integrative approach based on the combination of morphological characterizations and molecular analysis. Contrasting morphological hypotheses with molecular data provided rapid detection of six species, specifically *P. goodeyi, P. hamatus, P. pedrami, P. tenicaudatus, P. variabilis*, and *P. veruculatus*. *Paratylenchus* nematodes were detected in the 15.9 % of the soil samples analysed. The population densities of *Paratylenchus* spp. ranged between 10 to 250 nematodes/500cm3 of soil. No disease symptoms were observed on aboveground plant parts even though a heavy soil infestation occurred.

Keywords: D2-D3 - *Paratylenchus* - Plant-parasitic nematodes - rRNA - Vitis.

References:

- [1] Teliz et al 2011. Plant Disease 91, 1147–54.
- [2] Ghaderi et al 2019. J. Crop Prot., 8 (3): 243-257.
- [3] Macara A.M. 1988. Bol San. Veg. Plagas 14, 185–225.
- [4] Clavero-Camacho et al. 2021. Animals 2021, 11, 1161.
- [5] Clavero-Camacho et al. 2021. Plants 2021, 10, 1454.

Natural rootstock resistance to control *Xiphinema index* and RKNs in Vines.

Daniel Esmenjaud (daniel.esmenjaud@inrae.fr)

INRAe, Sophia Antipolis, France

No abstract submitted.

Resistance durability to *X. index* in grapevine.

Van Chung Nguyen (nchung.ppri@gmail.com)

Plant Protection Research Institute, Ha Noi, Vietnam

No abstract submitted.

FORUM 1

Young nematologists Network

Young nematologists Network.

No abstract submitted.

7th International Congress of Nematology
ICN 2022
Antibes Juan-les-Pins - France

"Crossing borders: a world of nematode diversity and impact to discover"

European Society of Nematologists

ABSTRACTS THURSDAY 5 MAY

PLENARY SESSIONS

Caenorhabditis elegans and other nematodes as biological models.

Lindy Holden-Dye (l.m.holden-dye@soton.ac.uk), Vincent O'Connor, Fernando Calahorro, Helena Rawsthorne, James Dillon, Emily Feist

University of Southampton, Southampton, United Kingdom

All animals must have the capability to detect environmental cues in order to drive appropriate behavioural responses. Understanding the intricacies of these mechanisms, and in addition how they can be modified in the context of past experience, is one of the fundamental questions in biology. The nematode *C. elegans* has provided an excellent experimental system to deliver insight into this fundamental neurobiology that has trans-phyla relevance. We have developed a paradigm that reveals inter-organismal signalling between *C. elegans* larval progeny and parental adult worms that is dependent on oxytocin-like signalling, a neuropeptidergic pathway related to social behaviour in mammals. Intriguingly, the experimental observations suggest that in the presence of abundant food i.e. a replete bacterial lawn, larvae signal to adult worms inducing them to leave the food patch and forage away from the primary food source [1]. In further experiments, we have shown that this social behaviour is regulated by neuroligin, a synaptic protein for which mutations are a rare highly penetrant determinant implicated in human autism spectrum disorder [2]. We envisage this genetic determinant of social circuits will have important roles in other classes of nematodes where social behaviour may impact on fitness.

References:

- [1] Scott E, Hudson A, Feist E, et al. An oxytocin-dependent social interaction between larvae and adult C. elegans. Sci Rep. 2017;7(1):10122. Published 2017 Aug 31. doi:10.1038/s41598-017-09350-7.
- [2] Rawsthorne H, Calahorro F, Feist E. et al. Neuroligin dependence of social behaviour in C. elegans provides a model to investigate an autism associated gene bioRxiv 2020.02.03.931592; doi: https://doi.org/10.1101/2020.02.03.931592.

Using *A. suum* to study the intestine of animal/human parasitic nematodes.

Makedonka Mitreva (mmitreva@wustl.edu)

Washington University School of Medicine, St. Louis, United States

The biological and molecular complexity of nematodes has impeded research on development of new therapies for treatment and control. We have focused on the versatility of the nematode intestine as a target for new therapies. To that end, it is imperative to establish a broad and deep understanding of the molecular architecture underlying intestinal cell functions at the pan-Nematoda level.
Multi-omics data form *Ascaris suum* were generated to uncover the evolutionary principles underlying both conserved and adaptable features of the nematode intestine. Whole genomes were used to reveal the functional potential of the nematodes, tissue-specific transcriptomes provided a comprehensive understanding of genes that are expressed in the adult nematode intestine, and comparison of selected core species was used to determine the pan-Nematoda intestinal transcriptome. Differentially expressed transcripts were also identified among intestinal regions, with the largest number expressed at significantly higher levels in the anterior region, identifying this region as the most functionally unique compared to middle and posterior regions. Profiling intestinal miRNAs targeting these genes identified the conserved intestinal miRNAs. Ans single-cell RNAseq has identified intestinal cell subpopulations. Proteomics of intestinal cell compartments assigned proteins to several different intestinal cell compartments (intestinal tissue, the integral and peripheral intestinal membranes, and the intestinal lumen). Finally, applying advanced bioinformatic approaches delineate intestinal functional categories of seminal importance to the parasite survival. The targets were linked to drug-like compounds that were experimentally tested and validated.
The data provide the most comprehensive compilation of constitutively and differentially expressed genes, gene regulators and proteins of the nematode intestine providing knowledge that is essential to understand molecular features of nematode intestinal cells and functions of fundamental importance to the intestine of all parasitic nematodes. The identified nematode intestine toxicants target different proteins involved in various processes and as good candidates has been further explored.

Keywords: Parasitic nematodes - Intestine - Genomes - Transcriptomes - Proteomes.

Legislation and Regulatory aspects of plant nematodes.

Manuel Mota (mmota@uevora.pt)

NemaLab-MED, University of Évora, Évora, Portugal

Plant parasitic nematodes (PPN) are responsible for heavy economic losses, estimated to reach over USD$ 100 billion annually, in agricultural crops as well as forestry. Examples include the root knot nematode (*Meloidogyne* spp.), cyst nematodes (*Heterodera* and *Globodera* spp.), root lesion nematodes (*Pratylenchus*spp.) and *Bursaphelenchus xylophilus*, the pinewood nematode. The spread of PPN constitute a serious economical, ecological as well as a biosecurity issue. The increase in world globalization and trade of agricultural commodities has become a challenge to food security and plant health services worldwide. Example: the import of ornamentals (e.g. *Ficus* species) from China to Europe, which has grown from 2 to 4 million plants in the last decade. International cooperation regarding management of plant health threats began in 1951, with the creation of the International Plant Protec tion Convention (IPPC). Other organizations collaborate with IPPC, namely the European and Mediterranean Plant Protection Organization (EPPO), National Plant protection organizations (NPPs) and Regional Plant Protection Organizations (RPPs). Internationally, 15 nematode species are regulated by more than 20 countries in international quarantine legislation. The EPPO has established a list and grading system regarding recommendations for nematode regulation as quarantine species; a few examples are: *Meloidogyne enterolobii*, *Aphelenchoides besseyi*, *Bursaphelenchus xylophilus*, Potato cyst nematodes (PCN) *Globodera rostochiensis* and *G. pallida*, *Nacobbus aberrans*. The European Comission Council Directive 2000/29/EC, which replaced and consolidated Directive 77/93 EEC, is the main guideline in the EU for plant health regulation. Case studies, including the *A. besseyi* species complex which damages soybean in Brazil and leguminous plants in Costa Rica, *Meloidogyne enterlobii* in the United States and Europe and *M. ethiopica* in South America and Europe, as well as the pinewood nematode, *Bursaphelenchus xylophilus*, will be presented. In this last case, and following Council Directive 2000/29/EC, Portugal informed the European Commission (EC) and implemented a major phytosanitary strategy with the purpose of controlling and eradicating the nematode.

Keywords: Regulation - Legislation - Plant parasitic nematodes - Quarantine.

ORAL SESSION 17

Integrated nematode management

Integrated management of cyst nematodes in agricultural important crops in China.

Deliang Peng[1] (pengdeliang@caas.cn), Ricardo Holgado[2], Huan Peng[1], Wenkun Huang[1], Jingwu Zheng[3], Shulong Chen[4]

[1] *Institute of Plant Protection, CAAS, Beijing, China*; [2] *Norwegian Institute of Bioeconomy Resear, Oslo, Norway*; [3] *Zhejiang University, Hangzhou, China*; [4] *Institute of Plant Protection, HAAFS, Daoding, China*

The family Heteroderidae contains cyst forming plant parasitic nematodes that are known to cause serious crop yield losses. Surveys showed the occurrence of four genera of cyst nematodes belonging to Heteroderidae (*Heterodera*, *Globodera*, *Cactodera*, *Cryphodera*), and 20 species were recorded. At present, the species found are *Heterodera avenae (= Heterodera sturhani)* and *H. filipjevi* on wheat and barley. *H. glycines* and *H. sojae* on soybean. *H. elachista* on rice, *H. cruciferae* on oil rape, *H. zeae* on maize, *H. goettingiana*, *H. vallicola*, *H. ripae*, *H. sinensis*, *H. hainanensis*, *H. fengi*, *H. guangdongensis*, *H, koreana*, *Globodera artemisiae*, *Cactodera cacti*, *C. estonica*, *C. thornei* and *Cryphodera sinensis*. Integrated management relies on the knowledge of species/ pathotype, the relationship between initial nematode density and yield. Our studies showed that Cereal cyst nematodes (CCN) *H. avenae*, *H. filipjevi* occurs in 16 provinces [1]. *H. filipjevi* occured in Henan, Anhui, Shandong, Qinghai and Ningxia Hui Autonomous Region. The dominant species was *H. avenae*, with high population densities indicated that CCN are serious threats for wheat production. Estimates of yield loss are Henan 18-35%, Hebei 15-20%, Beijing Suburb 11-18%, and Qinghai 28-24% [1, 2]. To elucidate the status of Chinese CCN, morphological and molecular studies were conducted. The studies showed that morphological characters of *H. sturhani* are similar to *H. avenae.* The ITS sequences of *H. avenae* was shown to be heterogeneous and to differ from other e.g. European and Indian populations. The molecular disparities are intra-species variation of *H. avenae*. Consequently, *Heterodera sturhani* is a junior synonym of *Heterodera avenae.* One generation of *H. avenae* is completed during a growing season. For diagnostic of *H. avenae* and *H. filipjevi* methods based on SCAR-PCR and LAMP were developed. *H. avenae* pathotypes Ha 43 and Ha 91 were distinguished. Integrated management systems, based on careful nematode identification, in combination with crop rotations using non-hosts, and knowledge of tolerance or resistance in varieties, for different ecotypes, are essential for CCN management. Twenty cultivars with resistance to CCN were screened and applied at field level. For CCN control a seed coating “Gannong III” was developed. We acknowledge the support of the Chinese Special R & D Funds for Public Benefit Agriculture (201503114), and The Agricultural Science and Technology Innovation Program.

Keywords: *Heterodera avenae* - Diagnosis - Integrated management - Pathotype - Synonymization.

References:

- [1] Peng, D.L.,et al.,2009, Cereal cyst nematodes: status, research and outlook, pp. 29-34.
- [2] Smiley, R. W. et. al., Plant Disease, 2017, 101:1692-1720.

Bridging the vegetable IPM GAP in Africa: from here to sustainerty.

Danny Coyne (d.coyne@cgiar.org)

East Africa, IITA, Nairobi, Kenya

Urban and peri-urban vegetable production systems in sub-Saharan Africa tend to be highly intensive, high value and with high pest and disease pressures. These systems, which supply fresh, often perishable, produce to urban centres attract a high reliance on the use of synthetic pesticides towards the rapid suppression of pest and disease invasions. Among these various pests and diseases, root knot nematodes (RKN; *Meloidogyne* spp.) are ubiquitous across sites and crops, but paradoxically are probably the least considered or recognized constraint. They are responsible for high levels of damage and can ultimately result in reduced yields and premature host-plant death. However, RKN infection also impacts plant host health and is associated with higher levels of pest and disease incidence. Damaged roots are also less able to access nutrients and water. Damaged, inefficient root systems are therefore a drain on limited water resources and lead to waste and nitrate contamination through excessive fertilizer applications. Raised pest and disease levels on RKN-infected plants create greater pesticide reliance, all of which negatively impacts the sustainability of the system. Studies showed that using healthy seedlings, free from nematode infection, improved crop health and yields of tomato and pepper. By observing good agricultural practices (GAP), pesticide use was reduced to 25% of normal farmer practice, while further improving crop yields. Resistance screening identified cultivars with resistance, which when combined with GAP has the potential to substantially reduce losses due to RKN infection. Management of RKN in these intensive production systems, using healthy seedling systems and GAP can reduce losses, waste and chemical contamination, improving their safety and sustainability.

Keywords: Sub Saharan Africa - Food security - Climate change - Food safety - Soil health.

IPM options in cotton production with plant parasitic nematodes in the USA.

Kathy Lawrence (lawrekk@auburn.edu)

Entomology & Plant Pathology, Auburn University, Auburn, AL, United States

Across the cotton belt, it is estimated that 0.1 to 9.5% of the cotton crop was lost to plant- parasitic nematodes which is estimated at an economic loss of nearly 26 million dollars in 2019. Specifically, cotton yields over the last five years in a reniform nematode infested field with an average initial population of 5,000 reniform/100 cm3 of soil, had 50% less yield compared to an identical field that had no detectable reniform nematodes. Research trends are the use of **crop resistance** for effective control of nematodes. *Meloidogyne*-resistant cotton cultivars suppress nematode reproduction and several *Meloidogyne*-resistant cultivars are available. *Rotylenchulus reniformis* resistance is expected to be available in 2021. Additionally, **rotation** with non-host crops have shown potential for plant-parasitic nematode management. Rotations of cotton, soybean and corn in a *M. incognita* race 3 field found that the nematode populations continued to increase when the host crop was consistent over years. However, rotations between susceptible crops suppressed the *M. incognita* population keeping the nematodes reproductive factors to half of that produced on continuous cotton. Cover crops generally increase soil microbial activity and biological diversity; they can suppress some populations of plant-parasitic nematodes. The use of cover crops for suppression of *M. incognita* and *R. reniformis* requires caution due to the broad host range of nematodes and susceptibility of cover crops may increase nematode populations. Nematicides have been proven effective for management of plant-parasitic nematodes. Aldicarb is the granular nematicide available in cotton production. Seed applied nematicides such as abamectin, thiodicarb, fluopyram, and fluazaindolizine are accepted by growers and provide management in fields with lower populations. In-furrow spray nematicide fluopyram combined with imidacloprid is also available. Oxamyl is a foliar applied pesticide that provides additional management of *R. reniformis* and *M. incognita*, often in conjunction with previously mentioned nematicides. Additional options are biologicals such as *Bacillus firmus* (Poncho/VOTiVO) and *Bacillus amyloliquefaqciens* strain PTA-4838 (Aveo), seed applied formulations that have efficacy against nematodes. In most cases, a stand-alone option for control of plant-parasitic nematodes is not sufficient and a combination of management practices is needed to keep nematode numbers below the economic threshold.

Keywords: Cover crops - Crop resistance - Nematicides - Rotations.

Can *Meloidogyne* and *Helicotylenchus* populations be manipulated using soil nutrient levels?

Mieke Daneel[1] (mieke@arc.agric.za), Lesego Matlala[2], Hendrika Fourie[3]

[1] *Crop Protection, ARC - Tropical and Subtropical Crops, Nelspruit, Mpumalanga, South Africa*; [2] *Natuurboerdery Research Centre, ZZ2 Bertie Van Zyl, Mooketsi, Limpopo, South Africa*; [3] *Integrated Pest Management, North West University, Potchefstroom, North West, South Africa*

Plant-parasitic nematodes (PPN) cause serious problems in tomato production, especially in protected production conditions, resulting in considerable yield losses if not controlled. Because of consumer demand to use less toxic chemicals, considerable efforts are exploited to investigate alternative control strategies. One such strategy is to create an environment in the soil rhizosphere that is unsuitable for *Meloidogyne* and highly suitable for *Helicotylenchus*. In a survey conducted in 26 nethouses used for tomato production in Limpopo Province, South Africa, it was observed that *Meloidogyne* was absent or found in lower proportions when *Helicotylenchus* was present and vice versa. Soil samples were collected in 6 nethouses, with nematode samples taken 8 weeks after planting over a 3-year period of. Soil data included Ca, K, Na, S, P, Mg, pH, density, acid saturation, Na:K, Mg:K, Ca:Mg, %Mg, %Ca, %Na and %K, while soil texture was also determined. Coinertia analysis used to determine the relationship between soil parameters and nematode numbers, identified Ca, K and acid saturation as being positively correlated with low *Meloidogyne* numbers and high numbers of *Helicotylenchus* and Criconematidae while Mg:K, Na:K and %Mg were positively correlated with high *Meloidogyne* levels and low levels of *Helicotylenchus* and Criconematidae. Significant differences were found between the high and low values of the above-mentioned elements and *Meloidogyne* except for Mg:K. When data were reorganized in three categories namely high, medium and low *Meloidogyne* infections, Ca, K and acid saturation values increased between the three categories while values of %Mg, Mg:K and Na:K decreased. When data were grouped according to rootstocks, irrespective of the tolerance level of these rootstocks, the presence of high levels of Ca, K and acid saturation were associated with lower *Meloidogyne* and higher *Helicotylenchus* levels. XY projections also showed that low *Meloidogyne* numbers were found in the high Ca, K and acid saturation plots. *Helicotylenchus* was found to be present in these plots, while it was often absent in the other plots. This information confirms that soil environment can influence *Meloidogyne* infection and that manipulating the balance between Ca, K and acid saturation levels can be exploited to reduce nematode damage.

Keywords: Alternative control strategies - Protected production - South Africa - Tomato.

Pathogenic variability of the population of *Pratylenchus brachyurus* in cover crops.

Lais Fontana[1] (laisffontana@hotmail.com), Bruna Orlandini Toninato[2], Glaucia Leticia Sete da Cruz[3], Ana Cristina Da Silveira[4], Angélica Sanchez Melo[3], Guilherme Tarini[3], Cláudia Regina Dias-Arieira[3]

[1] *Fitopatology, Maringá State University, Umuarama, Brazil*; [2] *Fitopatology, Maringá State University, Umuarama, Brazil*; [3] *Maringá State University, Umuarama, Brazil*; [4] *Universidade Paranaense, Umuarama, Brazil*

The great damage that *Pratylenchus brachyurus* causes in soybean and corn-producing regions in Brazil is already known, however, the pathogenic variability of populations of this species has not been studied. In this way, eight populations of *P. brachyurus* were collected in different locations in the Brazilian territory and evaluated for pathogenicity in soybeans, and three cultivars of millet (*Pennisetum Americanum)* recommended for crop rotation for this nematode. The populations were collected in the field, in areas producing soybeans and corn, with the population «1» being collected in the city of Campo novo do Parecis, population «2» in Lucas do Rio Verde, population «3» in Vale do Araguaia, population "4" in Sorriso, population "5" in Luis Eduardo Magalhães, population "6" Primavera do Leste, population "7" in Rondonópolis and population "8" in Umuarama.The populations were multiplied separately in a greenhouse. The experiment consisted of a completely randomized design, with 7 replications and 8 treatments (populations). For that, 500 juveniles of each population were inoculated in soybean (Monsoy 6210), and millet (ADR300, ADR500, and BRS 1501), and evaluated after 70 days, regarding the total number of nematodes, nematodes per gram of root, and reproduction factor (RF). In the soybean culture, the RF varied from 1 to 5, with the population 4 and 6, differing statistically from the others, and presenting the highest number of nematodes. For millet, the number of nematodes varied in relation to cultivar. For the cultivar BRS 1501, population 8 also showed greater multiplication (RF = 2), while the other populations had RF below 1, and some 0. For ADR300, the RF varied from 0 to 0.8, with the population 2 differed statistically from the others, and showed greater multiplication, even so, the RF was below 1 (RF = 0.8). Thus, it was possible to observe that the RF for *P. brachyurus*, varies according to the location of the population, indicating that some cultivars may be resistant in certain regions and susceptible in others, leading to an increase in the population of this nematode.

Keywords: Root-lesion nematode - *Pennisetum Americanum* - Crop rotation - Millet - Reproduction factor.

A complex between the plant parasitic nematode and a fungus - reevaluating *Pratylenchus capsici* disease etiology.

Sigal Braun Miyara[1] (sigalhor@volcani.agri.gov.il), Patricia Bucki[1], Qing Xue[1], Svetlana Duvrinin[2], Ohad Avraham[1], Nathalia Sichov[1], Abraham Gamliel[3]

[1] *The Entomology and Nematology Department, Agricultural Research Organization Volcani Center, Rishon LeZion, Israel*; [2] *Extension Service (Shaham), Ministry of Agriculture & Rural Development, Rishon LeZion, Israel*; [3] *The Department Agricultural Engineering, Growing, Production and Environmental, Agricultural Research Organization Volcani Center, Rishon LeZion, Israel*

During the last two years we have identified a new nematode species *Pratylenchus capsici* in Israel [1] leading to devastating damage on pepper crop resulting in stunted growth and significant yield reduction. Molecular phylogeography suggests that contemporary gene flow is prevented among different agricultural farms, while population dispersal from weeds (*Chenopodium album* and *Sonchus oleraceus*) to pepper occurs on a relatively small scale. Metabarcoding analysis of soil microbial community from *P. capsici* infested roots indicated that *Olpidium* species are widely presented in *Pratylenchus* introduced root-lesion, and might be a faithful companion associated with roots infected by *P. capsici*. We have established a system based approach to manage *P. capsici* in two commercial pepper farms. The treatments include root destruction of the previous crop before removal, followed by soil disinfection approaches using different soil fumigants, and combination with soil solarization. Application of soil fumigants together with soil solarization of fumigants showed effective nematicidial and fungicidial ability and greatly inhibited *P. capsici* when compared with the other treatments. The integrated approach enabled a healthy crop and commercial pepper production. The Migratory ability of *P. capsici* was studied in commercial by sectioning soil layers for the presence of nematodes. We found that nematodes reside in deeper soil layers, although they were controlled in the upper soil layer by the disinfestation process. The migratory potential of these nematodes is still under study. Altogether, results obtained through our research will facilitate developing innovative management strategies through tailoring them within the agricultural practices and according to *P. capsici* etiology and characteristics.

Keywords: *Olpidium* - *Pratylenchus* - Interaction - Management - Control.

References:

- [1] Qing, X., Bert, W., Gamliel, A., Bucki, P., Duvrinin, S., Alon, T., Braun Miyara, S. 2019. Phytopathology 109(5):847-858.

S17-PF1

Synergies between climate change impacts and conservation tillage practices on agricultural soils functionality.

Paula Lillo[1] (paula.lillo@inia.es), Mª Ángeles Hernández[2], Miguel Ángel Porcel[1], Sara Sánchez-Moreno[1]

[1] *Department of Environment and Agronomy, INIA-CSIC, Madrid, Spain*; [2] *Department of Environmental Biology, University of Navarra, Pamplona, Spain*

According to the predictions of the sixth IPCC assessment report, temperatures might increase by 2.7°C by the end of this century. In the Mediterranean region, droughts will become more frequent and precipitation will decrease. The aim of this project is to assess the effect of climate change on soil nematodes, soil functioning, and crop yield in a semi-arid, rain-fed cereal system in central Spain. Three experimental treatments were established: 1) open-top chambers were used to increase soil temperature (IT) by 2.2 °C, 2) rain-out shelters were used to reduce rainfall (RR) by 30%, 3) both IT and RR simultaneously (ITRR), and 4) a regular climate control (CK). Also, to test if reduced tillage might mitigate the effects of climate change in the soil, two tillage systems were applied (minimum vs standard). Each tillage system was replicated 4 times so 8 field plots were established, in which all treatments plus the CK were set up. At the end of the first cropping cycle, nematode-based indices indicated a poor successional stage and a stressed and nutrient-depleted system. Bacterivores reduced their % abundance in the RR compared to the other treatments, while % of herbivores increased. Soil respiration rate increased in the IT and ITRR treatments compared to CK. Rising temperatures altered wheat phenology, leading to a 20-day earlier anthesis and increased shoot biomass. Wheat grain weight was reduced by RR. Tillage affected soil properties, and total nematode biomass, bacterivore and composite nematode footprints, phosphatase enzyme activity, and organic carbon were higher in the minimum tillage compared to standard tillage plots. In general, our results show that both the increase in the ambient temperature and the reduction in precipitations had an impact on nematode trophic groups, CO2 emissions, and crop yield, while key elements in soil functioning such as organic carbon and phosphatase activity are promoted by conservation tillage practices.

Keywords: Climate change - Soil food web - Nutrient and carbon cycling - Crop performance - Agricultural system.

S17-PF2

A new mode of action classification scheme for nematode control agents ("nematicides") - nematode working group of IRAC.

John A. Wiles[1] (john.wiles@corteva.com), Ionit Iberkleid[2], Huazhang Huang[3], Tim C. Thoden[4], Ralf Nauen[5], Andrew Crossthwaite[6], Ekaterini Riga[7], Marc Rist[5], Matthias Gaberthueel[8], Russell Eldridge[9]

[1] *Corteva Agriscience, Cambridge, Cambridgeshire, United Kingdom*; [2] *Adama Agricultural Solutions, Airport City, Israel*; [3] *BASF Corporation, Research Triangle Park, North Carolina, United States*; [4] *Corteva Agriscience, Munich, Germany*; [5] *Crop Science Division, Bayer AG, Monheim, Germany*; [6] *Jealotts Hill International Research Centre, Syngenta Crop Protection, Bracknell, United Kingdom*; [7] *Stine Research Center, FMC Agricultural Solutions, Newark, Delaware, United States*; [8] *Syngenta Crop Protection AG, Basel, Switzerland*; [9] *Valent BioSciences, Libertyville, Illinois, United States*

Plant-parasitic nematodes continue to be recognized as an important threat to food security and crop production globally. Researchers worldwide remain very active to find and develop new solutions to be used in integrated nematode management programs. With recent developments and commercialization of several nematode control agents (both chemical and biological) it was considered an appropriate time to create a mode of action (MoA) classification scheme for nematode control agents (often generically termed "nematicides") in the same manner as those already successfully created for insecticides and other disciplines. Therefore, under the auspices of IRAC (Insecticide Resistance Action Committee) a new working group was formed "the Nematode Working Group". MoA Classification schemes have proven very useful in offering local, regional and global government agencies, growers, advisors, consultants, universities and extension staff clarity on different control options. They also facilitate IRM (Integrated Resistance Management) and IPM (Integrated Pest Management) in alternation or rotation-based programs by providing a harmonized platform for improved labelling of commercial products acquired by farmers. The newly proposed "Nematicide MoA Classification" incorporates a wide range of active ingredients, organisms, conventional chemical nematicides, fumigants and agents of biological origin that have demonstrated nematode control. The classification follows the same principles as for other schemes, and the groups names start with the letter N to denote "nematicide/nematode control agent". The proposed groupings include; Carbamates (Group N-1A), Organophosphates (Group N-1B), Avermectins (abamectin, Group N-2), Pyridinylmethyl benzamides (fluopyram, Group N-3), Tetramic acids (spirotetramat, Group N-4), a group of novel compounds (Group N-UN) with unknown MoA's, including fluensulfone, fluazaindolizine and tioxazafen, and fumigants (Group N-UNX). The various biologicals for nematode control have been divided into three groups; Bacteria (Group N-UNB), Fungi (Group N-UNF), and botanical/animal Extracts (Group N-UNE). As with other MoA Classification schemes, when new information becomes available, the nematicide MoA Classification scheme will be revised and updated to incorporate this information. This poster will show the proposed classification scheme, which will also be available by open access on the IRAC website - https://www.irac-online.org/teams/nematodes/.

Keywords: Nematicides - Mode of action - Plant-parasitic nematodes - Biological control agents - Bionematicides.

S17-PF3

Damage threshold, population dynamics and host-status of *Meloidogyne chitwoodi* on five selected crops.

Misghina Goitom Teklu[1] (Misghina.goitomteklu@wur.nl), Fantahun F Addisu[1], Thomas H Been[1], Corrie H Schomaker[2], Johnny Visser[3], Leendert P.G Molendijk[3]

[1] *Plant Sciences Group, Wageningen University and Research, Wageningen, Netherlands*; [2] *Schomaker Quantitative Solutions, Randwijk, Netherlands*; [3] *Plant Sciences Group, Field crops, Wageningen University and Research, Lelystad, Netherlands*

A pot experiment was carried out to investigate the host-status of five selected crops to *Meloidogyne chitwoodi* in a climate-controlled glasshouse. A susceptible (Urselina) and a resistant (MEL-2) sugar beet *(Beta vulgaris)*, Black oat *(Avena strigosa)*, Alfalfa *(Medicago sativa)*, Chicory and Belgian endive (*Cichorium intybus* L) were sown in 8 and 6 kg pots at a range of 9 nematode densities (Pi) to assess damage and host-status. Crops were harvested after 3 or 4 months. Extra replications at Pi = 16 and 32 J2 (g dry soil)-1 were used for those crops with an extended growing period and were harvested after 6, 9 and 12 months. Natural decline of nematodes was monitored by inoculating pots without hosts at Pi = 16 J2 (g dry soil)-1 which were assessed after 4, 6 and 9 months. Fresh weight of roots, haulm and produce of each crop was measured to estimate yield. Final population densities (Puff) were estimated as the sum of nematodes extracted from the soil and the roots. The effect of Pi on the total fresh weight (TFW) and Pf, after 3 and 4 months, was modelled using Seinhorst's yield and population dynamics, respectively. Except for sugar beet Urselina and Alfalfa at Pi >16 J2 (g dry soil)-1, no yield losses were observed. Sugar beet Urselina and Black oat can be considered good hosts, compared to the other crops tested, with maximum multiplication rates (a) = 9.78 and 29.51 and maximum population densities (M) = 7.71 and 34.36 J2(g dry soil)-1, respectively. Sugar beet (MEL-2) had both a lower a = 0.001 and M = 0.01 J2(g dry soil)-1, providing a resistance level of > 99% compared to Urselina. In addition, Alfalfa, Chicory and Belgian endive proved to be poor hosts with parameter values for a ≤ 0.14, and M ≤ 0.19 J2 (g dry soil)-1; at every Pi¸ the Puff was reduced. The reduction under sugar beet MEL-2 and Chicory was equal to the reduction caused by the population decline in the absence of a host. At Pi = 16 and 32 J2 (g dry soil)-1 the Pf seemed hardly to be affected by the different growing periods. As a result, resistance levels of sugar beet MEL-2 did not change during the growing period which suggests that testing for resistance can be done after 4 months. The resistant sugar beet MEL-2 and the poor hosts, Alfalfa, Chicory and Belgian endive, can be used in crop rotations for the management of M. chitwoodi, reducing population densities to grow crops susceptible to quality reduction like potato, carrots and black salsify.

Keywords: Modelling - Non-linear regression - Resistance.

ORAL SESSION 18

Nematodes as bioindicators

Historical perspective on nematodes as indicators.

Deborah Neher (deborah.neher@uvm.edu)

Department of Plant and Soil Science, University of Vermont, Burlington, VT, United States

As an *ad hoc* reviewer for journals, I find many authors unaware of the multi-decadal history of nematode indices. I partially blame the transition from publishing in paper to electronic in the period of 1985-1995. We need to build on the past rather than repeat it. For example, we know nematode communities are useful indicators and that they correlate with ecosystem function, especially nitrogen mineralization. Groundbreaking research in the 1990's empirically verified the statistical reliability of index variations, sample sizes, appropriate references, and calibration by ecosystem and land use type [4]. Sampling designs, high throughput molecular tools, and databases of trophic group assignments are available. The current hurdle is ecological interpretation. Interpretation requires calibration and quantification of response effects. Interpretation becomes achievable if we identify and evaluate suites of sentinel genera or species rather than extrapolate from coarse identification, such as trophic groups. Sentinel taxa are those that respond consistently in response to specific types of soil disturbance [1,2]. Focusing on sentinel taxa is a potential solution for reduction of the notorious spatio-temporal variation of soil nematodes and reduction of the sheer numbers of different taxa to identify in each sample. With the renewed interest in soil health and expanding databases of amplicon sequences, it is time to complete the research to identify and verify sentinel taxa [2] that predictably respond to specific disturbance types so tool kits can be accessible to non-specialists [3]. Large-scale implementation of nematodes as soil biological indicators requires both a critical labor pool and an empirically based interpretation.

Keywords: Biomarkers - Community structure - Ecological function - Ecological succession - Maturity index.

References:

- [1] Fiscus and Neher, 2002. Ecol. Appl. 12: 565-575.
- [2] Li et al., 2005. Ecotoxicology 14: 419-429.
- [3] Neher and Stürzenbaum, 2006. Comp Biochem Phys C 144: 279-285.
- [4] Neher, 2010. Annu. Rev. Phytopathol. 48: 371-394.

Nematode responses to oak forest dieback in Mediterranean landscapes.

Sara Sánchez-Moreno[1] (sarasm@inia.es), Jorge Curiel Yuste[2]

[1] *Department of Environment and Agronomy, INIA-CSIC, Madrid, Spain*; [2] *IKERBASQUE Basque Foundation for Science, Basque Centre for Climate Change, Bilbao, Spain*

The current increase of severe droughts associated with climate change is one of the main causes of the observed dieback in Mediterranean holm-oak (*Quercus Ilex* subs *ballota*) forests. Effects of forest dieback in soils are greatly variable and depend on a number of local factors, but generally include increased soil organic C due to increased litter inputs, alterations in soil nutrient contents, changes in the nitrogen cycle, and detriment effects on soil microbial communities and their functioning. There is little evidence, however, of the effects of forest dieback on soil faunal diversity. In this study, we assessed the effects of holm-oak dieback and induced nutrient shifts on soil microfaunal communities and their relationships with soil properties and functioning. We studied 13 Holm oak forest sites affected by tree dieback in the Iberian Peninsula and assessed the relationship between soil properties, microbial functions, and nematode diversity under healthy, defoliated, and dead trees at each site. We found that nematode abundances were variable and significantly patchy across sites. Bacterivore and herbivore nematodes were the most abundant nematode trophic groups. Nematode abundance increased with increasing levels of soil C only if soil P was available, indicating that nematode abundances might be P-limited in these semi-arid systems. Bacterivore nematodes were especially affected by tree death, since tree dieback switched the relationship between bacterivore abundance and soil nutrient contents. Opportunistic bacterivores, able to exploit ephemeral bacterial resources blooming after soil organic enrichment, seemed more resilient to tree death than generalist ones, while fungivores did not clearly respond to forest dieback. We found complex and unexpected effects of tree dieback on soil microfaunal communities, which should receive further attention.

Keywords: Dieback - Diversity - Richness - Drought.

Nematodes communities used for soil health indication in midwestern United States agricultural systems.

Carmen Ugarte (cugarte@illinois.edu)

Department of Natural Resources & Environmental Sciences, University of Illinois at Urbana-Champaign, Urbana, Illinois, United States

Nematode communities have been widely proposed as integrative indicators of soil health because they are present in all positions in the soil food web and reflect shifts in the community as influenced by management. Our team has been evaluating nematode communities in two different agricultural settings; urban agriculture systems in old industrial cities that are challenged by concerns of soil pollution, and large-scale organic grain production (corn, soybean, and wheat) systems with varying levels of management intensity. We used multiple approaches for analysis and interpretation including analysis of the nematode community structure based on the abundance of free-living and plant-parasitic families, and evaluation of metrics that describe the status of the food web in relation to soil function. The discussion will expand on the use of these metrics to inform management that enhance soil biological activity and functional diversity and that optimize production and environmental goals.

Keywords: Nematode community indicators - Nematode community analysis - Organic farming systems - Soil health - Urban agriculture.

Characterization of the biological state of soils by studying nematofauna as bio-indicators: ELISOL methodology.

Cecile Villenave (cecile.villenave@elisol.fr), Anne Jimenez, Manon Trambolho, Hélène Cérémonie, Camille Chauvin

ELISOL environnement, Congénies, France

Since the 1990s, numerous studies have shown that, due to their taxonomic and functional diversity, nematodes are the quantitative and qualitative reflection of the soil food web from both biodiversity and of functioning points of view and can be used as indicators [1, 2].
The main functional types of nematodes found in the soil are:
(1) microbivore nematodes (bacterial- and fungal- feeders) which provide information on the microbial compartment, the soil organic matter dynamics and the nutrients recycling; (2) nematodes of higher trophic levels (omnivores and predators) which reflect physical or chemical disturbances of the environment; (3) phytophagous nematodes (facultative or obligate) which provide information on the nature and state of the plant cover and, also, the risk of yield loss.
The soil health diagnosis given by ELISOL environnement is based on nematofauna analysis and takes into account the abundances of free-living nematodes (microbivores & carnivores) and phytophages (+ main phytoparasitic nematodes genera) found in soils as well as several nematofaunal indices: structure index (SI), enrichment index (EI), nematode channel ratio (NCR) and classical diversity indices (Shannon).
During the DANE project (2016-2018- PIA) [3], ELISOL environnement, using a database of more than 5,000 analyses of nematofauna (which now reaches 7,000 analyses), developed a references system dedicated to the diagnosis of the biological soil state in different contexts of agricultural and natural soils: field crops, vines, meadows, vegetable crops, forests. Prediction models of the main nematofauna parameters have been developed.
The soil physico-chemical characteristics, as well as geographical locations, which are also included in the database, are still under study leading soon to their integration into the interpretation standards.
Moreover, a specific interpretation framework for anthroposols has been developed, taking into account the initial constituents used for the creation of technosol, its age and its use (Biotubes Project).

Keywords: Nematofauna - Soil biological activity - Bio-indicators.

References:

- [1] Villenave et al., 2018. Innovations Agronomiques. 69, 47-60.
- [2] Ferris et al., 2001. Applied Soil Ecology. 18, 13-29.

Zinc effects on terrestrial nematodes: from field studies towards a toxicokinetic approach.

Péter Nagy[1] (nagy.peter@mkk.szie.hu), Krisztina Hrács[1], Lola Virág Kiss[1], Zoltán Sávoly[2], Gergely Boros[1], Katalin Posta[3]

[1] *Department of Zoology and Animal Ecology, Szent István University, Faculty of Agricultural and Environmental Sciences, Gödöllő, Hungary*; [2] *Individual Researcher, Budapest, Hungary*; [3] *Institute of Genetics and Biotechnology, Microbiology Research Group, Szent István University, Faculty of Agricultural and Environmental Sciences, Gödöllő, Hungary*

Free-living nematodes are widely used indicators of disturbances, including heavy metal pollutions. Zinc is an essential microelement with potential toxic effects. Its range of environmental exposure spreads from agriculture, used as fertilizer and animal fodder additive, through communal sources to industrial activity. From ecotoxicological aspects, nematological effects of Zn can be quite diverse, ranging from beneficial to toxic impacts, as concluded from the results of various field and laboratory studies. Moreover, effects of nanoparticles, like ZnO, may considerably differ from those of bulk forms. In particular, little is known of the fate of these compounds in the individuals affected. Therefore, we performed a set of monospecific experiments attempting to clarify acute toxicity and uptake patterns of Zn in nematodes belonging to different feeding types and c-p groups. Mortality tests and TXRF spectrometric measurements were performed on *Panagrellus redivivus* (c-p1 bacterivore) and *Xiphinema vuittenezi* (c-p5 plant feeder) individuals. Although increasing Zn-concentrations caused greater mortality and higher internal concentrations in both species, their stress responses and uptake patterns were different. *Panagrellus* took up a significantly higher amount of zinc than *Xiphinema.* Particle size hardly influenced the internal zinc content of nematodes, only in the highest concentration did *Xiphinema* show higher uptake for nano ZnO than for its bulk counterpart. For *P. redivivus,* both particles were similarly toxic, while in *X. vuittenezi* nano ZnO caused much higher mortality than the bulk form. Apart from this, *X. vuittenezi* was not more sensitive to the treatments than *P. redivivus.* It is notable that initial potassium content of the Zn-treated *P. redivivus* specimens was always significantly lower ($p < 0,01$) than that of the untreated individuals. In *X. vuittenezi*, there was no significant decrease along the treatments in general, but untreated *Xiphinema* specimens had lower potassium levels than *Panagrellus*. Potassium content is important as this metal is considered to play a role in zinc detoxification. Our results confirm the importance of bridging the gap between studies focusing on supraindividual and infraindividual organisation levels. Revealing toxicokinetics and elementary interactions of the most relevant substances may help tackle pollution effects.
The work was partly supported by the research project No. 2017-1.3.1-VKE-2017-00001.

Keywords: Nano ZnO - Mortality - *Panagrellus redivivus* - *Xiphinema vuittenezi* - TXRF spectrometry.

Regenerative agriculture and its ability to restore soil ecosystem health and functioning.

Gerhard Du Preez (gerhard.dupreez@nwu.ac.za), Driekie Fourie, Ané Loggenberg

Unit for Environmental Sciences and Management, North-West University, Potchefstroom, South Africa

Regenerative agriculture is endorsed as a system that promotes environmental health, agroecosystem resilience, and sustainable food production. Central to this is healthy and functioning soil ecosystems. However, minimal information is available on the potential of soil ecological restoration after transitioning from conventional systems. We therefore aimed to investigate soil ecosystem health and functioning in regenerative-based crop production systems in the Eastern Free State of South Africa. Replicated soil samples were collected during the summer and winter growing seasons of 2019/2020 from multiple regenerative croplands, as well as from a conventional cropland (negative reference) and undisturbed grassland (positive reference). Nematode community data were used to calculate maturity, food web diagnostic, and metabolic footprint indices, while active carbon, soil respiration, total organic matter, and physico-chemical parameters (pH, electrical conductivity, metal and nutrient content, and texture) were also measured. Our results showed that the regenerative croplands presented significant ecological restoration with structured food webs and fertile soils, as well as increased ecosystem functioning. Redundancy analyses confirmed that the observed effects mainly resulted from the change in systems (i.e., from conventional to regenerative). The undisturbed grassland also consistently presented mature and structured food webs, supporting the idea of using these habitats as positive reference sites for agroecosystem restoration. Inorganic nitrogen, copper, and nickel content, as well as pH, also presented significant effects. For example, a negative, linear relationship was evidenced between inorganic nitrogen and soil food web structure ($R^2=0.51$; $p<0.0001$), as well as between copper and soil respiration (microbial activity) ($R^2=0.6$; $p<0.0001$). This is indicative of the potential negative effects of agrochemicals on soil ecological restoration. These results clearly show that regenerative agriculture can restore the health and functioning of soil ecosystems and ultimately promote service delivery.

Keywords: Sustainable food production - Soil health - Ecosystem services - Agroecosystem resilience.

S18-PF1

A study on the ecological impact of recycling derived fertilisers (RDFs) using nematodes as environmental bioindicators.

Anna Karpinska (anna.karpinska@itcarlow.ie), Thomaé Kakouli-Duarte

Department of Science and Health, Institute of Technology Carlow, Carlow, Ireland

Soil is the main source of nutrients needed for plant growth. The three main such nutrients, nitrogen (N), phosphorous (P) and potassium (K), are routinely applied by farmers via artificial mineral fertilisers, whose production, mining and transportation require large amounts of energy, disturb and pollute the environment. At the same time, via food consumption, nutrients end up in waste without their placement back to food production. Nutrient recovery technologies are employed to produce recycling derived fertilisers (RDF) from three large waste streams, sewage sludge, food waste and manure, in the form of struvite or ash that can replace usage of conventional mineral fertilisers in farms. However, before these RDF are applied to the soil and used as the main source of nutrients, they need to undergo an ecological risk assessment. Nematodes are the most abundant and widespread soil roundworms, involved in nutrient cycling and enabling these to be utilised by plants. These organisms are sensitive to pollutants and environmental disturbance and therefore ideal as biological indicators of environmental change. This project is an investigation on the ecological impact of RDF by studying their effects on nematodes in single species ecotoxicology experiments and terrestrial nematode community analyses via the following approaches: investigating the effects of RDF on (1) beneficial entomopathogenic nematodes (*Steinernema feltiae*) in microcosm experiments looking at sub-lethal end points such as nematode infectivity and reproduction, (2) *Caenorhabditis elegans* in toxicity bioassays observing nematode mortality and (3) nematode communities in RDF testing field trials in three different locations in North West Europe, in Ireland, Belgium and France, as part of the ReNu2Farm project (www.nweurope.eu/renu2farm). For community analyses nematode DNA was extracted from soil samples, and the 18S rRNA gene was analysed, using suitable primers [1], via subsequent sequencing and further bioinformatic analysis. Absolute nematode abundance expressed in Molecular Operational Taxonomical Units (MOTUs) is currently examined using Nematode INdicator Joint Analysis (NINJA) to exclude any adverse effects of RDF on soil nematode diversity compared to that in control sites. This work is currently ongoing and its results will be presented and discussed at ICN 2020.

Keywords: Free-living nematodes - Ecotoxicology - Environmental risk assessment - Phosphorous - Food security.

References:

- [1] Bhadury et al., 2006. Development and evaluation of a DNA-barcoding approach for the rapid identification of nematodes. 320: 1-9.

S18-PF2

Evaluating the environmental risks of microplastics using nematodes as bioindicators.

Sebastian Höss[1] (hoess@ecossa.de), Marie-Theres Rauchschwalbe[2], Hendrik Füser[2], Walter Traunspurger[2]

[1] *Ecossa, Starnberg, Germany*; [2] *Animal Ecology, Bielefeld University, Bielefeld, Germany*

Microplastic (MP) pollution is a serious threat for aquatic and terrestrial ecosystems as high loads of mismanaged plastic waste have been dumped to land and rivers and finally reached marine ecosystems. Due to the environmental persistence of most polymers and fragmentation processes, the size spectrum of differently shaped plastic particles shifts more and more towards micro- and nanoplastics (< 5mm). Moreover, due to biofilm formation even low-density polymers are forced to sediment in aquatic ecosystems, accumulating in sediments. Thus, benthic organisms, such as nematodes, are often exposed to high concentrations of MPs.
We assessed the impacts of MPs on the model organism *Caenorhabditis elegans*, on several other freshwater nematode species as well as on whole benthic nematode communities. We studied the ingestion and ecotoxicological effects of polystyrene beads in standardized test systems (ISO 10872) and population growth set-ups, using single-species approaches in both water and sediment matrix. Moreover, ingestion and effects of various microplastic types (shape and polymer type) were assessed in microcosm systems with an indigenous benthic freshwater community.
The ingestion of polystyrene beads was dependent on the size of the beads and the mouth opening (and thus also the feeding-type) of the nematode species [1]. Considering permanent exposure to MPs, particle ingestion and a simultaneous egestion by the nematodes resulted in constant body burdens of beads [2]. Independent of the ingestion, *C. elegans* responded dose-dependently to polystyrene beads with a decrease of reproduction, whereas the effects were related to the size and surface area of the beads [3]. Mechanistical studies revealed that the effects of the polystyrene beads were caused by the disturbance of food bacteria consumption [4]. The lower food availability was found to result in an allocation of energy reserves from reproductive output to stored lipid droplets, as revealed by Raman microscopy [5]. This indirect effect mechanism was partly confirmed by a microcosm study, showing a microplastic-induced shift in nematode feeding-types (decrease of bacterial feeders), though there was no indication for a direct chemical toxicity by the pollution-sensitive NemaSPEAR[%]-index. This comprehensive study stresses the inclusion of food-web related and indirect ecotoxicological effects on benthic invertebrates for a realistic risk assessment of microplastics in aquatic ecosystems.

Keywords: Microplastic - Toxicity - Ingestion - Microcosms - NemaSPEAR.

References:

- [1] Fueser et al., 2019. Environ Pollut. 255: 113227.
- [2] Fueser et al. 2020 Chemosphere. 261: 128162.
- [3] Mueller et al., Environ Sci Technol. 54 (3): 1790–1798.
- [4] Rauchschwalbe et al., 2021. Environ Pollut. 273: 116471.
- [5] Fueser, et al., Environ Pollut. 294: 118662.

S18-PF3

Nematodes of argan biosphere: Biodiversity and assessment of soil quality.

Amina Braimi[1] (aminabraimi7075@gmail.com), Amine Idhmida[1], Abdolhadi Ajerrar[2], Hinde Benjlil[1], Mohamed Ait hamza[1], Fouad Msanda[1], Zahra Ferji[3], Abdelhamid El mousadik[1], Thierry Mateille[4], El Hassan Mayad[5]

[1] *Laboratory of Biotechnologies and Valorization of Natural Resources, Faculty of Sciences Agadir, Ibn Zohr University, Agadir, Morocco*;
[2] *Biotechnology and Environmental Engineering Team, National School of Applied Sciences, Ibn Zohr University, Agadir, Morocco*;
[3] *Laboratory of nematology, Department of Plant Protection, Agronomy and Veterinary Institute Hassan II, Ait Melloul, Morocco*;
[4] *Biology Center for Population management, Research and development institute, Montpellier, France*; [5] *Laboratory of Biotechnologies and Valorization of Natural Resources, Ibn Zohr University, Faculty of Sciences, Agadir, Morocco*

Biodiversity is increasingly becoming an issue in ecological debates because of its significant role especially in soil ecosystems functioning. The Argan biosphere as an ecosystem is experiencing a massive qualitative and quantitative regression due to the several environmental constraints that affect its functioning, particularly via the response of its biodiversity. Soil nematodes have multiple biological and ecological characteristics that make them relevant bioindicators of soil functioning. They provide a wealth of information on the state of the soil food web, which is responsible for the decomposition of organic matter, nutrient mineralization and, consequently, soil quality. This study aimed to (i. Characterize the taxonomic and functional diversity of nematode communities in Argan soils in Souss- Massa region, (ii. assess soils quality in this region. In this context, nematode communities were analyzed in 27 natural soil samples collected from 8 different geographical areas from argan forest. Nematode extraction was carried out by elutriation. The extracted nematodes were counted and identified to the family and genus level. The results obtained showed a very rich nematofauna with 44 genera (free nematodes and plant parasitic nematodes) belonging to 23 families. These nematodes are distributed within five trophic groups: bacteriovores, herbivores, fungivores, omnivores and predators with a remarkable prevalence of bacteriovores, followed by herbivores indicating significant differences between some surveyed areas. The Cephalobidae family is the most represented among the bacteriovores, while the herbivore family is dominated by Dolichodoridae. The calculated nemato-fauna structural and enrichment indices indicated that the prospected soil show differences in the degree of disturbance and maturity on the one hand and in the type of dominant mineralization with their fertility on the other hand.

Keywords: Argan Forest - Nematodes - Trophic groups - Bioindicators - Soil quality.

ORAL SESSION 19

Future of nematology: legislation, education and training

Teaching nematology – what do students need to know?

Cynthia Gleason (cynthia.gleason@wsu.edu)

Plant Pathology, Washington State University, Pullman, Washington, United States

Plant-parasitic nematodes are becoming increasingly problematic in agriculture, but awareness of nematodes is still relatively low, even amongst agricultural students. Although most students do not aspire to be nematologists, there is demand for scientists with at least some plant nematology background. With this in mind, the question is raised: what are the basic tenets of plant nematology that all undergraduate agricultural/science students should know? Part of the difficulty in answering this question is that plant nematology is a multidisciplinary science that combines fields like genetics, plant physiology and molecular biology, in addition to nematode biology. Here we will discuss whether, in an -omics world, students still need to learn nematode morphology or other nematode basics. Can a person call themselves a nematologist without ever having stepped into a field? In answering these questions, we will visit the two camps of nematology research - "basic" and "applied," and advocate that a better balance and communication is needed between the two. In addition to these philosophical issues, there are practical issues in teaching nematology that should be addressed. For example, in United States, nematology departments have almost all disappeared, and as a result, there are fewer and fewer classically trained plant nematologists to teach higher level courses. This predicates the need for a consolidated approach for updated, online nematology learning platforms. Moreover, there are currently a limited number of short intensive courses in nematology. These courses should be expanded as they are critical for graduate students and professionals who need more intensive nematology training but do not have instructors nearby. Lastly, phytonematology is still a small discipline, and in the USA, its scientific society is dwindling in numbers. Looking at the larger picture, I present a question to the audience– how can we better promote nematology to the public and encourage students to pursue an active and sustained career in nematology?

What is next? Nematology Education from a Ghent Perspective: together we create impact.

Inge Dehennin[1] (Inge.Dehennin@UGent.be), Laura Cortada-Gonzalez[1], Aäron Plovie[1], Danny Coyne[2], Solveig Haukeland[3], Hugues Baimey[4], Antoine Affokpon[5], Awol Seid Ebrahim[6], Beira Hailu Meressa[7], Nasamu Bawa[8], Christopher Okolo[9], Biodun Claudius-Cole[10], Julius Bulus[9], Hendrika Fourie[11], Antoinette P. Malan[12], Herbert Talwana[13], Shahasi Athman[13], Francis Onyilo[14], Njira Njira Pili[15], Shem Nchore[16], Pierre Abad[17], Matthew Back[18], Richard Sikora[19], Mieke Daneel[20], Raymond Collett[11], Elvire Line Sossa[5], Alfred Alumai[14], Dora Scott[1], Paul Sule Chindo (in memoriam)[9], Wim Bert[1]

[1] *Ghent University, Ghent, Belgium*; [2] *International Institute of Tropical Agriculture, Nairobi, Kenya*; [3] *International Centre of Insect Physiology and Ecology, Nairobi, Kenya*; [4] *University of Parakou, Parakou, Benin*; [5] *University of Abomey-Calavi, Cotonou, Benin*; [6] *Haramaya University, Haramaya, Ethiopia*; [7] *Jimma University, Jimma, Ethiopia*; [8] *Ahmadu Bello University, Zaria, Kenya*; [9] *Ahmadu Bello University, Zaria, Nigeria*; [10] *University of Ibadan, Ibadan, Nigeria*; [11] *North-West University, Potchefstroom, South Africa*; [12] *Stellenbosch University, Stellenbosch, South Africa*; [13] *Makerere University, Kampala, Uganda*; [14] *Muni University, Arua, Uganda*; [15] *Moi University, Eldoret, Kenya*; [16] *Kenyatta University, Nairobi, Kenya*; [17] *INRAe/Université Côte d'Azur, Antibes, France*; [18] *Harper Adams University, Newport, Shropshire, United Kingdom*; [19] *University of Bonn, Bonn, Germany*; [20] *Agricultural Research Council, Pretoria, South Africa*

The MSc in Agro- and Environmental Nematology (IMaNema) has trained more than 450 nematologists over the past 30 years, supported by the Belgian government (VLIR-UOS, International Course Programme - ICP) with operational funds and scholarships for students from countries in Africa, Asia and Latin-America. To address the needs and challenges related to nematology expertise in tropical and sub-tropical countries, a new IMaNema curriculum was implemented in 2016 and included the establishment of strategic partnerships with institutes in sub-Saharan Africa (SSA) to increase the outreach in the region and the creation of an open-access Nematology Digital Learning Platform (NDLP). A survey in 2017 conducted among SSA interviewees demonstrated that there had been a positive impact of several educational initiatives, showing an increase in the number of nematology lecturers at SSA universities and a significant number of nematology graduates returning to their countries to occupy qualified positions. However, notwithstanding many improvements, major challenges remain to increase the visibility of nematology in SSA, such as 1) a better inclusion of nematology as a discipline in the curricula of African universities; 2) the organization of trainings and promoting nematology in languages other than in English; and 3) reinforcing the nematologists' networks, both within SSA and with other research and academic groups around the world. To achieve these goals, a consortium of SSA and EU universities with the active engagement of (inter)national research institutions and private companies, collaborate in the Erasmus+ Capacity Building in Higher Education project *Nematology Education in Sub-Sahara Africa (NEMEDUSSA)*, running from 2021 until end 2023. Because the IMaNema programme will no longer be sponsored after 2027, the NEMEDUSSA project and the final 5 year VLIR-UOS ICP Connect project are part of its exit strategy for transferring nematology education to HEIs in SSA and for building up an active and strong Pan-African Nematology Network (PANEMA). Hence, the ICP Connect project foresees further identification of partner HEIs to set-up a joint programme through staff exchange, student mobility, local scholarships, joint course units in SSA, and co-sponsoring of PANEMA workshops. These actions aim to create impact and ensure continuation of a longstanding tradition of nematology education at Ghent University by jointly creating new centres of excellence in SSA.

Keywords: Capacity building - Education - Sub-Saharan Africa - International Master of Science in Agro- and Environmental Nematology.

Nematode legislation contributes to food sustainability by management of *Globodera rostochiensis* and *G. pallida* in Norway.

Ricardo Holgado[1] (ricardo.holgado@nibio.no), Christer Magnusson[1], Randi Knudsen[2]

[1] *Division of Biotechnology and Plant Heal, Norwegian Institute of Bioeconomy Resear, Ås, Norway*; [2] *Norwegian Food Safety Authority, Ås, Norway*

In Norway (PCN) *Globodera rostochiensis* and *G. pallida* are quarantine pests. PCN was recorded in 1955, generating the first regulation in 1956, requiring crop rotation with non-host a 4-years rotation of potato, and prohibiting movement of infested soil. The 2010 regulation prescribes surveillance and official control of infested and non-infested fields. Nematicides were never used so management relied on phytosanitary measures, resistant cultivars and crop rotation with non-hosts. Farmers must know the PCN-status of their fields, management options, and regulations. Surveys revealed a PCN infestation of about 5%. *G. rostochiensis* occurs in southern counties including Trøndelag, while *G. pallida* occurs locally in the southern counties Agder and Rogaland. *G. rostochiensis* contains the pathotypes Ro1, Ro2, Ro3 and Ro4, and *G. pallida* the pathotypes Pa1, Pa2 and Pa3. The most common is Ro1 with 98% of the finds [1]. Management of non-virulent *G. rostochiensis* (Ro1/Ro4) relies on 4-year crop rotation with non-hosts, and alternating susceptible and resistant certified seed potatoes. Detection of *G. pallida* or virulent *G. rostochiensis* results in 40-years ban on growing potato. Continuous cropping of resistant cultivars decimate Ro1 rapidly, so it is tempting to use resistant cultivars too often. Growing such cultivars in field trials resulted in Ro3 overcoming Ro1 after 10 years [1]. Growing resistant cultivars in soil with Ro1/Ro3 mixtures resulted in high populations of Ro3 after 5 years [1]. The decline in the absence of plant host, were studied in quarantine fields of *G. rostochiensis* (Ro3), *G. rostochiensis* (Ro1) and *G. pallida* (Pa 2/3) for 32, 18 and 12 years respectively. Baiting the soils with susceptible potato detected PCN in all soils after 4 months, demonstrating that *G. rostochiensis* survived for 32 years and *G. pallida* for at least 12 years without hosts [2]. Both species complete a second generation of J2s. Female development takes about 35 days, and cysts are formed in 40 days. Some populations formed cysts in 29 days [2]. Since 1956 fields with certified seed potatoes, have been monitored and found free from PCN. Norwegian regulations have been alert to new information, and have contributed in preventing PCN infestations in seed potato areas. Also further spread of *G. pallida* and virulent *G. rostochiensis* were prevented as each find was placed under quarantine [1, 2]. Norway has a register of PCN infested land that is free for access and use [3].

Keywords: *Globodera* - Managements - Norway - Potato - Quarantine.

References:

- [1] Holgado, R. & Magnusson, C. 2010. Aspects of Applied Biology, (103):87-92
- [2] Holgado, R. et al., 2015. Aspects of Applied Biology (130): 57-63.
- [3] Wesemael, W. et al., 2014. Potato Research. 57 (3): 365-366.

Nematology 101 - A collection of lectures for plant nematology as slide presentations, videos, and textbook chapters.

Jonathan Eisenback (jon@vt.edu)

School of Plant & Environmental Sciences, Virginia Tech, Blacksburg, VA, United States

The problem is, "How can I find the time and resources to do a better job of teaching nematology?"
Since only a few students are required to take a course in nematology which may be taught just every other year, there are very few resources that are widely available. Yet new information is revealed every year making it difficult to keep up. My light teaching load makes it hard to justify time spent on preparing for class, especially since there is no adequate textbook to teach an introductory course, and more advanced courses are even more taxing because there are even fewer students. The objectives of Nematology 101 are as follows: 1) To develop resources that are useful for teaching nematology - from a brief introduction to advanced courses, 2) To make these resources available on the internet, and 3) To use these resources to enhance the teaching of nematology around the world. The proposal is to produce PowerPoint presentations for numerous topics in nematology that can be used as is or changed according to the individual needs of the instructor, and to record the screen presentations to narrated video, and to make them available for free to anyone in the world from an internet video hosting service. Finally, each lecture will be developed into a chapter in a textbook, and printed on demand. Members of the International Federation of Nematological Societies may encourage the use of these resources to enhance the teaching of nematology around the world by stimulating the production of presentations in several languages. This collection of resources is projected to become the Open Access Library of Nematology Teaching. The videos will be free to everybody on the internet, the PowerPoints will be available as an added value of membership to Society of Nematologists, and the chapters from the textbook will be printed on demand. Several authors have already volunteered to write chapters, and since more than 125 chapters have been proposed, additional volunteers are needed.

Keywords: PowerPoint - Teaching - Lectures - Courses - Resources.

Best4Soil Nematode database tool to design clever crop rotations.

Leendert Molendijk (leendert.molendijk@wur.nl), Paulien van Asperen

Field Crops, Wageningen University and Research, Lelystad, The Netherlands

Healthy soils are of major importance for the future of the European horticultural and agricultural crop production. Especially in intensive production systems, nematodes and soil borne diseases are a major factor with a negative impact on soil health. Newly developed best practices and sound crop rotations permit the maintenance, improvement or restoration of soil health in Europe. The Best4Soil project creates factsheets, data bases, videos and organizes network activities. This network promotes knowledge that is ready for use on best practices for the control of nematodes and soil borne diseases: 1) the design of optimized crop rotations as a basis to prevent build-up of soil borne diseases, 2) measures that have a preventive effect, such as the use of compost, organic amendments, cover crops and green manures and 3) measures that reduce soil borne diseases such as (bio)solarisation and anaerobic soil disinfestation (ASD). In 22 European languages, Best4Soil provides open-access databases with information on the range of nematodes and soilborne pathogens that affect vegetable, arable and cover crops to help practitioners to build appropriate crop rotations and to choose innovative control strategies. For the nematode database the Dutch nematode scheme www.aaltjesschema.nl has been used as a starting point. For 20 arable crops, 29 vegetables and 21 green manure crops an extensive literature search resulted in an updated database for 32 nematode species and 77 fungal pathogens. Farmers and advisors can select the crops of their interest. The tool shows which nematodes and soil pathogens have to be taken into account and provides background information on biology and possibilities to prevent or to control these nematodes and soilborne pathogens. The information is the input to design clever crop rotations.
In this contribution the functionalities of the information tools are demonstrated and the possibilities of implementation and further development will be discussed.
www.best4soil.eu

Keywords: Host status - Crop rotation - Knowledge transfer - Decision support.

Regulation of potato cyst nematodes in Kenya.

Solveig Haukeland[1, 2] (shaukeland@icipe.org), Miriam W. Mbiyu[3], Moses Nyongesa[3], George Ngundo[4]

[1] *Plant Health, International Centre of Insect Physiology and Ecology, Nairobi, Kenya;* [2] *The Norwegian Institute of Bioeconomy Research (NIBIO), Norway;* [3] *Potato Research Centre, KALRO, Nairobi, Kenya;* [4] *KEPHIS, Nairobi, Kenya*

Potato cyst nematodes (PCN) were first detected in Kenya in 2015 (Mwangi et al. 2015). In 2016 - 2018 extensive surveys in the country found that PCN was widespread, affecting at least 20 potato growing regions (counties). In sampling and analysis activities involving more than 1300 farmers, 71% of the farms sampled were found infested with PCN. Incidence levels ranged from 12 to 100% . Both species were identified, *Globodera rostochiensis* and *G. pallida*, although the latter species from only one locality so far. Potato is a valuable food and cash crop in the East African region, and in Kenya it is ranked second in importance to maize. These findings therefore have serious implications for potato production and food security in the region. In Kenya potato yields have declined by around 50% in the last 15 years despite an increase in the area grown, current yields are about 7 - 9/t ha. PCN infestations are likely to be one of the contributing factors for these declining low yields. In Kenya, 90% of potatoes produced are grown by about 800,000 smallholder farmers often on less than an acre of land, hence crop rotation is a challenge. Currently there are 15 registered seed companies in Kenya dealing with certified potato seed, much below the current demand for clean seed. Soil analysis for PCN is available with focus on seed producers (conducted by KEPHIS - Kenya Plant Health Inspectorate Service), however capacity is currently lacking to tackle the large number of samples from small holder potato farmers requiring PCN testing for further decision making. In this presentation we will present current regulations being implemented to tackle the wide spread occurrence of PCN in Kenya and discuss the challenges encountered, as well as provide an update on PCN occurrence in the neighbouring countries.

Keywords: Potato cyst nematode - Kenya - Regulations - *Globodera*.

References:

- Mwangi, J. M., Kariuki, G. M., Waceke, J. W., and Grundler, F. M. 2015. First report of Globodera rostochiensis infesting potatoes in Kenya. New Dis. Rep. 31:18.

Pine wilt disease and the pinewood nematode in Portugal and Europe: it was 20 years ago today; what have we learned?

Manuel Mota (mmota@uevora.pt)

NemaLab - MED, University of Évora, Évora, Portugal

Pine Wilt Disease (PWD) is a worldwide conifer disease affecting native species from East Asia and Europe forestlands, and constituting a serious threat to countries with large areas of conifer species, namely of the genus *Pinus*. In May 1999, the pinewood nematode (PWN) was detected for the first time in Portugal (as well as in Europe), in dead maritime pine (*Pinus pinaster*) stands located in the Setúbal Peninsula. Portugal informed the European Commission (EC) and implemented a major phytosanitary strategy with the purpose of controlling and eradicating the nematode. Since 1999, several EC inspection missions have been carried out (DG SANCO). Despite a clear cut zone established around the initial detection, in 2006-2007, the PWN was able to spread to other areas, further North. The Portuguese Forestry Authority (ICNF) has nevertheless acted swiftly and intensly, by implementing road checks as well as annual surveys from the entire country, collecting several thousands of wood samples, especially from the 20-km buffer zone separating Portugal from Spain. These samples have been carefully analyzed. A clear picture is available of the presence and evolution of the PWN in Portugal. New approaches to controlling the nematode and its insect vector (*Monochamus galloprovincialis*) have recently been implemented by ICNF. Knowledge regarding the pine-PWN interaction at the cellular and molecular level is now also available, with some potential to use recent advanced methods of gene silencing. The complex interaction with the insect vector and the microbes present is also being studied. Some biocontrol methods (for the nematode and insect vector) have been also tested. The main control measures, however, are still centered around inspection (interception of the PWN and insect vector), intense national surveys, and treatment of infested wood, using heat methods. Some potential methods include the use of infra-red radiation. A good collaborative effort between Portuguese and Spanish authorities, as well as research institutions, has also been essential for the control of this disease.

Keywords: Pine wilt disease - Pinewood nematode - Control.

ORAL SESSION 20

Natural Products as nematicides

Attraction of *Meloidogyne* juveniles to essential oil constituents and derivatives.

Yuji Oka (okayuji@volcani.agri.gov.il)

Nematology Unit, Agricultural Research Organization, M. P. Negev, Israel

Nematode attractants may be useful for nematode control as traps or by confusing nematodes during their host-finding process.

Objectives:

To find attractants for second-stage juveniles (J2) of root-knot nematode species (*Meloidogyne* spp.) by screening essential oil constituents and their derivatives, and test their efficacy to prevent nematode infection in plants.

Methods:

Attraction of four *Meloidogyne* species, *M. hapla*, *M. incognita*, *M. javanica* and *M. marylandi*, to 60 aromatic compounds, mainly carboxylic acid, alcohols, aldehydes and phenols, was evaluated based on J2 densities in seven areas in an agar plate in 8.5-cm Petri dishes. J2 attraction was also evaluated by using trap tubes in 5.0- and 8.5-cm Petri-dishes filled with sand and trap sand balls in 50-cm3 test tubes. Some attractants were applied to the soil after inoculating lettuce seedlings with *Meloidogyne* J2.

Results:

Thirty-five compounds attracted *Meloidogyne* J2; however, *M. javanica*, *M. marylandi* and *M. hapla* were highly attracted to *o*-anisaldehyde, 2-methoxicinnamaldehyde, carvacrol and *o*-vanillin. No compound attracted *M. incognita* J2 in the same assay. Trap tubes containing *o*-anisaldehyde, *m*- and *p*-anisic acid and 2-methoxycinnamaldehyde highly attracted and killed J2 of all the four *Meloidogyne* spp. in 5-cm Petri-dishes filled with sand. However, *m*- and *p*-anisic acid and salicylic acid in the trap tubes attracted more J2 of the four *Meloidogyne* spp. than other attractants when these compounds were placed with other compounds in the 8.5 cm Petri-dish, except for salicylic acid that hardly attracted *M. hapla* J2. *p*-Anisic acid placed 4 or 8 cm above the nematode inoculation point in the test tube attracted more *M. incognita* and *M. javanica* J2 than did salicylic acid or *m*-anisic acid. 3-Methyl-*p*-anisaldehyde, *m*-anisyl alcohol and *m*-anisic acid reduced the number of lettuce root galls caused by *M. javanica* and *M. incognita*.

Conclusions:

The agar plate assay was not useful for *M. incognita* J2. Attractants would be better tested in soil. Several compounds were found to attract *Meloidogyne* spp. J2 in the sand and reduce the nematode infection. These attractants seem to have potential for nematode control.

Keywords: Aromatic compounds - *Meloidogyne* - Nematode attractants.

Nematotoxicity of vetiver extracts against root-knot nematodes by systemic defense activation in sa-pathway.

Kansiree Jindapunnapat[1] (Kansji@kku.ac.th), Buncha Chinnasri[2]

[1] *Dept. of Entomology and Plant Pathology, Khon Kaen University, Mueang, Thailand*; [2] *Dept. of Plant-Pathology, Kasetsart University, Bangkok, Thailand*

Activation of inherent plant resistance is one of the most promising measures widely used to control plant pests in the current situation of rising environmental and health concerns over the contamination of pesticides in the world society. In our studies, the nematocidal activity of vetiver compounds from vetiver grasses, *Vetiveria zizanioides,* on *Meloidogyne* spp. was extensively studied. The most active compounds were derived from 2-month-old vetiver aqueous and ethanol extracts. The extracts caused high second-stage juvenile (J2) mortality and possessed the property of nematode repellency as compared with French marigold (*Tagetes patula* L.) extract. Its nematicidal activity led to the analysis of their compounds. Gas chromatography – Mass Spectrometry (GC-MS) was used to identify the component of ethanol extracts and two major components including sesquiterpene acid 3,3,8,8 tetramethyltricyclo[5.1.0.0(2,4)] oct-5-ene-5-propanoic acid, and the sesquiterpene alcohol 6-isopropenyl-4,8a-dimethyl-1,2,3,5,6,7,8,8a-octahydronaph- thalen-2-ol. The acid was present in higher amounts in the extracts than in the oil. The aqueous extracts showed high polar fractions (fraction no. 2 and 3 from root and shoot respectively) that caused J2 mortality. Major monosaccharide type of both fractions was glucose. Aqueous shoot extract (VSA) showed higher percent of total polysaccharides but aqueous root extract (VRA) showed higher percent of total uronic acids. Foliar application of VSA revealed the delay of root-knot nematode stage development and induction of OsWRKY45 and OsPR1a which was SA signalling and pathogenesis gene in rice, respectively. VSA exerted both direct (killing and repellent) and indirect effects (delayed development and immunostimulation) on root-knot nematodes. VSA might be of interest to use, as a bio-nematicide, for controlling root-knot nematodes in the future or to use in crop rotation or as soil amendment to reduce root-knot nematode population. Additionally, vetiver grass may be useful as an alternative application in nematode-suppressive mulch or as a source for nematotoxic compounds.

Keywords: Vetiver extracts - *Meloidogyne* - Nematotoxic - Vetiver components - Defense gene.

References:

- Jindapunnapat et al., 2018. J. Nematol. 50(2): 147-162.
- Nahar et al., 2011. Plant Physiol. 157:305-316.
- Wiratno et al., 2009. The Open Natural Products J. 2: 77-85.

Biostimulants or Bionematicides: experience with seaweed extracts.

Bruno Ngala[1] (bruno.ngala@fnpppt.fr), Nicolas Mariette[2], Melina Ianszen[1], Pauline Dewaegeneire[1], Marie-Christine Denis[2], Catherine Porte[2], Christophe Piriou[2], Emilie Robilliard[3], Antoine Couetil[3], Eric Nguema-Ona[4], Virginie Gobert[1], Amélie Beury[1], Anne-Claire Le Roux[2]

[1]*Achicourt, Pas-des-Calais, France*; [2] *Agrocampus-Ouest, INRAe, Université de Rennes, IGEPP, Le Rheu, France*; [3] *Société d'Investissement Légumière et maraîchère de Basse Normandie (SILEBAN), Gatteville-le-Phare, France*; [4] *CMI-Roullier, Saint-Malo, France*

This study evaluated two seaweed extracts codded SW1 and SW2 for their potential to supress root infectivity and reproduction of potato, sugar beet and carrot cyst nematode species under repeated glasshouse experiments. Plants were assessed for development and yield following application of the respective seaweed extracts. Extracts of SW1 significantly ($P < 0.001$) reduced newly formed cysts for all three groups of cyst nematodes species at 2000 ppm and above, except for PCN where effective control was observed at 8000 ppm. Likewise, tuber yield plant^{-1} assessed at 90 days post-application of treatment doubled in pots treated with extracts of SW1 at 8000 ppm. Extracts of SW2 demonstrated phytotoxicity at concentrations above 5000 ppm and were therefore, excluded from further assessments. At 4000 ppm, SW38 boosted root production, but did not reduce *G. rostochiensis* reproduction relative to control treatments. We therefore concluded that plant stimulation and nematode suppression depended on the type of seaweed extracts and concentrations applied, as well as the nematode species. This study provides a pathway to environmentally sound and cost effective solutions to sustainable management of plant parasitic nematodes in vegetable production. Field validation and studies to understand the mechanism of action of the seaweed extract are on-going.

Keywords: Seaweed extracts - Cyst nematodes - *Globodera* - *Heterodera* - Concentrations.

Entomopathogenic nematodes, a powerful weapon to control foliar insect pests.

Kay Moisan[1] (kmoisan@koppert.nl), Magda Galeano[2], Mireya Baños[2], Kelly Arkoumanea[1], Stavroula Louka[1], Olga Kostenko[1], Roxina Soler[1], Jose E. Belda[2]

[1] *Microbiology - Agronomical Development, Koppert B.V., Berkel en Rodenrijs, Netherlands*; [2] *Microbiology - Agronomical Development, Koppert Spain, Vícar (Almeria), Spain*

Most terrestrial nematodes thrive in soil, a dark and moist environment that is favorable to their survival and movement [1, 2]. In contrast, leaves display more adverse conditions where nematodes can be exposed to multiple environmental stresses that may threaten their survival [3, 4]. Among its microbial portfolio, Koppert has developed over the past decade a range of entomopathogenic nematodes (EPNs) products as biological control agents. Against all odds, we have explored the potential of EPN products in targeting foliar insect pests by applying EPNs directly onto the crop canopy. Remarkably, the results show that two of the EPNs products based on *Steinernema feltiae* and *S. carpocapsae* can successfully control a wide range of foliar pests. EPNs withstood the foliar environmental conditions and survived up to a couple of days. Only a few hours were needed for the these EPNs to enter the insect pests, and upon infection insects died within a few days. Thorough field research validated these results and confirmed the vast potential of these EPNs to control foliar pests not only for horticultural crops but also for outdoor crops in achieving more than 70% control. Compatible with most common grower's practices, these EPNs represent an efficient weapon to complement the IPM portfolio and to successfully control soil and foliar pests.

Keywords: *Steinernema* spp. - Caterpillars - Foliar application - Plasticity - Field trials.

References:

- [1] Neher, 2010. Annu. Rev. Phytopathol. 48: 371-394.
- [2] Kaya, 1990. In Gaugler & Kaya (Eds): 93-116. CRC Press.
- [3] Gaugler et al., 1992. J. Invertebr. Pathol. 59(2): 155-160.
- [4] Shapiro-Ilan et al., 2006. Biol. Control. 38(1): 124-133.

Nematicidal plant secondary metabolites with plant and soil enhancement properties.

Ntalli Nikoletta (nntali@agro.auth.gr)

Department of Pesticides Control and Phytopharmacy, Benaki Phytopathological Institute, Athens, Attica, Greece

Root-knot nematodes (RKN; *Meloidogyne* spp.), are obligate sedentary endoparasites of plant roots, parasitizing almost every species of vascular plants. They are among the most important agricultural pests due to their worldwide distribution and economic impact. By attacking the root system, RKN remove photo-assimilates, diminish the absorption capacity of water and nutrients. They can enhance secondary host infections by soil pathogens that may lead to severe crop damage.
In the past, RKN suppression has been based on fumigants, such as methyl bromide, applied to soil for the production of high-value crops. After the phase-out of methyl bromide, environmental toxicity and high risks for applicators have led to the ban of other fumigants in some countries. Currently, new eco-friendly tools for RKN management are developed and the EU legislation introduces low risk plant protection products and basic substances to highlight ecotoxicity concerns (EC 1107/2009 Article 22, 47; EC 1107/2009 Article 23).
Herein we present some findings of our 15 years of research, during which we have studied more than 20 plant species for their nematicidal properties and have discerned for activity amongst constituent ingredients with some significant indications for synergism effects. We shed light on efficacy matters (lab and field scale), the mode of action (contact and fumigant toxicity, second stage juveniles' paralysis capacity, egg hatch inhibition activity and parasite biological cycle arrest in host roots) and behavioral sub-lethal effects (attraction and repellence) contributing to the overall parasite management. In parallel, we present the secondary beneficial effects of some botanical nematicides on the non-target organisms constituting soil communities as well as on the plant physiology. Last, we present our first steps into formulation issues.

Keywords: *Meloidogyne* sp - Efficacy - Synergism - Attraction - Cuticle.

References:

- Ntalli N.G., Vargiu S., Menkissoglu-Spiroudi U., Caboni P. 2010. Nematicidal carboxylic acids and aldehydes from Melia azedarach fruits. J. Agric. Food Chem., 58, 11390–11394.
- Ntalli N, Oplos C, Michailidis M, Thanasenaris A, Kontea D, Caboni P, Tsiropoulos N. G, Menkissoglu-Spiroudi U, Adamski Z. 2016. Strong synergistic activity and egg hatch inhibition by (E,E)-2,4-decadienal and (E)-2-decenal in Meloidogyne species. J Pest Science 89, 565-579.
- Ntalli N, Ratajczak M, Oplos C, Menkissoglu-Spiroudi U, Adamski Z. 2016. Acetic Acid, 2-Undecanone and (E)-2-Decenal Ultrastructural Malformations on Meloidogyne incognita. J Nem. 48, 248-260.
- Ntalli N, Monokrousos N, Rumbos C, Kontea D, Zioga D, Argyropoulou DM, Menkissoglu-Spiroudi U, Tsiropoulos GN. Greenhouse biofumigantion with Melia azedarach controls Meloidogyne sp. and enhances soil biological activity. J Pest Sci. 91, 29-40.
- Sobkowiak R, Bojarska N, Krzyzaniak E, Wagiel K, Ntalli N. Chemoreception of Meloidogyne incognita and Caenorhabditis elegans on botanical nematicidals J. Environ. Sci. Health, Part B. 53, 493-502.

What it takes for development and launch of a successful biological nematicidal product.

Sebastian Hartmann-Wittulsky (sebastian.wittulsky@bayer.com)

CropScience, Bayer, Monheim Am Rhein, Germany

Fungus-based concepts for biological pest control are long known, and many fungal strains with insecticidal activity have been identified. But not all identified strains become commercial products. The journey from an active strain to a commercially available product which fits the needs of growers is long and has to overcome many different obstacles. As an example, nematopathogenicity of the fungus Purpureocillium lilacinum strain 251 has been well documented in the literature. In 2013, Bayer began the development of BioAct DC, a novel liquid formulation of *P. lilacinum* strain 251 and worked extensively on the production, fermentation, and integration into commercial nematode solutions. This presentation is an overview of the work required in the lab, production, formulation development, compatibility testing, field work and launch preparation to end up with a biological product suitable for the needs of sustainable agricultural production. Highlighted are the key challenges faced during the process, and an outlook into the future of biological nematode control.

S20-PF1

Nematicidal plants for root-knot nematode management in tomato agrosystems.

Cliven Njekete[1] (cliven.njekete@inrae.fr), Anne-Violette Lavoir[2], Antoine Bedel[3], Vincent Michel[3], Caroline Djian-Caporalino[2]

[1] *ISA, INRAe, Université Côte d'Azur, CNRS, Sophia Antipolis, France*; [2] *INRAe, Université Côte d'Azur, CNRS, Sophia Antipolis, France*; [3] *LIDEA, Lescar, France*

Alternatives to nematicide use are required to increase the sustainability of vegetable crop protection strategies. Identifying biocontrol plants (BCPs), which is the aim of this work, is a promising and complex challenge [1]. To control endoparasites such as root-knot nematodes (RKN), BCPs could act before the juveniles penetrate the roots by involving a biocidal allelopathic effect (release of repellent or toxic root exudates, inhibiting egg-hatching or being lethal to juveniles) [2]. They could also attract and trap the juveniles, then disrupt their life cycle, involving toxic secondary compounds in the roots, constitutive substances or produced in reaction to infestation. In both cases, BCPs are non-host (no gall or egg mass) or poor host (few egg-masses). Twenty one summer and winter BCP candidates that can be used in tomato agrosystems were tested against *Meloidogyne incognita* and *M. arenaria*. Six replicates of each BCP were kept in climatic chambers at 24°C or 16°C. One month-old seedlings were inoculated with 2000 eggs and analysed after one cycle for galls, egg-masses, and juveniles (J2) in the soil. The whole experiment was replicated twice (n=6+6); with a susceptible tomato being the control. All Tagetes (spp. and varieties) tested were non-host for both *Meloidogyne* spp., suggesting that no J2 penetrated the roots or reached the vascular cylinder to initiate its feeding site. For the other plants, significant varietal effects as well as different responses to the two species of Meloidogyne were observed. P*hacelia tanacetifolia* and *Avena strigosa* were non-hosts of *M. arenaria* while *Foeniculum vulgare* cv. Rondo, *Fagopyrum esculentum*, and *Crotalaria spectabilis* were non-host for M*. incognita.* The rest were poor hosts of *M. incognita* and *M. arenaria*. In sorghums, if no or few galls were seen, egg-masses were found inside the roots as already seen in some previous work [3]. Post one cycle, *Meloidogyne* J2s were found in the soil except in non-host plant modalities. Species of saprophagous nematodes multiplied in all pots, especially those with *Foeniculum rondo*, *Diplotaxis tenuifolia*, and *Crotalaria*. In conclusion, the non-hosts T*agetes, Fagopyrum, Phacelia,* and *Foeniculum* seemed the most sanitising- eliminating *Meloidogyne* in the soil. Further studies are ongoing to identify the mechanisms behind and the modes of action of such nematicidal plants including metabolomic analysis of the active compounds in the root exudates and extracts.

Keywords: *Meloidogyne* spp. - Biocontrol plant - Trap-plant - Poor host plant - Non-host plant.

References:

- [1] Parolin et al., 2012. International Journal of Pest Management 58, (4) 369-377.
- [2] Djian-Caporalino et al., 2008. Biopesticides of plant origin: phytosanitary potentialities. Ed. Tec & doc, Lavoisier, Paris, 125-185.
- [3] Djian-Caporalino et al., 2019. Crop Protection 122: 142-150.

S20-PF2

Investigation on the effectiveness of some plant extractions against *Ditylenchus dipsaci* and *Meloidogyne incognita.*

İbrahim Halil Elekcioğlu[1] (halile@cu.edu.tr), Ece Börtecine Kasapoglu Uludamar[1], Adem Taskın[2]

[1] *Plant Protection Department, Agricultural Faculty, Cukurova University, Adana, Turkey*; [2] *Ant Protection Department, Agricultural Faculty, Cukurova University, Adana, Turkey*

The effects of 1%, 2.5%, 5% and 10% concentrations of aqueous plant extracts obtained from *Vitex agnus-castus, Ficus carica, Nerium oleander, Eucalyptus camaldulensis, Melia azedarach, Eruca sativa, Mentha piperita, Juglans regia, Anethum graveolens, Asphodelus aestivus, Zingiber officinale, Laurus nobilis* plants against *Ditylenchus dipsaci* and *Meloidogyne incognita* were examined. As the concentration and time increased, the mortality rate of nematodes increased in trials. As a result of the 6 h, 12 h and 24 h observations of the aqueous plant extracts, the effectiveness increased against *Meloidogyne incognita*. *Asphodelus aestivus* had the highest mortality rate on all concentration in comparison to the other aqueous plant extracts. Mortality rate of 56% was determined after 6 hours in the aqueous concentration of 5% of *A. aestivus*. When plant extracts were compared in terms of efficacy, it was found that higher concentrations and time (%10 and 24 h) only provided higher efficacy against *M. incognita.* After 24 hours of inoculations of 5% concentration of aqueous extracts of *Nerium oleander*, *Eucalyptus camaldulensis, Melia azedarach, Eruca sativa, Mentha piperita, Juglans regia, Anethum graveolens, Asphodelus aestivus, Zingiber officinale, Laurus nobilis* were resulted 80-100% mortality rates on *M. incognita*. 1% concentration of *Ficus carica, Melia azedarach, Eruca sativa, Mentha piperita, Juglans regia, Anethum graveolens* and *Asphodelus aestivus* caused 100% mortality rate on *D. dipsaci* after 24 hours. All of the plants included in the experiment at 10% concentrations caused 85-100% mortality rate on *D. dipsaci* and *M.incognita* in 24 hours. It was observed in this study that plant species, concentration and exposure time had a significant effect on nematodes.

Keywords: Mortality rate - Time - Concentrations - Aqueous plant extracts.

S20-PF3

Effects of synthetic and phytonematicides on the reproductive potential of *Meloidogyne* species on potato plants.

Kgabo Pofu (pofukgabo@gmail.com)

Green Biotechnologies Research Centre of Excellence, University of Limpopo, Polokwane, South Africa

The damaging effects that *Meloidogyne* species have on potato (*Solanum tuberosum*) had been underestimated prior to the withdrawal of synthetic nematicides from the agrochemical markets. All available potato genotypes do not have resistance to *Meloidogyne* species, which have been shown to have the capability of reducing yield by as high as 50%, with incidents of complete crop failure. The efficacy of Nemafric-BL phytonematicide and Velum, with fenamiphos as a standard, were compared for suppression of population densities of *Meloidogyne* species on potato cv. 'Mondial G3' under field conditions. All products were applied once and assessed using the reproductive potential (RP) at 56 days after initiating the treatments. Relative to untreated control, fenamiphos and Velum reduced RP values of root-knot nematodes by 74 and 61%, respectively, whereas Nemafric-BL phytonematicide reduced RP values by 100%. In conclusion, the efficacy of the phytonematicide on nematode suppression was comparable to those of the two synthetic nematicides.

Keywords: Velum - Potato - Phytonematicides - *Meloidogyne* species - Synthetic Nematicides.

ORAL SESSION 21

Nematode-plant interactions

Strigolactones enhance rice susceptibility to root-knot nematode infection by antagonizing jasmonate based defense.

Godelieve Gheysen[1] (godelieve.gheysen@ugent.be), Zobaida Lahari[1], Chhana Ullah[2], Tina Kyndt[1], Jonathan Gershenzon[2]

[1] *Ghent University, Gent, Belgium*; [2] *MPI for Chemical Ecology, Jena, Germany*

Previously it has been shown that jasmonate (JA) is important for basal defense to root-knot nematodes in rice (*Oryza sativa*). Crosstalks with other hormone pathways mainly converge on JA as key player in rice defense. In contrast to findings in *Arabidopsis thaliana*, JA is not working antagonistically to salicylate, but both are reducing susceptibility of rice to rootknot nematodes. Until recently, very little was known on the role of strigolactones (SL) in plant defense and therefore, this role was analyzed in the rice-*Meloidogyne graminicola* interaction.
We studied *M. graminicola* infection of rice mutants in SL biosynthesis and signaling and complemented these analyses with SL applications. Measurement of hormones and applications of inhibitors of JA and SL biosynthesis were also used.
The SL biosynthesis and signaling mutants were shown to be less susceptible to *M. graminicola* and to contain higher levels of JA and JA-isoleucine than the wild type control plants.
Application of the strigolactone analogue GR24 on the leaves increased nematode susceptibility of rice roots while the SL biosynthesis inhibitor TIS108 reduced nematode infection.
The SL level was found to antagonistically influence the levels of JA and this relation appeared to be pivotal in influencing the susceptibility level. For instance, the SL-mutants were less susceptible and have higher JA levels while the application of GR24 reduced JA and increased nematode infection. Furthermore, application of TIS108 which lowered the SL level, caused the accumulation of JA and lowers nematode susceptibility. Importantly, inhibition of the JA-pathway by ETYA abolished the effect of TIS108.
In conclusion, the strigolactone pathway in rice was antagonistic to the jasmonate pathway in basal defense to rootknot nematodes, further reinforcing the key role of jasmonate in rice defense.

Keywords: *Meloidogyne* - Rice - Jasmonate - Strigolactones.

References:

- Nahar et al., 2011. Plant Phys. 157: 305-316
- Kyndt et al., 2014. Annu. Rev. Phytopathol. 52: 135-153
- Lahari et al., 2019. New Phytol. 224: 454-465

Copper microRNAs modulate the formation of giant feeding cells induced by the root-knot nematode *Meloidogyne incognita* in *Arabidopsis thaliana.*

Stéphanie Jaubert[1] (stephanie.jaubert@inrae.fr), Yara Noureddine[1], Joffrey Méjias[1], Martine Da Rocha[1], Sébastien Thomine[2], Pierre Abad[1], Bruno Favery[1]

[1] *INRAe, Université Côte d'Azur, CNRS, ISA, Sophia Antipolis, France*; [2] *CNRS/CEA/Univ. Paris Sud, Université Paris-Saclay, I2BC, Gif-sur-Yvette, France*

The redifferentiation of root parenchyma cells into giant feeding cells induced by Root knot nematodes (RKN) is the result of a massive transcriptional reprogramming of host cells. Since RKN induce similar feeding cells in roots of thousands of plant species, RKN are thought to manipulate conserved plant molecular pathways. MicroRNAs are small non coding RNAs (20-22 nucleotides), with many microRNA families highly conserved, that regulate gene expression at the post-transcriptional level. Our work aims to investigate the role of plant microRNAs in the control of the massive transcriptional reprogramming observed during formation of RKN induced feeding cells.
We identified two microRNA families, miR408 and miR398, as upregulated in *Arabidopsis thaliana* and *Solanum lycopersicum* roots infected by root-knot nematodes. In plants, the expression of these two conserved microRNA families is known to be activated by the SPL7 transcription factor in response to copper starvation. By combining functional approaches, we deciphered the network involving these microRNAs, their regulator and their targets. *MIR408* expression was located within nematode-induced feeding cells in which it co-localised with *SPL7* expression and was regulated by copper. Moreover, infection assays with *mir408* and *spl7* KO mutants or lines expressing targets rendered resistant to cleavage by miR398 demonstrated the essential role of the *SPL7*/*MIR408*/*MIR398* module in the formation of giant feeding cells. Our findings reveal how perturbation of plant copper homeostasis, *via* the *SPL7*/*MIR408*/*MIR398* module, regulate the development of nematode-induced feeding cells.

Keywords: *Meloidogyne* - Epigenetic - Tomato - MicroRNA - Gene expression.

References:

- Medina C. et al., 2017, New Phytol., 882-896.
- Medina C. et al., 2018, BMC Genomics., 943.

Two sides of the same coin: *Serendipita indica* alters different development of sedentary plant-parasitic nematodes in *Arabidopsis*.

Michael Opitz[1] (michael.opitz@boku.ac.at), Virginia Ruiz-Ferrer[2], Fernando Evaristo Díaz-Manzano[2], Carolina Escobar[2], Roshanak Daneshkhah[1], Roland Ludwig[3], Cindy Lorenz[3], Siegrid Steinkellner[1], Krzysztof Wieczorek[1]

[1] *Institute of Plant Protection, University of Natural Resources and Life Sciences, Vienna, Tulln, Austria*; [2] *Facultad de Ciencias Ambientales y Bioquímica, Universidad de Castilla-La Mancha, Toledo, Spain*; [3] *Institute of Food Technology, University of Natural Resources and Life Sciences, Vienna, Austria*

The root endophyte *Serendipita indica* (= *Piriformospora indica*) is an orchid mycorrhiza that forms mutualistic relationships with many different plants including the model plant *Arabidopsis thaliana*. During this interaction, the endophyte promotes plant growth and development, increases biomass and seed production [1,2]. Furthermore, it significantly enhances the resistance to abiotic (*e.g*. drought, salt) and biotic stresses (*e.g*. pathogens). Hence, it can be speculated that, similar to arbuscular mycorrhizal fungi, *S. indica* receives carbohydrates (preferably hexoses) from the host in exchange for this service [3]. Therefore, in this study qPCR of sucrose synthase and invertases as well as analyses of multiple *sus* and *inv* lines of *A. thaliana* were carried out to assess importance of sugar metabolism during this endophyte-plant interaction. The results show general upregulation in directly colonized roots and initial downregulation followed by an upregulation of these genes in shoots. Typical growth promotion was only observed in colonized wild type, whereas multiple *sus* and *inv* mutants showed no such effects indicating the importance of these genes for successful interaction. To elucidate the impact of *S. indica* root colonization on sedentary plant-parasitic nematodes, development assays with two important species, *Heterodera schachtii* (cyst nematode) and *Meloidogyne javanica* (root-knot nematode), were performed. For the analysis of the direct effects, the fungus and nematodes competed for the same root system. Indirect effects were analyzed in three-chamber dish with one half of the root system inoculated with *S. indica* and the other half infected with nematodes. Interestingly, direct and indirect fungal effects showed opposite results. Whereas direct contact with the fungus resulted in significant reduction in nematode number, systemic effects lead to significant increase in nematode numbers. Additional analyses revealed the downregulation of some defense-related genes as well as slightly increased sucrose levels in non-colonized half of the split roots. Thus, we speculate that nematodes benefit from these changes triggered by the endophyte in the systemic part of the root what enables their significantly better development. With this work, we show the complexity of this multilayered tripartite relationship and deliver new insights into sugar metabolism and plant defense responses in *S. indica*-nematode-plant interaction.

Keywords: *Serendipita indica* - Plant-parasitic nematodes - Sucrose metabolism - Synthases and invertases - Plant defense.

References:

- [1] Varma et al., 1999. Appl Environ Microbiol. 65(6): 2741-4.
- [2] Weiß et al., 2016. New Phytol. 211(1): 20-40.
- [3] Schüssler et al., 2006. Nature. 444(7121): 933-6.

Molecular markers for cell damage induced by root-knot nematodes.

Janice de Almeida Engler (janice.de-almeida@inrae.fr)

SPE, INRAe, Sophia Antipolis, PACA, France

Feeding sites within galls, induced by the plant parasitic root-knot nematodes, are located in the root vascular cylinder and originate from cells with competence for cell cycle activity. Herein, we questioned if during the process of nematode penetration and infection, cell damage might occur, finally triggering cellular dedifferentiation and division. It emerges that the Ethylene Response Factor 115 (ERF115 of the ERF family), together with the Phytochrom A Signal Transduction 1 (PAT1 of the GRAS family) transcription factors (TFs) are able to activate a regeneration program through the induction of a stem cell-like fate in gall cells. The ERF115 TF is a potent element controlling the plant regeneration process. Upon stem cell death, cells that co-express *ERF115* and *PAT1* were found to engage recovery cell divisions [1]. Moreover, plants lacking a functional ERF115 or PAT1 showed a reduced ability to perform recovery divisions and displayed a lower regeneration frequency [2]. Our studies demonstrate that root tissue injury caused during nematode infection induces *ERF115*, *ERF114* as well as *PAT1*expression in cells immediately adjacent to damaged root cells and hereby potentially activating cell division to replace damaged cells as part of a regeneration program. These functional studies lead us to conclude that both ERF115 and PAT1 are most likely involved in gall homeostasis sensing the damage caused upon nematode infection and working in its replenishment. Using these observations as starting point, we aim to further understand the pathways that activate plant regeneration following wounding by nematodes, and map the signaling cascades operating downstream of ERF115. Overall, this knowledge might help us to understand their parasitic success and to fight against these plant pests.

Keywords: Cell damage - Tissue regeneration - Root-knot nematodes - Galls - *Arabidopsis thaliana*.

References:

- [1] Heyman et al., 2013. Science 342: 860 – 863.
- [2] Heyman, et al., 2016. Nature Plants 2: 1 – 7.

PCN hijack carbon-for-nutrient exchange between potato and arbuscular mycorrhizal fungi to enhance reproduction.

Christopher Bell[1] (c.a.bell@leeds.ac.uk), Emily Magkourilou[2], P Urwin[1], Katie Field[2]

[1] *University of Leeds, Leeds, United Kingdom*; [2] *University of Sheffield, Sheffield, United Kingdom*

Plants simultaneously interact with a range of biotrophic symbionts, from mutualists such as arbuscular mycorrhizal fungi (AMF), to parasites such as the potato cyst nematode (PCN). The exchange of mycorrhizal-acquired nutrients for plant-fixed carbon is well-studied, however the impact of competing symbionts remains under-explored.

In this study, we examined mycorrhizal nutrient and host resource allocation in potato with and without AMF and PCN using radioisotope tracing, whilst determining the consequences of such allocation.

The presence of PCN disrupted carbon for nutrient exchange between plants and AMF, with plant-C overwhelmingly obtained by the nematodes. Despite this, AMF maintained transfer of nutrients to PCN-infected potato, ultimately losing out in their carbon for nutrient exchange with the host. Although PCN exploited the greater nutrient reserves to drive population growth on AMF-potato, the fungus imparted tolerance to allow the host to bear the parasitic burden.

Our findings provide important insights into the below-ground dynamics of plant-AMF symbioses, where the nutritional benefits conferred by AMF to hosts and their parasites are seldom considered in plant community dynamics. Our findings suggest this may be a critical oversight, particularly in the consideration of carbon and nutrient flows in plant and soil communities.

Keywords: Potato cyst nematode - Arbuscular mycorrhizal fungi - Host pest tolerance - Symbiosis.

Unravelling the mechanisms underlying cyst nematode hatching.

Lemeng Dong (l.dong2@uva.nl), Kristyna Flokova, Alessandra Guerrieri, Harro Bouwmeester

Plant Hormone Biology group/SILS, University of Amsterdam, Amsterdam, The Netherlands

Cyst nematodes worldwide attack agricultural crops such as potato, sugar beet, rice and soybean and cause large yield losses. There is a great need for developing new control methods for cyst nematodes because resistance in crops is overcome, crop rotation is problematic and many nematicides have been banned. Plant roots secrete triterpenoid-like signaling molecules, called hatching stimulants (HSs), into the soil that are known to induce hatching of cyst nematodes. So far, HSs were identified in two plant species only: glycinoeclepin A, B and C in kidney bean [1,2] and solanoeclepin A in potato [3]. Due to the extremely low concentrations of these HSs and corresponding challenge to detect them, the knowledge about HSs is limited and progress in this area has been hampered for decades. In our lab, we have developed a highly sensitive method to detect solanoeclepin A in the root exudate of a single solanaceous plant. Using this method, we found that there is substantial natural variation in the amount of solanoeclepin A production between potato genotypes. In addition, we were able to show that other, wild, Solanaceae species produce solanoeclepin A, thus, production of this HS is widespread in the Solanaceae suggesting that it originates from a common solanaceous ancestor. In addition, we found that conditions such as developmental stage, nutrient availability and temperature variation affect HS production. This important breakthrough in the work on HSs now allows us to also unravel the biosynthetic pathway of the HSs and its regulation. Together this research should result in a better understanding of the mechanisms that mediate the communication between cyst nematodes and their host plant. In addition, we expect to facilitate the development of novel cyst nematode control methods by blocking or altering the signaling relation between the host and the parasitic nematodes.

Keywords: Cyst nematodes - Hatching stimulants - Chemical communication - Triterpenoid - Solanaceae.

References:

- [1] Masamune et al., 1982. Nature. 297(5866):495.
- [2] Fukuzawa et al., 1985. Tetrahedron Lett. 26(45):5539-5542.
- [3] Mulder et al., 1996. Google Patents.

S21-PF1

Trade-offs between virulence and breaking resistance in root-knot nematodes.

Alison Coomer[1] (alcoomer@ucdavis.edu), Henok Zemene Yimer[2], Valerie Williamson[1], Shahid Siddique[2]

[1] *Siddique Lab/Plant Pathology, University of California, Davis, California, United States*; [2] *Siddique Lab, University of California, Davis, California, United States*

Root knot nematodes (RKNs), *Meloidogyne spp.*, are among the most devastating pathogens of crops, causing substantial yield and economic losses worldwide. These plant parasitic nematodes can infect over a hundred plant species. For decades the resistance gene *Mi-1* has provided effective resistance to RKNs in tomatoes, but the underlying mechanisms of how it is able to detect these pathogens remain largely unknown. However, resistance-breaking populations have been found in both greenhouse and in field settings, threatening the effectiveness of the *Mi-1* gene. We used two strains of *M. javanica*, one strain, VW4, which is recognized by *Mi-1,* and another strain, VW5, which was selected from VW4 and is able to overcome resistance mediated by *Mi-1*[1]. To compare reproduction of the two strains, infection assays on tomato not containing *Mi-1,* plants were inoculated with 500 J2s and harvested 5 weeks post inoculation. Eggs were collected, counted, and roots were washed and stained. After staining, galls were counted and dissected, and nematode stage development was recoded. We found that the number of eggs produced by VW5 was significantly lower than for VW4. We also see a lower number of females inside the roots and reduced gall formations in plants inoculated with VW5 when compared to VW4. Reproduction of VW4 and VW5 was then compared on cucumber and rice; for both of these hosts we found a 3-fold decrease in egg number for VW5 compared to VW4. Penetration assays on tomato not containing *Mi-1*, show that the numbers of juveniles of VW4 and VW5 inside the root were not significantly different suggesting that the reduced reproduction of VW5 was due to post infection factors. Together these results suggest that although strain VW5 can overcome *Mi-1*, it is compromised for reproduction on tomato and other plant species.

Keywords: Resistance-breaking - Mi-1 - Resistance Gene - Root-Knot Nematode - *M. javanica*.

References:

- Gleason CA, Liu QL, Williamson VM, 2008, Mol Plant Microbe Interact, 576-85

S21-PF2

Parasitic worms redirect host metabolism via NADPH oxidase-mediated ROS to promote infection.

M. Shamim Hasan[1] (s7mdhasa@uni-bonn.de), Divykriti Chopra[1], Christiane Matera[1], Oliver Chitambo[1], Sina Valerie-Mahlitz[1], Ali Naz[2], Slawomir Janakowski[3], Miroslaw Sobczak[4], Axel Mithöfer[5], Tina Kyndt[6], Florian Grundler[1], Shahid Siddique[7]

[1] *Molecular Phytomedicine, Bonn University, Bonn, Germany*; [2] *Plant Breeding, Bonn University, Bonn, Germany*; [3] *Department of Botany, Warsaw University of Life Sciences, Warsaw, Poland*; [4] *Poland, Warsaw University of Life Sciences, Warsaw, Poland*; [5] *Bioorganic Chemistry, Max Plank Institute for Chemical Ecology, Jena, Germany*; [6] *Biotechnology, Gent University, Gent, Belgium*; [7] *Entomology and Nematology, University of California, Davis, United States*

Reactive oxygen species (ROS) generated in response to infections often activate immune responses in eukaryotes. In plants, ROS are primarily produced by plasma membrane-bound NADPH oxidases called respiratory burst oxidase homolog (Rboh). Surprisingly, Rbohs can also promote the infection of plants by certain pathogens, including parasitic nematodes. However, little is known of the mechanism and downstream components by which Rbohs promote infection. Here, we identified WALLS ARE THIN 1 (WAT1), an auxin transporter, as a downstream target of Rboh-mediated ROS during parasitic infections. We found that WAT1 is required to redirect host indole metabolism in infected cells and this reprogramming is necessary for successful establishment of parasite. In summary, this work clarifies a unique mechanism that enables nematodes to use host ROS for their own benefit.

S21-PF3

Meta-analysis of wild *Arachis* transcriptome data unravels candidate genes for combined nematode and drought resistance.

Patricia M Guimaraes (patricia.guimaraes@embrapa.br), Ana Paula Zotta Mota, Roberto Coiti Togawa, Ana Claudia Guerra Araujo, Ana Cristina Miranda Brasileiro

Biotechnology, EMBRAPA-Cenargen, Brasilia, Distrito Federal, Brazil

In nature, plants are exposed simultaneously to many stresses including drought, heat, salinity and pathogens. Although most studies focus in plant responses to one stress at a time, the combinatory biotic and abiotic stress responses are often quite different to those stresses separately. Root-Knot nematodes (RKN-*Meloidogyne* spp.) and drought are two of the major constraints to agricultural production worldwide. *Arachis stenosperma* is a peanut wild relative highly resistant to *M. arenaria* and moderately tolerant to drought. Here, we conducted a meta-analysis on 22 cDNA libraries previously produced from *A. stenosperma*, to identify core stress responsive genes differentially regulated (DEGs) upon biotic (nematode) and abiotic (drought) stresses. We also compared this dataset with DEGs obtained from a new plant bioassay combining nematode and drought stresses simultaneously. RNA-Seq libraries were sequenced using Illumina HiSeq-4000, in triplicates, from roots: (i) control, (ii) nematode infection, (ii) drought and (iv) combined stresses. All reads were mapped to the closest available genome (*A. duranensis)* and estimated read counts conducted for each library. Overall, a network of core stress responsive genes was identified, with roots submitted to RKN infection showing mainly upregulation of genes involved in the jasmonic acid (JA) hormonal via, while those submitted to drought displayed a robust enrichment on genes in the abscisic acid (ABA) pathway, and those submitted to both stresses simultaneously exhibiting a clear shift to the ethylene (ET) signaling route. Contrastingly, the functional annotation of meta-DEGs resulting from the overlapping of nematode and drought stresses was significantly distinct from that obtained from the combined stress experiment, indicating specialized responses to unique and combined stresses. This network of *Arachis* core stress-responsive genes can be further exploited for legume improvement aiming crop tolerance to multiple environmental stresses.

Keywords: Meta-analysis - *Arachis* - Resistance - Combined stress - Core genes.

ORAL SESSION 22

Metabolism & Physiology of nematodes and host plants

Tricky parasites: How nematodes take their vitamins from plants.

Clarissa Hiltl[1] (clarissa.hiltl@uni-bonn.de), Shahid Siddique[2], Zoran S. Radakovic[1], Anna Gioran[3], Muhammad Shahzad Anjam[1], Esther Riemer[1], Samer S. Habash[1], Syed Jehangir Shah[1], Julia Holbein[1], Divykriti Chopra[1], Miroslaw Sobczak[4], Daniele Bano[3], Sebastian Eves-van den Akker[5], Alexander Graf[6], Florian M. W. Grundler[1]

[1] *INRES, Molecular Phytomedicine, University of Bonn, Bonn, Germany*; [2] *Department of Entomology and Nematology, University of California, Davis, CA, United States*; [3] *DZNE, Bonn, Germany*; [4] *Department of Botany, Warsaw University of Life Sciences, Warsaw, Poland*; [5] *Department of Plant Sciences, University of Cambridge, Cambridge, United Kingdom*; [6] *Max Planck Institute of Mol. Plant Phys., Potsdam, Germany*

Heterodera schachtii is a plant-parasitic nematode with an economically important impact on sugar beet production. The second-stage juveniles invade the root of their host and move intracellularly towards the vascular cylinder, where they induce the formation of a plantderived syncytium and become sedentary. The hypertrophic and hypermetabolic syncytium serves as the sole nutritional source for the developing juveniles. Due to this dependency, it is crucial for *H. schachtii* to successfully initiate and maintain the syncytium in order to complete its lifecycle and produce progeny [1,2]. Transcriptome data of *Arabidopsis thaliana*derived syncytia compared to uninfected root tissues revealed an increase in expression of genes involved in metabolic processes, including the biosynthetic pathways of several vitamin Bs (VBs) [3]. These water-soluble vitamins are essential nutrients, as they cannot be stored or synthesized by humans and, presumably, all other animals. The important function of the increase in transcript abundance of VB biosynthetic genes during cyst nematode infection was confirmed for VB 5, the precursor of co-enzyme A. The first enzymatic step in the *de-novo* VB 5 biosynthesis is encoded by *AtPANB1*, which is significantly upregulated in the syncytium. *AtPANB1* knockout mutants were less susceptible to infection by *H. schachtii*. The last enzymatic step is performed by AtPANC [4], which is not differentially expressed in the syncytium, and the loss-of-function mutation had no effect on the parasitism of *H. schachtii*. Notably, our work identified a nematode *PANC* gene (*HsPANC*), showing that nematodes are able to perform the last step of the VB 5 biosynthesis using HsPANC. We assume that this compartmentalization between nematodes and syncytia circumvents feedback/feed-forward inhibitions to support continuous supply of VB 5 to nematodes. Taken together our results indicate that *H. schachtii* regulates its VB availability through the syncytium in a highly sophisticated manner.

Keywords: Vitamin B metabolism - Nutrient availability - Cyst nematodes - Parasitism.

References:

- [1] Wyss and Grundler, 1992. Neth. Journal of Plant Pathology. 2: 165-173.
- [2] Golinowski et al., 1996. Protoplasma. 194: 103-116.
- [3] Szakasits et al., 2009. The Plant Journal. 57: 771-784.
- [4] Ottenhof et al., 2004. The Plant Journal. 37: 61-72.

Extending the survival of *Heterorhabditis bacteriophora* through phenotype selection and marker-assisted breeding.

Carlos Molina[1] (c.molina@e-nema.de), Nanette Hope Nellas Sumaya[1], Giulia Godina[1], Rolish Singh[2], Carlotta Kirsch[1], Bart Vandenbossche[1], Verena Dörfler[1], Mike Barg[1], Ralf-Udo Ehlers[1]

[1] *e-nema GmbH, Schwentinental, Germany*; [2] *Ghent University, Ghent, Belgium*

The utilization of the entomopathogenic nematode (EPN) *Heterorhabditis bacteriophora* to control insect pests in large scale agriculture is often limited by environmental stresses. Breeding for nematode strains with enhanced stress resistance is thus of outmost priority for this biocontrol agent. For this purpose, we largely enhanced the repertoire of phenotypic and genotypic information available on this nematode concerning stress tolerance and survival. Within this frame, more than 80 *H. bacteriophora* WT strains and inbred lines were extensively characterized for their DJ-longevity and EMS-mutants with extended survival have been generated through selection. Concerning genomic information, RNA-seq analyses assessed the expression of more than 22000 transcripts in long- and short-living nematodes and a large set of WT-strains and inbred lines was genotyped by sequencing (GBS) yielding more than 700 reproducible single nucleotide polymorphisms (SNPs). All the generated phenotypic and genotypic information was subsequently combined to determine genes, DNA polymorphisms, and genotypes with high potential for improvement of DJ-longevity in *H. bacteriophora*. The resulting hybrid strains, selected inbred Lines, and EMS-mutants with extended survival were tested for their general performance with satisfactory results. In future steps, combination of traits will be aimed at incorporating high throughput genotyping screens for EPN breeding. Interestingly, several of the identified genes through our approach had not been characterized in the model nematode *Caenorhabditis elegans*. This fact suggested that the direct extrapolation of information from model organisms encounters limitations when nematodes with different habits are investigated, stressing the high value of new alternative approaches.

Keywords: Stress-tolerance - SNP - Association analysis - Genotyping by sequencing - Transcriptomics.

Reaction of genotypes of *Phaseolus vulgaris* L BAT-306 and Triunfo-70 to *Meloidogyne incognita* and the behavior of enzymes related to the defense of both cultivars.

Dainé Hernandez-Ochandía[1] (daineh@censa.edu.cu), Yailén Arias-Vargas[1], Ivonne González-Marquetti[1], Susana Gorrita-Ramírez[1], M.G. Rodriguez[1], I. Miranda[1], Belkis Peteira Delgado-Oramas[1], R. Holgado[2]

[1] *Centro Nacional de Sanidad Agropecuaria (CENSA), Grupo de Plagas Agrícolas, San José de las Lajas, Mayabeque, Cuba*; [2] *Norwegian Institute of Bioeconomic Research (NIBIO), Ås, Norway*

The common bean (*Phaseolus vulgaris* L.) is an important food for the population of Latin America and is affected by numerous pests, including *Meloidogyne incognita*. In Cuba, studies of relationships between bean-*M. incognita* are very few. The objective of this work was to know the behavior (resistant/susceptible) to *M. incognita* of the BAT-306 and Triunfo-70 cultivars, as well as to establish the behavior of four enzymes related to its defense. Pots with 1 kg of substrate were used and the plants were inoculated with a geometric series of six initial population densities of the nematode (Pi) (0.125; 0.25; 1; 4; 16 and 64 eggs and second stage juveniles (J2) per gram of substrate-1). At 60 days the final population (Pf) was determined and the reproduction factor (RF) was calculated. Seinhorst's Equation was fitted to data to determine the tolerance limit (T) and minimum yield (m). For the analysis of the enzymatic systems, samples were taken from the leaves at 1, 3, 5 and 7 days after inoculation and the activities of the enzymatic systems peroxidase (PO), phenylalanine ammonium lyase (PAL), chitinase and glucanase, as well as the PO isoenzymes were determined. The results were compared through an Analysis of Variance and Duncan's Multiple Range test, using the InfoStat program. The cultivar Triunfo-70 behaved as resistant in the presence of Pi < 64 eggs and J2 per gram of substrate-1, with RF between 0.23 - 0.9; however, BAT-306 allowed the reproduction of the nematode with RF between 6.1 and 8. The cultivars BAT-306 and Triunfo-70 could be used in studies of bean cultivars against Cuban populations of *M. incognita*, as susceptible- and resistant-controls, respectively. All systems studied showed induction in both cultivars. In 'Triunfo-70', the cultivar resistant to the nematode, considerable increases were induced in the activity levels of all enzyme systems and, in some, these increases were more sustained over time, of greater magnitude or occurred earlier than in the cultivar BAT-306 (susceptible to the nematode).

Defining the combined stress response in wild *Arachis:* the whole is not the sum of its parts.

Ana Paula Zotta Mota[1, 2] (ana-paula.zotta-mota@inrae.fr), Ana Cristina Miranda Brasileiro[2], Bruna Vidigal[2], Thais Nicolini Oliveira[2], Andressa da Cunha Quintana Martins[2], Mario Alfredo de Passos Saraiva[2], Ana Claudia Guerra de Araújo[2], Roberto C. Togawa[2], Maria Fatima Grossi-de-Sá[2, 3], Patricia Messenberg Guimaraes[2]

[1] *ISA, INRAe, Université Côte d'Azur, CNRS, Sophia Antipolis, France*; [2] *EMBRAPA, Brasilia, Brazil*; [3] *Universidade Católica de Brasília (UCB), Brasilia, Brazil*

Root-knot nematodes together with other plant-parasitic nematodes are the major constraints in tropical agriculture, however, often they occur simultaneously with the action of natural events, such as drought. Plant responses to these stresses are complex and require crosstalk between biotic and abiotic signaling pathways. In this study, we explored the transcriptome data of wild *Arachis* species subjected to drought (Abiotic-metaDEG) and the root-knot nematode *Meloidogyne arenaria* (Biotic-metaDEG) via meta-analysis, to identify core-stress responsive genes to each individual and concurrent stresses in these species. Transcriptome analysis of a nematode/drought bioassay (cross-stress) showed that the set of stress responsive DEGs to concurrent stress is distinct from those resulting from overlapping A- and B-metaDEGs, indicating a specialized and unique response to combined stresses in wild *Arachis*. Whilst individual biotic and abiotic stresses elicit hormone-responsive genes, most notably in the jasmonic and abscisic acid pathways, combined stresses seem to trigger mainly the ethylene hormone pathway. The overexpression of a cross-stress tolerance candidate gene identified here, an endochitinase-encoding gene (*AsECHI*) from *Arachis stenosperma*, reduced up to 30% of *M. incognita* infection and increased post-drought recovery in *Arabidopsis* plants submitted to both stresses. The elucidation of the network of cross-stress responsive genes in *Arachis* contributes to better understanding the complex regulation of biotic and abiotic responses in plants facilitating more adequate crop breeding for combined stress tolerance.

Keywords: M. arenaria - M. incognita - Arabidopsis - Arachis.

Loss of the ERGO-1 small RNA pathway in *Caenorhabditis inopinata.*

Vicky Hunt[1] (v.l.hunt@bath.ac.uk), Akemi Yoshida[2], Ryusei Tanaka[3], Kazunori Murase[3], Taisei Kikuchi[3]

[1] *Department of Biology & Biochemistry, University of Bath, Bath, United Kingdom*; [2] *Univeristy of Miyazaki, Miyazaki, Japan*; [3] *University of Miyazaki, Miyazaki, Japan*

The ERGO-1 small RNA (sRNA) pathway is best characterised in *C. elegans* where it is involved in regulating the expression of tandemly duplicated genes and pseudogenes. We investigated the sRNAs expressed in *C. inopinata*, the closest known relative of *C. elegans*. Many of the sRNA pathways and the associated sRNAs were conserved between these two species. However, orthologues of the genes coding for the ERGO-1 Argonaute protein and other genes involved in the ERGO-1 pathway were lost from *C. inopinata*. This loss was likely to be the result of high levels of transposase activity in the *C. inopinata* genome. The 26G small interfering RNAs associated with the ERGO-1 pathway were also absent in *C. inopinata* and the sRNA expression profiles in these nematodes that were similar to *C. elegans ergo-1* mutants. We identified Argonaute protein-coding genes and siRNAs in *C. inopinata* that are candidates for an alternative sRNA pathway to ERGO-1. To understand the conservation and diversification of the ERGO-1 pathway in nematodes more generally, we identified ERGO-1 pathway orthologues across diverse nematode species. We found that although the *ergo-1* Argonaute gene was well conserved amongst nematodes, other genes involved in this pathway were absent from nematodes outside of the *Caenorhabiditis* clade. We hypothesise that the ERGO-1 pathway found in *C. elegans* is unique to this clade of nematodes but has been lost in *C. inopinata*.

Keywords: *C. elegans* - *C. inopinata* - Small RNA - Argonuate - siRNA.

References:

- Kanzaki et el (2018) Nature Communications (9): 3216

A Comparative study of the development and reproduction of *Meloidogyne enterolobii* and other thermophilic *Meloidogyne* sp.

Raymond Lesley Collett[1] (22847650@student.g.nwu.ac.za), Mieke Stefanie Daneel[2], Mariette Marais[3], Hendrika Fourie[1]

[1] *Environmental Sciences and Management, North-West University, Potchefstroom, South Africa*; [2] *Nematology, ARC-Tropical and Subtropical Crops, Mbombela, South Africa*; [3] *Nematology, ARC-Plant Health and Protection, Queenswood, South Africa*

Little is known about the life-stage development of South African *M. enterolobii*, compared to that of its thermophilic counterpart species *M. incognita* and *M. javanica*. Second-stage juveniles (J2) of *M. enterolobii*, *M. incognita* and *M. javanica* were inoculated on roots of susceptible maize, soybean, and tomato seedlings, and placed in a glasshouse. Three days after inoculation (DAI) the first sampling interval occurred, while the rest of the infected seedlings were transplanted into 400-ml tubes and their roots uprooted 5, 10, 15, 20, 25 and 30 DAI. The sodium-hypochlorite-acid-fuschin method [2] was used to stain the roots of crop seedlings and determine the life-stage development [1,3] of each *Meloidogyne* spp. Differences among the three species in terms of life-stage development, and abundance thereof, were substantial for the different sampling intervals. Life stages of all three *Meloidogyne* spp. 3 DAI was dominated by vermiform, second-stage juveniles (J2) in roots of all three crops. However, *M. enterolobii* was the only species that also had swollen J2 at this sampling interval. Swollen third-stage juveniles (J3) of *M. enterolobii* only, was furthermore evident 5 DAI, while those for the other two species occurred only 10 DAI. Interestingly, 10 DAI swollen fourth-stage (J4) juveniles occurred for all species in roots of all the three crops while females were recorded for all three crops 15 DAI. However, *M. enterolobii* was the only species to possess single eggs 15 DAI for all three crops, indicating a quicker life-stage development than that of the other two species. Although all the species produced egg masses 20 and 25 DAI, the number of eggs per root system and per egg mass did not differ significantly. Of particular interest, however, was that vermiform and swollen second-generation J2 were present for *M. enterolobii* 20 DAI, while swollen second-generation J2 of *M. javanica* were recorded five days later (25 DAI). These results suggested that *M. enterolobii* had a quicker life-stage development compared to its thermophilic counterpart species. Such information is crucial and will be important when designing an integrated management approach to substantially reduce population densities of this particular species and optimizing yield and food security.

Keywords: Life-cycle - Thermophilic - *Meloidogyne entrolobii* - South Africa.

References:

- [1] Tritantaphyllou & Hirschmann, 1960. Annales de l'Institut Phytopathologique, Benaki. 3(1):1-11.
- [2] Byrd et al., 1983. Journal of Nematology. 15(1):142-143.
- [3] Abad et al., 2009. CAB International: Wallingford. 163-181.

Steroidal alkaloids as hatching factors for the potato cyst nematode, *G. rostochiensis*.

Juliet Ochola[1] (jochola@icipe.org), Laura Cortada[2], Margaret Ng'ang'a[3], Ahmed Hassanali[3], Danny Coyne[2], Baldwyn Torto[1]

[1] *Behavioural and Chemical Ecology Unit, International Centre of Insect Physiology and Ecology, Nairobi, Kenya*; [2] *International Institute of Tropical Agriculture, Nairobi, Kenya*; [3] *Chemistry Department, Kenyatta University, Nairobi, Kenya*

Potato is a widely consumed staple food crop worldwide and the second most important food crop in Kenya, commonly grown by small-holder farmers. In the recent years, Kenya has experienced a major decline in potato production even though the cultivation areas had been increasing [1]. One of the key constraints to potato production in the country is due to potato cyst nematodes (PCNs). Management strategies developed so far, in the context of intensive potato farming systems in Europe, have limited applicability in sub-Saharan Africa. Small-holder farmers in this region lack adequate extension services, support and appropriate national phytosanitary policies to combat PCN infestation. Therefore, there is a pressing need to develop innovative solutions for PCN management. A promising avenue is induction of 'suicide hatch' in the soil using naturally occurring plant-derived compounds by exploiting the chemical ecology of rhizosphere interactions between PCN and potato. The current study sought to investigate the PCN (*G. rostochiensis)* hatching response to three steroidal aglycones; solanidine, solasodine and tomatidine detected in the root exudate profile of potato plant. The known PCN hatching factors, the steroidal glycoalkaloids (SGAs) α-solanine and α-chaconine [2], were used as positive controls. In laboratory assays, the steroidal aglycones solanidine and solasodine stimulated hatching in PCN which compared favorably to the positive controls. Tomatidine elicited the lowest hatching compared to the aglycones. Interestingly, 33%-83% of the hatched juveniles failed to emerge from the cyst, which was compound- and dose-dependent. These results suggest that SGAs and their aglycones play a role in the chemo-ecological interactions of PCN and can potentially be exploited in management options.

Keywords: Glycoalkaloids - Hatching factors - Steroidal alkaloids - *Globodera rostochiensis* - Semiochemicals.

References:

- [1] FAOSTAT 2017. Food and Agriculture Organization Statistical Database, http://www.fao.org/faostat/en/#data/QC
- [2] Devine et al., 1996. Annals of Applied Biology, 129 (2), 323-334.

ORAL SESSION 23

Integrated nematode management
(continued)

Integrated nematode control options for cereal and leguminous crops in South Africa.

Hendrika Fourie[1] (driekie.fourie@nwu.ac.za), Raymond Collett[1], Nancy Ntidi[2], Mieke Daneel[3]

[1] *Unit for Environmental Sciences and Management, North-West University, Potchefstroom, South Africa*; [2] *Crop Protection, Agricultural Research Council-Grain Crops, Potchefstroom, South Africa*; [3] *Crop Protection, Agricultural Research Council-Tropical and Subtropical Crops, Nelspruit, South Africa*

Nematode pests adversely impact crop production in Africa, both in the developing and commercial agricultural sectors. Yield losses as a result of nematode parasitism are grossly underestimated and tend to increase as a result of intensified cropping systems, the limited availability of resistant crop cultivars, changes experienced in climatic conditions and the continuous withdrawal of red-band synthetically-derived nematicides. Researchers, producers and related crop industries are therefore investing in research that increasingly focuses on alternative, environmentally-friendly nematode management strategies. Results of such research efforts conducted under South African climatic conditions are presented and encompass laboratory, glasshouse and field experiments in which standard nematode inoculation, extraction, counting and identification protocols were applied. Highlights of these research initiatives, mainly focusing on *Meloidogyne* spp., include i) the identification of resistant and/or poor-host soybean, maize and *Amaranthus* (African Leafy vegetable) genotypes to commonly occurring species (including *M. enterolobii*); ii) adverse effects of biological agents (e.g. *Bacillus*) and plant-derived products (e.g. Vetiver root and leaf exudates) on the biology of *Meloidogyne* spp. second-stage juveniles (J2); iii) efficacy of soil amendments (animal manures; Brassicaceae aerial parts) as well as crop rotation sequences in reducing *Meloidogyne* spp. densities. Using genetic host plant resistance, the reduction of *Meloidogyne* spp. densities by up to 90% (compared to a susceptible genotype) has been demonstrated and should be a preferred, first line of defence strategy to effectively control this nematode pest in local cereal- and leguminous-based cropping systems. However, the susceptibility of local soybean and maize genotypes to *M. enterolobii* indicates that combatting this species in local crop fields will be a challenge. Furthermore, the use of secondary metabolites of biological agents (being >90% effective in paralyzing *Meloidogyne* spp. J2), plant-derived products (inhibiting *Meloidogyne* spp. J2 respiration up to 94%), soil amendments (reducing *Meloidogyne* spp. egg and J2 densities >50%) and the manipulation of cropping sequences are foreseen to play an important role in the future management of *Meloidogyne* spp. in local crop fields under harsh, changing environmental conditions.

Keywords: Alternative control - Host plant resistance - Maize - *Meloidogyne enterolobii* - Soybean.

Reaction of *Phaseolus vulgaris* accessions from EMBRAPA core collection as to resistance to *Heterodera glycines.*

Mara Rubia Da Rocha[1] (darochamararubia@gmail.com), Daniela Patricia Balduino[1], Cristiano Augusto Mendes Junior[1], Thiago Livio Pessoa Oliveira De Souza[2], Rosana Pereira Vianello Brondani[2], Paula Pereira Torga[2]

[1] *Escola de Agronomia/Laboratorio de Nematologia, Universidade Federal de Goias, Goiania, Goias, Brazil*; [2] *Embrapa Arroz e Feijao, Santo Antonio, Goias, Brazil*

Common bean (*Phaseolus vulgaris*) is one of the basis of Brazilian food and its planting and production has been increasing is some areas as a crop succession after soybeans. This practice may increase the soybean cyst nematode (*Heterodera glycines*) populations leading to yield loss for both crops. Thus, finding genetic resistance in commom bean genotypes that may be used in plant breeding programs for developing resistant cultivars should be one of the tasks to have management alternatives. This study had the purpose to evaluate the reaction of 55 accessions of common bean from Embrapa Core Collection regarding its resistance to *H. glycines*. Experiments were carried out under greenhouse using completely randomized design with 6 replications. At 7 days after sowing, the seedlings were inoculated with 4.000 eggs + J2 of *H. glycines*. Shoot and root fresh weight, total number of females, number of females per 10 grams of roots, number of eggs per female and reproduction factor (RF) were evaluated around 38 and 39 days after inoculation. Plants were removed from pots and their roots used for extraction of *H. glycines* females. After counting the number of females, they were popped on a set of sieves (100 and 500 mesh) for eggs release, which were counted under microscope with the aid of a Peters slide. Experiment 1 testing 25 accessions showed a number of females/g of root ranging from 15 to 151 while the soybean plants used as control presented an average of 461 females/g with RF = 5.13. All 25 bean accessions were resistant according to Oostenbrink (1966), with RF ranging from 0.08 to 0.66. Experiment 2 tested 13 accessions, all resistant to *H. glycines* with RF ranging from 0.0 to 0.53. Experiment 3 with 17 bean accessions had only one resistant accession (RF = 0.6). The others had RF ranging from 1.09 to 7.11. Resistant accessions besides presenting RF lower than 1.0 also had lower numbers of eggs per female compared to soybean. Even though most accessions tested were considered resistant all of them hosted *H. gycines* at certain level.

Keywords: Soybean cyst nematode - Genetic resistance - Common bean - Reproduction factor.

Detrimental impact of soil fumigants on nematode suppressiveness.

Tristan Watson[1] (TWatson@agcenter.lsu.edu), Johan Desaeger[2], Tom Forge[3], Louise Nelson[4], Denise Neilsen[3], Gerry Neilsen[3]

[1] *Plant Pathology and Crop Physiology, Louisiana State University, Baton Rouge, Louisiana, United States*; [2] *University of Florida, Wimauma, Florida, United States*; [3] *Agri-Food and Agriculture Canada, Summerland, British Columbia, Canada*; [4] *University of British Columbia, Kelowna, British Columbia, Canada*

In North America, soil fumigants have historically been used to reduce crop damage by soilborne pests, including plant-parasitic nematodes. There are growing concerns regarding the negative effects of these products on soil health, including the ability of soil to suppress nematode invasion or population growth. The objective of this study was to evaluate the impact of soil fumigants on nematode suppressiveness in fruit and vegetable production systems in Canada and the United States. At a *Pratylenchus penetrans*-infested orchard site in British Columbia, Canada, soil was fumigated with dazomet (Basamid®) prior to replanting with sweet cherry. The fumigation treatment was compared to an untreated control, soil incorporated compost, bark chip mulch, and a combination of compost and bark chip mulch. Although fumigation reduced *P. penetrans* population densities to negligible levels at the time of planting, by the end of the first growing season *P. penetrans* population densities in fumigated soil equalled or exceeded those in other treatments. A laboratory assay of soil suppressiveness indicated that fumigation reduced soil suppressiveness relative to the organic amendments. In Florida, United States, chloropicrin (Tri-Pic 100®) and chloropicrin + 1,3-dichloropropene (PicChlor 60®) were drip applied to soil prior to planting with a tomato crop in spring. Cucumber was directly seeded into the previous tomato planting holes the following fall. Both soil fumigants reduced root parasitism by *Meloidogyne javanica* on the tomato crop; however, the double-cropped cucumber plants were more heavily parasitized in previously fumigated soil than in untreated soil. In a subsequent survey on the distribution of nematode suppressive soils in Florida, United States, commercial strawberry farms under organic management possessed soil that was more suppressive to nematode invasion relative to a commercial strawberry farm under conventional management consisting of annual soil fumigation. Soil from the conventionally managed strawberry farm also had lower fungal diversity relative to that of the organically managed strawberry farms. Collectively, these studies indicate that while soil fumigation provides short-term nematode control, it also often results in greater nematode invasion or population growth later in the growing season or in subsequent seasons, likely as a result of loss of microbially-mediated nematode suppressiveness.

Keywords: Soil fumigation - Root-knot nematode - Lesion nematode - Soil suppressiveness.

Effects of some spice extracts on *Melodiogyne arenaria*.

Hissein Mahamat Haroun[1] (koubawi.hisseine@gmail.com), Gökhan Aydınlı[2], Sevilhan Mennan[1]

[1] *Plant protection dep., Ondokuz Mayıs University, Samsun, Turkey*; [2] *Department of Crop and Animal Production, Ondokuz Mayıs University, Bafra, Samsun, Turkey*

Plant-based natural approaches have always attracted attention for the management of RKN, since their effects on the environment and humans are less compared to synthetic nematicides [1]. It is known that many plants including spicy plants have components that negatively affect the nematodes by inhibiting egg hatch, causing immobility of second-stage juveniles (J2) or lethal effects [2, 3]. In this study, laboratory and pot experiments were designed to test the effects and effect types of extracts obtained from 13 different spice plants (*Anethum graveolens*, *Capsicum annuum, Cuminum cyminum, Coriandrum sativum, Curcuma longa, Ocimium basilicum, Helichrysium italicum, Piper nigrum, Prunus mahlep, Rhus coriaria, Syzygium aromaticum, Thumus vulgaris, Zingiber officinale*) on *M. arenaria*. Laboratory assays were conducted in 48-well Elisa plates with 8 replicates. Extracts at concentrations of 0.5%, 1% and 2% were tested for egg hatch inhibition, mortality and immobility of J2. The highest rate of egg hatch inhibition was detected in *C. annuum* extract with 82.5% at 0.5%, 86.13% at 1%, and 93.13% at 2%. The highest immobilization of J2 was determined in the *S. aromaticum* extract with 16.75%, 27.87% and 39.50% at concentrations of 0.5%, 1% and 2%, respectively. The five extracts (*O. basilicum, S. aromaticum*, *C. cyminum*, *C. sativum* and *C. longa*) having the highest effects on nematodes in laboratory assays were tested in the pot experiment. For this purpose, extracts at a concentration of 2% were mixed with 200 g of nematode-infested soils (Pi=3000 eggs) in plastic bags. Water and a nematicide suspension (200 g/l Ethoprophos) were used as negative and positive controls, respectively. Each plastic bag was sealed and incubated for 7 days at 24±2°C. After incubation, soils were transferred to pots and nematode-susceptible tomato seedlings were transplanted. Pots were arranged in a randomized complete block design with 8 replicates in the greenhouse (25±3°C). Forty-five days after nematode inoculation, gall index (according to 0-5 gall scale) and nematode egg number (Pf) were determined on roots. All extracts except *C. cyminum* had lower root gall indices than the negative control. Besides, all extracts caused lower reproductive factor (Pf/Pi) than negative control, but extract of *O. basilicum* showed the highest reduction in nematode reproduction (84.03%), followed by *C. longa* (78.94%), *S. aromaticum* (75.64%), *C. cyminum* (68.90%) and *C. sativum* (49.58%) extracts.

Keywords: *M. arenaria* - Spice extracts - Egg hatch inhibition - Immobility - Control.

References:

- [1] Chitwood, D. J. 2002. Annual review of phytopathology. 40(1): 221-249.
- [2] Oka et al., 2000. Phytol. 90(7): 710-715.
- [3] Claudius-Cole et al., 2010. Mycopath. 8(2): 53-60.

Biotic factors of mulch-induced soil suppressivity against *Meloidogyne incognita* may depend on soil microclimate.

Krisztina Boziné Pullai[1] (kriszti.pullai@gmail.com), Renáta Petrikovszki[1], Franciska Tóthné Bogdányi[2], Péter Nagy[3], Zoltán Mayer[4], Katalin Posta[4], Rozália Tóth[5], Ferenc Tóth[1]

[1] *Plant Protection Institute, Szent István University, Gödöllő, Hungary*; [2] *2FKF Zrt., Budapest, Hungary*; [3] *Department of Zoology and Animal Ecology, Szent István University, Gödöllő, Hungary*; [4] *Institute of Genetics, Microbiology and Biotechnology, Szent István University, Gödöllő, Hungary*; [5] *Szent István University, Hungary*

Soil suppressivity against root-knot nematodes (RKNs) is a complex soil trait, governed by several abiotic and biotic factors. One of our long-term, on-going experiments with various types of organic mulch suggests that mulching may induce suppressivity against RKNs. Since mulching lowers soil temperature in summer, we set up an experiment to test the influence of this particular abiotic factor. We tested the hypothesis that lowered temperature due to mulching was the most important factor to induce soil suppressivity.
There were 12 mulched and 12 unmulched plots in our long-term field study. We sampled all the 24 plots after 4 years. Samples were then halved and put into small pots measuring 11×11 cm. Half of the pots were artificially infested with root galls and with infested soil to ensure the survival of the RKN larvae. Tomato seedlings were planted individually into the pots and kept in a plant growth room. We made sure that all pots, regardless of the treatments were kept at the same temperature throughout the experiment. We monitored plant growth for two months. At the end of the experiment, we evaluated root galls and plant vigour parameters, and determined the enzymatic activity level within the plants and in the soil.
The 4-year mulching history of soil increased plant vigour significantly in the laboratory, but soil samples seemed to have lost their former in-situ suppressivity. While root galling index was significantly lower on plants in mulched plots in the field, in the laboratory we found that RKN symptoms were equally severe (Zeck scale 5-6) in both soil sample groups. This finding may emphasize the role of the temperature differences we experienced under mulching in the field. While the role of biotic factors still needs further studies, our results suggested that the biotic factors of mulch-induced suppressivity may depend on soil microclimate.
The work is supported by the EFOP-3.6.3- VEKOP-16- 2017-00008 project and the New National Excellence Program of the Ministry of Human Capacities (ÚNKP-19-3-I-SZIE-17). The project is co-financed by the European Union and the European Social Fund.

Keywords: Southern root-knot nematode - Integrated nematode control - Leaf litter mulch - Abiotic and biotic factors - Tomato.

Management of *Meloidogyne paranaensis* and *M. exigua* in coffee with chemical and biological nematicides.

Andressa Cristina Zamboni Machado[1] (andressaczmachado@hotmail.com), Daniel Sala Faria[2], Santino Aleandro Silva[1]

[1] *Plant Protection, Instituto Agronômico do Paraná, Londrina, Paraná, Brasil*; [2] *ADAMA do Brasil, Londrina, Paraná, Brasil*

Meloidogyne paranaensis (Mp) and *M. exigua* (Me) are important to Brazilian coffee crops, reducing the longevity and leading to plant mortality. Management is done with the resistant rootstock cv. Apoatã and, in lesser proportion, with the application of nematicides. The objective of this work was to verify the effect of the application of nematicides in the control of Mp and Me in coffee cv. Catuaí, under greenhouse conditions. For this, coffee plants with six pairs of leaves were transplanted to 25 L-capacity pots and inoculated with 1,000 eggs of Mp or Me. Treatments with fluensulfone and *Purpureocillium lilacinum* were done 24 hours before the inoculation. Inoculated plants without nematicides were used as controls. Evaluation was done 12 months after the inoculation by extracting nematodes from the entire root systems and calculating the final population (FP). Fresh top weight (FTW) of plants were measured. Data were analyzed considering a double factorial arrangement: nematicide and nematode species. We used the Scott-Knott test at 5% of significance and observed the linear model assumptions. Results showed that there was interaction between the analyzed factors in relation to FTW; while to Me no differences were observed between treatments, for Mp the non-treated plants showed lower FTW and other treatments did not differ between them. For treatments, independently of chemical or biological, Me inoculated plants showed higher FTW. In relation to FP, there was interaction between the factors; for Me, non-treated check showed higher FP in relation to nematicide treatments, which did not differ between them. For Mp, no differences between treatments were observed. In relation to nematode, Mp showed higher FP than Me, both in the chemical and biological treatments, but no differences were observed in FP of both nematodes in the absence of nematicides. We can conclude that application of chemical or biological nematicides has a positive impact in the vegetative development of plants inoculated with Mp and this reduced the FP of Me. In the absence of nematicides, Mp causes higher reduction in the vegetative development of plants than Me.

Keywords: Root-knot nematodes - Coffea arabica - Control.

S23-PF1

Case studies of root-knot nematode (*Meloidogyne* spp.) control in protected vegetables in Hungary.

Anita Gódor[1] (agodor@gmail.com), Franciska Tóthné Bogdányi[2], Ferenc Tóth[3]

[1] *Plant Protection Institute, Szent István Egyetem, Gödöllő, Hungary*; [2] *FKF Nonprofit Zrt., Budapest, Hungary*; [3] *Szent István Egyetem, Gödöllő, Hungary*

One of the most challenging soil-related pest management issues of protected crops in Hungary is the control of root-knot nematodes (RKNs). We started to monitor changes to RKN control technology in pepper and cucumber in 2004, and the monitoring and data collection for the purpose of case studies is still ongoing. The method of monitoring consists of in-situ interviews with growers and surveys of greenhouse conditions and the root system and vigour data of plant stands. After the withdrawal of Metabrom 980 (methyl-bromide) in 2005 in Hungary, chemical plant protection products (PPPs) used by growers against RKNs were the fumigants Basamid G (dazomet), Ipam (metam-ammonium) and Nemasol 510 (metam-sodium); and the systemic Nemathorin 10 G (fosthiazat) and Vydate G (oxamyl). During the study period, moderate changes were observed in the chemical control of RKNs due to new restrictions in the use of some registered products, new registrations (Nemacur 240 CS – fenamiphos), or off-label uses of other PPPs. Examples of biological control including the use of Artis (*Arthrobotrys oligospora*), or of resistant or tolerant varieties, hybrids or grafted plants were found only in a few locations. Although the benefits of increasing the organic matter content of the soil (by the addition of compost, organic manure or green manure) are widely known by growers, and the addition of organic matter is considered a potentially promising, sustainable solution against RKN damage, its use in the practice is strongly limited by the high logistic costs and availability problems in the case of organic manure and compost, and the lack of biological service period (active soil regeneration period) between two crop cycles in the case of green manure. Based on the findings of our extended survey we suggest economically sound solutions for RKN control: the registration of new fumigants that offer residue-free protection (e.g. dimethyl-disulfide – DMDS), and the regular use of locally available municipal green waste compost as a soil amendment in greenhouse production.
The publication was supported by the EFOP-3.6.3-VEKOP-16-2017-00008 project. The project is co-financed by the European Union and the European Social Fund.

Keywords: Integrated nematode control - Protected vegetables - Fumigation - Organic soil amendment - Municipal green waste compost.

S23-PF2

Ozone treatments for the management of *Meloidogyne* sp. in greenhouse tomato cultivation in southeastern Spain.

Caridad Ros Ibáñez (caridad.ros@carm.es), Mª Ángeles Hernández Colucho, Daniel Soler Cárceles, José Luis Lozano González

Crop protection, Instituto Murciano de Investigación y Desarrollo Agrario y Medioambietal (IMIDA), La Alberca (Murcia), Spain

The tomato (*Solanum lycopersicum*) is a monoculture in most of the greenhouses in the Region of Murcia, the reiteration of its cultivation causes the accumulation of pathogens such as *Meloidogyne* sp. that motivate the need to disinfect the soil prior to transplanting, in order to ensure the health and productivity of the crop. Currently, the use of most soil fumigants is prohibited or under the authorization of exceptional uses. In order to find alternatives to the usual chemical disinfestation of the tomato production area for the control of nematodes, a comparative trial was conducted in an experimental greenhouse whose soil is naturally contaminated by *Meloidogyne incognita*. The following treatments were evaluated: i) 1,3 dichloropropene + Velum in post-transplantation, ii) ozone gas + 2 irrigations with ozone in post-transplantation, iii) ozone gas + irrigations with ozone throughout the crop, iv) control without disinfection. The population of the nematode was measured before and after soil disinfestation and during crop cycle, the incidence of the nematode by means of the gall index and the percentage of affected plants. No juveniles were found in the soil after disinfestation, neither in the plots with 1,3 dichloropropene nor in those with ozone. The application of ozone gas + ozone irrigation during cycle crop reduced the incidence of damage and the percentage of affected plants by 61.9 and 42.4%, respectively, compared to the control. With respect to the treatment of 1.3 dichloropropene + Velum in post-transplantation, no significant differences were found in the percentage of affected plants, but were found in the gall index (0.2 for dichloropropene and 0.8 for ozone gas + irrigation with ozone during cultivation). The treatment with ozone gas + 2 irrigations with ozone did not show differences with respect to the control

Keywords: Ozonated water - Soil disinfection - Root Knot - *Solanum lycopersicum*.

S23-PF3

Interaction between *Fusarium* spp and root lesion nematode *Pratylenchus capsici* on pepper crops in the Arava (Israel).

Patricia Bucki[1] (pbucki@agri.gov.il), Maoz Aizikowitz[2], Marcel Maymon[3], Marina Benichis[4], Ohad Abraham[4], Qing Xue[1], Abraham Gamliel[4], Sigal Brown Miyara[1]

[1] *Plant Protection, Entomology and the Nematology and Chemistry units, ARO-Volcani Center, Rishon LeZion, Israel*; [2] *Department of Biotechnology Engineering, ORT Braude College, Carmiel, Israel*; [3] *Plant Protection, Plant Pathology and Weed Research, ARO-Volcani Center, Rishon LeZion, Israel*; [4] *Agricultural Engineering, Growing, Production and Environmental Engineering, ARO-Volcani Center, Rishon LeZion, Israel*

During the past 50 years, Arava rift – South of Israel has evolved into a unique agro-ecosystem with advanced agricultural technologies, producing highly profitable fresh vegetables, herbs and cut flowers which are mainly export oriented. However, in recent years, severe root infections caused by a root-lesion nematode were detected in pepper from several agriculture farms in Arava rift, suggesting a highly specialized nematode-plant interaction. The root lesion nematodes *Pratylenchus capsici* are prospering in pepper fields in the Arava- Israel [1]. The occurrence of the phytopathogenic fungi, *Fusarium solani* and *Fusarium oxysporum* [2], on pepper infected by *P. capsici* raised the question whether there is a synergistic effect among these pathogens. The objective of this work is to determine whether the root lesion nematodes, *P. capsici,* might serve as a carrier for phytopathogenic fungi, facilitating their entry into pepper roots.

In order to confirm our hypothesis, we first identified genetically the soil fungi population associated with *P. capsici,* and performed *in vivo* Koch postulates test. By using *Fusarium oxysporum* spores expressing the GFP reporter gene we studied the direct interaction between *P. capsici* and *F. oxysporum* spores through *in vivo* and *in vitro* tests. Next, we checked whether the infection of nematodes together with fungi spores has any synergistic effect on symptoms observed on pepper.

In terra experiments indicate that mutual inoculation led to increased stunted growth and wilting symptoms observed on pepper plants compared with inoculation by nematodes or fungus alone or to the non-inoculated control. These results further illustrate the potential of disease complex occurrence in which *Pratylenchus capsici* is involved.

Keywords: Root lesion nematode - *Pratylenchus capsici* - Pepper crops.

References:

- [1] Xue et al., 2019 Phytopathology. 109 (5): 847-58.
- [2] Cha et al. 2007 Mycobiology. 35 (2): 91-96.

WORKSHOP 8

Nematode-fungal interactions and complex diseases

Nematode-fungal interactions and complex diseases

Richard Sikora[1] (rsikora@uni-bonn.de), Ann MacGuidwin[2], Driekie Fourie[3], Danny Coyne[4], Holger Heuer[5], Matthias Gaberthüel[6]

[1] *University of Bonn, Bonn, Germany*; [2] *University of Wisconsin, Madison, Wisconsin, USA*; [3] *North-West University, Potchefstroom, South Africa*; [4] *IITA, Nairobi, Kenya*; [5] *Julius Kuehn Institute, Braunschweig, Germany*; [6] *Syngenta Crop Protection, Basel, Switzerland*

The objectives of this workshop are to review and anticipate future impact of nematode interaction with fungal and bacteria pathogens on root and crop health. The workshop will look at problems on a wide array of food crops affecting food security on a worldwide basis. The presentations will summarize: where we are, where we have problems, and where research is required in the future.
The presentations - listed in the order of the authors of this abstract - will cover:
1) Nematode-disease complexes: a neglected factor in improving crop health
2) *Verticillium / Pratylenchus* interaction for potato - both do the damage, but only one gets the credit
3) Cereal root rot - nematode interrelationships: vision for the future?
4) Nematode-fungal root-rot complexity on banana: a perennial problem of unknown importance?
5) Metagenomics: the future to disentangle complex diseases?
6) Present and future management systems for complexity management
Following the presentations ample time will be available for audience-speaker discussions on
1) the impact of complex diseases on root and plant health
2) new approaches for research in the soil and rhizosphere and
3) development of integrated management approaches.
We anticipate rigorous discussion on future nematological research on this important crop health problem.

Keywords: Root and soil health - Complex diseases - Integrated management - Metagenomics - Fungal and bacterial pathogens.

Nematode-disease complexes: a neglected factor in improving crop health.

Richard Sikora[1] (rsikora@uni-bonn.de), Ann MacGuidwin[2], Driekie Fourie[3], Danny Coyne[4], Holger Heuer[5], Matthias Gaberthüel[6]

[1] *University of Bonn, Bonn, Germany*; [2] *University of Wisconsin, Madison, Wisconsin, USA*; [3] *North-West University, Potchefstroom, South Africa*; [4] *IITA, Nairobi, Kenya*; [5] *Julius Kuehn Institute, Braunschweig, Germany*; [6] *Syngenta Crop Protection, Basel, Switzerland*

No abstract submitted.

Verticillum / Pratylenchus for potato: both do the damage, but only one gets the credit.

Ann MacGuidwin (aem@plantpath.wisc.edu)

Plant Pathology, University of Wisconsin, Madison, USA

No abstract submitted.

Cereal root rot - nematode interrelationships: vision for the future?

Driekie Fourie, Amer Dababat

No abstract submitted.

Nematode-fungal root-rot complexity on banana: a perennial problem of unknown importance?

Danny Coyne (d.coyne@cgiar.org)

IITA, Nairobi, Kenya

No abstract submitted.

Metagenomics: the future to disentangle complex diseases?

Holger Heuer (holger.heuer@julius-kuehn.de)

Julius Kuehn Institute, Braunschweig, Germany

No abstract submitted.

Present and future management systems for complexity management.

Matthias Gaberthüel (matthias.gaberthueel@syngenta.com)

Syngenta Crop Protection AG, Basel, Switzerland

No abstract submitted.

WORKSHOP 9

Slime time: Nematodes associated with terrestrial slugs

Slime time: Nematodes associated with terrestrial molluscs

This workshop aims to showcase current research, technology and development on nematodes associated with terrestrial molluscs. This subject area has gained significant interest in the last decade, with numerous advancements, as well as surveys on the occurrence of mollusc-parasitic nematodes from around the world. Join us to learn more about this subject area, with a number of key invited speakers, followed by a discussion on how we can drive this area of research forward.

Survey of slug-parasitic nematodes in East and West Flanders, Belgium, and description of *Angiostoma gandavense* n. sp. (Nematoda: Angiostomatidae) from arionid slugs.

Phougeishangbam Rolish Singh (phougeishangbamrolish.singh@ugent.be), Marjorie Amoto, Marjolein Couvreur, Wilfrida Decraemer, Wim Bert

Nematology Research Unit, Ghent University, Ghent, Belgium

Slug- and snail-associated nematodes were surveyed in different locations in Belgium which revealed the presence of one new, nine known and four unidentified nematodes species in the bodies of the collected mollusc samples. *Angiostoma gandavense* n. sp. (Angiostomatidae) was discovered from arionid slugs (Arion ater and Arion flagellus) and is described based on light microscopy and scanning electron microscopy data [1]. The new species can be readily distinguished from related species based on both sequence differences and morphological characters such as the distinctive mucronate structures at the tail tip of both sexes, presence of lateral ala, reflexed female ovaries and the number and arrangement pattern of male genital papillae. The known nematode species detected in the survey include *Cosmocerca longicauda, Agfa flexilis, Alloionema appendiculatum, Angiostoma dentiferum, A. limacis, A. norvegicum, Angiostrongylus vasorum, Phasmarhabditis californica* and *P. hermaphrodita*. Remarkably, C. longicauda and A. vasorum are also well-known parasites of amphibians and domestic/wild canids, respectively. The unidentified mollusc-associated nematodes include *Panagrolaimus* sp., *Rhabditis* sp., *Tetrameres* sp. and *Troglostongylus* sp. Several cases of co-infection of slugs by the detected nematodes species were found. Three species namely *A. norvegicum, P. californica and P. hermaphrodita* are recorded for the first time in Belgium. All these species have been documented by light microscopy microphotographs and informative DNA sequences of partial ITS, 28S and 18S of rDNA and partial COI mtDNA.

Keywords: Diversity - Molluscs - Mollusc-parasitic nematodes - Systematics - Taxonomy.

References:

- [1] Singh et al., 2020. J. Helminthol. 94: E35.

Looking for nematodes in the apple snail *Pomacea canaliculata* - a recent invasive in Kenya

Solveig Haukeland (Solveig.haukeland@nibio.no), Fernadis Makale, Ivan Rwomushan

Norwegian Institute of Bioeconomy Research (NIBIO), As, Norway

The apple snail, also named ´golden apple snail´(GAS), *Pomacea canaliculat*a, is widely considered as one of the most invasive invertebrates of waterways and irrigation systems worldwide, already causing havoc throughout much of south-east Asia. The snail was recently reported in Kenya (2020) from the expansive Mwea irrigation scheme. This was the first confirmed record of an established population in continental Africa. Given the impact of this species in Asia, there is need for a rapid and coordinated response to contain and mitigate the risk to other rice schemes, as well as neighbouring countries. Currently CABI and KEPHIS (The Kenya Plant Health Inspectorate Service) are addressing the problem. In tandem there is a need to investigate the presence of parasites of the snail for potential biocontrol as part of an integrated control strategy. We have started such investigations by collecting and dissecting snails. We have also conducted preliminary trials with Slugtech (DUDUTECH product of *Phasmarhabditis hermaphrodita*). In this presentation, the snails biology, distribution and impact in Kenya will be discussed as well as preliminary results of on-going studies on parasites.

Molluscs and EPNs: Symbiotic bacteria may help with slug control.

Jiří Nermuť (nermut@entu.cas.cz), Victoria Weijler, Jakub Savula, Vanessa Bachinger, Shakeel Zahid, Jana Konopická, Jiří Borák, Martin Janouch, Vladimír Půža

Institute of Entomology, Biology Centre CAS, České Budějovice, Czechia; Faculty of Agriculture, University of South Bohemia, České Budějovice, Czechia

Molluscs, in Europe mainly slugs, are serious pests of many crops and ornamentals. The most harmful species are members of *Deroceras* and *Arion* genera. Currently, there are two ways of their control: chemical and biological. Conventional baiting pellets are based on methiocarb (banned in EU), metaldehyde (banned or will be banned soon in most of EU countries) or iron phosphate while biological methods are based on nematode *Phasmarhabditis hermaphrodita* in combination with *Moraxella osloensis* bacterium. Among other, often limited methods such as salt, beer traps, slug bots etc., one could have a potential to become a new effective approach. Entomopathogenic nematodes of the genus *Steinernema* live in symbiotic association with bacteria of the genus *Xenorhabdus*. Bacterial symbiont is not able to live and infect insect without its nematode vector that transport bacteria directly to the new suitable host. On the other hand, nematode developing inside the cadaver needs protection for several days to finish the life cycle. This protection is possible thanks to the ability of bacteria to produce wide range of different metabolites that act e. g. as antibiotics, bactericides, fungistatics, repellents or antifeedants etc. Using of bacteria and their secondary metabolites in biological pest control is recently quite popular and seems to bring very promising results. Even the metabolites produced by bacteria *Xenorhabdus* spp. are known to be effective against some plant fungal pathogens, insects, protozoans and nematodes. In this contribution we would like to show how these metabolites can influence behaviour of slugs.

SlugBot: Developing an autonomous monitoring and biocontrol system for slugs.

Jenna Ross (jenna.ross@abdn.ac.uk), Tom Ashford, Archita Barua, Faye McDiarmid, Ben Scott-Robinson, Andy Hall, Pat Barretto, Tom Watsham, Tom Walters, Sam Herring, Rowan Duckworth, Michael Alcock, Tim Knott, Geoff Osmond, Daniel Rowe

Crop Health and Protection (CHAP), York., United Kingdom

Slugs are major economic pests in agricultural and horticultural crops in the UK. Historically, methods of slug control have been reliant on chemical molluscicide pellets, such as metaldehyde or Iron (Ferric) phosphate, however due to the negative impact on UK water systems, as well as on non-target organisms, metaldehyde has subsequently been banned in the UK. Bio-molluscicides are also commercially available as nematode-based products containing *Phasmarhabditis hermaphrodita*, however these are not economical for use in arable and oilseed rape crops. Therefore, the slug control toolbox in the UK is limited. The aim of this project was to develop an innovative game-changing prototype technology for autonomous slug monitoring and precision bio-molluscicide treatment. This presentation will provide high level results of the development and testing of the aforementioned technology.

FORUM 2

Nematology Education Forum: the current situation and the ideal way forward

Nematology Education Forum: the current situation and the ideal way forward.

Inge Dehennin, Driekie Fourie and Wim Bert

Given the immediate concerns over food security combined with the drive towards a more sustainable world, it is more important than ever that education in nematology is actively promoted in academia and elsewhere to ensure adequate nematological capacity building. The landscape of nematology education at Higher Education Institutions ranges from nematology as a rather marginalised discipline in agricultural oriented curricula in institutions around the world to a full Master of Science devoted entirely to nematology. However, this situation is changing due to new investments in education projects in sub-Saharan Africa (Erasmus+ CBHE NEMEDUSSA), but also due to crucial changes in financial support from long-term donors (VLIR-UOS). This workshop is conceived as an interactive forum with two components. Firstly, sharing experiences with nematology education from the perspective of a diverse group of stakeholders such as educators, students, but also potential employers. Secondly, to discuss the design of a future-proof curriculum in nematology, starting from a case study of the MSc in Agro- and Environmental Nematology at Ghent University, and then moving on to different options and opportunities around the world.

7th International Congress of Nematology
ICN 2022
Antibes Juan-les-Pins - France

"Crossing borders: a world of nematode diversity and impact to discover"

European Society of Nematologists

ABSTRACTS FRIDAY 6 MAY

PLENARY SESSIONS

New perspectives on nematode – bacteria interactions, their causes and consequences.

Tom Moens (tom.moens@ugent.be)

Biology Department, Marine Biology Lab, Ghent University, Gent, Belgium

Free-living nematodes can affect soil biogeochemistry and associated ecosystem processes, thereby contributing to important ecosystem services. The mechanisms through which they do so, however, remain poorly understood. One of the commonest explanations for the nematode – biogeochemistry link is that nematodes (micro)bioturbate soils and sediments, thereby influencing the fluxes of oxygen and nutrients, which in turn may enhance or suppress the activity of microbial communities. Here, I provide a brief overview of evidence for bioturbation by nematodes, and use radiography and CT scans to demonstrate that particle mixing but not bioirrigation is indeed enhanced by aquatic nematodes, the effect being more pronounced for larger and more motile species. Alternative explanations for the effects of nematodes on microbes include selective grazing and mucus production, both of which can mainly affect the community structure of microbial communities, which in turn could potentially translate in different microbial activity patterns and different roles in ecosystem processes. In this presentation, I use metagenomics data on the microbiomes from individual nematode specimens to discuss issues of niche differentiation between nematode species through, for instance, differential feeding between even closely related nematode species. Such patterns may, however, be blurred by large intraspecific variability in individual nematode microbiomes, a phenomenon which I show to be – among other things – dependent on the level of intraspecific competition experienced by nematodes. This may explain why in several recent field studies, various bacterial-feeding nematodes obtained from different (micro- and macro)habitats do not exhibit significantly different microbiomes. It may also hamper the detection of cases of nematode-bacteria co-evolution. Irrespective of the exact underlying method, the influence of nematodes on microbial activity in soils and sediments deserves further attention. I provide an example based on a recent study in which the microbial metatranscriptomes of bacterial communities in the presence and absence of a marine nematode community are compared, highlighting the power of such an approach to increase our understanding of the shifts in microbial assemblages induced by nematodes.

From human to plant diseases, scientific approaches of pest control in the big data era.

Corentin Barbu (Corentin.barbu@inrae.fr)

UMR Agronomie, AgroParisTech site de Grignon, Thiverval-Grignon, France.

Big Data has been defined as large, rapidly growing, and diverse data sets. While these characteristics are relevant to management and marketing, they are less relevant to understanding the challenges posed by Big Data in science. Manipulating geographic, qualitative, and quantitative data together requires a good understanding of their nature, but this understanding is generally within the reach of researchers. Statistical tools such as R or Python are commonly mastered in research teams and are sufficient to handle most data sets. Finally, we rarely have to manage the continuous flow of data ourselves. On the other hand, since these data are usually not collected for scientific purposes or at least not for a specific scientific purpose, researchers must be aware that big data are often “dirty”: biased, heterogeneous and that the increased statistical power brought by large volumes requires more rigorous statistical tests to avoid erroneous conclusions. Here, we will focus on approaches to overcome these difficulties through examples of scientific studies using big data. In particular, we will see 1) how citizen reports were used to understand the origin of re-infestations by a deadly pest in Latin America 2) that systematic door-to-door studies can be highly biased and auto-correlated and how this has been used to improve pest control 3) how different national level datasets collected for non-scientific purposes were merged to understand the impact of landscape composition on cereal crop pests in metropolitan France. These examples and very simple data simulations will also give us the opportunity to highlight the vastly underestimated risk of multiple testing and how rigorous cross-validation and p-value corrections can protect us from false conclusions.

Keywords: Pest control - Big Data - Dirty data - Statistical modelling - Multiple testing.

References:

- Delaune T. et al., 2021. Ecography. 44 (10) 1429-1442. doi.org/10.1111/ecog.05433.
- Hong A et al., 2015. Proceedings of The Royal Statistical Society Series A. 2015, 178, 641-658.
- Barbu C.M. et al., 2014. Emerging infectious disease. 20, 12, 2055-2063.

IFNS Global Capacity building programs in nematology.

Ernesto San-Blas (esanblas@yahoo.com)

Lab of Nematology, Universidad de O`Higgins, San Fernando, Chile

Scientific societies play a fundamental role in the science community by supporting researchers and students, fostering collaborations and giving opportunities to share knowledge and resources. In today's world, information and digital resources can be reached by anyone in any part of the globe making the traditional structures of the scientific societies strict. Also the participation and democratization of the information is forcing the governmental bodies to be more open to include many other views to make their objectives more attractive to students, young researchers and important non-academic actors (private companies, farmers, etc.). IFNS encompasses 17 nematological societies with different backgrounds and interests. However, the basic role of the Federation is circumscribed to foster the communication of it's society members, and the only factual activity is to organize the International Congress of Nematology (ICN) every 6 years. According to the membership rolls, more than 2500 members are under the umbrella of the IFSN and several self-motivated people act as unofficial collaborators in many tasks. The lack of more diverse structures on the IFSN inhibits the development of new and varied activities within the federation for building stronger programs. To improve the performance of the IFSN, different perspectives have been analyzed and will be proposed. The inclusion of more official chairs to oversee diverse tasks will be discussed and with the goal of creating structural changes to modernize IFNS in ways that make the federation more inclusive and effective.

Keywords: Interantional Federation of Nematological Societies - Fostering communication - Governing board - Inclusion - Social media.

ORAL SESSION 24

New challenges in nematodes taxonomy and evolution

Constraints on nematode evolution: how to conserve a Bauplan constructed from the substrate of an ever changing genome.

Philipp Schiffer (p.schiffer@uni-koeln.de)

Worm-Lab, University of Cologne, Köln, Germany

Among animal phyla the hyper conserved vermiform body plan of the nematodes is intruiging. This conservation is particularly astounding as the nematodes are ecologically at least as widespread and as successful as the insects, and have diversified into 1 million or more species. At the same time, they display very high rates of molecular evolution. Thus we need to ask: which factors might underly the strict conservation of the nematode Bauplan?
Recent studies of gene expression in the model nematode *Caenorhabditis elegans* have supported the idea of a constrained mid developmental period. These data show that gene expression is variable early in development, then becomes more and more constrained, and subsequently becomes variable again, resembling the shape of an hourglass. Here I will describe my own, contrasting results: comparing *C. elegans* to 2 other nematodes, I found a funnel shaped progression of gene expression variance, seeming to provide a possible mechanism for the convergence on the conserved adult Bauplan.
While I have expanded our knowledge beyond the *C.elegans* with this study, it is still far from clear if the hourglass or funnel model correctly describes developmental progression across the phylum.
In this talk, I will describe a study system to analyse developmental gene expression in time-course assays across several nematode taxa, in particular in the drastically under-sampled early branching nematodes. This I will then couple to our past and current genomic analyses profiling which parts of the *C. elegans* developmental toolkit are conserved and which vary throughout the phylum.
As I will show, these data appear to indicate that many genes important in *C. elegans* development are absent in other nematode species, while at the same time we have evidence that basally branching nematodes utilise important developmental regulators, which are lost in the crown clade model species.
In summary, my talk will give first insights into variability (or evolvability) of early stages of development on the genetic level across the phylum, and provide a hypothesis for the constrained Bauplan disparity in Nematoda.

Keywords: EvoDevo - Hourglass - Genome evolution - Constraints - Bauplan.

Analysis of *Meloidogyne hapla* in soil using Loop-mediated Isothermal Amplification technique.

Zahra Omer[1] (zahra.omer@hush.se), Maria Viketoft[2], Ann-Charlotte Wallenhammar[3], Stina Andersson[4]

[1] *Rural Ecomomy and Agricultural Society, Uppsala, Sweden*; [2] *Department of Ecology, Swedish University of Agricultural Scien, Uppsala, Sweden*; [3] *Rural Ecomomy and Agricultural Society, Örebro, Sweden*; [4] *HIR Skåne, Bjärred, Sweden*

The prevalence of root-knot nematodes (RKN) is increasing in Sweden, especially *M. chitwoodi* and *M. fallax* recently found in potatoes, causing severe problems for growers. Soil analysis is extremely important to differentiate between these new RKN species and the prevalent species *M. hapla*. It is also extremely important to be able to quantify RKN at the tolerance limit (T), ≤4 juveniles 250 g^{-1} soil, which means that the adopted soil analysis method must be sensitive enough to detect RKN at this low T-value. We have previously developed a DNA method based on the Loop-mediated Isothermal Amplification (LAMP) to detect and quantify *M. hapla* using both colorimetric and real-time LAMP. The next step is to implement the developed LAMP assay for analyzing *M. hapla* in soil. DNA extraction from soil is the first step in molecular diagnostic and it is also the most costly part when commercial DNA kits are used. The objective of this study was to develop a simple procedure for direct extraction of *M. hapla* DNA from soil samples. The developed extraction procedure mimics commercial kits in binding DNA to silica particles. In order to decrease the extraction time, two silica products were tested. In addition, different volumes of sodium dodecyl sulfate (SDS) and different concentrations of skim milk were tested, in order to increase DNA concentration and purity. The results showed no significant differences in DNA quality and concentration between the two tested silica products, however, gel electrophoresis showed that product A dried faster from ethanol than product B. Selecting product A reduced air drying of silica particles by 15 min. Using 100 µl SDS produced the highest DNA concentration as confirmed by gel electrophoresis. Different concentrations of skim milk did not result in different DNA concentrations. In conclusion, a simple and cheap procedure was developed for direct extraction of *M. hapla* DNA from soil. We were able to detect 0,5 juvenile 500 mg^{-1} soil.

Keywords: Root-knot nematode - LAMP - DNA extraction - Soil analysis.

Nematode species: their evolution and description in theory and practice.

Mike Hodda (mike.hodda@csiro.au)

Insect & Nematode Biosecurity, CSIRO National Research Collections, Canberra, ACT, Australia

Numerous species concepts exist, with several applied to nematodes, albeit implicitly. Most nematode species are assumed to be under the biological species concept—that is the population of interbreeding individuals—but this is seldom tested explicitly. In the few genera where experimental crosses were tested, results have not always aligned with the assumed and named species. A thorough analysis of the criteria for diagnosing species has shown that there are many requirements for unambiguously delineating a species. Very few species fulfil all of the requirements. This is not restricted to nematodes, but in many cases is a result of some characteristic of the nematodes themselves, for example their rarity or wide distribution. Comparing the information available with the ideal criteria can give an insight into how nematode species are continually evolving, and why there needs to be a close examination of species in nematodes. Without this, nematode species diagnosis may be trickier than is apparent because of nematodes' small size, large numbers, rapid life cycles, patchy habitat and adaptability. What are termed species in nematodes matter for many different reasons, from biosecurity to pest management, and considering nematode species closely may also point to where research may be most usefully deployed to improve our knowledge of nematodes and how they interact with other organisms.

Keywords: Species - Evolution - Delineation.

Satellitome evolution illuminates complex species history and satellite DNA transcriptomes show coordinated expression in *Meloidogyne* nematodes.

Evelin Despot-Slade[1] (evelin.despot.slade@irb.hr), Saša Širca[2], Brankica Mravinac[1], Philippe Castagnone-Sereno[3], Miroslav Plohl[1], Nevenka Meštrović[1]

[1] *Ruđer Bošković Institute, Zagreb, Croatia*; [2] *Agricultural Institute of Slovenia, Ljubljana, Slovenia*; [3] *INRAe, CNRS, Université Côte d'Azur, Sophia Antipolis, France*

Satellitome represents the collection of different satellite DNAs (satDNAs) in a genome. SatDNAs are tandemly repeated non-coding DNA sequences that constitute the most abundant and the fastest evolving part of the eukaryotic genome. Due to their extreme diversity and methodological difficulties to characterize and compare satDNA collection in complex genomes, knowledge on their putative functional constraints and capacity to participate in genome evolution remains rather elusive. SatDNA transcripts have been detected in many species. New studies have shown that deregulation of satDNA expression has role in cancer progression. However, how satDNA transcripts are regulated on the genome-wide scale is poorly understood.

Here, we conducted a genome-wide survey and comparative analyses of satelitomes among different closely related *Meloidogyne* nematodes. The evolutionary trends of satDNA on the genome-wide scale show that each round of proposed polyploidization in the evolutionary history of the investigated species is concomitant with the addition of a new subset of satDNAs in the satellitome of any particular *Meloidogyne* species. Successive incorporation of new sets of satDNAs in the genome along the process of polyploidization suggests multiple hybridization events as the main factor in the formation of these species. In addition, comparative expression analyses of complete satDNA set show similar transcription profile of satDNAs among different *Meloidogyne* species which is not dependent on satDNA genome abundance indicating similar mechanisms that operate in regulation of satDNA expression in related genomes. Furthermore, stage-specific expression pattern of satDNAs through the *Meloidogyne incognita* development supports the view that satDNA expression is not a stochastic event but is subject to coordinated control. Comparative analyses of 83 satDNA sequences detected in *Meloidogyne* satelitomes revealed conserved sequence features indicative of evolution under selective pressure and their putative functional potential.

Our results demonstrate the power of comparative analysis of the non-coding genome part for the successful elucidation of the origin of species with a complex history. Whereas satDNAs generally evolve extremely quickly and their functional role is largely difficult to be proved, comparative analyses of satellitomes and satDNA transcriptomes in related genomes could be a way to address satDNA regulation and their putative functional potential.

Keywords: Repetitive DNA - Satellitomes - Phylogenomics - Satellite DNA transcriptome - Nematode.

Species diagnosis, hosts and distribution of cyst nematodes of the genus *Heterodera* (Tylenchida: Heteroderidae) with a focus on species of concern for Australia

Daniel C. Huston (manda.khudhir@csiro.au), Manda Khudhir, Mike Hodda

Insect & Nematode Biosecurity, CSIRO National Research Collections, Canberra, ACT, Australia

Cyst nematodes of the genus *Heterodera* include around 80 species of plant-parasites, a number of which are significant pests of important crops. Many species of *Heterodera* are differentiated by subtle morphological differences making diagnoses difficult. Here, we discuss diagnostics of these nematodes and describe our ongoing studies on the species richness, distribution, and host range of cyst nematodes in Australia, which includes putative endemics, potentially undescribed natives, and quarantine exotics. Because of the complexities of cyst nematode taxonomy, diagnostics is becoming increasingly reliant on molecular data. However, around 40% of species have no molecular data available, and sequences of some of the more commonly used gene regions routinely fail to distinguish between some species. We will discuss our analysis of publicly available barcoding data and the utility of these data for species diagnoses. We will also report on several new records of cyst nematodes for Australia, updated distributions for known species and our efforts to recollect a putative endemic. Lastly, we will report on our efforts to build a new reference collection for research on cyst nematodes. Although multiple species are listed as priority plant pests in Australia, few specimens are held in Australian collections. Many species of *Heterodera* will only be distinguishable with more data, especially regarding intraspecific and interspecific morphological and molecular variation, so we are seeking collaborators for collecting specimens to facilitate acquisition of said data, and ultimately to better define what is likely to make a diagnosable species in the genus.

Keywords: Species diagnosis - Evolution - Cyst nematodes - *Heterodera*.

New cyst nematode species from the Tepeaca Valley, Puebla, Mexico

Ilia Mariana Escobar Avila[1] (mariana_miss140@hotmail.com), Alejandro Tovar Soto[1], Sergei Subbotin[2, 3]

[1] *Laboratorio de Nematología Agrícola, Departamento de Parasitología, Instituto Politécnico Nacional, Escuela Nacional de Ciencias Biológicas, Mexico City, Alcaldía Miguel Hidalgo, Mexico*; [2] *Plant Pests Diagnostic Center, California Department of Food and Agriculture, Sacramento, California, United States*; [3] *Center of Parasitology of A.N. Severtsov Institute of Ecology and Evolution, Russian Academy of Sciences, Moscow, Russia*

The Tepeaca Valley in Puebla State (Mexico) is known as an important vegetable producing area. The aim of this study was to identify the cyst nematode species (CN) that parasitize horticultural crops in the Tepeaca Valley by morphological, morphometrical and molecular methods. Several nematological surveys and samplings were made from 2008 to 2016 in six localities of the Tepeaca Valley. Cysts and white females were collected from beetroot, broccoli and carrot roots. Cysts, second-stage juveniles, and males were extracted from each soil sample by Fenwick's can and centrifugal flotation methods [1,2]. Thirty specimens of each development stage were measured and genomic DNA was extracted from single cysts, ITS rRNA and partial *COI* genes were amplified and sequenced [3]. The CN parasitizing beetroot and broccoli was identified as *Heterodera schachtii*, and *H. carotae* in carrots. As for the cysts obtained from one of the soil samples, morphological and molecular analyses revealed that it belonged to a new species, which was described as *Cactodera solani*. Cyst nematodes are the second most important plant-parasitic nematodes worldwide but in Mexico they are poorly studied. In Mexico *H. schachtii* has only been reported in sugar beet but not in beetroot and broccoli, whereas *H. carotae* was not reported in this country. *Cactodera solani* is the first species of the genus *Cactodera* that parasitizes an important crop (tomato). Additional surveys must be made to determine the distribution of these nematodes in Mexican agricultural areas and implement control measures to stop the spreading of these species.

Keywords: Cyst nematodes - taxonomy - molecular biology - Mexico.

References:

- Fenwick, 1940. J. Helminthol. 18: 155.
- Jenkins, 1964. Plant Disease Reporter. 48: 692.
- Subbotin et al., 2018. Nematol. 20: 671-702.

S24-PF1

Morphological and molecular characterization of several *Paratylenchus* spp. from Belgium.

Phougeishangbam Rolish Singh[1] (phougeishangbamrolish.singh@ugent.be), Gerrit Karssen[2], Catherine Malike Etongwe[1], Marjolein Couvreur[1], Wim Bert[1]

[1] *Nematology Research Unit, Ghent University, Ghent, Belgium*; [2] *Wageningen Nematode Collection, National Plant Protection Organization, Wageningen, Netherlands*

Pin nematodes of the genus *Paratylenchus* spp., are globally distributed plant parasites of a wide range of plants including several economically important crops. However, morphological identification of *Paratylenchus* species is problematic due to low number of diagnostic features and high intraspecific character variability. In addition, up to four different *Paratylenchus* species can be found in a single soil sample augmenting the complexity of the species diagnosis problem. Relatively recently available sequence data, mostly 18S and 28S rDNA, provide a powerful tool for their identification. However, the vast majority of morphospecies remain unlinked to DNA sequences and a substantial part of the existing sequence data appears to be mislabeled.
In the present study, a survey of *Paratylenchus* from different locations in Belgium revealed more than ten different *Paratylenchus* populations. Four different DNA fragments; including partial 18S, 28S and ITS of rDNA, and COI of mtDNA were sequenced from every population and these molecular data were linked to comprehensive morphological data, which included both light and scanning electron microscopy. The resulting phylogenetic framework in combination with the morphological information revealed insufficient resolution of 18S rDNA for *Paratylenchus* species identification, while COI of mtDNA sequences appeared to be easy to generate and very informative in species delimitation. Furthermore, several undescribed species of *Paratylenchus* were discovered and a comprehensive morphological and molecular characterization of these species is provided.

Keywords: Belgium - COI - Morphology - *Paratylenchus* - rDNA.

S24-PF2

An update to the identification compendium of *Aphelenchoides* Fischer, 1894 (Aphelenchoidea).

Samira Fadakar (sm_fadakar@yahoo.com), Majid Pedram

Department of Plant Pathology, Faculty of Agriculture, Tarbiat Modares University, Tehran, Iran

The formerly developed species list and the identification compendium of the genus *Aphelenchoides* [1] was updated by including 60 species described after 1995, and species recently transferred to the genus from other genera. During the recent years, some species from other genera like *Tylaphelenchus* and *Laimaphelenchus* have transferred to *Aphelennchoides*, and some species formerly under *Aphelenchoides* have transferred to other genera like *Bursaphelenchus, Sheraphelenchus, Potensaphelenchus, Robustodorus, Tylaphelenchus, Laimaphelenchus* and *Ruehmaphelenchus.* Some species were also added to the genus, all sharing the same feature: a warty mucro at the tail terminus of females, well visible in scanning electron microscopy (SEM). This latter species group might deserve establishing a new morphospecies group, facilitating identification of species using classic criteria.

Keywords: Aphelenchoididae - Morphological - Morphospecies group - Tylenchomorpha.

References:

- [1] Shahina et al., 1996. Pakistan Journal of Nematology. 14:1-32.

S24-PF3

Unravelling the race complex: A first look Into the population genetics of the stem nematode *Ditylenchus dipsaci.*

Dennie te Molder (dennie.temolder@wur.nl), Stefan van de Ruitenbeek, Joost Riksen, Jaap Bakker, Geert Smant, Mark Sterken

Laboratory of Nematology, Wageningen University, Wageningen, Netherlands

The stem nematode *Ditylenchus dipsaci* is an obligate endoparasite of higher plants and is mainly found in the stems, bulbs/tubers, and leaves of its host. It is a cosmopolitan species mostly prevalent and damaging in temperate regions across the world. Stem nematodes are problematic in bulbous and tuberous crops/ ornamentals (e.g. union, garlic, narcissus, hyacinth, tulip, sugar-beet), but also in grains and legumes (rye, oat, maize, lucerne, field bean, clover), with host ranges extending to many weeds and green manure crops. The species has a long history, being among the first plant pathogenic nematodes to be documented, during which over 30 distinct 'races' have been described that together are estimated to infect over 500 different host species. *D. dipsaci* host races are morphologically indistinguishable, but exhibit highly variable – and overlapping – host ranges that are inconsistent with host phylogeny and only have a limited usefulness in prediction and management of disease. The current definition of *D. dipsaci* seems to describe a species complex which harbours among the most excessive intraspecific variation of any nematode species. To unravel this race complex we have started an ongoing effort to collect and catalogue a large number of *D. dipsaci* populations, which will be genetically characterised using whole-genome sequencing, a first for the species. So-far we have collected over 22 distinct populations and sequenced 13 of these. Furthermore, we have resequenced 2 populations after cultivation on a different host, to asses shifts in population composition. Early results indicate the existence of defined subclusters within *D. dipsaci*, giving some credit to the idea of races, although they might not be linked directly to the primary host. By presenting these results we hope to create awareness for the project and spark a wider interest in the species.

Keywords: *Ditylenchus dipsaci* - Stem nematode - Genomics - Population genetics - Adaptation.

ORAL SESSION 25

Metabolism & Physiology of nematodes and host plants
(continued)

Identification of a *Globodera rostochiensis* eggshell annexin and analysis of its potential role in the control of hatch.

James Price[1] (jp203@st-andrews.ac.uk), Terry Smith[1], John Jones[2]

[1] *University of St. Andrews, St. Andrews, United Kingdom*; [2] *James Hutton Institute, Dundee, United Kingdom*

Hatching of juvenile PCN occurs in response to host derived hatching factors. Little is known about the molecular mechanisms that underpin hatching in response to these host diffusates. Our work was aimed at developing a better understanding of PCN hatching mechanisms by characterising the protein and lipid components in the nematode eggshell.
We have developed methods that allow extraction of proteins and lipids from large numbers of isolated PCN eggshells. These extraction methods have permitted identification of a calcium dependent phospholipid-binding annexin in the eggshell of *Globodera rostochiensis*. The protein was further analysed due to the role that calcium plays in hatching of PCN. Antiserum raised specifically against this annexin confirmed that the protein was localised to the eggshells of *G. rostochiensis*. To our knowledge, that makes this annexin the first eggshell protein to be identified and localised in any plant-parasitic nematode. Recombinant annexin was used to test the predicted calcium and lipid binding interactions of this protein. A conformational change in the recombinant annexin occured in the presence of calcium that was modified by the presence of host root diffusates that stimulate PCN hatch. A transgenic potato line that expressed double-stranded RNA specific to the eggshell annexin was produced. RNAi data showed that reducing expression of the eggshell annexin gave rise to a phenotype suggesting an impact on the ability of the nematode to control the permeability of the eggshell. This included a change in hatching patterns in response to host hatching factors. These data suggested a direct place for this protein within the PCN hatching cascade.

Keywords: PCN - Eggshell - Annexin - Hatching - Proteomics.

Modulation of anaphase-promoting complex genes impacts root-knot nematode gall development.

Aline Kohn Carneiro[1] (alinekohn@hotmail.com), Kercia Siqueira[2], Danila Cabral[2], Nubia Eloy[3], Harald Keller[4], Paulo Ferreira[5], Adriana Hemerly[5], Janice de-Almeida Engler[4]

[1] *ISA/ID/LAB. C218, INRAE, Sophia Antipolis, France*; [2] *INRAE, Antibes, France*; [3] *ESALQ/Universidade de São Paulo, Piracicaba, Brazil*; [4] *INRAE, Sophia Antipolis, France*; [5] *Universidade Federal do Rio de Janeiro, Rio de Janeiro, Brazil*

Root-knot nematodes (*Meloidogyne* spp.) are obligate plant parasites capable of forming galls that are caused by a hyper activation and imbalance in the plant cell cycle. Both the mitotic cycle and endocycle machinery are modulated to establish a compatible plant/nematode interaction. The cell cycle is divided into four distinct phases (G1, S, G2, and M) and is a highly regulated process, in which CDKs and cyclins are major players driving mitosis and endocycle progression. The destruction of cyclins by the 26S proteasome allows G1/S and G2/M phase switches, and cyclins are marked for destruction by a very particular protein complex called the Anaphase-promoting Complex/Cyclosome (APC/C). In this work, we demonstrate that by inhibiting the 26S proteasome by the treatment of plants with MG132 had a significant impact on gall morphology, leading to the formation of giant cells with large vacuoles containing virtually no cytoplasm, finally leading to a delay in nematode development. It was also found that Arabidopsis plants overexpressing the C-terminal region of subunit seven of the APC/C (*APC7-CTOE*) were more resistant to nematode infection with a reduction in gall number with smaller sizes, and presented less egg masses compared with wild-type infected plants, as well for other APC subunits (*APC5OE*, *APC10OE*, and *APC7-FULLOE*) overexpressing lines. Reconstruction of 3D projections of *APC7-CTOE*gall nuclei by confocal microscopy illustrated small nuclei with lower ploidy levels. Infection of the *APC7-CTOE*line by the oomycete *Hyaloperonospora arabidopsidis* (Hpa) resulted in greater resistance suggesting a common resistance behaviour as seen for root-knot nematodes. Together, our data suggest that APC/C is an important player for cell cycle progression and gall homeostasis, and that ectopic expression of *APC7-CTOE*in Arabidopsis was effective in controlling two important plant parasites.

Keywords: Plant cell cycle - Proteolysis - Plant-nematode interaction.

References:

- De Almeida Engler J, Kyndt T, Vieira P, Van Cappelle E, Boudolf V, Sanchez V, Escobar C, De Veylder L, Engler G, Abad P, Gheysen G, 2012. Plant Journal 72 (2):185-198.
- De Almeida Engler, J., De Vleesschauwer, V., Burssens, S., Celenza, J.L. Jr, Inzé, D., Van Montagu, M., Engler, G. and Gheysen, G., 1999. Plant Cell 11 : 793-808.
- De Almeida Engler, J. and Gheysen, G., 2013. Mol. Plant-Microbe Int. 26: 17-24.
- De Almeida Engler J, Vieira P, Rodiuc N, Grossi de Sa MF, Engler G, 2015. Advances in Botanical Research, vol 73.
- Eloy NB, de Freitas Lima M, Ferreira PCG, Inzé D, 2015. Critical Reviews in Plant Sciences 34 (5):487-505.

Nematode derived modular metabolites and their functions in chemical ecology.

Stephan Von Reuss (stephan.vonreuss@unine.ch)

Laboratory for Bioanalytical Chemistry, University of Neuchâtel, Neuchatel, Switzerland

Chemical communication in nematodes has been known since the 1960's but the underlying molecular basis has remained elusive for many decades. Recent advances in analytical techniques, especially the development of highly selective and sensitive mass spectrometric screens, has enabled the identification of hundreds of glycolipids, the ascarosides, that contain the 3,6-dideoxysugar L-ascarylose connected to a homologous series of lipid-type aglycones that originate from the peroxisomal β-oxidation pathway of fatty acid metabolism. Ascaroside signaling is highly conserved in nematodes and involved in a diversity of intraspecies, interspecies, and even cross-kingdom interactions, demonstrating its major importance in nematode chemical ecology [1,2].
Comparative analysis of nematode metabolomes indicated that many of the simple ascarosides were conserved in a large variety of species. Aiming to elucidate the molecular basis for species-specific ascaroside signaling we characterized known and yet unidentified ascarosides in a diversity of bacterivorous *Caenorhabditis* nematodes using a combination of mass spectrometric (MS) and nuclear magnetic resonance spectroscopic (NMR) techniques along with total synthesis. Our results demonstrated how bacterivorous nematodes combine diverse strategies to generate species-specific ascaroside signals, including the hydroxylation of the fatty acid derived aglycones, epimerization of the L-ascarylose sugar unit, dimerization and oligomerization of conserved ascaroside building blocks, as well as the combinatorial assembly of additional building blocks from diverse primary metabolic pathways to generate complex modular libraries that form the molecular basis for chemical communication in nematodes.
Furthermore, our research demonstrated that the basic principle of modular assembly of canonical building blocks that is common for ascaroside biosynthesis, also extends to other classes of secondary metabolites to give rise to a plethora of nematode derived modular metabolites (the NDMMs), which are currently being explored with regard to their molecular structures and biological functions.

Keywords: Metabolism - Nematode derived modular metabolites - Glycolipids - Species-specificity - Chemical Ecology.

References:

- [1] Von Reuss, Schroeder, 2015. Nat. Prod. Rep. 32 (7): 994-1006.
- [2] Von Reuss, 2018. Chimia 72 (5): 297-303.

Tapping into signaling interactions between nematodes and aphids.

Jessil Ann Pajar[1, 2] (jessil_ann.pajar@idiv.de), Nicole M. van Dam[1, 3]

[1] *Molecular Interaction Ecology, German Centre for Integrative Biodiversity Research (iDiv) Halle-Jena-Leipzig, Leipzig, Germany*; [2] *Max Planck Institute for Chemical Ecology, Jena, Germany*; [3] *Institute of Biodiversity, Friedrich Schiller University, Jena, Germany*

In a natural environment, plants are challenged by several organisms that simultaneously feed on them. These may be organisms of diverse species with varied feeding strategies. Systemically induced plant responses may cause interactions between spatially separated herbivores. Below-aboveground interactions were found in naturally-occurring herbivores of the black mustard plant, *Brassica nigra*. It is simultaneously attacked by several genera of plant parasitic nematodes (PPNs) belowground and the aphid *Brevicoryne brassicae* aboveground, among several other insects. Aphid population growth was affected by the presence of different PPN species infecting the plant belowground. Previous studies show that aphid population growth increased on plants infested with root knot nematodes (*Meloidogyne* spp.), yet was reduced on plants infested by root lesion nematodes (*Pratylenchus penetrans*) or cyst nematodes (*Heterodera schachtii*)[1,2,3]. While this was recently attributed to nematode-induced phytohormonal signaling that interferes with aphid-induced response, a broad list of possible mechanisms are still yet unexplored.

Using both targeted and non-targeted metabolomics, we show that shifts in chemical defense profile of *B. nigra* in response to nematode infection suggestively determines aphid infestation success aboveground. Glucosinolates, the main defense compound in brassica plants, differed in profiles and concentrations in response to nematodes of sedentary and migratory lifestyles. *P. penetrans* (*Pp*) infection caused a systemic increase of 4-hydroxyglucobrassicin –a glucosinolate previously reported to have aphid-deterring properties [4]. This is consistent with the reduced number of aphids in *Pp*-infected plants compared to control plants. *M. incognita* (*Mi*) on the other hand posed defense compound profile that favored aphid infestation success.

Overall, our study suggests that PPNs encompass ecological influence beyond direct damage to host plants. As an economically important crop and a genetic repository of many commercially grown *Brassica* species, our model system will be of particular importance to address challenges on the presence of multiple herbivores affecting related agricultural crops.

Keywords: Chemical ecology - Herbivory - Plant defense - Insects - Metabolites.

References:

- [1] Hol et al. 2013. J Chem Ecol, 39(9), 1193-1203.
- [2] Hol et al. 2016. Front Plant Sci, 7, 111.
- [3] van Dam et al. 2018. Frontiers in Ecology and Evolution, 6.
- [4] Kim et al. 2008. The Plant Journal, 54(6), 1015-1026.

The effects of female pheromone exposure on lethal fighting in *Steinernema carpocapsae* males.

Maria Cassells (maria.cassells@mu.ie), Christine Griffin

Behavioural Ecology and Biocontrol, Biology Department, Maynooth University, Maynooth, Kildare, Ireland

Males of certain species of the entomopathogenic nematode *Steinernema* spp. engage in intraspecific lethal fighting. Males wrap around and compress their opponent, resulting in paralysis or death of the target. Previously, it was found that this behaviour was increased in the presence of a virgin (but not a mated) female of the same species, supporting the hypothesis that males fight for access to females. It was also previously found that males that had mated were more successful in terms of killing than naïve males when the two were paired in controlled fights, but the reason for that advantage was unclear (Kapranas et al., 2020). Interpreting the advantage of mated over naïve males is complicated, since encountering a female results in physiological changes including sperm development in *Steinernema* males. To investigate the reason for mated male advantage in *Steinernema carpocapsae*, we included a third category, males that had been exposed to female pheromone. We set up pairs of males from the same or different categories and recorded incidence of fighting, number and duration of fights, as well as survival at 24hrs. Pheromone-treated males developed sperm and were bigger than naïve males, but did not have an advantage over naïve males in asymmetric fights, therefore the fighting advantage of mated males cannot be explained by the physiological effects of female pheromone. The results of these experiments will be discussed in the context of male nematode reproductive and competition strategies.

Keywords: Male-Male competition - Reproductive biology - Behaviour.

References:

- [1] Kapranas et al., 2020. Animal Behaviour, (164) :149-154.

Calcium signaling events modulate host immune responses during parasitic interactions.

M. Shamim Hasan[1] (shamim.hasan@uni-bonn.de), Chhana Ullah[2], Sakil Mahmud[3], Clement Pellegrin[4], Sharon Letia[1], Mirosław Sobczak[5], Ute C. Vothknecht[3], Sebastian Eves-van den Akker[4], Florian M.W. Grundler[1]

[1] *Molecular Phytomedicine, University of Bonn, Bonn, Germany*; [2] *Department of Biochemistry, Max Planck Institute for Chemical Ecology, Jena, Germany*; [3] *Plant Cell Biology, University of Bonn, Bonn, Germany*; [4] *Department of Plant Sciences, University of Cambridge, Cambridge, United Kingdom*; [5] *Department of Botany, Warsaw University of Life Sciences, Warsaw, Poland*

Calcium (Ca^{2+}) is a central second messenger that perceives pathogen-associated molecular patterns (PAMPs)-derived signals via cell surface-localized immune receptors to establish appropriate immune responses in plants. Each recognition event is encoded into Ca^{2+} signatures that are sensed by multiple intracellular Ca^{2+} binding proteins and decoded to distinct downstream responses, usually through transcriptional reprogramming. However, very little is known about the Ca^{2+}-mediated signaling responses during early stages of plant-nematode interactions. Here we show that host root infection by a parasitic cyst nematode induces cytosolic Ca^{2+} and activates transcription of several proteins involved in calcium sensing and defence responses. Our previous work suggested that the expression of several Ca^{2+}-signaling responsive genes in *Arabidopsis* roots is significantly upregulated at the early stage of nematode infection. In the present work, analysis with genetically encoded biosensors revealed a cytosolic Ca^{2+} burst during the early nematode infection process. This is the first example of nematode induced Ca^{2+} signatures recorded for nematode parasitism. A comprehensive microscopic, transcriptomic, biochemical, and genetic analysis reveals an inter-connection of Ca^{2+}-signalingnetwork and modulation of plant defence hormones during the early interaction of plant-nematode parasitism. Our findings provide new insights into the mechanisms of Ca^{2+} signaling-mediated molecular responses in plant-nematode interactions.

Keywords: *Arabidopsis* - Sedentary endoparasitic nematodes - Calcium sensory protein - Phytohormone - Immune response.

Silencing a new female-specific multi-gene family of *Pratylenchus penetrans* can reduce nematode propagation.

Cláudia Vicente[1, 2] (cvicente@uevora.pt), Jordana Branco[1], Margarida Espada[1], Manuel Mota[1], Paulo Vieira[3]

[1] *NemaLab, Mediterranean Institute for Agriculture, Environment and Development, MED, Évora, Portugal*; [2] *NemaINIAV, Instituto Nacional de Investigação Agrária e Veterinária, Oeiras, Portugal*; [3] *Mycology & Nematology Genetic Diversity & Biology Lab, United States Department of Agriculture (USDA), Beltsville, United States*

Ranking third among the most economically important plant-parasitic nematodes (PPNs)[1], root lesion nematodes (RLNs) from the genus *Pratylenchus* represent a significant limitation to global food security. Among the more than 70 species described so far for this genus, *Pratylenchus penetrans* is considered one of the most economically important species due to its cosmopolitan geographic distribution and severe impact in numerous important food crops (e.g., corn, potato and several fruit trees), as well as on ornamental plants (e.g., lily)[2,3]. Depending on each species, RLNs can reproduce by parthenogenesis or sexual dimorphism. *P. penetrans* is a sexual dimorphism species, which needs both females and males for reproduction. Investigation of the molecular reproductive processes in PPNs can have important implications for understanding fundamental aspects of nematode development/reproduction (e.g., gametogenesis, female-male attraction), but can also be seen as an opportunity to develop novel approaches for nematode control, through the interruption of the nematode life cycle and consequent propagation. In this work, we annotate and analyze a new multi-gene family of *P. penetrans.* During data mining of the transcriptome of *P. penetrans* [4, 5] we identified 11 transcripts coding for 7 different proteins without any functional prediction or annotation. Blast analyses suggested that this family of genes is (so far) specific to *P. penetrans.* A total of 6 DNA and 4 cDNA clones were sequenced, validating our *in silico* analyses. All genes encode putatively secreted proteins without any transmembrane domains. Gene expression was detected only in adult females, while *in situ* hybridization supported their female-specific localization along the vulva region of *P. penetrans*. RNAi gene silencing assays of this gene family resulted in 50% reduction of the total number of nematodes 6-weeks after plant infection, suggesting an important role of this family for nematode reproduction.

Funding: This work is funded by National Funds through FCT - Foundation for Science and Technology under the Project PTDC/ASP-PLA/0197/2020.

Keywords: Root lesion nematodes - Parasitism - RNAi - *In situ* hybridization.

References:

- [1] Jones et al. (2013). Mol Plant Pathol. 14:946-961.
- [2] Esteves et al. (2015) Eur J Plant Pathol. 141:397-406.
- [3] Rusinque, Vicente et al. (2020) Plant Disease Notes DOI: 10.1094/PDIS-03-20-0524-PDN.
- [4] Vieira et al. (2015) PLoS ONE 10: e0144674.
- [5] Vieira et al. (2018) MPP. 19: 1887-1907.

ORAL SESSION 26

EPN commercialization and application

Conservation biocontrol with entomopathogenic nematodes: biotic and abiotic factors driving its potential.

Raquel Campos-Herrera (raquel.campos@icvv.es)

In-Vid, ICVV-CSIC, Logroño, La Rioja, Spain

Conservation biological control (CBC) seeks to enhance the presence and efficacy of natural enemies by combining actions that favor their activity and limiting those harmful for them. Entomopathogenic nematodes (EPNs) are naturally present in agricultural soils, making them perfect candidates for CBC approaches. Unraveling the biotic and abiotic factors that affect their activity as well as the agricultural practices that modulate the EPN soil food web is key to implement a successful CBC. The evaluation of EPNs and selected soil organisms using traditional and molecular tools combined with the study of abiotic factors in various agroecosystems provided insight into potential use in CBC. Perennial systems were shown to be suitable for the CBC using EPNs, although their success depended on the specific scenario. For example, Florida citrus grove soils were characterized by the presence of belowground pest *Diaprepes abbreviatus*. In these groves, the EPN diversity was driven by variables associated with water content, which also modulated the soil food web assemblage. However, the introduction of novel cultural practices to fight against huanglongbing altered the soil food web in ways that decreased the potential CBC by EPNs and increased the severity of another critical pest–disease complex. On the other hand, annual crops subjected to changes every season without a stable rhizosphere might be less effective for CBC by EPNs. For example, in Switzerland, the EPN natural occurrence in various long-term field experiments involving annual crops (mainly maize and wheat) was low compared with their presence in natural areas. In these crops, the low numbers of EPNs and high numbers of certain natural enemies implied that CBC would not be satisfactorily suppressive against root-feeding pests. Finally, the introduction of ecological structures might help to support the EPN occurrence, but with a distinct effect depending on the crop. For example, in Swiss annual crops, the cover crops did not enhance the presence of natural EPNs, while a study conducted in Spanish vineyards concluded that spontaneous cover crops planted within lines favored the presence of EPNs compared with traditional tillage practice. Therefore, to make CBC by EPNs a reality it is necessary to advance our knowledge of the multitrophic interactions in the soil affecting EPN efficacy and to disentangle the best habitat modification that supports EPN activity under varying scenarios.

Keywords: Conservation biological control - Entomopathogenic nematodes - Natural occurrence - Soil food web - Ecological structures.

Host searching behavior of native and introduced entomopathogenic nematodes for control of *Aegorhinus superciliosus* (Coleoptera: Curculionidae) in blueberry crops.

Patricia Navarro[1] (patricia.navarro@inia.cl), Almendra Monje[1], Graciela Berrios[1], Ivonne Alvarez[1], Camila Herrera[2]

[1] *INIA Chile, Carrillanca, Chile*; [2] *Giddings Farm, Purranque, Chile*

The Raspberry weevil is the most important native pest affecting berries in Chile. The damage is caused by the larva which is located in the root, where insecticides cannot reach the target. The use of entomopathogenic nematodes (EPNs) is a promising tool because their ability to find the host, however commercial applications of *Steinernema feltiae* (introduced nematodes) have shown low efficacy of larval control. In this study two native species of EPNs *Steinernema unicornum* and *S. feltiae* (INIA strain) and two introduced *S. feltiae* (K strain) and *S. feltiae* (B strain) were evaluated to determine their ability to find and kill *A. superciliosus* larva, under laboratory and field (sarsaparilla crop) conditions. The efficacy of the EPN species was evaluated at 4, 13 and 20 CO. Horizontal movement was evaluated at 1,2 and 3 cm length every 10, 20 and 30 minutes, and vertical movement was evaluated at 5, 10, 20, 30 and 40 cm depth after 72h IJs infection. Data were analyzed using ANOVA. Under laboratory conditions all species showed 40% mortality aprox at temperatures ranging between 15-20^{0}C. The relation vertical/horizontal movement was 5-10 cm/low for *S. unicornum*, 5 cm/high for *S. feltiae* (INIA strain), 5-20 cm/low for *S. feltiae* (K strain), and 20 cm/low for *S. feltiae* (B strain). None of the evaluated species showed nictation behavior. Results under field conditions showed 0% mortality for all species evaluated at 10^{0}C of soil temperature, time of the year that matches with larvae availability. In conclusion *S. unicornum* and all strains of *S. feltiae* evaluated were poor candidates for larval control of *A. superciliosus*.

Keywords: *S. unicornum* - *S. feltiae* - *Aegorhinus superciliosus* - Efficacy.

Entomopathogenic nematodes for insect pest management: progress and prospects for commercialization in South East Asia.

Nanette Hope Sumaya[1] (nanettehope.sumaya@g.msuiit.edu.ph), Prakaijan Nimkingrat[2]

[1] *Department of Biological Sciences/PRISM Biodiversity Laboratory, MSU-Iligan Institute of Technology (MSU-IIT), Iligan, Lanao del Norte, Philippines*; [2] *National Biological Control Research Center (NBCRC) Northeast Region, Khon Kaen University, Khon Kaen, Thailand*

Entomopathogenic nematodes (EPNs) from the two genera *Steinernema* and *Heterorhabditis* are obligate parasites and are widely used as biological agents against various insect pests. They possess unique combinations of traits making them promising biopesticides. To date, more than 90 and 20 species of *Steinernema* and *Heterorhabidtis,* respectively have been described worldwide. Despite this number, only very few species are produced commercially and these species also receive a profuse attention for research. In some developed countries, progress is made towards increasing efficacy through breeding programs using classical and molecular approaches to improve the EPN's unique attributes. Additionally, studies are focused on the improvement of methods of production, formulation, delivery and application in the field. However, in developing countries like in South East Asia (SEA), the use of EPNs either local or imported products are not yet prevalent. In fact, only 5 out of 11 countries in SEA *viz* Thailand, Philippines, Malaysia, Indonesia and Vietnam have existing and published EPN studies (local surveys, characterization and virulence tests). Researchers in other SEA countries like Brunei, Cambodia and Laos have received training on EPN isolation, identification and production. With a strengthened agricultural sector, only Thailand commercialized *S. siamkayai* and *S. carpocapsae* for control against pests like flea beetles, *Spodoptera exigua, Plutella xylostella, Cossus* sp. and *Microchlora* sp. In the Philippines, there is an ongoing feasibility study for potential EPN markets. To further strengthen the use of EPNs, an increased effort and a robust support from the government and private sectors are required. Also, the contribution of policies on the regulation of EPN products to SEA countries and issues that influence import regulations must be addressed. Should we choose to directly import and use commercially available EPNs from Europe for instance? Enhanced EPN products with extended shelf-life and increased tolerance to environmental stress like desiccation and heat plus higher infectivity must be produced. The use of local EPNs on the other hand is still limited due to several factors. Potential non-target effects, and the utilization of commercial or local EPNs should be comprehensively taken into account. These and other strategies will contribute to the expanded and successful use of EPN-based products in SEA countries.

Keywords: Biocontrol agents - EPN application - Importation - Regulation - South East Asian countries.

Evaluating locally isolated entomopathogenic nematodes against *Thaumatotibia leucotreta* in laboratory bioassays.

Francois du Preez (fdupreez@sun.ac.za), Antoinette Malan, Pia Addison

Department of Conservation Ecology and Entomology, Stellenbosch University, Stellenbosch, South Africa

False codling moth (FCM), *Thaumatotibia leucotreta*, is a major pest of citrus, and an important pest of stone fruit, pomegranates and table grapes in South Africa. It is of significant concern to export markets due to its phytosanitary status and invasion risk. Different from some other tortricid moths, final instar larvae exit the fruit and drop to the soil to pupate. This provides an opportunity to treat the soil with biopesticides, such as with entomopathogenic nematodes (EPN). In this study, locally isolated EPN species were evaluated for their biological control potential against FCM larvae and pupae, using laboratory bioassays. Insects were sourced from a local mass production facility, while nematodes were obtained from the collection kept at Stellenbosch University. *Heterorhabditis* species were cultured *in vivo*, while *Steinernema* species were cultured using an *in vitro* liquid culture technique. Insects were placed in alternate wells of 24-well bioassay trays, with a total of 60 insects per treatment. Larvae and pupae received 50 IJs and 200 IJs each, respectively, after which they were incubated for 48 h. Mortality by EPN infection was confirmed diagnostically or by dissection. Treatments were repeated at least twice. At a dose of 50 IJ/larva, *Heterorhabditis noenieputensis* performed the best (100%), followed by *H. zealandica* (green variant) (97%), *Steinernema yirgalemense* (93%) and *H. indica* (89%). Treatments were considerably less effective against pupae, with the best performing species being *H. baujardi* (28%), *H. noenieputensis* (26%), *H. indica* (23%) and *H. zealandica* (blue variant) (23%). The EPN species with the best potential for mass production and insect control were selected for dose-response assays, using a log-dose probit model. For larvae, LD_{50} estimates were calculated at 7.33 (*H. indica*), 14.55 (*S. yirgalemense*) and 47.76 (*S. jeffreyense*), while for pupae, they were calculated at 296.48 (*S. yirgalemense*) and 811.51 (*S. jeffreyense*), nematodes per insect. The potential of these EPN species to reproduce within FCM cadavers were also investigated. The results of this study will form the basis for upcoming field trials, and are part of a larger collaborative effort to develop local EPN species into a commercially-viable product in South Africa.

Keywords: False codling moth - Entomopathogenic nematodes - Integrated pest management.

Exosome-like vesicles are a key process of non-canonical protein secretion in *S. carpocapsae*.

Duarte Toubarro[1] (duarte.nt.tiago@uac.pt), Jorge Jorge[1], António Marcilla[2], Nelson Simões[1]

[1] *Centro de Biotecnologia dos Açores (CBA), Universidade dos Açores, Ponta Delgada, Portugal*; [2] *Dpto. Farmacia y Tecnología Farmaceutica y Parasitología Facultat de Farmacia, Farmacia Universitat de Valencia, Valencia, Spain*

Entomopathogenic nematodes are able to kill the host shortly after contact. Currently, the pathogenicity of these organisms is ascribed to excretory/secretory products (ESP), released by the infective nematode. In an attempt to identify the pathogenic effectors, we noticed the presence of exosome-like vesicles for the first time in *Steinernema carpocapsae*. Exosome-like vesicles (EVs) were isolated from the ESP by size-exclusion chromatography and the size distribution determined by nanoparticle tracking analysis (NTA). The purity and size of EVs were confirmed by TEM and the protein cargo analyzed by Ms-Ms. Based in the number of EVs counted per ml of ESP we calculated a production of 5.4 e+3 vesicles per IJ. Two groups of EVs were identified by NTA and TEM. One with a mean size of 146.7 ± 6.4 nm and a mode of 160.7 ± 3.1 nm corresponding to 85 % of total, and a second group of larger vesicles with a mean of 245 ± 5.2 nm and a mode of 271 ± 7.4 nm. TEM analysis of ultra-thin sections of infective stage showed a concentration of EVs below the cuticle layer along nematode lateral fields at the level of pharynx and mid-gut. However, future studies are needed to completely disclose the biogenesis of the vesicles population present ESPs. We identified 88 proteins in EVs including typical exosomal proteins like anoctamin (TMEM16) and ferlin (C2A-F) and neprilysin (CD10) and non-secretory proteins including cytoskeletal proteins, heat shock proteins, membrane transporters, membrane fusion proteins. Forty-six percent of proteins identified in vesicle cargo lacked the signal peptide thus evidencing the importance of vesicles as a process of "non-canonical" secretion in *S. carpocapsae*. Concerning the molecular function of EV proteins they were essentially divided into catalytic activity, binding activity and protease inhibitors. The catalytic group was mainly constituted by proteases, particularly serine proteases, aspartic and metalloproteases. In an "*in vitro*" assay we showed that EVs were internalized by hemocytes of *G. mellonela*, an insect susceptible to this nematode. Our findings revealed for the first time that exosomes are another mechanism by which EPNs interact with the host providing a way for the delivery of molecular effectors.

Keywords: *Steinernema carpocapsae* - Excreted secreted products (ESP) - Exosome-like vesicles - Internalization - Pathogenicity.

Diabrotica v. virgifera management using genetically improved strains of *Heterorhabditis bacteriophora*.

Ralf-Udo Ehlers[1] (ehlers@e-nema.de), Carlos Molina[1], Bart Vandenbossche[1], Stefan Toepfer[2]

[1] *e-nema GmbH, Schwentinental, Germany*; [2] *Plant Protection Directorate, CABI, Hodmezovasarhely, Hungary*

The Western Corn Rootworm (*Diabrotica virgifera virgifera;* Coleoptera: Chrysomelidae) is one of the most damaging pests of maize. Since the ban of neonicotinoid seed treatments most European farmers rely on the application of less effective granular soil insecticides. The sustainable, non-toxic alternative, the entomopathogenic nematode, *Heterorhabditis bacteriophora*, has been tested for a decade in plot trials with maize plants artificially infested with insect eggs to ensure an even population density. In addition, field trials using standard farming machineries were conducted in different European countries. Commercial nematodes application was at $2x10^9$ ha^{-1} with 200 l water ha^{-1} into the furrow together with the maize seeds, using special injectors mounted on the single-seed drilling machine. At the time of application, *Diabrotica* eggs were still in diapause. Nematodes survived and remained virulent until larvae hatch approximately 2-6 weeks later. With this precise fluid application method, nematodes achieved a mean reduction of the pest population of 65% (ranging from 33-82%) and outperformed results obtained with the chemical standards in 11 of 16 trails. Frequent nematode applications led to a reduction of the insect population. More virulent and persistent *H. bacteriophora* lines were obtained through genetic improvement using classical breeding technology. An improved line performed better than the commercial strain, justifying a reduction of the application density from 2 to $1x10^9$ ha^{-1} and bringing application costs into the range of synthetic chemicals.

Keywords: Maize - Dianem - Application technology - Breeding.

S26-PF1

Efficacy of species mixtures of entomopathogenic nematodes against different larval stages of cockchafers.

Bart Vandenbossche[1] (b.vandenbossche@e-nema.de), Mike Barg[1], Helge Postel[2], Edith Ladurner[3], Mauro Piergiacomi[3], Ralf-Udo Ehlers[1]

[1] *e-nema GmbH, Schwentinental, Germany*; [2] *e-nema GmbH, Klausdorfer Straße 28-36, Germany*; [3] *Area Tecnica Biogard, CBC (Europe) Srl, Cesena, Italy*

In a laboratory experiment the efficacy of entomopathogenic nematodes against third instar larvae of the common cockchafer (*Melolontha melolontha*) was assessed. The experiment comprised of *Heterorhabditis bacteriophora* alone and in combination with either *Steinernema feltiae* or *S. carpocapsae* at a nematode concentration of 50 dauer juveniles (DJs) per cm^2. Highest infection (76 %) of white grubs was obtained with the combination of *H. bacteriophora* and *S. carpocapsae* rather than with the combination of *H. bacteriophora* and *S. feltiae* (59%) and *H. bacteriophora* alone (57%). In a second lab experiment, efficacy was tested against first instars at a concentration of 50 DJs per cm^2. Highest mortality (75 %) of first stage larvae was obtained for a combination of *H. bacteriophora* and *S. carpocapsae* followed by *H. bacteriophora* and *S. feltiae* (59%) and *H. bacteriophora* alone (58%). Control mortality of white grub larvae was 25%. In a subsequent field test, the combination *H. bacteriophora* (500.000 DJs/m^2) and *S. carpocapsae* (250.000 DJs/m^2) was tested in an apple orchard in Italy. Efficacies calculated based on recovered white grub larvae in nematode and control plots were 81, 24 and 19% for first, second and third instar white grub larvae, respectively.

Keywords: *Melolontha melolontha* - white grubs - *Steinernema* - *Heterorhabditis*.

S26-PF2

Early season use of *Heterorhabditis bacteriophora* increases strawberry yield in fields infested by the white grub *Temnorhynchus baal*.

Ibrahim Shehata[1] (Ib_shehata80@yahoo.com), Mostafa Hammam[2], Fahiem El- Borai[3], Larry Duncan[4], Mahfouz Abd-Elgawad[2]

[1] *Pests and plant protection, National Research Centre, Cairo, Egypt*; [2] *Plant Pathology Department, National Research Centre, Cairo, Egypt*; [3] *Gulf Coast Research and Education Center, University of Florida, Florida, USA*; [4] *Citrus Research and Education Center, University of Florida, Florida, USA*

The importance of safe methods to manage strawberry pests has become more evident with increasing strawberry production and export in Egypt. Root herbivory by white grubs (*Temnorhynchus baal* larvae) inflicts severe damage to the growing plants, causing wilt and eventually plant mortality. Growers often rely on a few selective chemical insecticides to control *T. baal* larvae, minimizing pesticide residues in order to comply with maximum residue limits for export. Entomopathogenic nematodes (EPNs) that invade and kill grubs in the soil may be as effective as insecticides in some cases. We evaluated the profitability of mulching strawberry with commonly used cow and/or chicken manure, with and without application of commercial *Heterorhabditis bacteriophora* on two farms for three consecutive seasons. All plots did not receive chemical fertilizers. Timing of EPN application varied due to import regulatory guidelines. All manure mulch treatments increased strawberry yield each year at a farm in Al-Qalyubia governorate, while having no effect on yield at the farm in El-Beheira governorate. Compared to EPN-treated plots, 70% more (P=0.06) insects were recovered in soil beneath dead plants in plots that did not receive EPN. *Heterorhabditis bacteriophora* increased fruit yield ($P < 0.05$) in the second year at the El-Beheira farm and in all three years at the Al-Qalyubia farm. Mulches had no measurable effect on EPN performance. The greatest yield enhancement by EPN occurred in the second year when the nematodes were applied shortly after planting (October), enabling early season control of the pest.

Keywords: Strawberry yield - Entomopathogenic nematodes - Mulching - Egypt.

S26-PF3

The undetectable killer: *Steinernema carpocapsae* avoid recognition when infecting *Drosophila suzukii* larvae.

Anna Garriga[1, 2] (anna.garriga.oliveras@uab.cat), Duarte Toubarro[1], Nelson Simões[1], Ana Morton[2], Fernando García-del-Pino[2]

[1] *Centro de Biotecnologia dos Açores, Universidade dos Açores, Ponta Delgada, S. Miguel - Açores, Portugal*; [2] *Unitat de Zoologia, Universitat Autònoma de Barcelona, Bellaterra, Spain*

Drosophila suzukii is a pest of global concern in which several biological control agents are being tested. Among these, parasitoid eggs were strongly encapsulated by *D. suzukii* larvae [1] while entomopathogenic nematodes (EPNs) achieved high infection rates without an apparent immune response. For this reason, to assess the activation of humoral and cellular mechanisms through variations of gene expression, larvae were infected with the complex *Steinernema carpocapsae – Xenorhabdus nematophila*. Responses were evaluated in three time points of the infection course to determine the impact of nematode entry (90 min), establishment into hemocoel (4h) and bacterial release (14h) through the analysis of 16 immune-related genes with q-RT-PCR. The results revealed that at 90 min and 4 h none of the analysed genes presented variations compared with non-infected larvae, thus, the larval immune system failed to recognize nematode presence. Nevertheless, after *X. nematophila* were released into hemolymph, Toll and Imd pathways were activated leading to an upregulation of antimicrobial peptides. However, genes belonging to Jak/STAT and pro-phenoloxidase pathway remained invariable to the presence of both pathogens even at 14 h of infection. These results evidenced a lack of activation of cellular and melanization response which permitted an undetected and successful infection. These data with previous physiological observations on phenoloxidase, antimicrobial peptide activity, phagocytosis, and encapsulation ability of infected larvae [2], provide a clear understanding on nematode-bacterial ability to modulate the defense response of *D. suzukii*.

This study was supported by the projects FEDER (AGL2017-86770-R) and FCT (PTDC/BAA/30609/2017).

Keywords: Immune response - *Drosophila suzukii* - Gene expression - *Steinernema* - *Xenorhabdus*.

References:

- [1] Kacsoh BZ, Schlenke TA (2012). PLoS One 7. Doi: 10.1371/journal.pone.0034721
- [2] Garriga A, Mastore M, Morton A, García-del-Pino F, Brivio MF (2020). Insects 11. Doi: 10.3390/insects11040210

ORAL SESSION 27

Next-generation nematicides

Reklemel™ active: a novel highly selective nematicide.

Tim Thoden[1] (tim.thoden@corteva.com), Marta Garcia[2], John Wiles[3]

[1] *Corteva Agriscience, München, Germany*; [2] *Corteva Agriscience, Indianapolis, United States*; [3] *Corteva Agriscience, Cambridge, United Kingdom*

Plant-parasitic nematodes remain a significant threat and source of yield reduction in numerous important crops around the world. Over the last two decades many synthetic nematicides used for protection against these soil dwelling pests have come under significant regulatory as well as public pressure due to a range of toxicological and environmental issues. In response, the crop protection & seed industry has initiated an intensive effort directed at the discovery and development of new biological and chemical nematicides as well as native and transgenic traits in crops. Reklemel active, developed by Corteva Agriscience, is a selective molecule for the control of plant-parasitic nematodes with a favourable toxicological and ecotoxicological profile. It is the first member from the novel chemical class of sulfonamide nematicides. Commercial formulations primarily include a liquid suspension concentrate (Salibro™, 500SC), with granular formulations also under development for certain markets around the world. Salibro™ has been extensively tested in laboratory, greenhouse, micro-plot and field trials in North America, Latin America, Europe, Africa, the Middle East and Asia. In those trials Salibro™ was proven to be extremely effective against a range of important plant-parasitic nematode species. An introduction to the discovery of Reklemel, its chemical and biological properties as well as an overview of its field performance in key crops will be presented.

Nematicidal or nematistatic? New insights into the mode of action of fluopyram in plant-parasitic nematodes.

A. Sylvia S. Schleker[1] (sylvia.schleker@uni-bonn.de), Marc Rist[2], Christiane Matera[1], Arunas Damijonaitis[2], Katja Twelker[2], Corinna Saalwaechter[2], Sven Geibel[2], Oliver Gutbrod[2], Svend Matthiesen[2], Koichi Matsuoka[1], Samer S. Habash[1], Ute Schlee[1], Florian M. W. Grundler[1]

[1] *Molecular Phytomedicine, University of Bonn, Bonn, Germany*; [2] *CropScience Division, Research & Development, Bayer AG, Monheim am Rhein, Germany*

Plant parasitic nematodes (PPNs) cause severe yield losses in agricultural crop production worldwide. This highlights the need for efficient control which is challenging as potent means are rare. Recently, it was discovered that soil applications of the succinate dehydrogenase (SDH) inhibitor fluopyram can be effective against PPNs. Here we show that fluopyram selectively binds to the target in nematodes but not in mammals and insects. Adenylate energy charge values clearly indicated that fluopyram impairs ATP generation in *Meloidogyne incognita*, *Heterodera schachtii* and *Caenorhabditis elegans*. Our investigations revealed that the compound caused paralysis of *M. incognita* and *H. schachtii* second stage juveniles (J2s) as well as *C. elegans*. Although transient exposure to micromolar concentrations of fluopyram was nematicidal for *M. incognita* J2s, *H. schachtii* J2s completely recovered even at about a 100 times higher concentrations of the active substance. Accordingly, Fluopyram efficiently reduced gall formation on lettuce after pre-incubation of *M. incognita* J2s with the compound, whereas pre-incubation of *H. schachtii* J2s was not effective in inhibiting nematode parasitism of *Arabidopsis thaliana*. Sequence comparison of the target protein highlights an unique amino acid exchange within subunit C of the *H. schachtii* SDH. In line with the above observations, a *C. elegans* strain with a point mutation at this site was far less sensitive towards fluopyram than the respective wildtype. Further, docking studies indicated an influence of this amino acid mutation; a biochemical study for validation is currently ongoing. Although fluopyram was only nematistatic for *H. schachtii* J2s, permanent contact of the nematodes with the compound in a micromolar to nanomolar range completely prevented nematode infection of *A. thaliana* or considerably reduced nematode development at the root in a concentration dependent manner.

Keywords: Nematode control - Fluopyram - Mode of action - Root-knot nematode - Cyst nematode.

NIMITZ®, efficacious, safer and selective nematode management tool: technical review.

Bruno Lovato[1] (bruno.lovato@adama.com), Pablo Navia[2], Morten Boisen[3]

[1] *Product Development, Adama Agricultural Solutions, Schaffhausen, SH, Switzerland*; [2] *Product Development, Adama Agricultural Solutions, Raleigh, United States*; [3] *Product Strategy, Adama Agricultural Solutions, Schaffhausen, SH, Switzerland*

Plant-parasitic nematode (PPN) are key issues globally because they cause significant yield reduction in many crops around the world and, consequently, result in large economic losses. The increasing regulatory pressure on traditional nematicides, which eventually leads to severe restrictions and bans, has limited the number of chemical control alternatives to manage nematode populations in their crops for growers around the world.
Fluensulfone, discovered in 2001, launched a new era of chemical nematicides containing trifluoro (3-F) group in their molecular structures. Many publications around the world showed fluensulfone intrinsic potency against key PPNs species at low concentrations and its ability to cause permanent paralysis after 24-48 hours exposure. Along with its high intrinsic potency against PPNs, low toxicity against soil beneficial organisms including beneficial nematodes contributed to soil health and overall sustainability of food production systems. Recent results suggested that Fluensulfone has a unique mode of action on PPNs as it acted as fatty acid beta oxidation inhibitor that has a distinct profile of effects on PPNs compared to other nematicides.
More than one thousand field trials were conducted since the development in 2007 of Nimitz 480 EC and Nimitz 20 GR, both formulations containing fluensulfone as the active ingredient. More recently, external and independent field trials showed field efficacy of fluensulfone-based products against a wide range of nematode species and in several crops around the world. Results demonstrated the products' ability to reduce the severity of nematode damage, improve overall plant health and increase marketable yield when used following label instructions. Nimitz 480 EC and Nimitz 20GR are currently registered in 29 countries for use in several crops and represent an alternative in managing plant-parasitic nematodes in agricultural crops.

Keywords: Fluensulfone - Nimitz - Chemical control - Nematicide.

A Pipeline for the Discovery of Modulators of Neuromuscular Function with Potential Utility as Nematicidal Leads.

Sean Harrington[1] (sean.harrington@mail.utoronto.ca), Jacob Pyche[1], Jess Knox[2], Aaron Au[3], Inga Zasada[4], Ken-Loon Choo[5], Cassandra D'Amata[2], Mark Lautens[5], Peter Roy[2]

[1] *Dept. Pharmacology & Toxicology, University of Toronto, Toronto, Canada*; [2] *Dept. Molecular Genetics, University of Toronto, Toronto, Canada*; [3] *Institute of Biomaterials and Biomedical Engineering, University of Toronto, Toronto, Canada*; [4] *Agricultural Research Service, United States Department of Agriculture, Corvallis, United States*; [5] *Dept. Chemistry, University of Toronto, Toronto, Canada*

Nematode parasites of humans, plants and animals are a major barrier to human development posing a considerable threat to economic and food security. Plant-parasitic nematodes (PPNs) alone are estimated to account for >$100 billion/year in lost crop production globally [1]. Warranted regulation on the use of chemical control agents that are now understood to have environmental toxicity issues has beckoned the development of novel nematode control methods. A majority of chemical nematicides act through the disruption of neuromuscular function; this strategy is effective as even at sub-lethal concentrations neuromuscular disruption can inhibit parasitism through the inhibition of feeding, reproduction and host tissue attachment. The non-parasitic nematode *Caenorhabditis elegans* has proven useful for understanding the mechanism of many nematicides through its facile genetics and conserved biology with parasites [2].

Here, I present my work that identifies small-molecules that disrupt *C. elegans* neuromuscular function using a custom screening pipeline. One identified chemotype ('PR72') demonstrates activity against several animal and PPNs while lacking activity against non-target vertebrate models. With the Lautens lab (U-Toronto) we have generated a series of 40 PR72 analogs that demonstrate differential activity across free-living, animal and PPNs. We have demonstrated that several PR72 analogs are effective at inhibiting *Meloidogyne incognita* infection of tomato plant roots in mock soil trials (Inga Zasada group - USDA).

Mechanistic studies of PR72 have revealed that it induces *C. elegans* locomotor phenotypes through the agonism of neurotransmitter release pathways governed by calmodulin-dependent kinase II. Studies to understand the target of the PR72 are ongoing. Importantly, genetic resistance to PR72 cannot be generated in the lab suggesting that resistance is unlikely to develop in wild populations. It is our hope the PR72 family will prove useful to manage various PPNs.

Keywords: Nematicides - Small-molecule screening - Plant-parasitic nematodes - *Caenorhabditis elegans*.

References:

- [1] Abad et al., 2008. Nature Biotechnology, 26(1), 909–915.
- [2] Holden-Dye & Walker, 2011. WormBook, 1-29.

Sensitivity of *Pratylenchus vulnus* to Salibro™ nematicide *in vitro* and *in planta.*

Yu-Chen Wang[1] (yuchenw@ucr.edu), Tim Thoden[2], Zin Thu Zar Maung[1], Andreas Westphal[1]

[1] *Department of Nematology, University of California, Riverside, Parlier, United States*; [2] *Corteva Agriscience TM, Munich, Germany*

Root lesion nematode (*Pratylenchus vulnus*) is a migratory endoparasite affecting many plant species, with a wide range of distribution. It is particularly damaging in nut crop orchards in California. A large proportion of walnut and almond orchards are expected to be infested with this nematode. It is one of the main production limiting factors. Currently, it is effectively suppressed by preplant soil fumigation with 1,3-D-containing materials. Environmental and human health concerns make this approach non-sustainable, and frequently the protection does not cover the lifetime of the orchard. Alternative additional tools are needed. Reklemel™ is a novel selective and effective active substance (a.s.) in managing plant-parasitic nematodes [1,2] that will mainly be available as 500SC formulation (Salibro™). It was the objective of this study to determine the efficacy of Salibro in controlling *P. vulnus*. In *in vitro* studies, the impact of Salibro on the motility and vitality of *P. vulnus* was evaluated by exposing vermiform stages of the nematode to a concentration series of 0, 5, 20, 25, 50, 125, 250 ppm (a.s.). Batches of nematodes were incubated in these suspensions, and subsamples were evaluated after 24, 72, 120, and 168 hours of exposure. In in planta pot experiments, sandy loam soil was infested with either 2,774 or 3,779 vermiform of *P. vulnus* via infected walnut roots. Pots (2L) were drenched with 250 ml of an aqueous concentration series of 0, 5, 20, 25, 50, 125, 250 ppm (a.s.) of Salibro. One week after the treatments were applied, seedlings of peach rootstock 'Nemaguard' were transplanted. Plants were cultivated for two months when the plant tops were collected, and nematodes were extracted from the roots and soil. In the *in vitro* study after 24 hours exposure, Reklemel exhibited high toxicity to *P. vulnus*, with the average LC_{50} value of 40 ppm (a.s). After 168 hours, the average LC_{50} value was 7 ppm. In the greenhouse pot experiments, the soil drenches at 5 to 50 ppm (a.s.) reduced the numbers of extractable nematodes (both from roots and soil). Plant growth was unencumbered at these rates, but some stunting was observed at a concentration of 250 ppm (a.s.), which may be much higher than intended label rates. These results showed that Salibro has potential for managing root lesion nematode in almond production.

Keywords: Salibro - Root lesion nematode - Infectivity - Toxicity - Motility.

References:

- [1] Thoden and Wiles. 2019. Nematology 21:625-639.
- [2] Thoden et al., 2019. Nematology 21:889-893.

Biological Activity of TYMIRIUM™ molecule as a Soil- and Seed-Applied Nematicide.

Matthias Gaberthueel[1] (matthias.gaberthueel@syngenta.com), Shannon Morsello[2], Jon Hamill[2], Dale Ireland[2], Jeff Simmons[3], André Bachiega[4]

[1] *Syngnta Crop Protection AG, Basel, Switzerland*; [2] *Syngenta Crop Protection LLC, Greensboro, United States*; [3] *Syngenta Crop Protection LLC, Vero Beach, United States*; [4] *Syngenta Proteção de Cultivos Ltda, São Paulo, Brazil*

Plant-parasitic nematodes are hard to eradicate once a field site has been infested. The management of nematodes is very challenging and practices like crop rotation and cultural measures are difficult for growers to implement. A growing world population and a limited production area coupled with a demand for higher yields in existing production areas has led to an increased intensification which favours nematode multiplication. An efficient way to control nematodes is to apply a nematicide prior to or at sowing/transplanting. This is one of the key elements to protect the root development of plants in an early growing phase. A chemical nematicide can actively protect the young seedling in the first critical weeks of early establishment and secure crop yield. The newest active substance with outstanding nematicidal efficacy is TYMIRIUM™. TYMIRIUM™ molecule is highly active against a broad range of plant-parasitic nematodes and can be applied either as soil- or a seed treatment. During the presentation we will cover the activity of TYMIRIUM™ molecule in key crops against different genera of plant-parasitic nematodes and the benefit this new chemical nematicide can bring to growers.
TYMIRIUM™ is a trademark of Syngenta Group Company

Keywords: TYMIRIUM™ molecule - Nematicide - Nematodes.

S27-PF1

Selective control of parasitic nematodes using bioactivated nematicides.

Andrew R. Burns[1] (andy.burns@utoronto.ca), Rachel Ross[2], Megan Kitner[3], Jessica Knox[1], Jack M. P. Castelli[1], Kirsten Yeung[2], Emily Puumala[1], Bruna M. Palmeira[4], Elizabeth M. Redman[4], Sagar Marwah[5], Jens Tiefenbach[6], Jamie Snider[1], Constance A. M. Finney[7], Igor Stagljar[1], Henry M. Krause[1], Sonya A. MacParland[5], John S. Gilleard[4], Leah E. Cowen[1], Hui Peng[2], Susan Meyer[8], Mark Lautens[2], Inga Zasada[3], Peter J. Roy[1]

[1] *Department of Molecular Genetics, University of Toronto, Toronto, Canada*; [2] *Department of Chemistry, University of Toronto, Toronto, Canada*; [3] *Horticultural Crops Research Laboratory, USDA-ARS, Corvallis, United States*; [4] *Department of Comparative Biology and Experimental Medicine, University of Calgary, Calgary, Canada*; [5] *Deparment of Immunology, University of Toronto, Toronto, Canada*; [6] *In Danio Bioscience Inc., Toronto, Canada*; [7] *Department of Biological Sciences, University of Calgary, Calgary, Canada*; [8] *Genetic Diversity and Biology Laboratory, USDA-ARS, Beltsville, United States*

Parasitic nematodes are a major threat to global food security, particularly as the world amasses 10 billion people amidst limited arable land. Most traditional nematicides have been banned due to poor nematode-selectivity, leaving farmers with inadequate controls. Using the free-living nematode *Caenorhabditis elegans* as a small-molecule screening system, we have identified a family of selective imidazothiazole nematicides, called selectivins, that undergo cytochrome p450-dependent bioactivation exclusively in nematodes. At low parts-per-million concentrations, selectivins perform comparably well with commercial nematicides to control root infection by *Meloidogyne incognita* – the world's most destructive plant-parasitic nematode. Tests against a wide range of phylogenetically diverse non-target systems demonstrate that selectivins are more nematode-selective than nearly all marketed nematicides. Thus, selectivins are first-in-class bioactivated nematode controls that provide efficacy as well as much-needed nematode selectivity.

Keywords: *C. elegans* - Pro-nematicide - Bioactivation - Cytochrome p450 - *M. incognita*.

S27-PF2

The effect of 1-Octen-3-ol and 3-Octanone on plant parasitic nematodes.

Arben Myrta[1] (myrta@certiseurope.com), Nicola Sasanelli[2], Astha Gurung[3], Pasqua Veronico[2], Salim Khoja[3], Alberto Troccoli[4], Ian Baxter[1], Tariq M. Butt[3]

[1] *Innovation and Development, Certis Europe B.V., Utrecht, Netherlands*; [2] *Nematology, Institute Sustainable Plant Protection, Bari, Italy*; [3] *Swansea University, Swansea, United Kingdom*; [4] *Institute Sustainable Plant Protection, Bari, Italy*

The nematicidal effect of two volatile organic compounds (1-Octen-3-ol and 3-Octanone) was assessed under *in vitro* conditions against *Meloidogyne incognita*, *Pratylenchus thornei* and *Xiphinema index.* The assays were carried out in Petri dishes with 3.8% water agar. Each Petri was divided into 6 circles which served as nematode sampling zones. Each circle was inoculated with 20 ml of a water nematode suspension. The number of the juveniles was variable according to the tested nematode. A 22 x 22 mm glass slide holding a round piece of filter paper was placed in the center of the Petri dish lid. Different amounts (2.5, 5, 10 and 20 mL) of each VOC were deposited onto the filter paper. Petri dishes were closed and sealed with parafilm. Five replications were considered for each combination concentration x exposure time. Only one control (0 concentration and 5 replications) was used for both VOCs and for each nematode species. Survival and mortality of the nematodes was assessed after 45 min, 90 min, 3, 6, 12 and 24 h. The percentage mortality was corrected by eliminating the natural death in the control according to the Schneider Orelli's formula. Probit analysis was used to calculate lethal doses (LD50) for each exposure time (from 45 min to 24 h). For the root-knot nematode *M. incognita*, LD50 ranged from 10.1 to 0.2 mL and from 8.5 to 0.6 mL for 1-Octen-3-ol and 3-Octanone, respectively. *P. thornei* was less susceptible to the VOCs in comparison to *M. incognita* showing values of LD^{50} ranging from 50.8 to 0.01 mL for 1-Octen-3-ol and from 389 to 1 mL for 3-Octanone. The dagger nematode *X. index* showed a LD^{50} from 12.1 to 0.5 mL and from 2.1 to 0.2 mL for 1-Octen-3-ol and 3-Octanone, respectively. Therefore, 1-Octen-3-ol resulted more effective than 3-Octanone in the control of *M. incognita* and *P. thornei*. By contrast, 3-Octanone was more effective against *X. index*.

Keywords: Fungal volatiles - Metarhizium - *Meloidogyne* - *Pratylenchus* - *Xiphinema*.

References:

- Khoja S. et al,, 2019. Pest Management Science, 75: 3392-3404. DOI 10.1002/ps.5578.

S27-PF3

Impacts of long-term SDHI nematicide use on turfgrass.

Christian Kammerer (kammech@ufl.edu), William Crow

Entomology & Nematology, University of Florida, Gainesville, United States

This research investigated reported reductions in efficacy of Succinate Dehydrogenase Inhibitor (SDHI) nematicides on turfgrass nematodes. Enhanced microbial degradation and resistance were the two mechanisms investigated in this study. Small field plots received fluopyram for four years at the maximum labeled rate, fluopyram for one year at the maximum labeled rate, or remained untreated. Golf courses across the state of Florida were organized into two groups: those with no history of fluopyram use and those with a long history of fluopyram use. A biosaasy was used to test for enhanced microbial degradation. Turf cores were collected from the field plots and golf courses and treated with different rates of fluopyram. At specific time intervals, tomatoes were planted in the soil and then inoculated with either *Meloidogyne incognita* (small plot study) or *M. enterolobii* (golf course study). The tomato roots were harvested after four weeks, with the galls and egg masses being quantified. To test for resistance, *Meloidogyne graminis* from the field plots and golf courses were exposed to concentrations of fluopyram in vitro. For three days, movement and inactivity in response to fluopyram were measured at specified time intervals. This data was recorded before and after a stimulant was added to each sample to stimulate nematode movement. Results from the bioassay experiment do not indicate that enhanced microbial degradation of fluopyram is occuring. Results from the in vitro study, indicate that *M. graminis* from plots or golf courses with prolonged history of fluopyram use were less sensitive to fluopyram in vitro. This suggests that continuous fluopyram applications in the field can lead to reduced nematode sensitivity to the nematicide. The longevity of this nematicide in the environment is cause for concern, making resistance more likely. With the increased use of SDHI nematicides, special attention needs to be paid to prevent further efficacy loss and to preserve the usefulness of these chemistries.

ORAL SESSION 28

Interactions of nematodes with micro-organisms

Microbiota associated with phytonematodes in the rhizosphere of cultured plants.

Holger Heuer (holger.heuer@julius-kuehn.de), Ahmed Elhady, Shimaa Adss, Johannes Hallmann

Epidemiology and Pathogen Diagnostics, Julius Kühn-Institut, Braunschweig, Germany

Microbes and plant-parasitic nematodes simultaneously colonize the plant rhizosphere. Plants influence the microbiome in their rhizosphere and thereby eventually acquire microbes rendering the holobiont more resistant to plant-parasitic nematodes. The modified microbiome is passed on to the plant subsequently growing in the same soil. We investigated the effect of plant-soil feedback of different pre-crops rotated with soybean in order to suppress root lesion nematodes (RLN). Transplanting the rhizosphere microbiome from different crops resulted in different degrees of suppressiveness against RLN on soybean roots. The inoculated microbiomes from soybean, Ethiopian mustard, and maize significantly reduced the invasion of RLN compared to the microbiomes from bulk soil or tomato rhizosphere. In the analogous experiment with tomato plants and either RLN (*Pratylenchus penetrans*) or root-knot nematodes (*Meloidogyne incognita*), the microbiomes from maize and tomato reduced root invasion of both nematodes compared to the microbiomes from soybean or bulk soil. In a split-root experiment, the suppressive effect of the microbiome on *P. penetrans* was mediated by the plant and depended on the plant species from which the microbiome was transplanted. The fungal and bacterial communities attached to the surface of RLN significantly differed in those rhizospheres. This implied that attached microbes antagonized the RLN, directly and/or by signals to the plant. Engineering the plant associated microbiome through pre-, cover- or inter-crops may lead to eco-friendly crop protection.

Cultivating relationships: genetics shape microbiome form and function in *C. elegans.*

Buck Samuel (buck.samuel@bcm.edu)

Center for Metagenomics and Microbiome Research, Baylor College of Medicine, Houston, TX, United States

The gut microbiome extends the capabilities of its host and alters its physiology. Together with diet, host genetic landscapes shape microbiome form and function in the animal gut. Despite its importance, the essential functions that drive microbiome assembly and stability remain largely elusive. To address this challenge, we leveraged the nematode *Caenorhabditis elegans* to explore how microbiomes assemble in different host genetic backgrounds. This system has several advantages including: (i) a simple microbiome that can rapidly be removed (bleaching) and replaced in high-throughput gnotobiotic experiments; (ii) highly conserved intestinal physiology, metabolism and innate immunity; and (iii) shared microbial functions for host gut persistence [1,2].
To examine the natural variation in acquisition of the microbiome in *C. elegans*, we first established the natural core microbiome [1] and assembled a functionally redundant, model core microbiome of bacteria (BIGbiom) in the lab. Then a panel of 38 fully genome sequenced *C. elegans* wild strains were made 'germ-free' and colonized with BIGbiom to assess strain-level microbiome composition (16S) and levels (CFU) longitudinally using a high-throughput pipeline. The strains clustered tightly into three distinct groups: (i) a highly-selective group that differed greatest from the surrounding environment [*Ochrobactrum*-dominant]; (ii) a 'dysbiotic' group [*Bacteroidetes*-dominant, 30-fold more CFUs]; and (iii) a 'non-selective' group. All strains tested retain deterministic selection and/or control, the two programs for microbiome regulation. By GWAS-, RNAi- and RNAseq-based approaches, we identified ~1000 candidate regulators in highly conserved pathways (>60%). Insulin signaling pathways specifically regulate *Ochrobactrum* colonization, as impaired daf-2/IGFR signaling (mutants or RNAi of wild strains) limited its colonization.
Since we saw dramatic host-to-host differences in microbiome composition, we next sought to examine alterations in microbiome function. To do this, we sequenced and annotated >100 bacterial genomes [BIGbiom plus others in natural microbiome]. Metabolic reconstructions were generated for each *C. elegans* microbiome and indicated broad microbiome functions were shared, but also point to many emergent functions in each host groups. Our study highlighted the potential of a robust platform to identify conserved host and microbial determinants that may underlie assembly and stability of the microbiome.

Keywords: Microbiome - *C. elegans* - Genomics - Ecology.

References:

- [1] Zhang, Berg et al., 2017, Frontiers in Microbiology, 8, 485.
- [2] Samuel et al., 2016, PNAS, 113 (27), E3941-E3949.

Can microbiomes protect phytonematodes against antagonists?

Olivera Topalovic[1] (otopalovic@agro.au.dk), Frederik Bak[2], Susana S. Santos[1], Mette H. Nicolaisen[2], Flemming Ekelund[3], Mette Vestergård[1]

[1] *Department of Agroecology, Aarhus University, Slagelse, Denmark*; [2] *Microbial Ecology and Biotechnology, University of Copenhagen, Copenhagen, Denmark*; [3] *Department of Biology – Terrestrial Ecology, University of Copenhagen, Copenhagen, Denmark*

Root-knot nematodes (RKN) are among the most devastating plant parasites. The infective second-stage juveniles (J2s) of RKN reside in soil before invading the roots. While in soil, the J2s are exposed to a large pool of microorganisms. Many of these microorganisms are nemato-antagonistic and the J2s need to overcome them in order to survive. Some microorganisms have been shown to protect nematode species that do not parasitize plants. The modes of microbial protection range from suppressing the immune system of the host to fighting against the nematode pathogens. So far, there is no evidence that microbiomes protect phytonematodes seek protection from microbiomes when under attack by antagonists. To study if J2s of two major RKN species, *Meloidogyne incognita* and *M. hapla*, acquire protective microbiomes on their surface, we incubated J2s with soil microbial suspensions supplemented or not with *Pseudomonas protegens* strain CHA0. After three days, we recorded a decreased J2 performance in microbial suspensions from two soils, while the presence of *P. protegens* CHA0 alleviated this response. We then collected 100 live and 100 moribund J2s from each treatment to study the composition of bacteria and fungi associated with the nematode surface using amplicon sequencing of the 16S rRNA and ITS genes. Our data revealed that the live and dead J2s of both nematode species differed in the composition and relative abundance of surface-attached bacteria, while the attachment of fungi was not prominent. In addition, the presence of *P. protegens* CHA0 in microbial suspensions caused shifts in J2-attached microbiomes. Altogether, our study is a step forward in understanding the role of microbiomes in RKN protection against their antagonists, adding a completely new dimension in the field of RKN ecology and biocontrol.

Keywords: *Meloidogyne* - Nematode protection - Microbiomes - Soil - 16S rRNA / ITS gene.

Microbiome analysis of malacopathogenic nematodes suggests no evidence of a single bacterial symbiont responsible for gastropod mortality.

Laura Sheehy (l.sheehy@ljmu.ac.uk), James Cutler, Gareth Weedall, Robbie Rae

School of Biological and Environmental Sciences, Liverpool John Moores University, Liverpool, United Kingdom

Nematodes and bacteria are prevalent in soil ecosystems, and some have evolved symbiotic relationships. In some cases, symbionts carry out highly specialised functions: a prime example being entomopathogenic nematodes (EPNs), which vector bacteria (*Xenorhabdus* or *Photorhabdus*) into insect hosts, killing them to provide a food source for the nematodes [1]. It is thought that the commercially available malacopathogenic (kills slugs and snails) biocontrol nematode *Phasmarhabditis hermaphrodita* vectors the bacterium *Moraxella osloensis* into slugs to kill them [2][3]. To investigate this, we used a metagenomic approach to profile the bacteria present in the commercial strain of *P. hermaphrodita*, a wild strain of *P. hermaphrodita* and two other *Phasmarhabditis* species (*P. californica* and *P. neopapillosa*), after they had killed their slug host (*Deroceras reticulatum*). We show that these nematodes do not exclusively associate with one bacterium but a range of species, with members of the phyla Pseudomonadota, Bacillota, Actinobacteriota and Bacteroidota the most prevalent. The commercial strain of *P. hermaphrodita* had the least diverse bacterial community. Furthermore, we found that the bacterium *P. hermaphrodita* has been cultured on for 25 years is not the expected species *M. osloensis* but is actually *Psychrobacter spp.* and the only strain of the *Phasmarhabditis* species to associate with *Psychrobacter spp.* was the commercial strain of *P. hermaphrodita*. In summary, we found no evidence to show that *P. hermaphrodita* rely exclusively on one bacterium to cause host mortality but found variable and diverse bacterial communities associated with these nematodes in their slug hosts.

Keywords: Metagenomics - Nematodes - Gastropods - Symbiosis - Biocontrol.

References:

- [1] Campos-Herrera R. et al, 2015, Ecology and Applied Technologies for Sustainable Plant and Crop Protection – sustainability in Plant and Crop Protection. Springer International Publishing.
- [2] Tan L. et al, 2001, J Parasitol, 87 (6):1349-1354.
- [3] Tan L. et al, 2002, Appl Environ Microbiol, 68(8):3943-3947.

Nematodes rely on microbial facilitation in extreme environments.

Jesse Jorna[1] (jjgjorna@outlook.com), Byron J. Adams[2]

[1] *Department of Biology, Brigham Young University, Provo, UT, United States*; [2] *BYU Life Science Museum, Provo, UT, United States*

In the McMurdo Dry Valleys of Antarctica (MDV) nematodes and other microinvertebrates make up the top levels of the food chain, while the biotic function of soils is dominated by microbial taxa performing most biogeochemically important functions in the ecosystem. Abiotic factors have been considered as the drivers for community composition in these systems, yet as this simple ecosystem undergoes the effects of climate change and the resulting abiotic changes affect individual species and their interactions, the underestimated role of biotic interactions is slowly coming to light. A long-term experiment in the McMurdo Dry Valleys of Antarctica (MDV) was established to identify the biotic components of community assembly and primary succession in this purely microbial ecosystem. Soils containing viable microinvertebrate communites and with known abiotic conditions were sterilized by autoclave and replaced into their original environment to measure the rate and order of microbial and microinvertebrate succession. Microinvertebrates were extracted from soils after 1, 4 and 15 years and the microbial communities were analysed by shotgun metagenomics to reveal the functional groups present in soils over time and between samples.
Strikingly, both one and four years after sterilization no live microinvertebrates were found to have colonized the sterilized soils, and after 15 years only five out of eight plots showed nematode colonization at numbers much lower than neighbouring control plots. Previous experiments have shown that dispersal is not a limiting factor in MDV environments, even if most metazoans in these ecosystems require more than one year to complete a single life cycle. While microbial communities are shown to have recovered to some extent as measured by respiration of the treated soils even after a single year, the metagenomic results reveal which specific metabolic or environmentally crucial functions are currently lacking in the microbial community profile. The specific functional groups absent in uncolonized soils provides direct insight into the reliance of these microinvertebrates on microbial facilitation in an extreme environment.

Nematode–microbe complexes play an essential role in apple replant disease.

Xorla Kanfra (xorla.kanfra@julius-kuehn.de)

Julius Kuhn Institute, Braunschweig, Niedersachsen, Germany

Apple replant disease (ARD) is a severe problem in apple production worldwide. It is caused by a complex of yet unknown soil biota, leading to small discolorated roots, as well as increased biosynthesis of phytoalexins, total phenolic compounds, andantioxidants. We investigated the contribution of nematodes to ARD by dissecting the soil biota from soil infested with ARD and non-infested control soil into a nematode and a microbe fraction. Their separate and synergistic effect on ARD symptoms of susceptible M26 apple plants was analysed in greenhouse assays after inoculation. In three independent experiments, the combination of nematodes from ARD soil with microbes from ARD soil had the strongest adverse effect on the plants, concerning growth parameters of shoots and roots, total phenolic compounds, phytoalexins in roots, and antioxidants in leaves. Nematodes from ARD soil together with microbes from control soil, or together with the microbiome associated with their bodies, also had significant effects on the plants but less pronounced. Microbes from ARD soil alone, or in combination with nematodes from control soil, induced only minor ARD symptomsthat were mostly not distinguishable from those of plants inoculated with microbes from control soil or uninoculated. Overall, a highly significant effect of the source of the inoculated nematodes, ARD or control soil, on ARD symptoms and a strong synergistic interaction between ARD nematodes and microbes was revealed. Microbes from control soil also induced strong but delayed ARD symptoms when inoculated together with nematodes from ARD soil. Nematode communities significantly differed between ARD and control soil as revealed by high-throughput sequencing of 18S rRNA. Plant-parasitic nematodes were too low in abundance to explain root damage. In conclusion, exploring the associations of nematodes and microbes in ARD soils will give the chance to unravel the etiology of ARD.

Keywords: Nematode-microbe interaction - Apple replant disease - Phytoalexins - Malus domestica - Soil microbiota.

References:

- Kanfra et. al., 2021. Horticulturae 7(11) 433.
- Winkelman et. al., 2019. Curr. Issues Mol. Biol. 30: 89-106.
- Kanfra et. al., 2022. Microorganisms. 10 (1), 157.

S28-PF1

Soil-born endophytic fungi antagonize plant-parasitic root-knot nematodes in tomato.

Polina Marchenko (polina.marchenko@students.boku.ac.at), Tina Austerlitz, Krzysztof Wieczorek

Institute of Plant Protection, University of Natural Resources and Life Sciences, Tulln an der Donau, Austria

Plant-parasitic nematodes, especially sedentary root-knot nematodes (*Meloidogyne* spp.), are a serious threat to many crop plants causing worldwide tremendous economic losses. Due to the banning of most chemical nematicides, there is a strong need for new environmentally friendly alternatives. One such promising option is the utilization of endophytic fungi, such as different species of Sebacinales family. There are already auspicious results from various studies showing considerable positive impact of those fungi on plant growth, yield and tolerance/resistance against different biotic and abiotic stresses [1, 2]. Our previous *in vitro* studies indicate that *Serendipita indica* acts against cyst nematodes in a model plant *Arabidopsis* [3]. Therefore, the objective of this work was more applied investigation of the possible application of two Sebacinales fungi, *S. indica* and its domestic relative *Serendipita williamsii*, against the root-knot nematodes in tomato (*Solanum lycopersicum* L.). For that, we inoculated tomatoes with *M. incognita* and both Sebacinales species *in vitro* and in pots. For both conditions, we demonstrated the significant increase in growth parameters in all plants colonized with both fungi irrespective of nematode infection in comparison to control plants and plants infected only with *M. incognita*. Further, we observed similar significant reduction in number of galls induced by *M. incognita* in plants colonized with both endophytes. These results clearly show that the fungal colonization counteracts the negative impact of nematode infestation in tomato. An additional analysis of the expression of defense and hormone-related marker genes at 3 and 7 days after inoculation showed significant differences between variants indicating expressional reprogramming of the host in this tripartite interaction. Our data demonstrate the beneficial effects of both *Serendipita* species on plant growth as well as their antagonistic properties against *M. incognita*. These findings can be used for the further development of new environmentally harmless nematicides based on these endophytes or on Sebacinales-derived substances.

Keywords: *Meloidogyne incognita* - *Serendipita williamsii* - *Serendipita indica* - Plant defense - Tomato.

References:

- [1] Varma A et al., 1999. Appl. Environ. Microbiol. 65, 2741-4.
- [2] Weiß M et al., 2016. New Phytol. 211, 20-40.
- [3] Daneshkhah R et al., 2013. J. Exp. Bot. 64, 3763-3774.

S28-PF2

External and internal microbiomes of Antarctic dry valley nematodes are distinct, but more similar to each other than the surrounding environment.

J. Parr McQueen[1] (jparr.mcqueen@ufl.edu), Kaitlin Gattoni[1], Eli Gendron[1], Pacifica Sommers[2], Steven Schmidt[2], Dorota Porazinska[1]

[1] *Department of Entomology and Nematology, University of Florida, Gainesville, Florida, United States*; [2] *Ecology and Evolutionary Biology, University of Colorado, Boulder, Boulder, Colorado, United States*

In prior research, we showed that internal microbiomes of nematode guts from Antarctica's McMurdo Dry Valleys were distinct from the surrounding environment and primarily driven by host identity. However, host identity encompasses a wide range of specific factors (e.g., body morphology, cuticle biochemistry, feeding behavior). We wanted to explore whether internal microbiome diversity and composition could be influenced by the external microbiome. Although the cuticle is itself impermeable, its morphology and function could influence what passes into the stoma. Specific bacterial strains adhering to the nematode cuticle (e.g*., Pasteuria penetrans*) are well known, but knowledge about external microbiome composition is limited. The objectives of this study were (1) to characterize the diversity of external nematode microbiomes and (2) determine their potential role in the assembly of internal microbiomes. We hypothesized that external microbiomes would be primarily driven by the environment rather than factors of the nematodes themselves. Nematode communities in the Dry Valley streams provide a simple model system as they contain only 2 distinct types of cyanobacterial mats each with 2 species of nematodes (*Plectus murrayi* & *Eudorylaimus antarcticus*). We collected 3 replicates of 2 types of cyanobacterial mats from each of 4 streams in Taylor Valley, Antarctica. For all 24 mats, ~14 individuals of each nematode species were hand-picked and half were moved through a series of sterile washes while the other half remained unwashed. Picking nematode individuals directly from mats rather than using typical soil extraction methods provides an opportunity to examine intact external microbiomes that otherwise would be washed off or altered. Both washed and unwashed individuals and the mats they inhabited were characterized for bacterial and eukaryotic communities using 16S and 18S rRNA metabarcoding. Our results showed that external bacterial microbiomes were significantly less diverse than mats but more diverse than internal microbiomes. Community composition variation was most explained by the identity of nematode species (13%) followed by the identity of stream (6%), suggesting a greater influence of host factors than the environment. No consistent trends were observed for eukaryotic microbiomes. Overall, results indicated the presence of a host specific external microbiome and a potential role of the cuticle in structuring the assembly of internal microbiomes.

S28-PF3

Characterization of nematode-bacteria associations with gastropods and their virulence towards crop pests in northern Mindanao, Philippines.

Veronica Tañan (veronicatanan1998@gmail.com), Nanette Hope Sumaya, PhD.

Department of Biological Sciences, College of Science and Mathematics, Mindanao State University - Iligan Institute of Technology, Iligan, Lanao del Norte, Philippines

Molluscs known to be successful land invertebrate invaders include the class gastropoda: slugs and snails. Nematodes are diverse metazoans that can be isolated from gastropods. The group of nematodes being recorded such as *Aelurostrongylus abstrusus* and *Troglostrongylus brevior* use gastropods as taxis, parasites which causes mortality to the hosts, and pathogens to humans and other invertebrates. Therefore, this study aimed at assessing the occurrence of nematodes found in the different gastropods within the provinces of northern Mindanao, Philippines. This region is ideal for the study because it is geologically diverse including plains, rolling hills, mountains, as well as fertile soil and agricultural resources. Sampling was done on the identified locations collecting a total of 10 individuals each of the snails *Pomacea canaliculata*, *Achatina fulica, Oxychilus alliarius* and 15 individuals of *Subulina octona,* another 10 individuals each of slugs *Laevicaulis alte* and *Sarasinula plebeia.* Nematodes were isolated using the nutrient agar medium and semi-solid NGG was utilized to obtain pure culture incubated at 20°C. Based on morphology and morphometrics, the nematode species were identified as *Rhabditis, Panagrellus,* and *Panagrolaimus*. Through DNA barcoding using 16S, SSU and D2/D3 of the 28S rDNA regions, bacterial and nematode isolates will be later confirmed and is ongoing. Nematode-bacteria associations will be utilized in the virulence test against common crop pests: *Spodoptera litura, Sarasinula plebeia, and Subulina octona.* This study could be beneficial in agricultural management, given that gastropods are used as vectors in the transmission of bacterial infections. In addition, no nematodes parasitic to humans were recorded in this study.

Keywords: Gastropods - Nematodes - *Panagrolaimus* sp. - Virulence test - Crop pests.

References:

- Coughlan, L. et al., 2015. Evolutionary and Genomics Microbiology 6(672).
- Diano, M. et al., 2022. Biologia 77: 469-478.
- Lambshead, P. et. al., 2007. An improved molecular phylogeny 42(3): 622-636.
- Tandingan-De Ley, I. et al., 2020. Gastropod parasitic nematodes 18: 175–193.

ORAL SESSION 29

Trade and market access implications of plant parasitic nematodes

Nematology in support of plant health – role of EPPO.

Françoise Petter[1] (petter@eppo.int), Madeleine McMullen[1], Charlotte Trontin[1], Fabienne Grousset[2], Camille Picard[1], Muriel Suffert[1], Rob Tanner[1], Baldissera Giovani[3], Anne-Sophie Roy[1]

[1] *EPPO, Paris, France*; [2] *EPPO consultant, Paris, France*; [3] *Euphresco, Paris, France*

One of the main roles of the European and Mediterranean Plant Protection Organization (EPPO) is to help its member countries to prevent entry or spread of dangerous pests. The Organization has therefore been given the task of identifying pests which may present a risk for the region (early warning), evaluating them and making proposals on the phytosanitary measures which can mitigate the risk (Pest Risk Analysis, PRA). PRAs are available in the platform on PRAs (https://www.eppo.int/RESOURCES/eppo_databases/pra_platform). Once a pest has been identified as presenting a risk for the EPPO region, EPPO recommends that their members countries regulate it as a quarantine pest, and develop guidance on how to detect and identify it (procedures for phytosanitary inspection and diagnostic protocols) as well as recommendations on how to control and eradicate this pest. To perform these activities, EPPO has established a number of Panels composed of experts of individual countries. The Panels develop Standards e.g. Diagnostic Protocols, Regulatory Control Systems, to harmonize the work of the plant protection organizations across the region. The EPPO Secretariat also collects information and makes it available to its member countries, for example on pest distribution (in EPPO Global Database, gd.eppo.int) and on expertise available for diagnostic (http://dc.eppo.int/). In addition to pest-specific activities, EPPO also develops recommendations for quality assurance in laboratories (including guidance on how to perform validation of tests), in order to harmonize procedures in the EPPO region and improve diagnostic quality. EPPO is also hosting the Euphresco network, which is a network of organizations funding research projects and coordinating national research in the phytosanitary area. The different activities conducted in this framework are presented with a special focus on activities in nematology.

Keywords: Standard - Diagnotic - Pest Risk Analysis - Official Control - Euphresco.

Nematodes associated with damage on *Taxus media* cv. Hillii in Norway.

Christer Magnusson[1] (christer.magnusson@nibio.no), Dag Ragnar Blystad[1], Zahra Ben Hellal[2]

[1] *Norwegian Institute of Bioeconomy Research, Aas, Norway*; [2] *Norwegian University of Life Sciences, Aas, Norway*

Taxus media cv. Hillii is currently becoming increasingly popular as an ornamental plant in Norway. *Taxus* spp. are expected to be winter-green and endurable because of their tolerance to many plant diseases and pathogens. Many ornamental plantings with *Taxus media* cv. Hillii have developed yellow to brown needles in autumn. This persists during the winter, but in the spring many plants regain their green colour. However, some bushes remain chlorotic, drop their needles, show die back of branches and finally wilt. This leaves the hedges in a poor shape having yellow dying plants with thin growth. Samples were received at the Plant Clinic of NIBIO from one plantation in Oslo, which for many years had experienced this type of plant damage. Two out of four hedges had growth disturbances. No insects or fungi were detected that could explain this damage. Edema was present on the needles. Soil sampling revealed the presence of stubby root nematodes *Trichodorus* sp. This nematode was almost the only plant parasitic nematode present in the soil and occurred in density of 430 ind./250 ml of soil in one of the hedges. This is well above damaging thresholds for *Trichodorus* spp. on many plants. However, the strong aggregation of the nematodes made a clear quantitative correlation between nematode densities and damage of isolated bushes difficult to establish. Using tobacco (*Nicotiniana debneyi)* as a bait plant followed by root sap inoculations from this plant on *Chenopodium quinoa* demonstrated the presence of Tobacco Rattle Virus (TRV) in the soil from the two damaged hedges. *Trichodorus* spp. are well-known as vectors of this virus. Both hedges had received drip irrigation. *Táxus* does not thrive well in moist soil, and high soil humidity will allow *Trichodorus* to increase its population density and its efficiency in virus transmission. Therefore, we speculated that the damage to the bushes in these two hedges may have been the result of an interaction between nematodes, virus and water. Further studies are needed to clarify this potential interaction in more detail. This case demonstrates the value of having competence in and paying attention to plant protection in ornamental landscapes. In order to avoid damage it is necessary to increase the level of control to avoid the introduction of pests and diseases with plants, growth media, mulch and native soil. It is important that retailers ensure pest freedom of plants by obtaining the material from safe sources.

Keywords: *Trichodorus* - Yew - Ornamentals - Hedge plantings.

Movement of plant-parasitic nematodes associated with the turf industry.

Valeria Orlando[1] (Valeria.Orlando@fera.co.uk), Thomas Prior[2], Rebecca Lawson[2], Lesley Jutson[2], Margarida Correia[2], Paul Taylor[2], James Stubbs[2], Charlotte Howard[2]

[1] *Crop & Environment Sciences, Newport, United Kingdom*; [2] *Nematology department, Fera science Ltd., York, United Kingdom*

The turf industry is a multi-million pound business, comprising such sports facilities as golf courses, football and rugby stadiums and racecourses. For golf courses and sports stadiums, the move to sand-based construction to improve drainage and playability creates a perfect environment for soil-borne nematodes and as a consequence, has raised the typical levels of plant-parasitic nematodes in the soil base. This is an increasing issue within sports grounds, with an estimated 90% of all new football and golf courses with sand based constructions experiencing significant nematode damage (Fleming, 2011). Several key changes in modern turf grass construction and maintenance have also encouraged the spread of nematodes and has exacerbated the problem. Root damage caused by plant-parasitic nematode feeding creates distinctive patches and deterioration to the sward within a few months of introduction.
Movement of nematodes associated with turf, growing media and machinery with EU trade and within member states will be presented.

Keywords: Plant-parasitic nematodes - Machinery - Import - Turf.

References:

- Fleming, 2011. Pitchcare Magazine. 34.

Plant-parasitic nematodes pose a serious threat to the production and nutritional quality of popular biofortified cassava.

Aminat Oyetunde[1] (korede.akinsanya@augustineuniversity.edu.ng), Steve Afolami[2], Peter Kulakow[3], Danny Coyne[4]

[1] *Department of Biological Sciences, Augustine University, Ilara-Epe, Lagos State, Nigeria*; [2] *Department of Crop Protection, Federal University of Agriculture, Abeokuta, Ogun State, Nigeria*; [3] *Cassava Breeding Unit, International Inst. of Tropical Agric., Ibadan, Oyo State, Nigeria*; [4] *Nematology Unit, International Inst. of Tropical Agric., Ibadan, Oyo State, Nigeria*

African countries are not only faced with the problem of food security but that of nutritional deficiency due to limited micronutrients in the diet. Cassava plays a major role in efforts to alleviate the African food crisis because of its efficient production of food energy, year-round availability, tolerance to extreme stress conditions and suitability to smallholder farming. Consequently, the development of new cultivars with enhanced micronutrient contents offer greater prospects for food and nutrition security. Among the various constraints affecting cassava production are plant parasitic nematodes (PPN), especially root-knot nematodes (*Meloidogyne* spp.). In this study, six biofortified cultivars were evaluated for response to PPN in the field in Nigeria. Biofortified cassava growth and yield were measured in plots naturally infested with 11 genera of PPN but dominated by *Meloidogyne* spp., followed by *Pratylenchus* and *Helicotylenchus.* Plots treated with carbofuran (3 kg a.i./ha) were compared with untreated plots, which had significantly ($P \leq 0.05$) lower PPN densities at harvest and no galling damage, compared with significantly higher galling damage caused by root-knot nematodes in untreated plots. Plant height, stem girth, plant fresh weight, marketable storage root number and weight were significantly lower for most of the biofortified cultivars in untreated plots. Percentage yield losses of between 21.3 - 63.7% were recorded from two separate trials conducted for 12 months each, while lower nutritional quality (total carotenoid) and dry weight percentage was also associated with higher PPN densities in some biofortified cassava cultivars. This work demonstrates that natural field populations of PPN, especially *Meloidogyne* spp., can substantially affect yield and nutritional quality of biofortified cassava and should be considered when breeding new cultivars, as they are prevalent across the tropics and pose a significant threat to food security.

Keywords: Carotenoid content - Root-knot nematodes - Sub-Saharan Africa - Yield loss.

References:

- [1] Afolami, S.O., 2000. International Journal of Nematology 10, 94-100.
- [2] Coyne et al., 2004. Roots 9, 3-5.
- [3] Maroya et al., 2012. Journal of Life Sciences 6, 595-601.
- [4] Akinsanya et al., 2018. Nematropica 48, 50-58.

Plant-parasitic nematodes and export to non-European Union countries.

Anne Sophie van Bruggen (a.s.vanbruggen@nvwa.nl), Evelyn van Heese

National Reference Centre, Nematology, National Plant Protection Organization, Wageningen, Netherlands

The Netherlands export all kinds of plant products in considerable quantities all over the world. These products have to fulfil phytosanitary requirements in order to prevent the spread of harmful organisms. Trade inside and into the EU is harmonised for all EU countries by the Plant Health Regulation EU 2016/2031. For export to 3rd countries (outside the EU) the country of destination can demand additional specific requirements. The NPPO of the Netherlands has developed various inspection programs in cooperation with non-EU countries in order to facilitate export to those countries with specific requirements. Flower bulbs and perennials are important export products from the Netherlands. For example the USA and Canada are countries which have special inspection programs with the Netherlands for the import of flower bulbs. Most of the specific requirements concern virus diseases, but also nematodes. For export to the USA/Canada flower bulbs have to be free of 15 nematode taxa including *Meloidogyne* spp., *Aphelenchoides subtenuis*, *A. fragariae*, *A. ritzemabosi* and *Pratylenchus fallax*. Furthermore, the fields where the flower bulbs have been grown have to be free of *Globodera rostochiensis* and *G. pallida*. The perennials *Aconitum*, *Paeonia* and *Astilbe* have to be treated with a hot-water treatment to eradicate possible infestations with *A. fragariae* and *A. ritzemabosi*. Export of plants for planting to several non-EU countries is also enabled by a specific inspection program. For instance, for the export of host plants of the nematode *Radopholus similis* a system is developed to guarantee absence of this nematode in greenhouses of exporting growers. Every six months root samples are taken and analysed for absence of *R. similis*. If the nematode is not found the grower is registered in "the list of companies free of *R. similis*". For growers on this list it is allowed to export plants, without sampling of every consignment before export to countries with specific requirements. Similarly, there are lists for companies free of *Heterodera fici* and *Cactodera cacti*. These are some examples of special Dutch inspection programs to enhance export of plant material.

Keywords: Export - Trade - Plant-parasitic nematodes - Phytosanitary requirements.

Detection and diagnostics of tropical *Meloidogyne spp.* within the Euphresco project *MeloTrop.*

Saša Širca[1] (sasa.sirca@kis.si), Barbara Gerič Stare[2], Anne-Marie Chappe[3], Fabrice Olliver[3], Laurent Folcher[3], Maria L. Inácio[4], Filomena Nóbrega[4], Leidy Rusinque[4], Eugénia Andrade[4], Carla Maleita[5], Luci Conceição[6], Isabel Abrantes[6], Evelyn Y.J. van Hesse[7], Gerrit Karssen[7], Jasmina Bačič[8]

[1] *Plant Protection, Agricultural Institute of Slovenia, Ljubljana, Slovenia*; [2] *AIS, Ljubljana, Slovenia*; [3] *ANSES, Le Rheu, France*; [4] *INIAV, Oeiras, Portugal*; [5] *CFE/CIEPQPF, Coimbra, Portugal*; [6] *CFE, Coimbra, Portugal*; [7] *NPPO-NL, Wageningen, Netherlands*; [8] *Institute Tamiš, Pančevo, Serbia*

Tropical root-knot nematodes (RKN), *Meloidogyne* spp., are considered as an emerging phytosanitary problem within Europe for the future. The Euphresco initiative entitled «Global warming and distribution of root-knot nematode species of the tropical group (MeloTrop)» was launched as a result of the potential damage these species can cause on economically important crops, especially under a climate change scenario. Six research partners joined forces in the consortium to reach the following objectives: to generate distribution maps of the tropical RKN in Slovenia, France, Portugal and Serbia; to assess the survival ability of *M. incognita* and *M. arenaria* in the continental European climate conditions; to validate biochemical and molecular methods for the diagnosis of tropical RKN, and to generate geographical maps of the possible open field spreading for each tropical RKN species occurring in Europe. Species identification within the group was based on a combination of morphological, morphometrical, biochemical and molecular methods. However, distinguishing tropical RKN species is very difficult due to inter-specific morphometrical similarity and intra-specific morphological and molecular variability. Esterase (Est, EC 3.1.1.1) and malate dehydrogenase (Mdh, EC 1.1.1.37) isozyme phenotyping as well as molecular identification approach based on multi locus sequencing of four mtDNA genes (nad2, nad5, cox2 and cox3) were validated within the project with inter-laboratory test performance studies. Additionally, several tropical RKN species were detected in partnering countries at the open field plant production. Open field RKN occurrence represents additional risk for several agricultural crops, especially due to predicted climate change effects and the fact that infestations at larger acreages are much more difficult to manage. The data of tropical RKN species occurrence will be presented and discussed.

Keywords: *MeloTrop* - RKN - Europe - Occurrence - Validation.

S29-PF1

Negative binomial modeling of nematode count data yield more accurate mean and variance estimates.

Gideon Alake[1] (agideon@ufl.edu), Inga Zasada[2], Edzard van Santen[3], Peter DiGennaro[4]

[1] *Department of Entomology and Nematology, University of Florida, Gainesville, Florida, United States*; [2] *Horticultural Crops Research Laboratory, U.S. Depart. of Agriculture (USDA-ARS), Corvallis, Oregon, United States*; [3] *Statistical Consulting Unit, University of Florida, Gainesville, United States*; [4] *Department of Entomology and Nematology, University of Florida, Gainesville, United States*

Nematode count data regularly produces nonnormal, discrete, positive, and skewed data that often include valid '0' observations. Analyzing such data using classical statistical methods can yield inaccurate mean and variance estimates. The characteristic clustering of nematode populations, sampling techniques, sample size, and extraction efficiency can also influence variability in nematode counts, making traditional analyses of variance (ANOVA) inappropriate, as it requires the data have a normal and homogenous distribution. Log or square root transformation shortcuts are often applied to data to circumvent the inherently nonnormal and highly variable nematode counts, and to make data agreeable to parametric tests. However, log transformations commonly produce underestimated, and difficult-to-interpret mean and variance estimates for clustered distributions. The generalized linear mixed models (GLMM) approach provide a more adaptable model-based framework for analyzing nematode count data. Here we analyzed multiple data transformations and demonstrate that GLMM-based analyses of nematode counts with a negative binomial distribution yield more accurate mean and variance estimates. On the contrary, log-normal distributions produced downwardly biased mean and variance estimates, and Poisson distributions did not explain the high natural variability seen with nematode counts. Therefore, rather than log-transforming and forcing nematode count data into ANOVA-conforming analysis, we recommend the adoption of GLMM. Further, we present a framework within commonly used statistical packages to assess current and future nematode count data as a tool for the nematology community.

Keywords: GLMM - Negative binomial - Log transformation - Nematode count.

References:

- O'Hara, R. B. & Kotze, D. J., 2010. Methods in Ecology and Evolution, 1(2):118–122.
- Stroup, W. W., 2015. Agronomy Journal, 107:811–827.

S29-PF2

FAGUSTAT: Investigating Beech Leaf Disease, a threat to beech trees and forests in Europe.

Nicole Viaene[1] (nicole.viaene@ilvo.vlaanderen.be), Negin Ebrahimi[1], Annelies Haegeman[1], Ondrej Douda[2], Anne Sophie van Bruggen[3], Nikica Ogris[4], Saša Širca[5], Barbara Gerič Stare[5], Thomas Prior[6], Ana Pérez Sierra[7], Mariana Groza[8], Mihaela Coman[8], Marie-Jo Hurley[9], Déborah Lanterbecq[10], Simon Van Kerkhove[11], Quentin Leroy[12]

[1] *Flanders Research Institute for Agriculture, Fisheries and Food (ILVO), Merelbeke, Belgium*; [2] *Crop Research institute (CRI), Prague, Czech Republic*; [3] *Netherlands Food and Consumer Products Safety Authority (NVWA), Wageningen, the Netherlands*; [4] *Slovenian Forestry Institute (SFI), Ljubljana, Slovenia*; [5] *Agricultural Institute of Slovenia (KIS), Ljubljana, Slovenia*; [6] *Fera Science, York, United Kingdom*; [7] *Forest Research (FR), Surrey, United Kingdom*; [8] *National Phytosanitary Authority (NPA), Ilfov, Romania*; [9] *Department of Agriculture, Food and the Marine (DAFM), Dublin, Ireland*; [10] *Research Center for agronomy and agro-industry HEPH-Condorcet (CARAH), Ath, Belgium*; [11] *Ornamental Plant Research (PCS), Destelbergen, Belgium*; [12] *Observatoire Wallon de la Santé des Forêts (OWSF), Gembloux, Belgium*

Beech Leaf Disease (BLD), reported from the northeastern United States and Canada (Ontario), is causing severe damage to mainly American beech (*Fagus grandifolia*) but also to European beech (*F. sylvatica*) [1]. The disease is characterised by dark interveinal bands on the leaves, leaf deformation and bud abortion in spring, leading to canopy thinning and tree mortality in advanced cases. So far, the etiology of the disease is not fully understood. *Litylenchus crenatae* subsp. *mccannii*, a nematode species isolated from beech leaves and buds in America, is involved in BLD, although other microorganisms probably contribute to its development [2]. In Japan, *L. crenatae* was found in leaves of *F. crenata*, but has not been associated with damage. It is not known if BLD or *Litylenchus* spp. occur in Europe, therefore, it should be considered as a threat to the European beech trees and forests at this moment. Within the Euphresco project 2020-A-334, FAGUSTAT, we are increasing awareness of BLD in Europe and making the first assessment of its status in the region. In 2021, we performed surveys on the health status of *Fagus* spp. in the six participating countries (Belgium, the Netherlands, Romania, Slovenia, United Kingdom and Ireland). We collected about 250 samples of beech leaves, buds and nuts in forests, parks, botanical gardens and nurseries and extracted the nematodes using the Baermann technique. Although look-alike BLD symptoms were observed, only very few nematodes (e.g., *Aphelenchoides*sp., *Plectus* sp., *Panagrolaimus* sp.) were found; none belonging to *Litylenchus*. We drafted BLD information sheets to further increase public awareness. As pathways of BLD transmission are still unknown, we also explore possible ways of entry and spread. To study the biology of *L. crenatae* under controlled conditions, we are testing different culturing methods (carrot discs, callus tissue, fungal cultures and beech sapling inoculation) to obtain a large population of the nematode. In addition, we took subsamples of (a)symptomatic leaf and bud samples throughout the survey to determine their microbial community using high-throughput sequencing (HTS) technology. This will provide insight into the main drivers of the communities living in/on beech leaves and buds. We recommend countries not involved in the FAGUSTAT project to look out for BLD symptoms, as knowledge about the presence (or absence) of *Litylenchus* spp. is important to assess its threat to European beech trees.

Keywords: Alert organism - Beech leaf disease - Foliar nematode - *Litylenchus*.

References:

- [1] Carta et al., 2020. Forest Pathology 50 (2): e12580.
- [2] Ewing et al.,2021. Phytobiomes Journal 5(3): 335- 349.

S29-PF3

Plant-parasitic nematode: a potential threat to medicinal plants in Vietnam.

Nguyen Huu Tien[1] (tien.quelampb@gmail.com), Bert Wim[2], Trinh Quang Phap[3]

[1] *Nematology, Ghent University & IEBR, Ghent, Belgium*; [2] *Nematology, Ghent University, Ghent, Belgium*; [3] *Nematology, IEBR, Hanoi, Vietnam*

Plant-parasitic nematodes (PPNs) are among the most important pests and cause serious damages in many crops over the world. In Vietnam, plant-parasitic nematodes were reported to be the main cause of declining, yellowing leaves, root-gall, root lesion, stunting, and slow death diseases on thousands of hectare of coffee and black pepper [1-5]. Recently, medicinal plants have been growing in similar large scale monocultures in Vietnam, bringing significant economical value for the country. However, it is hypothesized that monoculture of medicinal plants will create favourable conditions for the development and multiplication of specific groups of PPNs. Indeed, a baseline study of plant-parasitic nematodes on medicinal plants revealed a high rate of nematode infection, i.e. none of the 23 studied medicinal plants in Vietnam was found free of PPNs and each medicinal plant was infected by at least two species of PPNs. Many symptoms such as declining, yellowing leaves, root-knot, root lesion, and stunting have been found associated with numerous valuable medicinal plants, including *Panax vietnamensis*, *Polyscias fruticosa*, *Salvia mitiorrhiza*, and *Morinda officinalis*. Extreme PPN-caused yield losses, up to 70%, were recorded on *Curcuma longa* and *Amomum longiligulare* in the Central Highlands in Vietnam. Ninteen genera of plant-parasitic nematodes associated with medicinal plants in Vietnam were recorded, including *Aphelenchoides*, *Criconemella*, *Discocriconemella*, *Ditylenchus*, *Helicotylenchus*, *Hemicriconemoides*, *Heterodera*, *Hirschmaniella*, *Meloidogyne*, *Paratrichodorus*, *Paratylenchus*, *Pratylenchus*, *Rotylenchulus*, *Scutellonema*, *Trichodorus*, *Trophonema*, *Trophorus*, *Tylenchus*, *Tylenchorhynchus*, *Tylenchulus*, and *Xiphinema*. Several are new PPN reports in Vietnam, and morphological and molecular analyses indicated two putative new species. This study underscores the need for further studies on both the diversity and damage assessment of plant-parasitic nematodes on medicinal plants, in order to ensure sustainable development of the cultivation of medicinal plants.

Keywords: Medicinal plants - Diversity - Systematic - Vietnam - Nematode.

References:

- [1] Nguyen, N.C., 1995. Plant protection journal. 1(139): 14-18.
- [2] Khanh, L.D., et al. 2013. Plant protection journal. 6: 24-30.
- [3] Bui , T.T.N., et al. Journal of Biotechnology. 2015. 13(4A): 1359-1367.
- [4] Trinh, P.Q., et al. Russian journal of Nematology, 2009. 17(1): 73-82.
- [5] Trinh, P.Q., T.K.D. Pham, and N.C. Nguyen. 2013. In Proceedings of the 2nd VAST-KAST workshop on biodiversity and bop-active compounds. 313-318.

ORAL SESSION 30

Nematode community assemblies

Impact of cultural practices and environmental conditions on plant-parasitic nematode communities through a long-term agroecological field trial.

Nathan Garcia[1] (nathan.garcia@anses.fr), Eric Grenier[2], Laurent Folcher[1]

[1] *Nematology, Anses - Plant Health Laboratory, Le Rheu, France*; [2] *IGEPP, INRAe, Le Rheu, France*

To avoid the use of chemical agents and find alternative methods to manage damaging plant-parasitic nematodes (PPNs), research studies have mainly focused on plant resistance genes and biocontrol methods involving host plants or natural enemies. Another method may consist in ecological regulation using native non-damaging species that could compete with the more damaging ones. For this purpose, knowledge about the biodiversity, structure, and functioning of indigenous PPNs communities is needed to carry out better risk assessments and to develop possible future management strategies. In a recently published study, we investigated 37 root crop fields in eight French regions over two consecutive years to describe PPNs diversity and assess the potential effects on communities of cultural and environmental variables. We observed that at a French national scale, environmental variables (mainly rainfall and heavy metals) and cultural practices (tillage practices or use of chemical products), seem to drive the structuration of the communities among the geographical areas. To describe the effects of profound tillage and crop rotation on PPNs communities, we are currently working on an ongoing long-term agroecological trial (between 75% and 100% of input reduction) that has begun at the end of 2018 and should last at least six years. The trial has been designed in a 3Ha field, divided into six sub-fields (0.5Ha each). For each sub-field, the crop rotation will be the same over the six-year period, but three of them will be systematically tilled before each crop while the remaining three will never be. For each sub-field and each crop, PPN communities will be identified and counted at four time points (before and after tillage/sowing, during crop growth and after harvest). PPNs communities' diversity before the trial showed seven species (5 genera). At this point, we can already observe the abundance decreasing for several PPN in tilled compared to non-tilled sub-fields, and some abundance variations depending on the crop used (*e.g.* with potato decreasing *Pratylenchus*, *Paratylenchus* and *Amplimerlinius* abundance or wheat/pea mix increasing *Paratylenchus* abundance). Samplings for PPN communities will continue until 2024.

Keywords: Biodiversity - Community ecology - Variable effects - Agroecological trial - Risk assessment and management.

A novel metabarcoding strategy for studying nematode communities.

Md Maniruzzaman Sikder[1] (mms@agro.au.dk), Mette Vestergård[1], Rumakanta Sapkota[2], Tina Kyndt[3], Mogens Nicolaisen[1]

[1] *Entomology and Plant Pathology, Aarhus University, Slagelse, Denmark*; [2] *Environmental Science, Aarhus University, Roskilde, Denmark*; [3] *Biotechnology, Ghent University, Ghent, Belgium*

Nematodes are the widely abundant soil metazoa, and often considered indicators of soil health. While the recent advances in next-generation sequencing technologies have accelerated research in microbial ecology, the ecology of nematodes remains poorly elucidated, partly due to the lack of reliable and validated sequencing strategies. Objectives of the present study were (i) to compare commonly used primer sets in literature and identify the most suitable primer set for metabarcoding of nematodes, and (ii) to validate and establish a high-throughput sequencing strategy for nematodes using Illumina paired-end sequencing. In this study, we tested four primer sets for amplicon sequencing: JB3/JB5 (mitochondrial, I3-M11 partition), SSU_04F/SSU_22R (18S rRNA, V1-V2 region), Nemf/18Sr2b (18S rRNA, V6-V8 region) from earlier studies [1-4], and MMSF/MMSR (18S rRNA, V4-V5 region) a newly developed primer set from this study. In order to test the primer sets, we used 22 samples of individual nematode species, 20 mock communities, 20 soil samples, 20 spiked soil samples (mock communities in soil), and 4 root/rhizosphere soil samples. We successfully amplified variable target regions (I3-M11 partition of the COI gene; V1-V2, V4-V8 region of 18S rRNA gene) from these 86 DNA samples with the four different primer combinations and sequenced the amplicons on an Illumina MiSeq sequencing platform. After thorough study, we found that the MMSF/MMSR and Nemf/18Sr2b were suitable primers for studying nematode communities in agricultural environments based on annotation of sequence reads at genus and in some cases at species level.

Keywords: Nematode diversity - Primer design - Next generation sequencing - Soil - Environmental.

References:

- Derycke et al., 2010. PLoS One. 5(10):e13716.
- Blaxter et al., 1998. Nature. 392:71–75.
- Sapkota and Nicolaisen. 2015. BMC Ecology. 5(3):1-8.
- Sapkota and Nicolaisen. 2018. Agric Ecosyst Environ. 257:120-131.

Community assembly in the wake of glacial retreat: a meta-analysis.

Satyendra K. Pothula[1, 2, 3] (spothula@email.arizona.edu), Byron J. Adams[1, 2, 3]

[1] *Department of Biology, Brigham Young University, Provo, UT, United States*; [2] *Evolutionary Ecology Laboratories, Brigham Young University, Provo, UT, United States*; [3] *BYU Life Science Museum, Brigham Young University, Provo, UT, United States*

Ecosystems shaped by retreating glaciers provide a unique opportunity to study the order and timing of biotic colonization, and how this influences the structure of successive ecological communities. In the last century glaciers across most of the cryosphere have receded at an unprecedented pace. Many studies have been published from different parts of the world testing hypotheses about how soil ecosystems are responding to rapid, contemporary deglaciation events. To better understand and draw general conclusions about how soil ecosystems respond to deglaciation, we conducted a global meta-analysis of 88 published studies focused on the succession of various organisms and soil physicochemical properties in glacier forefields along the chronosequence. Our global synthesis reveals that key soil properties and the abundance and richness of biota and followed two conspicuous patterns: 1) Some taxa demonstrate a persistent increase in abundance and richness over the entire chronosequence, 2) other taxa increase in abundance and richness during the first 50 years of succession, then gradually decline 50 years onward. The soil properties and soil organisms that are intimately tied to vegetation followed the first pattern, consistent with the idea that aboveground patterns of vegetation drive patterns of belowground biodiversity. The second pattern may be due to an initial increase and subsequent decline in available nutrients and habitat suitability caused by increased biotic interactions, including resource competition among soil biota. A consensus view of the patterns of historical and contemporary soil ecosystem responses to deglaciation provides a better understanding of the processes that generate these patterns and informs predictions of ongoing and future responses to environmental changes.

Exploring belowground biodiversity in Lincoln, Nebraska's tallgrass prairie corridor.

Abigail Borgmeier[1] (ab6240@student.byu.edu), Kaitlin Gattoni[2], Timothy Harris[3], Peter Mullin[3], Dorota Porazinska[2], Kirsten Powers[3], David Wedin[3], Thomas Powers[3]

[1] *Department of Biology, Brigham Young University, Provo, UT, United States*; [2] *Department of Entomology and Nematology, University of Florida, Gainesville, FL, United States*; [3] *Department of Plant Pathology, University of Nebraska, Lincoln, NE, United States*

Nebraska is in the heart of the North American tallgrass prairie ecoregion, of which over 95% has been converted for agriculture in the past 150 years. Once one of the most diverse grasslands on the planet, the ecosystem now exists as small remnant prairies, fragmented by residential development and agricultural fields. Lincoln, NE has created a habitat corridor of remnant tallgrass prairie sites to preserve and protect this degraded ecosystem. Each site, while never plowed, has been managed differently, which has caused changes in the plant diversity. The aim of this study is to document the nematode diversity within the tallgrass prairie and examine if the belowground nematode community reflects differences in management and plant diversity.

Fifteen unplowed prairie fragments, 1.2 hectares in size, were selected from the 16km long prairie habitat corridor, representing different management histories. Two sites that were under corn-soybean rotation were also sampled for comparison. Soil analysis determined that the soils were all either clay loam or silty clay and similar in soil organic matter, but plant diversity differed significantly between the prairie sites. A 500cc soil sample was collected from a 40x40m2 plot in each unit. Nematodes were extracted from 200cc of soil and the nematode community composition was analyzed using a traditional morphological analysis of 150 nematodes per site, a metabarcoding analysis using the 18S-V9 genetic marker, and a Sanger Sequencing DNA barcode analysis of selected nematodes using the COI and 18S genetic markers.

Morphologic, barcoding, and metabarcoding analyses revealed different measures of nematode diversity. The total diversity of nematodes identified morphologically from prairie sites was 80 genera, while 111 total genera were identified via metabarcoding. Only 34 genera of the 80 genera identified via morphology were represented by COI, as non-amplification events with some taxa reduced overall assessment of taxonomic diversity. There were few differences in the composition of the nematode community between the remnant tallgrass prairie sites, however, the nematode community differed significantly between the remnant sites and the corn-soybean fields. Genera which were most abundant in the remnant prairie sites were absent or largely reduced in the corn-soybean fields, indicating a unique tallgrass prairie nematode assemblage within the remnant sites which is lost when cultivated for agriculture production.

Keywords: Barcoding - Metabarcoding - COI - Tallgrass prairie - Ecology.

DNA barcoding individual specimens using COI and 18S in nematode community analyses: advantages of a combined approach.

Thomas Powers[1] (tpowers1@unl.edu), Dorota Porazinska[2], Byron Adams[3], Abigail Borgmeier[3], Rebecca Higgins[1], Timothy Harris[1], Peter Mullin[1], Kirsten Powers[1]

[1] *Department of Plant Pathology, University of Nebraska, Lincoln, NE, United States*; [2] *Department of Entomology and Nematology, University of Florida, Gainesville, FL, United States*; [3] *Dept of Biology and Evolutionary Ecology, Brigham Young University, Provo, UT, United States*

The acknowledged advantage of DNA barcoding with the mitochondrial Cytochrome oxidase subunit 1 (COI) gene is its greater taxonomic species resolution than traditionally applied nuclear ribosomal genetic markers [1]. The oft-cited limitation of COI DNA barcoding is that the high rate of nucleotide substitution in COI prevents the design of PCR primers that function across a broad taxonomic spectrum of nematodes [2]. The advantage of 18S is the increased support for deeper divergences on the phylogenetic tree. Here we present COI and 18S barcode analyses of *Plectus* in nematode communities from vastly different ecosystems and geographically distant locations. The depauperate continental nematode communities of Antarctica include two ecologically different *Plectus* species. *Plectus murrayi*, widespread on the continent and most often collected from glacial melt streams, is represented by a single COI haplotype suggesting recent colonization and widespread dispersal across the continent. In contrast, *Plectus frigophilus,* an inhabitant of the Taylor Valley glacial lakes, includes at least six COI haplotypes, possibly indicating a longer residence time on the continent. The 18S gene shows the two *Plectus* species are distantly related to each other and other plectids, suggesting independent continental introductions. Extending the taxonomic comparisons to native plant communities in North America, it becomes apparent that considerable *Plectus* COI haplotype diversity underlays a relatively conserved morphology. There is little evidence of recent long-distance dispersal in the form of shared haplotypes across large geographic scales. Haplotype clades within ecoregions exhibit greater similarity with each other. There is moderate support for haplotype groups that are specific to habitats. The application of *Plectus* species names, however, is confounded by the lack of well-defined diagnostic morphological features and conflicting GenBank Accessions with identical species names.

Keywords: Biogeography - Ecology - Phylogeography.

References:

- [1] Prosser et al., 2013. Mol Ecol Resour 6:1108-15.
- [2] Ahmed et al., 2019. Metabarcoding and Metagenomics 3: 77–99.

Agriculture land-use intensification causes biotic homogenization on phytonematode communities at regional scale.

Antonio Archidona Yuste[1] (aarchidona@ias.csic.es), Thorsten Wiegand[1], Nico Eisenhauer[2], Carolina Cantalapiedra-Navarrete[3], Juan E. Palomares Rius[3], Pablo Castillo[3]

[1] *Department of Ecological Modelling, UFZ, Leipzig, Germany*; [2] *iDiv, Leipzig, Germany*; [3] *Crop Protection, IAS/CSIC, Córdoba, Spain*

Land-use change due to agricultural intensification is a major driver of loss of soil biodiversity. Agricultural land-use intensification is well recognized as drivers of biotic homogenization. However, there is still a major knowledge gap regarding the effect of agricultural land-use intensification on the structure and composition of soil communities, particularly in soil nematodes. Biotic homogenization means a decrease in beta-diversity, and it might occur through two separately phenomena, the loss of specialized species or the gain and spread of generalist species, or a combination of both. We investigated how the diversity and community assembly of plant-parasitic nematodes responded to the dramatic land-use change from the conversion of complex natural to simplified managed ecosystems. To this end, our study was conducted within the olive growing area of southern Spain, which includes wild and cultivated ecosystems covering an extensive area. Within each ecosystem, 123 sites were chosen both in natural (wild olive trees) and agricultural ecosystems (cultivated olive trees). The sites were selected to represent the regional range of management intensity. Alpha and beta diversity indices were used to assess the effect of agricultural land-use intensification on soil nematode community assembly. Relative total biomass for each species present was determined for the estimation of beta diversity. Land-use intensity measure (LUI) for natural ecosystem was based on inventory data including different age-class and the presence of main trees species within the wild ecosystem. In cultivated olive ecosystem, LUI included different components describing age of the olive cultivation and different agricultural management strategies. Changes in alpha and beta diversity were detected among the natural and cultivated ecosystems suggesting the loss of biodiversity and causing homogenization of soil nematode communities. Here we also showed that even moderate increases in LUI cause biotic homogenization across nematode community assemblies given the significant effect on the average body size of the nematode community. This suggests a loss of specialized species and is a further evidence for biotic homogenization. Indeed, biotic homogenization rather than local diversity loss could prove to be the most substantial consequence of agricultural land-use intensification.

Keywords: Land-use - Biotic homogenization - Beta diversity.

S30-PF1

Nematode communities in organic and conventional rice production in Indonesia: a morphological and metabarcoding approach.

Muhammad Aksan[1, 2] (akhsanpulalo07@gmail.com), Prabowo Lestari[1, 3], Marjolein Couvreur[1], Antarjo Dikin[4], Johannes Helder[5], Wim Bert[1]

[1] *Nematology Research Unit, University of Ghent, Gent, Belgium*; [2] *Departement of Agrotechnology, Faculty of Science and Technology, Universitas Muhammadiyah Sidenreng Rappang, Sidenreng Rappang, Sulawesi Selatan, Indonesia*; [3] *Laboratory of Nematology, Wageningen University, Gerderland, Netherlands*; [4] *Directorate General of Estate Crop, Ministry of Agriculture, Jakarta, Indonesia*; [5] *Laboratory of Nematology, Wageningen University, Wageningen, Netherlands*

Nematode communities can be used to study the soil's health and disturbance level. This study focuses on a comparison between the nematode community of organic and conventional rice production by using two different methods, a morphological and a metabarcoding approach. Soils samples were taken from three different regions in Indonesia, Bantul, Klaten, and Sragen. Based on the morphological and metabarcoding approach, respectively, 19 and 23 taxa were identified; fourteen taxa were shared by both methods, including plant-parasitic nematode and free-living. Metabarcoding revealed more taxa than the morphological approach. However, the t-test revealed there is no significant difference (p-value > 0.05) between both management systems. Nematode indices, MI (Maturity Indices) between organic and conventional on both methods, are not significantly different. PPI/MI ratios were different on both systems, and values were classified as a natural habitat to slight nutrient disturbance, in accordance to stress value and points patterns through NMDS, both systems are different where morphological is ideal for interpreting while metabarcoding revealed misleading points. Therefore, for future work, other indices, such as soil type and micro-climate are necessary.

Keywords: Free-living nematodes - Indices - Plant-parasitic nematodes - Rice - Taxa.

S30-PF2

Association of beneficial terrestrial nematodes with glyphosate-tolerant and conventional soybean-based cropping systems.

Akhona Mbatyoti[1] (24088978@nwu.ac.za), Mieke Daneel[2], Antoinette Swart[3], Dirk De Waele[1], Hendrika Fourie[1], Rinus Knoetze[4]

[1] *Integrated Pest Management, North-West University, Potchefstroom, South Africa*; [2] *Nematology, Agricultural Research Council-Tropical & Subtropical Crops, Mbombela, South Africa*; [3] *Nematology, Agricultural Research Council-Plant Health & Protection, Queenswood, South Africa*; [4] *Plant Protection, Agricultural Research Council, South Africa, Stellenbosch, South Africa*

Little is known about the effect of glyphosate, a broad-spectrum systemic herbicide, on nematodes. The abundance and diversity of beneficial, terrestrial nematodes in commercial glyphosate-tolerant (genetically modified; GM) and conventional (non-glyphosate-tolerant) soybean fields as well as in natural vegetation (reference system) in South Africa were recorded during 2011/2012 and 2012/2013 growing seasons. In addition, the effect of glyphosate was investigated on beneficial nematodes associated with a soybean-maize cropping sequence in a small field trial. Thirty-two nematode genera were identified from soil samples with most of the genera being present in natural vegetation (28), less in conventional soybean (23) and the least in glyphosate-tolerant soybean (21) fields. Bacterivores were superior in terms of diversity during both seasons and for all ecosystems, while fungivores were more abundant in glyphosate-tolerant soybean fields during the second season. The soils of all ecosystems were disturbed and degraded reflecting a low abundance and diversity of omnivores and predators. For the small soybean-maize field trial, the 14 genera recorded were dominated by bacterivores in non-treated plots, while fungivores dominated in glyphosate-treated plots. Also, soils from glyphosate-treated plots were degraded, less enriched and fungal-mediated, and those from non-treated plots disturbed, enriched, and bacterial-mediated. Data from this study showed that glyphosate had no effect on the beneficial nematode assemblages prevailing in ecosystems investigated.

S30-PF3

Research on free-living terrestrial nematodes (Order Dorylaimida) from tropical rain forest in Vietnam.

Thi Anh Duong Nguyen[1] (nad2807@gmail.com), Joaquin Abolafia[2], Reyes Pena-Santiago[3]

[1] *Nematology Department, Institute of Ecology and Biological Resources, Hanoi, Vietnam*; [2] *Department of Animal Biology, Plant Biology and Ecology, University of Jaén, Jaén, Spain*; [3] *Department of Animal Biology, Plant Biology and Ecology, University of Jaén, Jaén, Spain*

We investigated the nematode fauna in soils of three nature reserves in Cao Bang, Lang Son and Cat Ba provinces in north-eastern Vietnam. Nematodes in the order Dorylaimida were identified to species level in order to get an estimate on the numbers and diversity of known and undescribed taxa. In total, more than 14,000 specimens were identified. These nematodes belonged to 105 genera, 42 families and 9 orders, of which 48 genera and 13 families belonged solely to the order Dorylaimida. The Order Dorylaimida is taxonomically particularly challenging because it is highly diverse, but species show only little differences in morphological characters. More than 1/3 of the identified species of Dorylaims were new to science, a proof of the much understudied nematode diversity existing in the poorly explored natural areas from Southeast Asia. Also molecular data confirmed that the diversity of dorylaims is little known, only 5 sequences obtained from Genbank could be assigned to the sequenced nematodes with a similarity higher than 95%.
The nematode assemblages and diversity were significantly different between different regions and among four land use intensities. Nematode assemblages in primary forests were composed mainly of large and long-lived predators and omnivores (46-74%), compared to a successive increase of short-lived bacterivores (5-14%), plant parasites (11-24%) and fungivores (3-15%) with land-use intensification. Our data indicated that nematode community analysis was a useful tool to predict functional changes in soil ecosystems.

Keywords: Nematodes - Free-living - Tropical forest - Dorylaimida - Vietnam.

ORAL SESSION 31

Effectors in plant parasitic nematodes

Effector gene birth in plant parasitic nematodes: neofunctionalization of a housekeeping glutathione synthetase gene.

Sebastian Eves-van den Akker[1] (se389@cam.ac.uk), Catherine Lilley[2], Abbas Maqbool[3], Duqing Wu[2], Hazijah Yusup[2], Laura Jones[2], Paul Birch[4], Mark Banfield[3], Peter Urwin[2]

[1] *University of Cambridge, Cambridge, Cambridgeshire, United Kingdom*; [2] *University of Leeds, Leeds, United Kingdom*; [3] *The John Innes Centre, Norwich, United Kingdom*; [4] *University of Dundee, Dundee, United Kingdom*

Plant pathogens and parasites are a major threat to global food security. Plant parasitism has arisen four times independently within the phylum Nematoda, resulting in at least one parasite of every major food crop in the world. Some species within the most economically important order (Tylenchida) secrete proteins termed effectors into their host during infection to re-programme host development and immunity. The precise detail of how nematodes evolve new effectors is not clear. Here we reconstructed the evolutionary history of a novel effector gene family. We showed that during the evolution of plant parasitism in the Tylenchida, the housekeeping glutathione synthetase (GS) gene was extensively replicated. New GS paralogues acquired multiple dorsal gland promoter elements (DOG boxes), altered spatial expression to the secretory dorsal gland, altered temporal expression to primarily parasitic stages, and gained a signal peptide for secretion. The gene products were delivered into the host plant cell during infection, giving rise to “GS-like effectors”. By solving the structure of GS-like effectors we showed that during this process they had also diversified in biochemical activity, and likely represented the founding members of a novel class of GS-like enzyme. To the best of our knowledge, these were the first three crystal structures of any kind for a plant-parasitic nematode. Our results demonstrated the re-purposing of an endogenous housekeeping gene to form a family of effectors with modified functions. We anticipate that our discovery will be a blueprint to understand the evolution of other plant-parasitic nematode effectors, and the foundation to uncover a novel enzymatic function.

Keywords: Plant-parasitic nematodes - Effectors - Gene birth - Glutathione synthetase.

A root-knot nematode effector targets the spliceosomal plant machinery allowing the giant cell formation.

Michaël Quentin (michael.quentin@inra.fr)

INRAe, Université Côte d'Azur, CNRS, ISA, Sophia Antipolis, France

Root-knot nematodes are obligate endoparasites that maintain a biotrophic relationship with their hosts over a period of several weeks and induce the differentiation of root cells into specialized multinucleated feeding giant cells. Nematodes use secreted proteins called effectors, synthesised in oesophageal glands and delivered within the host plant through a syringe-like stylet, to allow *de novo* organogenesis of these structures. *In planta*, effectors are addressed to different subcellular compartments and associate with specific host proteins to allow hijacking important processes for cell morphogenesis and physiology or immunity.
MiEFF18 was identified as such a putative secreted effector. We showed that MiEFF18 was delivered into host cells where it localized into the nucleus, and particularly within the nucleolus. Because MiEFF18 did not have any known function, a yeast two-hybrid approach was used to search for plant partners of this effector using a tomato root cDNA library. We validated the interaction of MiEFF18 with a core spliceosomal protein. We demonstrated the importance of MiEFF18's target in plant susceptibility to nematode pests, playing a key role in giant cell formation. We investigated the outcomes of MiEFF18 interaction with its target, and focused on the alternative splicing, as the probable cellular function hijacked by this effector.

Keywords: Root-knot nematode - *Meloidogyne* - Effector - Alternative splicing - Spliceosome.

Nematode-encoded RALF peptide mimics facilitate parasitism of plants through the FERONIA receptor kinase.

Xin Zhang[1] (toxinzhang@hnu.edu.cn), Huan Peng[2], Hongdong Liao[1], Deliang Peng[2], Feng Yu[1]

[1] *Hunan University, Changsha, Hunan, China*; [2] *Chinese Academy of Agricultural Sciences, Beijing, China*

The molecular mechanism by which plants defend against plant root-knot nematodes (RKNs) is largely unknown. The plant encodes the receptor kinase FERONIA and its peptide ligands, rapid alkalinisation factors (RALFs), to regulate plant immune responses and cell expansion, which are two important factors for RKN successful parasitism. Here, mutation of the FERONIA in *Arabidopsis thaliana* showed low susceptibility to RKN *Meloidogyne incognita*. In search for the underlying mechanisms, we identified eighteen novel RALF-like peptides from multiple RKNs, and confirmed two RALF-like peptides (i.e. *MiRALF1*, and *MiRALF3*) from *M. incognita* were expressed in the esophageal gland and had high expression during the parasitic stages of nematode development. These nematode RALF-likes also possessed the typical activities of plant RALFs and bound to the extracellular domain of FERONIA, modulating certain steps of nematode parasitism related immune responses and cell expansion. Genetically, Both *MiRALF1* and *FERONIA* were required for the RKN parasitism in *Arabidopsis* and rice. Our study suggests that nematode-encoded RALFs facilitate parasitism via the plant-encoded FERONIA and provides a novel paradigm for studying host-pathogen interaction.

Keywords: RALF - FERONIA - Root-knot nematodes - *Arabidopsis thaliana*.

NemaWAARS: A motif to unveil mechanisms of parasitism gene regulation in the pinewood nematode as a target for disease control and plant resistance.

Margarida Espada[1] (mespada@uevora.pt), Cláudia Vicente[1], Jorge Faria[2], Paulo Vieira[3], Sebastian Eves-van den Akker[4], Lurdes Inácio[2], Manuel Mota[5]

[1] *NemaLab, MED, IIFA, Polo da Mitra, University of Évora, Évora, Portugal*; [2] *INIAV - Instituto Nacional de Investigação Agrária e Veterinária, Oeiras, Portugal*; [3] *Mycology & Nematology Genetic Diversity & Biology Lab, United States Department of Agriculture - USDA, Beltsville, United States of America*; [4] *Department of Plant Sciences, University of Cambridge, Cambridge, Portugal*; [5] *NemaLab, MED, Department of Biology, University of Évora, Évora, Portugal*

The interaction of plant-parasitic nematodes (PPN) with their hosts are mediated by parasitism proteins (effectors) that interact and/or modify host proteins to promote infection. NemaWAARS project focuses on the regulation mechanisms of parasitism genes and their control expression of the migratory endoparasite *Bursaphelenchus xylophilus* (the pinewood nematode). Supported by genomic and transcriptomic data previously established for this pathogen we have identified a DNA motif - STATAWAARS - associated in the promotor region of secreted pharyngeal gland cells coding effector genes. Given that this non-coding genetic signature unifies many sequences of unrelated effectors, it implies the existence of a potential major regulator(s), that binds to this sequence to orchestrate the expression of downstream effector genes. A similar master regulator has been recently proposed for other PPNs (Tylenchida) non-related to migratory nematodes or to this clade of nematodes (Aphelenchida). We hypothesize that by disrupting this regulator(s), it will be possible to simultaneously disrupt the expression of a large number of associated effector genes. This could be a new attractive target for host induced gene silencing, as switching-off the regulator of numerous effectors at the same time can reduce and inhibit the performance of this PPN. This project aims to identify transcription regulators associated with STATAWAARS motif and understand the impact of silencing enriched gland cells transcription factors on the PWN interaction with the host. The strategy in NemaWAARS will include innovative approaches to develop a better understanding of how the effectors are regulated, their function in the cellhost and use their sequences as potential targets for genetic editing towards an effective nematode resistance in the plant. The ability to explore host-delivered mechanisms against the pathogens could have an impact from a biotechnology standpoint in important forestry species.

Funding: This work is funded by National Funds through FCT - Foundation for Science and Technology under the Project PTDC/ASP-PLA/1108/2021.

Keywords: Gene regulation - Effectors - Pinewood nematode - Parasitism.

Discovery of novel stylet-secreted NLS-containing effector candidates from the soybean cyst nematode *Heterodera glycines.*

Anju Verma[1] (anju.verma@uga.edu), Rick Masonbrink[2], Tom Maier[2], Thomas J. Baum[2], Melissa G. Mitchum[1]

[1] *Department of Plant Pathology and IPBGG, University of Georgia, Athens, GA, United States*; [2] *Department of Plant Pathology and Microbiology, Iowa State University, Ames, IA, United States*

The soybean cyst nematode (SCN) *Heterodera glycines* releases stylet secretions from esophageal gland cells into host root tissues to aid penetration, migration, and the formation of a feeding site called a syncytium. These stylet secretions include a suite of effector proteins that play key roles in parasitism, including a handful of effectors that localize to the host nucleus to alter nuclear functions [1]. Prior *in silico* effector mining employing a dual prediction strategy from the transcriptome of early life stages of SCN identified a group of novel, putatively secreted effectors with a predicted nuclear localization signal (NLS) [2]. We cross-referenced this list to a gland cell-specific RNA-seq dataset [3] to prioritize a list of nine potential stylet-secreted NLS-containing effectors for further characterization. We were able to clone the corresponding full-length cDNAs for seven of the nine candidates. Transient expression of these seven effectors as GUS-GFP fusions without their predicted signal peptides by agroinfiltration in *Nicotiana benthamiana* leaves confirmed five of these seven effectors can localize to the plant nucleus. In addition, site-directed mutagenesis of the predicted NLS motifs validated a functional NLS in three of the five effectors. *In situ* hybridization confirmed the spatial expression of these effector genes within the dorsal esophageal gland cell. The temporal expression pattern of these effectors throughout the life cycle of SCN was determined by quantitative RT-PCR. In a BlastP analysis, three of the putative effectors had no homology to any other proteins in the database, one shared sequence similarity with an unnamed protein from *Meloidogyne enterolobii,* and the other shared some homology to plant transcriptional repressor proteins. Functional characterization of these putative effectors has the potential to uncover new insights into how cyst nematodes alter nuclear function to parasitize their hosts.

Keywords: Effectors - Nucleus - Soybean cyst nematode.

References:

- [1] Mejias et al., 2019. Front. Plant Sci. 10: 970.
- [2] Gardner et al., 2018. Sci. Rep., 8(1): 1-15.
- [3] Maier et al., 2021. Mol Plant-Microbe Interact. 34(9): 1084-1087.

The *Meloidogyne incognita* effector MiL648 contributes to nematode virulence through its interactions with OPR2 in tomato.

Ava Verhoeven[1] (ava.verhoeven@wur.nl), Anna Finkers-Tomczak[2], Pjotr Prins[2], Debbie Valkenburg-van Raaij[2], Koen Varossieau[2], Hein Overmars[2], Erik Slootweg[2], Mark Sterken[2], Aska Goverse[2], Geert Smant[2]

[1] *Laboratory of Nematology, Wageningen University & Research, Wageningen, Netherlands*; [2] *Wageningen University & Research, Wageningen, Netherlands*

Insight in the molecular basis of virulence can help to identify new and more durable sources of resistance to the root-knot nematode *Meloidogyne incognita* in tomato. In nematode genomes, evidence of positive, diversifying selection can point to loci involved in a molecular arms race with other organisms. We used evidence of positive selection as a first criterion to identify novel virulence genes in the genome of *M. incognita*, one of which encodes the effector MiL648. Bioassays with *M. incognita* on tomato plants overexpressing an RNAi construct matching the MiL648 sequence showed that this gene is required for nematode virulence. Likewise, tomato plants overexpressing MiL648 were more susceptible to *M. incognita*, demonstrating that it functions as a bona fide effector. We identified six likely host targets of MiL648 in a yeast two-hybrid screen of nematode-infected roots of tomato, including the 12-oxophytodienoate reductase SlOPR2. We confirmed the interaction between SlOPR2 and MiL648 *in planta* by transient expression and co-immunoprecipitation in leaves of *Nicotiana benthamiana*. We therefore concluded that the effector MiL648 enhances the virulence of *M. incognita* through its interactions with SlOPR2.

Keywords: Effector - Jasmonic acid - 12-oxophytodienoate reductase - Diversifying selection.

S31-PF1

The root-knot nematode effector Mj-NEROSS suppresses plant immunity by interfering with the ROS production in plastids.

Boris Stojilkovic (boris.stojilkovic@ugent.be), Yujin Chen, Xiang Hui, Godelieve Gheysen

Molecular biotechnology, Faculty of Bioscience Engineering, Gent, Belgium

Plant-parasitic nematodes produce effectors to overcome plant immunity and fine tune plant cellular processes. Molecular mechanisms of how effector proteins co-opt plant processes, especially plant immunity to support nematode survival, have been intensively investigated, but they are still poorly understood. Identifying protein-protein interactions is crucial for understanding this cross-kingdom network.

Using the high throughput screening technique Y2H-seq, we have identified tomato proteins involved in various cellular processes interacting with *M. javanica* effectors. Among those, Mj-NEROSS (**N**ematodes **e**ffector involved in **ROS s**uppression, previously referred to as 4D01 or Msp3) was found to interact with a Heavy metal transport/detoxification superfamily protein and a Rieske iron-sulphur protein (ISP), one of the putative subunits of the cytochrome b6f complex. Furthermore, we confirmed direct interaction *in planta* and showed that ISP is a conserved dicot target of Mj-NEROSS.

We showed that the plastid localization of Mj-NEROSS plays a crucial role in its interaction with ISP. Once this interaction was established, a significant decrease in electron transport rate and subsequently in the host's reactive oxygen species production was observed. Furthermore, we revealed that the presence of the effector in the plastids leads to changes in genes expression. Importantly, we showed that the differentially expressed genes (DEGs) were involved in ROS production, protein folding recognition, and upregulation of oxidative phosphorylation.

We hypothesized that DEGs are most likely subsequent consequences of interaction of Mj-NEROSS with ISP and biochemical communication between plastids and the nucleus, leading to suppression of plant basal defense and attenuation of host resistance to the nematode infection.

Keywords: ROS - ISP - Plastid - Protein-protein interactions.

S31-PF2

Studying root-knot nematode *Meloidogyne javanica* MAP-1 variation and their implication in parasitism.

Anil Kumar (anilmalik58@gmail.com), Patricia Bucki, Sigal Braun Miyara

Department of Entomology and the Nematology and Chemistry Units, Agricultural Research Organisation, Rishon lezion, Israel, Israel

Root-knot nematodes (RKNs) are among the most detrimental plant parasites and present a major threat to crop productivity worldwide. These nematodes deliver the effector proteins into host cells to suppress plant immunity that play a key role in successful parasitism. However, their precise mechanism of action remains largely unknown. Previous studies showed that one such effector protein, *Meloidogyne* Avirulent Protein (MAP-1) produced within the amphids of *M. incognita* and was secreted into the plant apoplasm during early stage of infection, although was still lacking functional validation. In the present study, two orthologs of MAP-1 were identified in *Meloidogyne javanica (MjMAP-1a* and *MjMAP-1b)*. These proteins encoded highly conserved repeats of 58 and 13 aa motifs in its internal part. The 58 aa motif was repeated twice in *MjMAP1a*, and once in *MjMAP-1b*, respectively. The 13 aa motif was repeated four times in the *MjMAP-1a* and one time in *MjMAP-1b* sequence. Previously it was suggested that variations in the number and arrangement of *MAP-1* repeats correlated with nematode (a) virulence against the tomato *Mi-1* resistance gene. Fluorescence *In situ* hybridization studies showed localization of all *MjMAP-1* transcripts specifically to the subventral pharengeal gland cell of pre- parasitic J2. Furthermore, we performed gene expression studies in four different developmental stages (egg, J2, 2 and 10 dpi) of *M. javanica* using qRT-PCR and observed that *MjMAP-1a* and *b* were highly expressed in the J2 stage compared to other stages. Using an *Agrobacterium*-mediated transient expression system and plant immune response assays, we demonstrated that *MjMAP-1a* and *MjMAP-1b* localized in the plant cells endoplasmic reticulum and golgi organelles and could suppress Gpa2/RBP-1- induced cell death in *Nicotiana benthamiana*. An InterPro scan identified a Barwin-like endoglucanase domain in *MjMAP-1* protein, suggesting their function as pathogenicity factors involved in manipulating the host plant. Therefore, here we hypothesized that repetitive region of *MAP-1* may be involved in physical interactions with plant ligands and may play a role in the specificity of the plant-nematode interaction. Further studies are underway to test this hypothesis.

Keywords: MAP-1 - *Meloidogyne javanica* - Fluorescence *In situ* hybridization - Root knot nematode - Effector.

S31-PF3

The *M. javanica* effector Mj-10A08 downregulates ethylene receptors via protein-protein interactions to facilitate tomato parasitism.

Yujin Chen[1] (Yujin.Chen@Ugent.be), Boris Stojilković[2], Godelieve Gheysen[2]

[1] *Molecular Biology, Faculty of Bioscience Engineering-Ugent, Ghent, Belgium*; [2] *Faculty of Bioscience Engineering-Ugent, Ghent, Belgium*

Meloidogyne javanica is one of the most frequently reported species in the root-knot nematodes responsible for damage to many different crops worldwide, especially tomato (*Solanum lycopersicum*), which is a very important source of nutrition in the human diet. Recent studies have declared that root-knot nematodes modulate the host immune responses to enhance parasitism via hundreds of effectors secreted from their pharyngeal glands, while molecular mechanisms underlying the sophisticated protein-protein interactions remain poorly understood. Studying the interactome in tomato roots will provide crucial insight for unraveling these important biological issues across the plant and nematode kingdoms.

We have identified tomato proteins involved in diverse plant processes interacting with *M. javanica* effectors employing high throughput screening techniques such as Y2H-seq and TurboID-MS. Mj-10A08 was identified to interact with Unknown Protein (possibly ankyrin-like protein) as demonstrated by yeast two-hybrid. The proteins colocalize in the plasma membrane or endomembrane system in *Nicotiana benthamiana* leaves after agro-infiltration, which also matched with the localization of Mj-10A08 in *S. lycopersicum* when expressed in hairy roots. Moreover, Mj-10A08 suppresses INF-induced cell death and ROS production in plant immune response assays. With the help of RNA-Seq on hairy root lines overexpressing Mj-10A08 and quantitative reverse transcription PCR analyses, we found that Mj-10A08 is capable of interfering with the host hormone signaling pathways via downregulation of ET-responsive and JA-responsive genes.

We hypothesize that DEGs could be consequences of interaction networks between Mj-10A08, ankyrin-like proteins and ethylene receptors. In addition, based on the ectopic expression of Mj-10A08 in *E. coli* and hairy roots, we expected that Mj-10A08 could undergo potential post-translational modifications *in planta,* which could inhibit plant innate immune responses and hijack relevant signaling pathways for promoting parasitism.

PLENARY SESSION

Multi-criteria assessment in agriculture: how can agroecology help?

Anne Mottet (anne.mottet@fao.org), Abram Bicksler, Dario Lucantoni, Rassoul Sy

Food and Agriculture Organization of the United Nations, Rome, Italy

Agroecology is advancing globally as a science, a practice, and a social movement that is knowledge intensive, contextual, and incorporates co-creation and sharing of knowledge. The need for harmonized cross-cutting evidence on agroecology was a systematic recommendation from the various global and regional consultations on agroecology conducted by FAO between 2014 and 2018 and from FAO's governing bodies. To answer this need, FAO, in partnership with many different stakeholders, develop the Tool for Agroecology Performance Evaluation (TAPE), which is a comprehensive tool that aims to measure the multi-dimensional performance of agroecological systems across the different dimensions of sustainability. It applies a stepwise approach at the household/farm level but it also collects information and provides results at a community and territorial scale. To date, TAPE has been used in over 30 countries globally, by a range of stakeholders, and on over 4,500 farms/households. TAPE provides a multi-criteria diagnostic on how far systems are in the agroecological transition and what their impacts are on different dimensions of sustainability. The 10 elements of agroecology proposed by FAO and approved by member nations are used to establish the diagnostic. 10 core criteria of performance are then assessed to measure the impact of systems, namely: land tenure, productivity, income, added value, exposure to pesticides, dietary diversity, women's empowerment, youth employment, biodiversity, and soil health. Preliminary results show that in a variety of contexts and countries, production systems that are more advanced in their agroecological transition also have better scores on the different dimensions of sustainability. These results can also help identify similarities in how to support the transition, including in terms of governance and enabling environment in general.

Keywords: Agroecology - Multi-criteria analysis - Sustainability assessment - Global.

POSTERS

S1. Plant-nematode interactions

S1-P01

Daf16like and *Skn1like* genes are reliable targets to develop biotechnological tools for control of *Meloidogyne incognita.*

Inra Antipolis[1] (isabelatl@gmail.com), Isabela Lourenço-Tessutti[2], Marcos Fernando Basso[3], Reneida Mendes[4], Clidia Moreira Pinto[4], Caroline Bournaud[5], François-Xavier Gillet[5], Roberto Togawa[6], Leonardo Pepino De Macedo[6], Maria Cristina Mattar Da Silva[6], Janice Almeida Engler[2], Maria Fátima Grossi De Sá[3]

[1] *IPN, INRAe, Sophia Antipolis, France*; [2] *INRAe, Sophia Antipolis, PACA, France*; [3] *EMBRAPA, Brasilia, DF, Bresil*; [4] *EMBRAPA/UNB, Brasilia, DF, Bresil*; [5] *CNRS, Université de Grenoble, Grenoble, France*; [6] *EMBRAPA, Brasilia, Bresil*

Meloidogyne incognita is a plant-parasitic root-knot nematode (RKN, PPN) responsible for causing damage to several crops worldwide. In *Caenorhabditis elegans*, the DAF-16 and SKN-1 transcription factors (TFs) orchestrate aging, longevity, and defense responses to several stresses. Here, we report that *MiDaf16-like1* and *MiSkn1-like1*, which are orthologous to DAF-16 and SKN-1 in *C. elegans,* and some of their targets, are modulated in *M. incognita* J2 during oxidative stress or plant parasitism. RNAi technology was applied for the stable production of siRNAs *in planta* to downregulate the *MiDaf16-like1* and *MiSkn1-like1* genes of *M. incognita* during host plant parasitism. *Arabidopsis thaliana* and *Nicotiana tabacum* overexpressing a hairpin-derived dsRNA targeting these genes individually (single-gene silencing) or simultaneously (double-gene silencing) were generated. T2 plants were challenged with M. *incognita* and the number of eggs, galls, and J2, and the nematode reproduction factor (NRF) were evaluated. Data indicate that *MiDaf16-like1*, *MiSkn1-like1* and some genes from their networks are modulated in *M. incognita* J2 during oxidative stress or plant parasitism. Transgenic *A. thaliana* and *N. tabacum* plants with single- or double-gene silencing showed significant reductions in the numbers of eggs, J2, and galls, and in NRF. Additionally, the double-gene silencing plants had the highest resistance level. Gene expression assays confirmed the downregulation of the *MiDaf16-like1* and *MiSkn1-like1* TFs and defense genes in their networks during nematode parasitism in the transgenic plants. All these findings demonstrate that these two TFs are potential targets for the development of biotechnological tools for nematode control and management in economically important crops.

Keywords: Root-knot nematodes - Oxidative stress - siRNA crosstalk - Nematode management.

S1-P02

Determining genes conferring resistance against nematodes in *Capsicum* spp and relationship with capsaicin.

Refik Bozbuga[1] (refikbozbuga@gmail.com), Kazım Mavi[2]

[1] *Faculty of Agriculture, Plant Protection Department, Eskisehir Osmangazi University, Eskisehir, Turkey*; [2] *Horticulture Department, Mustafa Kemal University, Hatay, Turkey*

Root knot nematodes belonging to *Meloidogyne* genus are important parasites cause damage on numerous plant species including crops. The nematode causes cellular and molecular changes in the feeding site within the roots of hosts. It is difficult to control root knot nematodes economically, thus, growing resistant plant varieties is one solution to cope with them. Pepper is an important agricultural products and has high economic importance for the producers, but the nematode limits the production. Breeding studies are carried out in order to increase the quality and quantification of pepper production and the new plant hybrids obtained in the breeding works can be sensitive to some diseases and pests including *Meloidogyne* species despite the high yield. *Meloidogyne incognita* is one of the most common and damaging nematode species to pepper plants. Different resistance genes have been identified in pepper. Some pepper genotypes including wild types may carry resistance genes which can be determined using different molecular markers. However, it is not fully understood how carrying resistance genes in *Capsicum* varieties is related to capsaicin. This study was aimed at determining the resistance genes (*Mech1*, *Mech2*, *Me3*, *Me4* and *Me7*) against nematode using multiple molecular markers in *Capsicum* spp. In addition, capsaicin (8-methyl-N-vanillyl-6-nonenamide) analyses was achieved to determine the relationship with resistances genes. Results revealed that most of the *Capsicum* genotypes and varieties were susceptible to nematodes based on both molecular and screening assays. A relationship between capsaicin and nematode resistance was also derived in this study. It can be concluded that pepper species with resistance genes should be the main material of the next breeding activities.

Keywords: Resistance genes - Capsicum spp - Capsaicin - Molecular markers - Meloidogyne.

S1-P03

Vertical movement of *Meloidogyne enterolobii* as influenced by temperature and plant stimuli.

Ana Karina Oliveira[1] (janetebrito@fdacs.gov), Elvira Pedrosa[1], Diego Leitão[1], Janete Brito[2], Ênio Silva[1], Donald Dickson[3]

[1] *Agricultural Engineering Department, Federal Rural University of Pernambuco, Pernambuco, Recife, Brazil*; [2] *Nematology, Florida Dept. Agriculture and Consumer Services/University of Florida, Gainesville, Florida, United States*; [3] *Nematology and Entomology, University of Florida, Gainesville, Florida, United States*

Meloidogyne enterolobii is a highly virulent root-knot nematode species that affects major agricultural crops worldwide. Our objective was to determine the movement of second-stage juveniles (J2) of *M. enterolobii* vertically in 14-cm long segmented PVC soil columns (three 4-cm long ×4.4-cm internal diameter rings connected and placed on top of a 2-cm long injection ring) towards two plant stimuli, tomato 'Cobra' (*Solanum lycopersicum* L.) and French marigold 'Petite' (*Tagetes patula* L.) at two temperatures (20 and 26ºC). Seedlings were transplanted into Styrofoam cups attached to the top of each column; seedling free columns were used as a control. Freshly hatched J2 (1000/column) were injected into a hole located 1 cm from the bottom of each column and placed in environmental control chambers. The columns were dismantled at 3, 6, 9 or 12 days after nematode injection (DAI). At each harvest, J2 were extracted from soil of each ring of the columns and cups, then counted to compare their distribution; also root systems of both tomato and marigold were collected, weighed, and stained to determine the number of J2 inside roots, if any. *Melodogyne enterolobii* moved more than 13 cm vertically 3 DAI regardless of temperature and plant stimuli, even in the control. Nematode vertical movement was greater at 26ºC, where 60% of active J2 were found at distances greater than 13 cm 12 DAI. Temperature did not affect J2 root penetration. The number of J2 was ca. three-fold greater in tomato roots than in French marigold roots.

Keywords: Behavior - Meloidogyne enterolobii - Migration - Plant stimuli.

References:

- Dávila-Negrón and Dickson, 2013. Nematropica 43(2): 152-163
- Dutta et al., 2011, Nematology 13 (5): 509-520.

S1-P04

Microscopy and organismal associations of *Litylenchus crenatae* in beech leaf disease of *Fagus grandifolia* in Ohio, USA.

Lynn Carta[1] (lynn.carta@usda.gov), Zafar Handoo[1], Mihail Kantor[1], Gary Bauchan[2], Ronald Ochoa[3], David Burke[4]

[1] *Mycology & Nematology Genetic Diversity & Biology Laboratory, USDA-ARS NEA BARC, Beltsville, MD, United States*; [2] *Electron and Confocal Microscopy Unit, USDA-ARS NEA BARC, Beltsville, MD, United States*; [3] *Systematic Entomology Laboratory, USDA-ARS NEA BARC, Beltsville, MD, United States*; [4] *Holden Arboretum, Cleveland Botanical Garden, Kirtland, OH, United States*

American beech trees are a major component of many deciduous forest areas in eastern North America, and a new disease was discovered affecting beech near Cleveland, Ohio in 2012. Beech leaf disease (BLD) caused striped, galled leaf tissue, canopy thinning and sometimes young tree mortality. Scientists from the US and Canada demonstrated that invasive *Litylenchus crenatae* nematodes from diseased leaves could cause BLD symptoms on seedlings primarily through bud inoculation. Low-temperature scanning electron microscopy images of the nematodes from symptomatic bud and leaf tissue revealed that nematodes filled spaces around spongy parenchyma. Nematodes entwined around the bodies of mites were imaged, and an inventory of mites was made at the Holden Arboretum near Cleveland. Bird mites were also collected and imaged because a subtractive molecular microbiome survey showed different bacterial communities between symptomatic and asymptomatic leaves, including the presence of the intracellular endosymbiont *Wolbachia* on symptomatic leaves. Recovered *Wolbachia* sequences showed high affinity to bacteria isolated from a quill mite associate of the rose finch family of birds. One finch is known to consume beech buds in North America but not in Asia. Birds, mites and *Wolbachia* may be important for nematode distribution and disease etiology. The disease is spreading primarily eastward and identifying agents of transmission is critical to stop BLD progression by foresters and plant pathologists developing integrated controls against BLD.

Keywords: Anguinata - Fagaceae - Invasive species - Disease transmission - Scanning electron microscopy.

S1-P05

Characterising tolerance to root knot nematodes in robusta coffee (*Coffea canephora*).

Adam Casey[1] (bsacas@leeds.ac.uk), James McCarthy[2], Peter Urwin[1]

[1] *University of Leeds, Leeds, United Kingdom*; [2] *Nestlé Research, Tours, France*

A detriment to coffee production is the damage caused by plant parasitic nematodes, which reduce yields by up to 15% although individual growers may experience much higher reductions. The most damaging species are root-knot nematodes (*Meloidogyne* spp.) that modify and feed on living plant cells, causing a major nutrient sink that diverts resources from the host plant. We are investigating both the tolerance of new coffee varieties against root-knot nematodes and the molecular components which contribute to tolerance and susceptibility. We have characterised the physiological response, and susceptibility of new *Coffea canephora* (robusta) varieties to root-knot nematode infection. While all varieties tested were susceptible to *M. incognita* and *M. paranaensis*, only three of the five varieties suffered a significant impact on photosynthesis, as measured by a reduction in chlorophyll fluorescence, and a significant reduction in growth parameters. An RNA-Seq approach was then used to analyse differential gene expression between the two varieties with the most contrasting susceptibility and tolerance in the presence and absence of root-knot nematode infection. Different sets of genes in the two varieties were regulated in response to nematode infection, and within a variety there were also differences in gene regulation between short term (1 week post inoculation) and long term (12 weeks post inoculation) infection. Genes with annotated roles in general defence and hypersensitive responses were up regulated in the susceptible and intolerant coffee variety at both one and 12 wpi, but were down regulated in the tolerant coffee variety 12 wpi. Genes involved in cell wall biosynthesis and organisation were also differently regulated between the tolerant and intolerant varieties, suggesting a role for the cell wall in the host-plant pathogen interaction, as well as in maintaining health and vigour of the host plant despite infection. Finally, multiple genes that have putative disease resistance roles in other plant-pathogen interactions were upregulated in the tolerant coffee variety. Following functional analysis, these genes and their associated biological processes can offer insight as to what mediates resistance and tolerance to root-knot nematodes in coffee. These findings will better inform growers on the best attributes for new coffee varieties, minimising damage caused by plant parasitic nematodes.

Keywords: Coffee - Root-knot nematode - Susceptibility - Transcriptomics - Cell wall.

S1-P06

Nodal vine cutting technique for assessing nematode resistance in yams.

Abiodun Claudius-Cole[1] (bi_cole@yahoo.com), Temitayo Omotayo[1], Antonio Lopez-Montez[2]

[1] *Dept. of Crop Protection and Env. Biology, University of Ibadan, Ibadan, Oyo, Nigeria*; [2] *Yam breeding, International Institute of Tropical Agriculture, Ibadan, Oyo, Nigeria*

Identifying resistance to plant-parasitic nematodes in yams has been a major challenge in yam breeding programmes due partly to insufficient planting material (tubers) for evaluation. Effective inoculum level was evaluated on nodal vine cuttings of a susceptible yam cultivar, TDr 93-31 with 0 (control), 200, 500, 1000 and 2000 eggs/individuals each of *Meloidogyne incognita*, *Scutellonema bradys* and *Pratylenchus brachyurus*. The efficiency of nodal cuttings were evaluated on two cultivars each of *Dioscorea rotundata*, *D. alata* and *D. dumetorum* compared to conventional minisett method of evaluation. Planted vines and minisetts were inoculated with 500 and 5000 eggs/individuals respectively. The experiments were laid out in a randomized complete block and split plot design respectively, with four replications each. Data were collected on percentage vine survival, fresh root and tuber weight, and nematode population and damage. Vine survival was not significantly different between control and inoculated vines. Nematode population increased with increase in inoculum density. All inoculum levels induced damage in tubers. Maximum damage rating (>60%) was observed with initial inoculum of 1000 eggs of *M. incognita* and 200 individuals of *S. bradys* and *P. brachyurus*. The two cultivars of *D. alata* and *D. rotundata* developed visible symptoms and supported nematode reproduction while the *D. dumetorum* cultivars did not. The reaction of yam cultivars planted as vines were consistent with results from conventional minisett method. The lowest inoculum of 200 egg/juveniles was sufficient to elicit a reaction from vines in a vertical sac system.

Keywords: Dioscorea - Screening methods - Vine cutting - Resistance - Scutellonema.

References:

- Claudius-Cole et al., 2017. Journal of Agriculture and Rural Development in the Tropics and Subtropics 118(2):297-306.
- Coyne et al., 2006. Field Crops Research 96(1), 142-150.
- Shiwachi et al., 2005. Tropical Science 45 (4):163–69

S1-P07

Determining the distribution of *Pratylenchus quasitereoides* and *Pratylenchus curvicauda* in the WA Wheatbelt and understanding how they find host roots.

Rhys Copeland[1] (31553633@student.murdoch.edu.au), John Fosu-Nyarko[1], Michael Jones[1], Sarah Collins[2]

[1] *Crop Biotechnology Research Group, Murdoch University, Murdoch, WA, Australia*; [2] *Department of Primary Industries and Regional Development, South Perth, WA, Australia*

Root-lesion nematodes (RLN) (*Pratylenchus* spp.) are amongst the top three most economically important plant parasitic nematodes in the world. They have a wide host range and attack major crops such as wheat and barley, annually causing between 15 and 50% yield losses in Western Australia (WA) [1]. WA's major grain production area, known as the 'Wheatbelt', covers seven million hectares across its southwest. A 2017 survey found that 79% of soil samples from the WA Wheatbelt had significant levels of RLN infection, with 57% being yield-limited due to the presence of different species of the nematodes [2]. When designing and implementing effective pest management strategies for RLNs, it is important to identify the species present accurately. *Pratylenchus curvicauda* was recently characterised from soils in the Wheatbelt by Begum *et al*. (2019) [3]. The soil samples from which *P. curvicauda* was identified were presumed to also contain *Pratylenchus quasitereoides*. However, Begum *et al*. (2019) found that *P. curvicauda* was the predominant species. Most of the biology of either *P. quasitereoides* or *P. curvicauda*, for example, their distribution in the WA Wheatbelt, culturing under sterile conditions *in vitro* for easier experimentation and how they find their hosts, is still unknown. For the first time, this project has established that both *P. quasitereoides* and *P. curvicauda* exist in the WA Wheatbelt. Gas Chromatography-Mass Spectrometry analysis of root exudates from a susceptible and a resistant wheat cultivar grown in WA has indicated the presence of organic chemicals possibly responsible for attraction or repulsion of the nematodes to the roots or for successful infection. Also, to lay the foundations for developing a new approach to controlling RLNs in grain crops, Next Generation Sequencing and electron microscopy will be used to study how the neurons in the amphids are employed by nematodes during interactions with host plants.

Keywords: Pratylenchus - Quasitereoides - Curvicauda - Plant-nematode interactions - Host attraction.

References:

- [1] Collins et al. 2013. DAFWA Bulletin.
- [2] Chambers et al. 2018. GRDC Research Updates
- [3] Begum et al. 2019. Journal of Nematology. 51(1)
- [4] Hodda et al. 2014. Zootaxa. 3866(2): 277-88

S1-P08

HANDLER - Host adaptation and root-knot nematode selection by partially-resistant tomato rootstocks.

Sofia R. Costa[1] (sofia.costa@bio.uminho.pt), M. Clara Vieira dos Santos[1], M. Teresa M. Almeida[1], João M. Rocha[2], Isabel Conceição[3], Soraia Perpétuo[3], A. Rita Gonçalves[1]

[1] *CBMA - Centre of Molecular and Environmental Biology, Department of Biology, University of Minho, Campus de Gualtar, Braga, Portugal*; [2] *LAQV, REQUIMTE, Department of Chemistry and Biochemistry, Faculty of Sciences, University of Porto, Porto, Portugal*; [3] *Centre for Functional Ecology, Department of Life Sciences, University of Coimbra, Coimbra, Portugal*

Root-knot nematodes (RKN, *Meloidogyne* spp.) are responsible for significant yield losses worldwide, reaching up to 100% in tomato crops. Among management options, grafting is increasingly used by tomato farmers. Several commercially available rootstocks contain the *Mi* gene that confers resistance to *M. arenaria*, *M. incognita* and *M. javanica* but not to *M. hapla;* however, control success depends on temperature [1]. The HANDLER project (POCI-01-0145-FEDER-029283; PTDC ASP-PLA/29283/2017) aims to assess, in a quantitative method, the selective pressure exerted by tomato rootstocks and the dynamics of virulence development in RKN populations. The involvement of ascarosides, nematode communication signals [2] in these processes will be investigated. In the initial stage of HANDLER, we tested cv. Embajador, one of the most widely used rootstocks in Portugal to assess claims of intermediate resistance to *M. arenaria*, *M. incognita* and *M. javanica*. A pot trial was run in controlled conditions with single egg mass isolates of the 4 most common RKN species. Susceptible tomato cv Tiny Tim was used as control, and each treatment had 5 replicates. Sixty days after inoculation, plant damage, RKN reproduction factor (Rf), reproduction index (RI) and female fecundity (Ff) were determined. Contrary to expected effects, the rootstock was completely resistant to *M. arenaria* and susceptible to *M. javanica* (root damage and reproduction comparable to control). Furthermore, this rootstock was partially resistant not only to *M. incognita* but also to *M. hapla,* with the RI, RF and Ff being significantly lower than in the control. These results emphasize the need to assess host-RKN interactions to support informed decisions by seed companies, nurseries and farmers in rootstock selection.

Keywords: Grafting - Reproduction index - Root-knot nematodes - Rootstock - Virulence selection.

References:

- [1] Williamson, 1998. Annu Rev Phytopathol 36: 277-93.
- [2] Manosalva et al., 2015. Nat Commun 6: 7795.

S1-P09

Biochemical aspects of plant sterols in plant nematode interaction.

Paul Dahlin (paul.dahlin@agroscope.admin.ch), Laurin Müller, Alessandro Cabianca, Andrea Ruthes

Plant Protection, Agroscope, Wädenswil, Switzerland

Unlike mammals, plants produce a mixture of sterols, phytosterols, including sitosterol, stigmasterol and campesterol, differing from cholesterol by an alkylation at C24 at the side chain. These biomolecules belong to the family of isoprenoids and are synthesized from the five-carbon precursor isopentyl-pyrophosphate and its isomer dimethylallyl-pyrophosphate. Subsequent metabolic reactions and modifications generate an enormous diversity and complexity of sterols. For multicellular organisms, sterols represent essential organic compounds for growth and reproduction. Differing from plants that have cholesterol as a minor component, mammals and other vertebrates have cholesterol as the main cell membrane sterol, while fungi commonly synthesise ergosterol. For nematodes, sterols, in particular cholesterol, are necessary components of the membrane, which they are not able to synthesize *de novo* and need to acquire from their feeding source (Chitwood 1999). During our research, mainly on tomato plants, the phytosterol composition, after root-knot nematode (*Meloidogyne incognita*) infection was analysed by gas chromatography–mass spectrometry. Free sterols of nematode infested and non-infested plants revealed significant changes in plant sterol composition, with a relative reduction of stigmasterol and an increase of sitosterol after *M. incognita* infection. These results were enhanced in the root galls caused by the nematodes. Generally, the major plant sterols, sitosterol, stigmasterol and campesterol, are believed to have different physiological functions, e.g. stigmasterol biosynthesis in *Arabidopsis thaliana* was described to be stimulated after bacterial and fungal infection, which appears to be contrary to *M. incognita* infection on tomato plants.

Keywords: Sterol - Root-knot nematode - Sitosterol - Stigmasterol - Meloidogyne.

References:

- Chitwood, D. J. (1999). Critical reviews in biochemistry and molecular biology, 34(4), 273-284.

S1-P10

Molecular markers for cell damage induced by root-knot nematodes.

Cleberson Ribeiro[1] (janice.de-almeida@inra.fr), Helkin Forero Balesteros[1], Bruno Paes de Melo[1], Karla Gonçalves[1], Adriana Hermely[2], Maria Fátima Grossi de Sá[3], Lieven De Veylder[4], Janice de Almeida Engler[1]

[1] *IPN, INRAe, Sophia Antipolis, PACA, France*; [2] *LBMP, UFRJ, Rio de Janeiro, Rio de Janeiro, Brazil*; [3] *LIMPP, Embrapa - Cenargen, Brasília, DF, Brazil*; [4] *PSB, PSB-VIB Ghent University, Ghent, Belgium*

Galls induced by plant parasitic root-knot nematodes are predominantly situated in the root vascular cylinder containing cells which maintain the competence for division and differentiation to other cell types. Herein, we questioned if during the process of nematode penetration and infection, cell damage might occur, finally triggering cellular dedifferentiation and division. It appears that the Ethylene Response Factor 115 (ERF115 of the ERF family), together with the Phytochrom A Signal Transduction 1 (PAT1 of the GRAS family) transcription factors (TFs) are able to activate a regeneration program through the induction of a stem cell-like fate in gall cells. The ERF115 TF is a potent element controlling the plant regeneration process. Upon stem cell death, cells that co-express *ERF115* and *PAT1* were found to engage recovery cell divisions [1]. Moreover, plants lacking a functional ERF115 or PAT1 showed a reduced ability to perform recovery divisions and displayed a lower regeneration frequency [2]. Our studies demonstrated that using the propidium iodide stain cell damage was located nearby cells expressing the *ERF115* as well as *PAT1*. Both TFs also are present in cells adjacent to the nematode and of giant cells potentially activating cell division as part of a regeneration program. These functional studies lead us to conclude that both ERF115 and PAT1 are most likely involved in gall homeostasis sensing the damage caused upon nematode infection and working in its replenishment.

Keywords: Cell damage - Tissue regeneration - Root-knot nematode - Galls.

References:

- [1] Heyman et al., 2013. Science 342: 860 – 863.
- [2] Heyman, et al., 2016. Nature Plants 2: 1 – 7.

S1-P11

A multi-state fffort to contain and mManage *Meloidogyne enterolobii* in vegetable crops.

Jennifer Corbin[1] (jcorbin@clemson.edu), Paula Agudelo[1], Johan Desaeger[2], Adrienne Gorny[3], Zane Grabau[4], Zhengfei Guan[2], Abolfazl Hajihassani[5], John Mueller[1], Lina Quesada-Ocampo[3], Will Rutter[6], Phillip Wadl[6]

[1] *Department of Plant and Environmental Sciences, Clemson University, Clemson, SC, USA*; [2] *University of Florida Gulf Coast Research and Education Center, Wimauma, FL, USA*; [3] *Department of Entomology and Plant Pathology, North Carolina State University, Raleigh, NC, USA*; [4] *Department of Entomology and Plant Pathology, University of Florida, Gainesville, FL, USA*; [5] *Department of Entomology and Plant Pathology, University of Georgia, Tifton, GA, USA*; [6] *USDA-ARS, Charleston, SC, USA*

FIND*Me* (Focused Investigations on the Distribution and Management of *Meloidogyne enterolobii*) is a project sponsored by the USDA Specialty Crop Research Initiative. Research partners include Clemson University, North Carolina State University, the University of Georgia, USDA-ARS and the University of Florida. The spread of *Meloidogyne enterolobii* (*M.e.*), a highly virulent root-knot nematode (RKN) species, is potentially devastating to specialty crop production in the southeastern United States. This species can cause losses in yield and quality, and its quarantined status jeopardizes interstate and international trade. *M.e*. can infect and damage crop genotypes that are resistant to the other major species of RKN, including sweetpotato. Our goal is to reduce the vulnerability of growers to the emerging agricultural threat posed by *M.e.* by using a systems-based approach involving five research and extension objectives: 1) Study the prevalence and distribution of *M.e*. in vegetable crops in the Southeast, and characterize the genetic variability encountered; 2) Evaluate and develop vegetable germplasm with resistance to *M.e*.; 3) Evaluate the efficacy of nematicides, cover crops, and rotations as management strategies; 4) Assess the costs and returns of management tactics such as rotations, cover crops, and nematicides for the mitigation of *M.e*. on sweetpotato, cucumber, watermelon and tomato crops; 5) Develop print and web-based educational materials on management and containment strategies for *M.e*. It is critical for commercial growers and home gardeners to learn about *M.e.* so they can prevent and manage the problem.Systematic surveying of symptomatic crops has led to identification of *M.e.* in FL, GA, SC, and NC.

S1-P12

Acacia biochar promotes tolerance of tomato plants' to root-knot nematodes.

Zakheleni Dube[1] (zakheleni.dube@ump.ac.za), Lambert Letsoalo[2], Dineo Mcunu[2], Mbokota Khosa[3]

[1] *School of Biology&Environmental Sciences, University of Mpumalanga, Mbombela, South Africa*; [2] *School of Agriculture, University of Mpumalanga, Mbombela, South Africa*; [3] *Tropical and Subtropical Crops, Agricultural Research Council, Mbombela, South Africa*

Biochar is a carbon-rich product of thermochemical conversion of biomass in an oxygen-limited environment. Biochars has been developed and studied for their effects in promoting plant growth, soil mobilisation of heavy metals and currently, reduction of crop fungal and bacterial pathogens. The effectiveness of biochar in all the reports has been strongly linked to source of biomass used in its production. There is little information on the effects of biochar as a soil amendment on plant parasitic nematodes and growth of nematode infected plants. The objective of this study was therefore to determine the effects of acacia biochar on population densities of *Meloidogyne incognita* and growth of nematode infested tomato plants under greenhouse conditions. Acacia biochar rates of 0, 5, 10 and 20 t/ha were applied in 22-cm plastic planting pots containing loam soil two weeks before uniform two-week-old tomato cv. "Floradade" seedlings were transplanted. Two weeks after transplanting, tomato seedlings were each inoculated with 3 000 *M. incognita* second-stage juveniles. Treatments were replicated 8 times in a randomized complete block design. Fifty-six days after transplanting, plant height, stem diameter, shoot mass, root mass, root galls, nematodes in roots and soil were measured. Acacia biochar amendments increased both plant growth and nematode infestations. Relative to control of no biochar, plant growth was increased by between 44 and 78%, whereas, nematode infestation increased by between 29 and 43%. In conclusion, even though *M. incognita* increased with biochar application, plant where able to tolerate the increase by increasing their growth.

Keywords: Nematode suppression - Organic amendments - Pyrolysis - Plant resistance - Soil borne diseases.

S1-P13

Heat stability of resistance in tomato breeding lines to *M. incognita* and *M. javanica* populations under increasing soil temperature.

Awol Seid Ebrahim[1] (awolseid07@gmail.com), Chemeda Fininsa[1], Tesfamariam Mengistu Mekete[2], Wim M.L. Wesemael[3], Wilfrida Decraemer[4]

[1] *School of Plant Sciences, Haramaya University, Dire Dawa, Oromia, Ethiopia*; [2] *Departement of Entomology and Nematology, University of Florida, Florida, USA*; [3] *Crop Protection, ILVO, Ghent, Belgium*; [4] *Department of Biology, Ghent University, Ghent, Belgium*

In tomato, the only commercially available source of resistance to root-knot nematodes (RKN) is the *Mi-1* gene that confers resistance to *Meloidogyne incognita*, *M. javanica* and *M. arenaria*. However, its effectiveness was limited at higher soil temperatures. A study was initiated with the objective to check the durability of the potential resistance genes found in some tomato breeding lines after screening in controlled glasshouse conditions ≤ 27°C by exposing them to higher soil temperatures at 28, 32 and 36°C for 24 and 48 h periods. The aggressive Jittu and Babile *M. incognita* and Jittu and Koka *M. javanica* populations originally collected from Ethiopia were used. When seedlings reached the four-leaf stage, each tube was inoculated with 50 freshly (≤ 24 h) hatched infective second-stage juveniles (J2). Immediately after inoculation, the seedlings were exposed continuously for 24 and 48 h in a warm water bath at 28, 32 and 36°C, respectively. A control was kept separately in ambient temperature (24°C ± 2°C). The external ambient temperature and the soil temperature inside the tube while in the water bath were simultaneously recorded using a TESTO data logger. Temperature, tomato breeding lines and time had a significant effect on the number of J2 of Jittu and Babile *M. incognita* and Jittu and Koka *M. javanica* populations that penetrated the roots. The utility of the potential resistance found in the breeding lines during the controlled growth chamber resistance screening experiment was limited at higher soil temperatures, especially at 32 and 36°C. At 36°C there was no significant difference found on the mean number of penetrated J2 of Jittu and Babile *M. incognita* and Jittu and Koka *M. javanica* populations inside the roots of all the tested breeding lines compared to 'Marmande' (a susceptible control) after 48 h of heat exposure after inoculation. More J2 were found in the roots of the tested breeding lines after 48 h compared to 24 h heat exposure after inoculation for each soil temperature level tested and for both populations of *M. incognita*. It is clear from our observations that local tomato breeding lines with resistance potential can be used when soil temperatures remain below 32°C. Differences were observed between breeding lines depending on the RKN population used at higher temperatures and this knowledge can help in further optimising the development of sustainable resistance under local Ethiopian circumstances.

Keywords: Climate change - Exposure time - Management - Mi-1 - Meloidogyne species.

S1-P14

The development of root-lesion nematodes (*Pratylenchus thornei*, *P. neglectus*, *P. penetrans*) on some chickpea varieties.

Ibrahim halil elekcioğlu[1] (halile@cu.edu.tr), Ece börteçine kasapoğlu uludamar[1], Irem ayaz[1], Tohid Behmand[2]

[1] *Plant protection department, cukurova university, ADANA, Turkey*; [2] *Plant protection department, agriculture faculty, cukurova university, ADANA, Turkey*

Population development and reproduction rates of root lesion nematodes, *Pratylenchus thornei*, *Pratylenchus neglectus* and *Pratylenchus penetrans* on Bari 2, Bari 3 (*Cicer reticulatum*) and Cermi (*C. echinospermum*) chickpea varieties were investigated in the climatic chamber conditions. *Pratylenchus thornei* reached the highest population density on Cermi variety within 21 days, also on Bari 3 variety within 42 days, and *Pratylenchus thornei* did not grow on Bari 2 variety. *Pratylenchus neglectus* reached the highest population density on Cermi and Bari 3 varieties within 28 days. *Pratylenchus penetrans* reached the highest population density on the variety Bari 3 within 49 days while Bari 2 and Cermi cultivars showed low population densities during the trial period.Considering the reproduction rates of the root lesion nematodes in the tested varieties, Bari 2 cultivar is determined as resistant to *Pratylenchus thornei* and *Pratylenchus neglectus* and susceptible to *Pratylenchus penetrans*; Bari 3 variety is determined as resistant to *Pratylenchus thornei*, sensitive to *Pratylenchus neglectus* and *Pratylenchus penetrans*, while Cermi is sensitive to *Pratylenchus thornei* and *Pratylenchus neglectus* and moderately resistant to *Pratylenchus penetrans*.

Keywords: Root lesion nematodes - Population development - Chickpea.

S1-P15

Element levels in wood from maritime pine infected by *Bursaphelenchus xylophilus* in field conditions.

Jorge Faria[1] (jorge.faria@iniav.pt), Cláudia Vicente[1, 2], Pedro Barrulas[3], Dora Teixeira[3], Maria L. Inácio[1]

[1] *INIAV, I.P., National Institute for Agrarian and Veterinarian Research, Oeiras, Portugal*; [2] *MED, Mediterranean Institute for Agriculture, Environment and Development, Institute for Advanced Studies and Research, Évora University, Évora, Portugal*; [3] *HERCULES Laboratory, Évora University, Évora, Portugal*

The plant parasitic nematode *Bursaphelenchus xylophilus* (Tylenchida: Parasitaphelenchidae), commonly known as the pinewood nematode (PWN) is responsible for pine wilt disease (PWD). Infected pines often display reduced resin flux, pine needle yellowing and wilting, culminating in tree death as a result of embolism in the water transport vessels and affected photosynthesis [1,2]. In plants, essential nutrient uptake and translocation are dependent on a healthy vascular system. The effect of interruptions in the water column due to PWN infection on pine nutrient content has only been scarcely studied under field conditions. In the present work, wood samples from *Pinus pinaster* (maritime pine) were collected in Seia in central continental Portugal, an area with monitored PWD detection, and the levels of iron (Fe), manganese (Mn) and copper (Cu), elements with important roles in photosynthetic biochemical mechanisms, were determined through inductively coupled plasma mass spectrometry (ICP-MS) [3]. Infected pines displayed a yellowish to reddish canopy and were classified in Class III for PWN infection, with 50 to 200 PWNs per 100 g of wood. In unaffected pines, where no PWD symptoms were detected, no PWNs were found. Preliminary results revealed that, in wood from infected maritime pines, Mn and Cu showed a 3- and 2-fold increase, respectively, while Fe showed a 3-fold decrease, in comparison to wood from uninfected maritime pines. Infection of susceptible trees with the PWN leads to drastic metabolic and structural changes. Transmission of the PWN is accompanied by alterations in the tree's bacterial and fungal communities, which can influence shifts in the levels of wood essential elements. Future studies will focus on nutrient transport in PWN infected pine trees to determine overall influence in tree elemental status. This work was conducted in the national project PineEnemy: Exploring the NEmatode-MYcobiota interactions in Pine Wilt Disease (LISBOA-01-0145-FEDER-028724).

Keywords: Copper - Iron - Manganese - *Pinus pinaster*.

References:

- [1] Faria J. M. S. et al. (2021) Plants 10: 2614.
- [2] Vicente et al. (2012) Eur J Plant Pathol 133:89-99.
- [3] Faria J. M. S. et al. (2021) Applied Sciences 11: 8745.

S1-P16

Virulence in UK *Globodera pallida* populations in relation to resistance and durability.

Vivian Blok (Vivian.Blok@hutton.ac.uk), Ulrike Gartner, Kyriakos Varypatakis, John Jones

Cell and Molecular Sciences, The James Hutton Institute, Dundee, Perthshire, United Kingdom

Using host resistance to manage plant parasitic nematodes is a highly attractive control option. However, breeding resistant varieties that are also commercially successful is generally a long process relying on a few amenable sources of resistance. The durability of these resistances is thus crucial to the viability of this strategy. While resistance in potato to *Globodera rostochiensis* was achieved relatively quickly and remains durable in the UK after many decades, breeding varieties with resistance to *G. pallida* has taken much longer. The 2 main resistance sources used for *G. pallida* are from *Solanum vernei* and *S. tuberosum* ssp. *andigena* CPC2802. Resistance from both has been described as polygenic and quantitative, amplifying the difficulty in breeding for high levels of resistance in tetraploid potato. Increased virulence of *G. pallida* populations towards these resistance sources has been observed after repeated cycles of multiplication which has implications for their durability and could be related to the genetic and phenotypic complexity found in UK populations of *G. pallida*. Molecular comparisons of *G. pallida* populations that differ in their virulence towards *Solanum vernei* or *S. tuberosum* ssp. *andigena* CPC2802 resistances have revealed several potential avirulence candidate genes which are currently being investigated. Given the likelihood that commercial varieties with resistance from either *Solanum vernei* or *S. tuberosum* ssp. *andigena* CPC2802 are unlikely to remain durable with extended use, combining these resistances is the current breeding strategy. Additive and high levels of resistance have been demonstrated when these resistances are combined and was shown to be effective for range of different *G. pallida* populations with different virulence profiles. Advanced breeding lines with dual *G. pallida* resistance from the James Hutton Limited breeding program show high levels with a range of *G. pallida* populations ; however, their durability has yet to be assessed.

Keywords: Globodera - Resistance - Virulence - Durability.

S1-P17

Pathogenicity of *Aphelenchoides besseyi* populations from rice *(Oryza sativa)* to soybean (*Glycine max)* plants.

Angélica Halfen[1] (angelicahalfe.agronomia@gmail.com), Luciany Favoreto[2], Mauricio Conrado Meyer[3], Andressa Cristina Machado[4], Santino Aleandro Silva[4], Patrícia Pricilla França[5], Rafaela Loreto[5], Cesar Gomes[6]

[1] *Plant Protection Post Graduation Program, Universidfade Federal de pelotas, Pelotas, RS, Brazil*; [2] *Nematology lab, Epamig, Londrina, PR, Brazil*; [3] *Phythopathology and Nematology Lab, Embrapa Soja, Londrina, PR, Brazil*; [4] *Nematology lab, Instituto Agrônomico do Paraná, Londrina, PR, Brazil*; [5] *Nematology lab, Unifil- Centro Universitário Filadélfia, Londrina, PR, Brazil*; [6] *Phythopathology and Nematology Lab, Embrapa Clima Temperado, Pelotas, RS, Brazil*

The occurrence of green stem disturbance (Soja Louca II) on soybean crops was first detected in the central-north region of Brazil about fifteen years ago. However, the disease etiology was associated with *Aphelenchoides besseyi* only in 2017. The problems from this complex disease on soybean have increased in tropical regions characterized by a hot and rainy climate, resulting in significant yield losses especially in Pará, Maranhão, Mato Grosso and Tocantins states. In the extreme south of Brazil, soybean has been used systematically in crop rotation with irrigated rice in which is a good host of the white tip nematode (*A. besseyi*), a pathogen that has been found in this region frequently. Therefore, the objective of this study was to evaluate the pathogenicity of different populations of *A. besseyi* from rice and soybean. Three nematode populations from rice and just one from soybean were previously identified based on morphological and morphometric parameters. Subsequently these populations were characterized by PCR (Polymerase Chain Reaction) using species-specific primers for *A. besseyi*. In order to establish an assay at greenhouse conditions, firstly all nematode populations were multiplied *in vitro* and 50 days later each one was inoculated into 'BRS 284' soybean plants using two levels of inoculum (600 and 1200 nematodes/plant). Non-inoculated soybean plants were used as control. Fifty days after inoculation, it was observed that all *A. besseyi* populations multiplied in soybeans and caused symptoms of the green stem syndrome (leaves presenting sheathing, less hairiness, intense green color and thinning in the leaf blade). In addition, there was a reduction in the fresh mass of the aerial part of the inoculated plants compared to the control independently of nematode population. There was a significant interaction between populations and levels of inoculum for the reproduction factor (RF) ($P \leq 0.05$), where lower inoculum densities resulted in higher RF values. In general, the populations of *A. besseyi* from rice presented higher reproduction indexes. In this sense, areas of rice infested with this nematode and located in hot and rainy regions can be sources of inoculum for the manifestation of green stem syndrome in soybeans.

Keywords: Aphelenchoides besseyi - Soybean - Rice - Soja Louca II - Foliar retention.

S1-P18

Bifurcation of pathogen recognition specifity and convergence of immune responses by the potato *R* genes *Rx1* and *Gpa2.*

Aska Goverse (aska.goverse@wur.nl), Octavina Sukarta, Amalia Diaz Granados Munoz, Erik Slootweg, Geert Smant

Laboratory of Nematology, Wageningen University, Wageningen, Netherlands

The closely related potato resistance genes *Gpa2* and *Rx1* encode canonical intracellular CC-NB-LRR immune receptors. They belong to the same *R* gene cluster, but evolved to defend against two unrelated pathogens. Gpa2 detects specific GpRbp-1 effector variants secreted by the potato cyst nematode *Globodera pallida*, whereas Rx1 recognizes the viral coat protein (CP) of Potato Virus X. Despite their high sequence similarity (88%), these two *R* gene homologs confer distinct resistance responses. Whereas *Rx1* confers extreme virus resistance in the areal parts of the plant, *Gpa2* confers a mild nematode resistance response in the roots. This provides us with a model system to study the molecular and cellular mechanisms involved in pathogen recognition and *R* gene activation in plants. Using structure-informed approaches, we could demonstrate that exchanging the recognition moiety in the LRR is sufficient to convert extreme virus resistance in the leaves into mild nematode resistance in the roots, and vice versa. Apparently, the CC-NB-ARC can operate independently of the recognition specificities defined by the LRR domain. This shows the convergence of downstream immune responses to unrelated pathogens with completely different lifestyles and routes of invasion. Interestingly, both Rx1 and Gpa2 form a complex with the co-factor Ran GTPase Activating Protein 2 (RanGAP2). Here we present first evidence for the role of RanGAP2 as a common virulence target of two evolutionary distinct pathogens, which is guarded by two bifurcated intracellular CC-NB-LRR immune receptors in potato. This provides us with novel insights in the evolution of *R* gene recognition specificities in plants and the functioning of the encoding NB-LRR immune receptors, which can be exploited to select or engineer novel (broadspectrum) resistances to pathogens including plant-parasitic nematodes.

Keywords: Virulence - R gene - *G. pallida* - PVX - Immunity.

S1-P19

Root architecture plasticity in response to endoparasitic cyst nematodes is mediated by damage signaling.

Nina Guarneri[1] (nina.guarneri@wur.nl), Jaap-Jan Willig[1], Mark G. Sterken[1], Wenkun Zhou[2], Viola Willemsen[3], Aska Goverse[1], Geert Smant[1], Jose L. Lozano-Torres[1]

[1] *Laboratory of Nematology, Wageningen University & Research, Wageningen, Netherlands*; [2] *State Key Laboratory of Plant Physiology and Biochemistry, China Agricultural University, Beijing, China*; [3] *Laboratory of Molecular Biology, Cluster of Plant Developmental Biology, Wageningen University & Research, Wageningen, Netherlands*

Plant root architecture plasticity in response to biotic stresses has not been thoroughly investigated. Infection by the endoparasitic cyst nematodes induces root architectural changes that involve the formation of secondary roots at infection sites. However, the molecular mechanisms regulating secondary root formation in response to cyst nematode infection remain largely unknown. We first assessed whether secondary roots form in a nematode-density dependent manner by challenging wild type *Arabidopsis* plants with increasing numbers of cyst nematodes (*Heterodera schachtii*). Next, by using jasmonate-related reporter lines and knock-out mutants, we tested if tissue damage by nematodes triggers secondary root formation. Finally, we verified whether damage-induced secondary root formation depends on local auxin biosynthesis at nematode infection sites. Intracellular host invasion by *H. schachtii* triggers a transient local increase in jasmonates, which activates the expression of ERF109 in a COI1-dependent manner. Knock-out mutations in COI1 and ERF109 disrupt the nematode-density dependent increase of secondary roots observed in wildtype plants. Furthermore, ERF109 regulates secondary root formation upon *H. schachtii* infection via local auxin biosynthesis. Altogether, we conclude that host invasion by *H. schachtii* triggers secondary root formation via the damage-induced jasmonate-dependent ERF109 pathway. This points at a novel mechanism underlying plant root plasticity in response to cyst nematode infections.

Keywords: Auxin - Cyst nematodes - Damage - Jasmonates - Root plasticity.

S1-P20

Small RNAs (miRNAs) involved in wild *Arachis* resistance against *Meloidogyne arenaria.*

Patricia Messenberg Guimaraes[1] (patricia.guimaraes@embrapa.br), Ana Cristina Miranda Brasileiro[1], Matheus Correa de Paula[2], Roberto Coiti Togawa[1], Priscila Grynberg[3]

[1] *Biotechnology, EMBRAPA- Cenargen, Brasilia, DF, Brazil*; [2] *Biologia Celular, University of Brasilia, Brasilia, DF, Brazil*; [3] *Bioinformatics, EMBRAPA- Cenargen, Brasilia, DF, Brazil*

Root-knot nematodes (RKN-*Meloidogyne*spp.) are a major constraint to agricultural production worldwide. *Arachis stenosperma* is a peanut wild relative exclusive to South America that shows high levels of resistance against *M. arenaria*, including the Hypersensitive Response (HR). The genus *Meloidogyne* comprises sedentary endoparasites with complex strategies of parasitism promoting the differentiation of plant vessels into giant cells and formation of galls in the host roots. MicroRNAs (miRNAs) are small non-coding RNA molecules (usually between 20 and 22 nt) that play important roles in regulating gene expression at post-transcriptional level. In order to contribute to the repertoire of resistance genes, defense molecules and management measures utilized for RKN control, we explored the role of miRNAs in the incompatible *A. stenosperma/M.arenaria* interaction. We used sRNA-seq to survey the miRNAs produced during this interaction, and identified their predicted targets in the host control and RKN-inoculated samples. The miRNAs from infective second-stage juveniles (J2) of *M. arenaria* were also sequenced and annotated, and potential silencing targets for these molecules were identified in the reference genome of the susceptible species *A. hypogaea*. Overall, we identified 87 conserved miRNAs belonging to 29 families and 31 new miRNA candidates in resistant *A. stenosperma*, of which 14 were responsive to the RKN infection. The relative expression of these miRNAs and their respective targets, including a resistance gene (*AsNLR*), were validated in control and infected plants by qRT-PCR analysis, suggesting a role for these RNA molecules in this wild species resistance. The annotation of *M. arenaria* miRNAs enabled the identification in the susceptible *A. hypogaea* of dozens of target genes associated with defense, root growth, cell differentiation, response to oxidative stress and hormone homeostasis. These results strongly suggest the role of these RKN miRNAs in host genes silencing in order to install the infection.

Keywords: Gene silencing - Arachis - Resistance - miRNA - NLR.

S1-P21

The Plant Secretory Pathway During Cyst Nematode Infection.

Ashley Hayes (bsaha@leeds.ac.uk), Catherine Lilley, P.E. Urwin, Jurgen Denecke

Centre for Plant Sciences, University of Leeds, Leeds, West Yorkshire, United Kingdom

Cyst nematodes cause billions of dollars' worth of crop loss each year. To obtain nutrients from their host, cyst nematodes re-programme root cells to form highly specialised feeding sites named syncytia. Subcellular reorganisation occurs during syncytial development, including changes to key components of the plant secretory pathway. For example, several electron microscopy studies suggest that proliferation of the endoplasmic reticulum and Golgi apparatus, loss of the large central vacuole and formation of numerous smaller vacuoles occurs. We aim to further characterise the plant secretory pathway during syncytial formation, using a portfolio of novel double fluorescent reporter constructs. Each construct contains a 'housekeeping' Golgi marker and an additional plant secretory pathway marker, which labels either the endoplasmic reticulum, trans-Golgi network, prevacuolar compartment or the vacuole. Arabidopsis lines expressing these constructs have been infected with the beet cyst nematode *Heterodera schachtii.* Confocal microscopy has confirmed previous observations including changes to the vacuoles, and has provided information on previously uncharacterised organelles. We also aim to identify cyst nematode effectors which target the plant secretory pathway, in particular the endoplasmic reticulum (ER). The ER has several roles within the cell, including the production, folding and quality control of proteins, lipid biosynthesis and immune responses. The ER is also a major target of pathogen effectors. A common feature of ER-targeting plant pathogen effectors is the presence of a C-terminal transmembrane domain (TMD). However, cyst nematode proteins containing TMDs have typically been discounted from effector screens. Therefore, we identified putative cyst nematode effectors within *Globodera pallida* and *Heterodera schachtii* containing TMDs. From this, a set of 9 proteins was selected for *in situ* hybridization to confirm gland cell expression, and transient expression within tobacco leaves to characterise *in planta* subcellular localisation.

Keywords: Cyst nematode - Syncytium - Arabidopsis - Membrane Trafficking - Cell Biology.

S1-P23

Uncovering the role of the syncytia-forming nematode core effector GrGLAND11 in *Globodera rostochiensis.*

Kerry Leslie[1] (kl97@st-andrews.ac.uk), Sophie Mantelin[2], Sebastian Eves-van den Akker[3], John Jones[2]

[1] *Nematology, The University of St Andrews, St Andrews, Scotland*; [2] *The James Hutton Institute, Dundee, Scotland*; [3] *The university of Cambridge, Cambridge, England*

Plant parasitic nematodes infect all major food crops worldwide, causing damage valued at approximately 80 billion U.S. dollars per year (Nicol *et al.* 2011). Some nematodes form a feeding site called a syncytium in the roots of their host. Relatively little is known about how nematodes initiate and maintain these feeding structures. However, specialised proteins and small molecules, known as effectors, secreted into the plant by the pathogen, are thought to play critical roles in these processes. It is important to establish a greater understanding of the role of effectors in syncytium formation and maintenance. By exploiting the genomic and transcriptomic data available from four syncytium-forming species; *Globodera rostochiensis, Globodera pallida, Rotylenchulus reniformis* and *Nacobbus abberans,* we have identified a series of core effectors present and expressed in all these nematodes across their life cycle. One of these, the pioneer GLAND11 effector from *G. rostochiensis* (GrGLAND11), localises to the actin cytoskeleton when expressed as a fusion with fluorescent proteins in plant cells. Yeast two-hybrid screening has identified an arginine N-methyltransferase that interacts with GrGLAND11. We are currently investigating this interaction in more detail to define the role of GrGLAND11 in syncytium formation and maintenance.

Keywords: Plant-parasitic nematodes - Syncytium - Effector - Actin.

References:

- Nicol et al., 2011. Genomics and Molecular Genetics of Plant-Nematode interactions, 21-43

S1-P25

Delivery of macro molecules to plant-parasitic nematodes.

Olaf Kranse[1] (ok297@cam.ac.uk), Sebastian Eves - van den akker[1], Alper Akay[2], Helen Beasley[1]

[1] *University of Cambridge, Cambridge, United Kingdom*; [2] *University of East Anglia, Norwich, United Kingdom*

To cause disease, plant-parasitic nematodes inject proteins, or other molecules, termed “effectors” to differentiate plant biology to their benefit. A primary goal of the research field is to develop a mechanistic understanding of effector function. Variable efficacy and off-target effects limit the only method to interrogate nematode gene “loss-of-function” (RNA-interference). There is currently no method to interrogate nematode gene “gain-of-function”. Thus, elucidating true causal relationships is held back by a lack of transformation; we are unable to express transgenes to test function, nor edit the genome of plant-parasitic nematodes. Both these goals require delivery of macro molecules directly into the cell. Two methods were tested in parallel for the delivery of various macro molecules to plant-parasitic nematodes. One based on micro injection and the other, potentially a more accessible technique, based on lipofection. We designed a series of CRISPR-Cas9 guide RNAs, and donor template fragments to introduce targeted edits and specific base changes respectively. The outcomes of editing experiments were analysed by next-generation amplicon sequencing. We described the germ line morphology of several plant-parasitic nematode species. We were able to deliver fluorescent DNA oligos and dyes into the germline of *Meloidogyne hapla* males, *Heterodera schachtii* males, and *Bursaphelenchus xylophilus* females by microinjection, and the digestive tract of second-stage juvenile *H. schachtii* by lipofection. Combining these techniques had no obvious additional effect on the spread of fluorescent signal in these species. Lipofection aided mRNA delivery, however, and shows promising preliminary results in juveniles. Sequencing data showed that PCR-mediated recombination can be a cause for false positives when assessing homology directed repair following CRISPR experiments. Current efforts are aimed at optimising delivery of genome editing macromolecules and detection of genome editing events. We have made important strides towards understanding and circumventing the challenges associated with genetic modification of plant-parasitic nematodes, including delivery of macromolecules, and faithful detection of successful events.

Keywords: CRISPR-Cas9 - Lipofection - Micro-injection - Plant-parasitic nematode.

S1-P26

An insight into host-specific behavior of *Globodera spp.* hatched in root exudates from potato and its wild relative, *Solanum sisymbriifolium.*

Joanna Kud[1] (jkud@uidaho.edu), Syamkumar Sivasankara Pillai[1], Allan Caplan[2], Joseph Kuhl[2], Fangming Xiao[2], Louise-Marie Dandurand[1]

[1] *Department of Entomology, Plant Pathology and Nematology, University of Idaho, Moscow, Idaho, United States*; [2] *Department of Plant Sciences, University of Idaho, Moscow, Idaho, United States*

Understanding belowground chemical interactions between plant roots and plant parasitic nematodes is immensely important for sustainable crop production and soilborne pest management. Due to metabolic diversity and ever-changing dynamics of root exudate composition, the impact of only certain molecules, such as nematode hatching factors, repellents, and attractants, has been examined in detail. Root exudates are a rich source of biologically active compounds, which plants use to shape their ecological interactions. However, the impact of these compounds on nematode parasitic behavior is poorly understood. In this study, we specifically address this knowledge gap in two cyst nematodes, *Globodera pallida*, a potato cyst nematode and the newly described species, *Globodera ellingtonae*. *Globodera pallida* is a devastating pest of potato (*Solanum tuberosum*) worldwide, whereas potato is a host for *G. ellingtonae*, but its pathogenicity remains to be determined. We compared the behavior of juveniles (J2s) hatched in response to root exudates from a susceptible potato cv. Desirée, a resistant potato cv. Innovator, and an immune trap crop *Solanum sisymbriifolium* (litchi tomato – a wild potato relative). Root secretions from *S. sisymbriifolium* greatly reduced the infection rate on a susceptible host measured using acid Fuchsin two weeks post infection for both *Globodera spp*. Juvenile motility, measured as a dispersion over time on Phytagel plate, was also significantly influenced in a host-dependent manner. However, reproduction on a susceptible host from juveniles hatched in *S. sisymbriifolium* root exudates was not affected, nor was the number of encysted eggs from progeny cysts. Transcriptome analysis by using RNA-sequencing (RNA-seq) revealed the molecular basis of root exudate-mediated modulation of nematode behavior. Differentially expressed genes are grouped into two major categories: genes showing characteristics of effectors and genes involved in stress responses and xenobiotic metabolism. To our knowledge, this is the first study that shows genome-wide root exudate-specific transcriptional changes in hatched preparasitic juveniles of plant-parasitic nematodes. This research provides a better understanding of the correlation between exudates from different plants and their impact on nematode behavior prior to the root invasion and supports the hypothesis that root exudates play an important role in plant-nematode interactions.

Keywords: Globodera spp. - Root exudate - Potato - Solanum sisymbriifolium - RNA-Seq.

S1-P28

Effect of *Radopholus similis* and *Meloidogyne* spp. on plant growth and yield of *Musa* AAB 'Dominico Hartón.'

Horacio Lopez-Nicora[1] (lopez-nicora.1@osu.edu), Oscar Guzmán-Piedrahita[2], Carolina Zamorano-Montañez[2]

[1] *Department of Plant Pathology, The Ohio State University, Columbus, Ohio, United States*; [2] *Departamento de Producción Agropecuaria, Universidad de Caldas, Manizales, Caldas, Colombia*

Both in Colombia, and worldwide, there is scarce information regarding the effect of plant-parasitic nematodes (specifically, *Radopholus similis* and *Meloidogyne* spp.) on *Musa* AAB (subgroup plantain) under field conditions. The damage caused by these pathogens has previously been measured indirectly by the observation of toppled plants or low yield in infested areas [1]. The objectives of this study, therefore, were to evaluate the effect of *R. similis* and *Meloidogyne* spp. alone and in combination on the growth and yield of *Musa* AAB (subgroup plantain) 'Dominico Hartón'. The study was conducted under field conditions at the Montelindo Research Farm of the Universidad de Caldas, Colombia. Plantain seeds (corms) were planted in bags containing sterilized soil and placed on stainless steel tables 50 cm above the ground. Forty days after planting, these seedlings were infested with 1,250, 2,500, 3,750 and 5,000 *R. similis* alone. Similarly, using the same nematode densities, other seedlings were infested with *Meloidogyne* spp. In addition, seedlings concomitantly infested with 5,000 *R. similis* and 5,000 *Meloidogyne* spp were included as the interaction treatment. Uninfested seedlings were incorporated as control. One month post infestation, plants were moved to the field and arranged in a randomized complete block design. Each treatment was replicated ten times (with five plants per replica). Plant height, number of functional leaves and pseudostem circumference were recorded at flowering, while bunch weights were recorded at harvest. Toppled plants were also documented during the study. Nematode treatments had no effect on plant height, number of functional leaves or pseudostem circumference at flowering. Bunch weight, however, was significantly affected by nematode treatments and number of toppled plants. Moreover, when nematode treatments and toppled plants were included in the analysis, the uninfested control treatment had the highest yield. Compared to the control, concomitant infestation with *R. similis* and *Meloidogyne* spp., *Meloidogyne* spp. alone and *R. similis* alone reduced yield by over 23%, 20% and 16%, respectively. Yield was significantly reduced by *R. similis* or *Meloidogyne* spp. alone, but the greatest damage occurred when plants were simultaneously infested with both nematodes. Results from this study will aid in long-term plant-parasitic nematode management and evaluation of their interaction in reducing plantain production.

Keywords: Nematode interaction - Yield loss in plantain - Musa AAB - Radopholus similis - Meloidogyne spp..

References:

- [1] Moens and Araya, 2002. CORBANA 28(55): 43-56.

S1-P29

Multi-resistant reactions of new resistant sources of *Oryza* spp. to root-knot nematodes from the graminis group.

Raycenne Leite[1] (cennerosa@gmail.com), Vanessa Mattos[2], Jório Mendonça[2], Valácia Lobo[3], Juvenil Cares[4], Regina Carneiro[2]

[1] *Nematology, Universidade de Brasília/Embrapa Cenargen, Brasilia, Federal District, Brazil*; [2] *Nematology, Embrapa Cenargen, Brasilia, Federal District, Brazil*; [3] *Embrapa Arroz e Feijão, Santo Antônio de Goias, Goias, Brazil*; [4] *Nematology, Universidade de Brasilia, Brasilia, Federal District, Brazil*

The efficient management of *Meloidogyne* spp. in flooded rice should be based on genetic resistance and crop rotation with poor host plants, both not yet available to farmers. Our work evaluated the resistance reaction of four wild rice species (*Oryza alta*, *O. grandiglumis*, *O. glumaepatula*, *O. longistaminata*) and two domesticated rice species (*O. glaberrima* and *O. sativa* – susceptible control) to recently identified *Meloidogyne* species from Brazil (*M. oryzae* and *M. ottersoni*) and two variants of *M. graminicola* (esterase phenotypes (EST), EST SP2 and EST SP3). The rice plants were inoculated with 5,000 eggs in completely randomized design, with 6 treatments and 8 replications. The resistance was evaluated 90 days after inoculation, considering the reproduction factor (RF= final population/initial population) submitted to statistical analyses. The experiments were repeated twice. For *M. graminicola* (ESTsp2), the rice species *O. glumaepatula*, *O. glaberrima* and *O. alta* were highly resistant in both assays (FR<1.0). *Oryza grandiglumis* and *O. longistaminata* were moderately resistant. All rice species can be considered resistant to *M. graminicola* (ESTsp3), except the control and *O. longistaminata*, which were susceptible. For *M. oryzae*, *O. glumaepatula*, *O. glaberrima*, *O. alta*, and *O. grandiglumis* were considered highly resistant with RFs<1, while *O*. *longistaminata* was resistant. For *M. ottersoni*, *O. glumaepatula, O. glaberrima*, and *O. alta* were highly resistant in both assays, with RF<1; *O. grandiglumis* was considered resistant and *O. longistaminata* susceptible. Our findings revealed resistance of wild rice species to *Meloidogyne* spp. We also report *O. glumaepatula* as a promising genetic source of resistance since it belongs to the same genomic group (AA) as *O. sativa.*

Keywords: Wilde rice - Meloidogyne - Resistance - Genetic source.

S1-P30

The link between genetic diversity and pathogenicity variation in temperate root-knot nematode *Meloidogyne hapla*.

Muhammad Iqbal Maulana (muhammad.maulana@wur.nl), Denni M. te Molder, Stefan van Ruitenbeek, Aska Goverse, Jan E. Kammenga, Mark Sterken

Laboratory of Nematology, Wageningen University and Research, Wageningen, Netherlands

Root knot nematode (RKN) *Meloidogyne hapla* is one of the most damaging agricultural pests in temperate regions. Current control strategies focus on host-plant resistance genes inhibiting the ability of the nematode to establish feeding sites in the roots of host plants. Feeding site induction is a complex process involving: the modification/modulation of the cell wall, cell cycle regulation, plant hormones regulation, cytoskeleton regulation, and gene expression regulation. Natural variation plays an important role in the nematode-plant interaction (e.g. yield loss). However, the majority of research focusses on the diversity of plant resistance against the nematode, but little is known about the genetic- and pathogenicity diversity among natural strains of *M. hapla*. Here, we investigated the pathogenicity and genetic diversity of 23 *Meloidogyne hapla* strains collected from different locations and crops. The pathogenicity of these nematodes was characterized via *in vitro* infection assay in two tomato accessions: *Solanum lycopersicum* cv. Money Maker and *S. pimpinellifolium* (CGN14498). We found that *M. hapla* strains display a large spectrum of pathogenicity in the two tomato accessions. Analysis of whole-genome DNAseq of *M. hapla* also revealed ample genetic variation with more than 100,000 SNPs segregating among the strains. These results of pathogenicity variation in tomato and genotypic diversity of *M. hapla* make them suitable to be further used in quantitative genetic studies of plant-nematode interaction. Based on the results of this study, we will construct a RIL-like population of *M. hapla* and perform plant-nematode eQTL study using *M. hapla* RIL x tomato RIL.

Keywords: Meloidogyne hapla - Genetic diversity - Pathogenicity variation - Natural variation.

S1-P31

Inherited memories: Preparing the next generation for battle.

Anikó Meijer[1] (aniko.meijer@ugent.be), Mohammad Reza Atighi[1], Bruno Verstraeten[1], Klaas Vandepoele[2], Tina Kyndt[1]

[1] *Ghent University, Ghent, Belgium*; [2] *VIB, Ghent, Belgium*

Intergenerational acquired resistance (IAR) is a phenomenon where the offspring of stressed plants expresses higher resistance against the same or other (a)biotic stress factors. This is associated with epigenetic changes in the offspring. One possible epigenetic memory mark is methylation of the DNA, which is directed by 24nt small interfering RNAs (siRNAs) in the plant-specific RNA-directed DNA methylation (RdDM) pathway. Here, we present our findings in the monocot staple crop rice, infected by the devastating root-knot nematode *Meloidogyne graminicola*. IAR experiments and RNA-seq in plants impaired for 24nt siRNA biogenesis, indeed point towards a role of small RNAs in intergenerational memory. This is further consolidated by small RNA sequencing in the root and shoot of infected rice throughout plant development and in the embryo and endosperm. Further, our data seems to indicate that sRNA-dependent regulation of plant hormone pathways plays a role in IAR in rice.

Keywords: Root-knot nematode - Oryza sativa - Intergenerational acquired resistance - Epigenetics - Small RNA.

S1-P32

Host status of *Salvia hispanica* (chia) to *Meloidogyne incognita* and nematotoxic activity of root and shoot extracts.

Susan Meyer[1] (Susan.L.Meyer@usda.gov), Margaret MacDonald[1], Nathan Reetz[1], Mihail Kantor[1], Lynn Carta[1], Zafar Handoo[1], Mary Camp[2], Tim Phillips[3]

[1] *Mycology and Nematology Genetic Diversity and Biology Laboratory, USDA Agricultural Research Service, Beltsville, Maryland, United States*; [2] *ARS Statistics Group, USDA Agricultural Research Service, Beltsville, Maryland, United States*; [3] *Department of Plant and Soil Sciences, University of Kentucky, Lexington, Kentucky, United States*

Chia (*Salvia hispanica* L.) plant parts are used for traditional medicine, and the seeds for food, oil, and drinks. Plant-parasitic nematodes have been collected from other *Salvia* spp., but in a literature search, chia was not found to be listed as a host [USDA Nematode Collection,1,2,3]. It is not known if the lack of phytoparasitic nematodes reported from chia is due to resistance to multiple species, or because little attention has been paid to nematodes attacking chia. As chia production expands, this information is important for growers. The objectives of this research were as follows: 1) Chia lines were tested as hosts for the root-knot nematode *Meloidogyne incognita* (Kofoid & White) Chitwood (referred to here as RKN). 2) Chia shoots were amended into soil to determine whether RKN populations would be suppressed on a subsequent crop. 3) Extracts from plant parts were investigated for nematotoxic compounds. Six chia lines were used for the study: Brad's Organic, Cono, E2, G3, G5 and W13.1. Aqueous root and shoot extracts from all six lines were tested against RKN eggs and second-stage juveniles (J2) in microwell assays. Seeds from the same chia lines were planted in the greenhouse, inoculated with RKN, and harvested after 6 weeks to determine numbers of eggs and galls on the roots. Chia shoots were collected, chopped, and amended into soil in greenhouse pots (2.3% or 2.5% weight fresh shoots:weight dry soil). The soil was then inoculated with RKN, and cucumber `Sweet Slice' seedlings were transplanted into the pots and harvested 5-6 weeks later to determine effects on RKN population numbers. In the microwell assays, chia plant extracts could kill at least a third of RKN J2, but hatch was generally not affected. In greenhouse trials, all six chia lines were RKN hosts. Galling and egg production on cucumber roots were not suppressed by soil amendment with chia plant parts. This study demonstrated that chia is a host to RKN, and that chia roots and leaves produce compounds that can kill J2. However, at the tested rates, amending soil with chia shoots did not suppress RKN on susceptible cucumber. Surveys for *Meloidogyne* spp. and other phytoparasitic nematodes on chia may provide information about distribution and potential damaging effects of nematodes on this crop plant.

Keywords: Meloidogyne - Nematode host - Plant extract - Salvia hispanica - Soil amendment.

References:

- [1] Lisetskaya, 1971. Parasites of Animals and Plants. 7: 142-144.
- [2] Walker and Melin, 1998. J. Nematol. 30 (4S): 607-610.
- [3] Jaimand, 2013. J. Med. Plants By-Products 2: 13-16.

S1-P33

Extract of *Macrotyloma axillare* 'Java' on hatching, penetration and development of *Meloidogyne javanica* in soybean plants.

Angelica Miamoto[1, 2] (angelicamiamoto@gmail.com), Angelica Calandrelli[2], Monique Silva[2], Claudia Dias-Arieira[2]

[1] *Nematology, State University of Maringa, Maringa, Parana, Brazil*; [2] *State University of Maringa, Maringa, Brazil*

The alternative management of phytonematodes has been enhanced because this practice may reduce pathogen populations without harming the environment. One of these options is the application of vegetable bionematicides containing volatile compounds and secondary metabolites with the capacity to reduce the development of parasites. 'Java' (*Macrotyloma axillare*) is a plant little studied but presents great potential to control *Meloidogyne javanica*, to which it exhibits resistance [1]. Thus, this work aimed to investigate the action of 'Java' vegetable extract on the hatching, penetration and development of *M. javanica*. A hatching test evaluated crude methanolic extract (CME), hydromethanol (HMF), hexane (HEXF), dichloromethane (DCMF) and ethyl acetate (EAF) fractions under four doses (0, 0.05, 0.5 and 5%). After 10 days incubation, the hatching percentage of second stage juveniles (J2) was determined. Also, the penetration and development of *M. javanica* were evaluated in soybean plants treated with CME, HFM, HEXF, DCMF and EAF at 1%, with a water control at 10 and 30 days after inoculation (DAI). All the fractions and doses were able to reduce J2 hatching being stable from the dose of 0.05%. The fractions CMF, DCMF and EAF reduced the penetration of *M. javanica* from 10 DAI as well as female occurrence from 30 DAI. The extracted fractions of 'Java' displayed nematicide effects acting on hatching, penetration and development of *M. javanica*.

Keywords: Root-knot nematode - Alternative control - Plant extract - Leguminous.

References:

- [1] Miamoto et al., 2020. AJCS 14(06):940-946.

S1-P34

Efficacy of *Aphelenchoides sp.* against Spruce bark beetle *Ips typographus* from Bakuriani (Georgia).

Nona Mikaia[1] (n.mikaia@sou.edu.ge), Irina Khelisupali[2], Zaira Tkhebuchava[3], Kety Chxaidze[3]

[1] *Faculty of Natural Sciences, Mathematics, Technology and Pharmacy, Sokhumi State University, Tbilisi, Georgia*; [2] *LPL, Borjomi Municipality Tsagveri Borrow Public School, Tsagveri, Georgia*; [3] *Samtskhe-Javakheti State University, Akhaltsikhe, Georgia*

Ips.typographus, the European spruce bark beetle (*Coleoptera: Scolitidae*) is one of the most serious pests of the spruce (*Picea orientalis*) in the Georgia mountains of Bakuriani. Bark beetles are so named because they reproduce in the inner bark of living and dead phloem tissues of trees [1]. The beetles attack standing weakened and healthy trees, infesting mainly lower and middle parts of stems. Previous studies report nematodes associated with bark beetles [2] and the goal of this study was to determine the efficacy of the nematode A*phelenchoides sp.* against spruce bark beetle. Adult and larvae *I. typographus* were collected in forests near the town of Bakuriani, rinsed in distilled water and 10 adults and 10 larvae were placed on filter paper in individual Petrie dishes (80X15 mm). One ml of a suspension of 500 adult and larvae *Aphelenchoides sp*. were pipetted onto the filter paper. The nematodes were recovered from soil in a potato field and maintained refrigerated in a flask. Insects were monitored for 3 days at 250C and those that died were dissected to confirm nematode infection by *Aphelenchoides sp.* microscopically. The treatment was replicated twice, and the experiment repeated three times. The mortality of adult beetles was 15%, 26% and, 38% and larval mortality was 28%, 32% and 55 %. Mortality of the untreated control for adults was 0,03%, 0,02%, 0,02% and in larvae was 0,01%,0,02%, 0,03%. We observed *Aphelenchoides sp*. in all cadavers. Additional work is needed to identify the species of nematode used in this study.

Keywords: I.typographus - Aphelenchoides sp. - Spruce bark beetle.

References:

- [1] Branislav Hroššo et al., 2020. Forests 11: doi:10.3390/f11121290.
- [2] Jaroslav et al., 2017. Bulletin of Insectol. 70(2): 291-297.
- [3] Shakeel Ahmad and Zahid Hussain, 2002. Pakistan J. Biol Sci 5: 640-642.

S1-P35

Distribution and damage potential of plant parasitic nematodes on medicinal and aromatic plants in Germany.

Ilya Noskov[1] (ilya.noskov@julius-kuehn.de), Johannes Hallmann[1], Hanna Blum[2], Hansjörg Komnik[2]

[1] *Institute for Epidemiology and Pathogen Diagnostics, Julius Kühn Institute (JKI), Braunschweig, Germany*; [2] *Institute of Crop Science and Resource Conservation, University of Bonn, Rheinbach, Germany*

Yield reductions on medicinal and aromatic plants occur repeatedly in agricultural practices. Plant parasitic nematodes are often assumed to be the cause but concrete data is scarce. As part of the joint project NemaAG, funded by the Federal Ministry of Food and Agriculture and in collaboration with partners from research, consulting, and producers, we are investigating the distribution and damage potential of plant parasitic nematodes on medicinal and aromatic plants. The damage potential of economically important nematode species is also being studied on selected plants such as peppermint, parsley, marjoram, and valerian.

After a detailed evaluation of more than 300 soil samples collected in 2021, it can be noted that plant parasitic nematodes show widespread occurance on medicinal and aromatic plants in both conventional and organic field practices. *Pratylenchus* and *Tylenchorhynchus* have been observed in more than 80% of all examined fields, followed by *Helicotylenchus, Paratylenchus and Trichodorus / Paratrichodorus* observed in 30-50% of examined fields. The genera *Meloidogyne* and *Heterodera*, which are usualy present in high numbers in agricultural crops, have rarely been observed (< 10%). The densities of frequently occuring genera have schown strong fluctuations depending on cultivated plant and field location, e.g. *Pratylenchus*: 93 - 800 nematodes / 100 ml soil; *Tylenchorhynchus*: 70-3344 nematodes/100 ml soil; *Paratylenchus*: 133-1736 nematodes/100 ml soil; and *Meloidogyne*: 33-244 nematodes/100 ml soil.

First examinations on the damage potential of plant parasitic nematodes on peppermint and parsley have been conducted with *Meloidogyne hapla*, since this nematode species is known to be a serious pathogen for these plants. The experimemts confirmed that peppermint and parsley are host plants for *M. hapla*. However, the reproduction rates of *M. hapla* on these plants are clearly lower than on tomatoes. Furthermore, the conducted greenhouse experiments showed that even high nematode densities up to 25.000 *M. hapla* / plant have no negative impact on plant growth. Also, no symptoms were observed on aboveground plant material which are typical for nematode damage. It is possible that both cultures are much more tolerant to *M. hapla* infestation than other cultures.

S1-P36

Characterization of plantain *cv* Agbagba infection with *Radopholus similis* and *Meloidogyne* spp. using macropropagated plantlets.

Emmanuel Olajide[1, 2] (olajide.o.emmanuel@gmail.com), Laura Cortada[2], Delphine Amah[1], Danny Coyne[2, 3], Wim Bert[2], Rony Swennen[4], Yao Kolombia[1]

[1] *International Institute of Tropical Agriculture, Ibadan, Nigeria*; [2] *Nematology Research Unit, Ghent University, Ghent, Belgium*; [3] *International Institute of Tropical Agriculture, Nairobi, Kenya*; [4] *International Institute of Tropical Agriculture, Kampala, Uganda*

The sustainable production of plantain in Africa is threatened by pests and diseases especially plant-parasitic nematodes (PPNs) which cause snapping and toppling of mature plants. Host plant resistance is considered one of the most effective strategies to manage PPNs in a sustainable and environmentally friendly manner. Recently, a rapid and cheap technique of generating clean planting material is becoming widely adopted, prompting the need to conduct host plant response studies using macropropagated plantlets. In this research, we aimed at studying the most suitable time to evaluate plantain germplasm for host resistance to PPNs under screen house conditions, using macropropagated plantlets. The reaction of plantain *cv* Agbagba to *Radopholus similis* and *Meloidogyne* spp. was observed at 30, 60 and 90 days post-inoculation (dpi). Acclimatized macropropagated plantain *cv* Agbagba plantlets in 2-litre nursery bags were inoculated with 1000 mixed stages (no eggs) of *R. similis* and 1000 *Meloidogyne* spp juveniles. The total number of nematodes (Pf) and the fresh root weight (g) at the end of the experiment were recorded. For *R. similis*, results indicated no statistical differences in the root weight between inoculated and uninoculated plantlets at 30 days, although differences were observed at 60 and 90 days. The highest root weight for the inoculated plantlets was scored at 30 days, and lowest at 90 days with no visible functional root. The Pf was highest at 60 dpi and lowest at 30 dpi. For plantlets inoculated with *Meloidogyne* spp., the root weight and the total number of recovered nematodes increased over time. No significant difference were observed in the root weight between *Meloidogyne* spp. inoculated and uninoculated plantlets at 30, 60 and 90 dpi. Our research provides new insights to banana and plantain reactions during nematode infestation and it was concluded that *R. similis* should be evaluated in 2-litre nursery bags using macropropagated plants at 60 dpi before they damage the root too much. For *Meloidogyne* spp, we suggest that evaluation could be done 90 dpi when nematode reproduction is at maximum.

Keywords: Burrowing nematode - Musa - Nigeria - Root-knot - Screening.

S1-P37

Characterization of *T. semipenetrans* in South African citrus orchards and baseline data for beneficial nematode communities in *Citrus* tree rhizospheres.

Mathys C Pretorius[1] (MC@cri.co.za), N. Labuschagne[2], M. Daneel[3], H. Fourie[4], E. Shokoohi[5], A. McLeod[6]

[1] *Citrus Research International, Nelspruit, South Africa*; [2] *Department of Plant and Soil Sciences, University of Pretoria, Pretoria, South Africa*; [3] *Agricultural Research Council, Tropical and Subtropical Crops, Nelspruit, South Africa*; [4] *Unit for Environmental Sciences and Management, Integrated Pest Management, North-West University, Potchefstroom, South Africa*; [5] *Green Biotechnologies Research Centre of Excellence, University of Limpopo, Sovenga, South Africa*; [6] *Department of Plant Pathology, Private Bag X1, University of Stellenbosch, Stellenbosch, South Africa*

Despite that South African populations of *Tylenchulus semipenetrans* have been characterized morphometrically and morphologically, information about its molecular phylogeny lacks. In addition, ample information is available for plant-parasitic nematodes associated with citrus tree rhizospheres, while none exists for their co-inhabiting beneficial nematode counterparts. This study focused at determining i) the phylogenetic positions of six South African *T. semipenetrans* populations (from orchards situated in four provinces); and ii) the abundance and diversity of nematode communities from four Mpumalanga orchards and assessing whether associations exist between their densities and visually healthy-looking and declining citrus trees. Using the 18S, ITS and 28S rDNA molecular-based methods, South African *T. semipenetrans* populations grouped in a clade with exotic populations confirming their identity. Ten plant-parasitic nematode genera were identified, with *Tylenchulus semipenetrans* being predominant in soil samples, followed by Criconematidae. Of the 21 beneficial nematode genera and 3 families identified, *Acrobeloides* dominated, followed by *Anaplectus*. Relationships between PPN and category 1 trees (healthy) were evident for high abundance of *Criconema*, while high abundance of *T. semipenetrans* were associated with category 3 trees (severely declined): no relationships of this nature were evident for beneficial nematodes. Metabolic footprints of beneficial nematodes from all orchards plotted in the degraded and depleted quadrat of the soil food web. Abundance of bacterivores were positively correlated with pH, %C and Na:K, while PPN abundance was positively related with % clay, % silt and Mg:K. Although beneficial nematode abundance could not be associated with either healthy or declined trees, this study represents baseline information showing the poor status of soil health in the orchards studied.

S1-P38

Identifying molecular plant functions targeted by root knot nematode nuclear effectors.

Sarah Ranty-Roby[1] (sarah.ranty-roby@inrae.fr), Salomé Soulé[1], Claire Caravel[1], Nhat-My Truong[1, 2], Joffrey Mejias[1], Pierre Abad[1], Bruno Favery[1], Michaël Quentin[1]

[1] *INRAe-Université Côte d'Azur-CNRS, Sophia Antipolis, France*; [2] *Vietnamese - German Center for Medical Research (VG-CARE), Hanoi, Vietnam*

Root knot nematodes (RKN) are endoparasitic worms that invade plant roots causing economically important damages worldwide as they have a wide host range. Furthermore, RKN populations are predicted to geographically spread and to increase with climate warming. Through their stylet (a syringe-like organ), these pests inject proteins into plant cells to manipulate diverse functions to their advantage and to escape plant defenses. Indeed, RKN induce in the root, the formation of giant polynucleate cells, which constitute their feeding sites to drain plant nutrients thus affecting plant yield. Understanding the molecular dialog between plant and root knot nematodes is therefore of high interest to build new strategies of plant protection against these parasites. Transcriptomic analysis of the interaction between tomato and *Meloidogyne incognita* was performed in order to identify RKN putative secreted proteins, named putative effectors, more strongly expressed *in planta*. For these predicted effectors, we performed *in situ*hybridization in order to see whether they were expressed in the RKN salivary secreting glands suggesting effector injection *in planta*. Then, *in planta* localization of these candidates proteins by agro-infiltrating GFP-tagged effectors in tobacco was performed to select RKN effectors targeting the plant cell nucleus. . Finally, yeast two-hybrid screening was also undertaken in order to identify potential targets of these effectors in the tomato plant. Results will be presented for such putative effectors and discussed in the context of the biology of the interaction.

Keywords: Root knot nematodes - Effector - Giant cells - Susceptibility gene - Molecular dialogue.

S1-P39

Physiological and biochemical defense response of soybean cultivars parasitized by *Pratylenchus brachyurus.*

Leonardo Levorato Freire[1] (leolfreire@hotmail.com), Marta Cristina Corsi de Filippi[2], Anna Cristina Lanna[2], Mara Rúbia Da Rocha[1], Denner Robertfaria[3]

[1] *Programa de Pós-graduação em Genética e Melhoramento de Plantas, Universidade Federal de Goiás, Goiânia, GO, Brazil*; [2] *Embrapa Arroz e Feijão, Santo Antônio de Goiás, GO, Brazil*; [3] *Genetics and Crop Breeding, Universidade Federal de Goiás, Goiânia, GO, Brazil*

Pratylenchus brachyurus is one of the main plant parasitic nematodes present in Brazilian soybean crops. Its parasitism causes drastic reductions in the production of this crop and there is little information in the literature about how a plant interacts with this nematode in order to resist its parasitism. The objective of this study was evaluate the physiological and biochemical defense responses of soybean cultivars when parasitized by *P. brachyurus* and inoculated with the bacteria *Pseudomonas fluorescens* strain BRM 32111 (*PF*). The experiment was conducted under greenhouse in a completely randomized design in a 2 x 4 factorial scheme, for physiological variables, and 2 x 4 x 2 for biochemical variables. The treatments consisted of the combination of two soybean cultivars (BRSGO Caiapônia and BRSGO 8560 RR) with: 1) Control (plants without nematode and bacteria), 2) Pb (plants inoculated with *P. brachyurus*), 3) Pf (plants treated with *P. fluorescens*) and 4) PbPf (plants inoculated with *P. brachyurus* and treated with *P. fluorescens*). For biochemical analyzes the evaluations were performed in the leaves and the roots. The treatments with *P. fluorescens* were inoculated 24 hours after the nematode inoculation. Plants were evaluated weekly until 63 days after *P. brachyurus* inoculation. Plants with higher density of nematodes showed low photosynthetic rate, stomatal conductance, evapotranspiration and clorophyll content. Higher activity of chitinase, β-1,3-glucanase and lipoxygenase occourred in the roots and higher activity of phenylalanine ammonia lyase occourred in the leaves. In these conditions the nematode population was lower. The soybean cultivar BRSGO Caiapônia presented higher activity of chitinase, β-1,3-glucanase and lipoxygenase in the leaves and the roots than cultivar BRSGO 8560 RR. BRSGO Caiapônia also presented lower nematode population than BRSGO 8560 RR. The reproduction factor and nematode density in the roots were higher in plants treated with the *P. fluorescens*

Keywords: Induced resistance - Root lesion nematode - Photosynthesis - *Pseudomonas fluorescens*.

S1-P40

Pathogeneicity of *Meloidogyne incognita* populations from pepper crops in southeast Spain to resistant pepper.

Caridad Ros Ibáñez[1] (caridad.ros@carm.es), Adriana Esteban López[1], Mª Carmen Martínez Lluch[1], Mª Ángeles Hernández Colucho[1], Victoriano Martínez Alarcón[1], Celia Martínez Mora[2]

[1] *Crop Protection, Instituto Murciano de Investigación y Desarrollo Agrario y Medioambiental (IMIDA), La Alberca (Murcia), Spain*; [2] *Molecular Genetic Improvement, Instituto Murciano de Investigación y Desarrollo Agrario y Medioambiental (IMIDA), La Alberca (Murcia), Spain*

Meloidogyne incognita is one of the main soil pathogens in pepper greenhouses (*Capsicum annuum*) in the Campo de Cartagena. The use of genetic resistance is an effective method of nematode control. However, the overuse of genotypes carrying the Me3 gene in the same soil has caused the selection of populations capable of overcoming the resistance conferred by this gene. Thus, it is necessary to study the pathogenicity of pathogen populations to establish integrated strategies of control. Under controlled conditions, the incidence and severity of damage caused by populations of *M. incognita* to genotypes carrying Me1 and Me3 genes and a genotype with partial resistance was evaluated. The populations evaluated were isolated from roots of pepper plants without resistance, from greenhouses in which no resistance pepper plants had been cultivated except when grafted plants on Oscos was repeated during two consecutive years. The material used was HDA 330 (Me1 / Me1), HDA 149 (Me3 / Me3), Terrano (Me1 / me1 by Syngenta seeds), Osco (Me3 / me3 by Ramiro Arnedo), Alcos (partial resistance) and Lamuyo (without resistance) . The design was randomized blocks with five repetitions. The quantified parameters were the gall index, the number of egg masses and the percentage of affected plants. The results show that all populations are avirulent to the genotypes carrying the Me1 gene, one population affected the genotypes carrying the Me3 gene in addition to the population isolated from the greenhouse where Oscos had been grown; Alcos was infected at the same level as the susceptible variety by more than 40% of the nematode populations.

Keywords: Root-knot nematode - Virulence - Gene - Capsicum annum.

S1-P41

Unravelling the genetic and molecular basis of North-West European *Globodera pallida* populations overcoming resistance in potato.

Arno Schaveling (arno.schaveling@wur.nl), Mark Sterken, Geert Smant

Nematology, Wageningen University & Research, Wageningen, Netherlands

Potato cultivation is constantly being threatened by a range of microbial and animal pathogens. Persistent infections by the potato cyst nematode (PCN) species *Globodera rostochiensis*and *G. pallida* cause symptoms like stunting, leaf yellowing, and a reduction in tuber size in potato. The losses caused by PCNs amount to an estimated 9% of the global potato production. In PCN control, the use of nematode resistant cultivars plays a key role. However, since the first reports of resistance breaking *G. pallida* populations in Germany in 2014, the durability of PCN resistance in current potato cultivars has become a matter of concern. By using genome sequencing of offspring of *G. pallida* populations generated on a large set of potato cultivars with PCN resistance, we have identified a genome region in *G. pallida* significantly associated with virulence. We suspect that *G. pallida* virulence on current PCN resistant cultivars is determined by one or more genes located within this region. Our work focusses on the further functional characterization of genes encoding secretory proteins within this region. To this end, we combine spatial and temporal transcriptomics with gene silencing approaches to pinpoint the candidate gene(s) causal for breaking resistance. This will help us to resolve the genetic and molecular basis of virulence in *G. pallida* populations in North-West Europe. Importantly, this project will also enable making predictions about the durability of PCN resistance in current potato cultivars.

Keywords: Potato cyst nematodes - Globodera pallida - Secretory proteins - Effectors - Virulence.

S1-P42

What is the best inoculum density of *Meloidogyne incognita* to evaluate its reproduction factor in cotton?

Santino Aleandro Silva (santinoaleandro@gmail.com), Orazilia França Dorigo, Andressa Cristina Zamboni Machado

Nematology, IAPAR, Londrina, Paraná, Brazil

Cotton (*Gossypium hyrsutum*) is an important commodity and in Brazil occupies a significant area especially in Cerrado, which is the highest producer, primarily in Mato Grosso State. Abiotic and biotic stresses interfere in the development and yield of cotton plants and, among them, the phytonematodes are responsible for extensive losses. *Meloidogyne incognita* is widely distributed and causes significant losses to cotton in Brazil. Genetic breeding programs look for genotypes with resistance to pathogens, and the use of resistant cultivars is a well accepted technology to manage nematodes. The reproduction factor (RF = final population / initial population) is commonly used to phenotype responses to nematodes and, knowledge of appropriate nematode population densities to be inoculated is important, since high or low initial population densities could interfere in the correct classification of the resistance/ susceptibility of the genotype. Therefore, the objective of the present work was to observe the RF behavior of *M. incognita* in a susceptible cotton cultivar, FM 975 WS, under increasing initial population densities (1.000, 2.000, 3.000, 4.000, 5.000) and with evaluations at 30, 40, 60, and 80 days after the inoculation (DAI). In order to meet the assumptions of the model, Box-Cox procedure indicated that the transformation of RF values by log (y) was necessary. The variance analysis was significant for both factors and for the interaction between them and thus the population densities of nematodes were deployed within each evaluation period. According to data dispersion, for 30 DAI, no significant difference was observed between the initial population desities and at 60 DAI, the behavior of RF values linear; for 40 and 80 DAI, it was necessary to adopt nonlinear models, which are preferable due to the interpretation of the parameters. These models were reparametrizatied by the quadratic model. Based in the *Xi* parameter, initial population densities of 4.000 or more nematodes compromised the observation of the RF values while the initial density of 1.000 resulted in the higher RF values, after 40 DAI. We conclude that initial population densities varying from 1,000 to 2,000 *M. incognita* per plant should be used in experiments to phenotype cotton genotypes, with evaluation after 40 DAI, in order to guarantee an adequate classification of genotypes in relation to their resistance/ susceptibility to *M. incognita*.

Keywords: Root-knot nematode - Regression models - Phenotyping.

S1-P43

Functional characterization of root-knot nematode effectors and their host targets during giant feeding cell ontogenesis.

Salomé Soulé (salome.soule@inrae.fr), Bruno Favery, Michaël Quentin

INRAe, Université Côte d'Azur, CNRS, Sophia Antipolis, France

Root-knot nematodes (RKN) are microscopic parasitic worms able to infest roots of thousands of plant species and causing massive crop yield losses worldwide. Within host roots, RKN induce formation of galls by redifferentiating five to seven root cells into giant and multinucleated feeding cells. These giant cells supply water and essential nutrients for nematode development. The formation and maintenance of giant cells is the result of an extensive regulation of the gene expression within targeted root cells and manipulation of key host functions. Secreted RKN effectors are key actors of this cellular reprogramming. They are mostly synthesized in the esophageal glands of the nematode and injected into the plant cells via the syringe-like stylet. Recent progress in nematode genomics and transcriptomics, has allowed identifying a large panel of RKN-specific effectors, conserved in the genome of the five main RKN species: *M. enterolobii*, *M. arenaria*, *M. javanica*, *M. incognita* and *M. hapla*, and notably produced during the parasitic stages. By performing *in situ* hybridization, we demonstrated specific expression of some genes, e.g. EFFECTOR17 (EFF17), in the esophageal glands of diverse RKN species. To decipher the role of these candidate effectors during parasitism, we are characterizing their plant targets in tomato and Arabidopsis. I will present the cellular functions that may be hijacked by EFF17. The identification of plant targets of protein effectors will open up new perspectives for the control of these pests and will provide a better understanding of the molecular dialogue that is established between the pest and the host plant.

Keywords: Meloidogyne spp - Effector - Giant cells - Susceptibility genes - Molecular plant-pathogen interaction.

S1-P44

Arabidopsis thaliana phenotyping to *Meloidogyne paranaensis* interaction and identification of SNPs linked this trait.

Juarez Tomaz[1] (tomaz@iapar.br), Arthur Gouveia[1], Caroline Ariyoshi[1], Geovana Yop[1], Shahid Siddique[2], Andressa Machado[3]

[1] *Laboratory of Biotechnology, IAPAR, Londrina, PR, Brazil*; [2] *Department of Entomology and Nematology, University of California, Davis, CA, United Nations*; [3] *Laboratory of Nematology, IAPAR, Londrina, PR, Brazil*

Meloidogyne paranaensis is the most aggressive nematode for *Coffea arabica* crops, causing annual losses up to billions of dollars to the sector. Despite this importance, little is known about *C. arabica* and *M. paranaensis* molecular interaction. Time, space and labor needed to phenotype coffee plants lead to this lack of knowledge. An alternative to accelerate the knowledge about plant x nematode interaction could be the use of *Arabidopsis thaliana* as a host. This study aimed to identify genes possibly involved in *A. thaliana* and *M. paranaensis* interaction, by Genome-Wide Association Study (GWAS). For this, we phenotyped 21 *A. thaliana* ecotypes for reproduction factor (RF) and number of nematodes per gram root (NNRG), which were submitted to the Scott-Knott test at 5% of significance. The genotyping was available in the GWA-Portal, fullsequence database platform and were filtered with minor allele frequency (MAF)> 0.1, using only intragenic and non-synonymous SNPs. The genotypic and phenotypic data were associated using the Linear Model and Accelerated Mixed Model. The ecotypes were distributed in 6 and 7 groups of reaction according to RF and NNRG data, respectively. Two SNPs were highly associated, using RF as phenotype, in both models. A SNP was found in a gene corresponding to a Fantastic Four family member and the other in an Ankyrin repeat family protein. For the NNRG phenotype, 2 SNPs were also associated in both models, one corresponding to an unknown transmembrane protein and the other to a pectin methylesterase inhibitor superfamily member.

Keywords: Root-knot - Ecotype - Phenotyping - Model plant - Genotyping.

S1-P45

TIR-NBS-LRR genes in *Prunus* spp. offer a fantastic resource to decipher the molecular determinants involved in root-knot nematode resistance.

Cyril Van Ghelder[1] (cyril.van-ghelder@inrae.fr), Henri Duval[2], Caroline Callot[3], Emeric Dubois[4], Marianne Mazier[2], Daniel Esmenjaud[1], Simon Saucet[1]

[1] *IPN, INRAe, Sophia Antipolis, France*; [2] *GAFL, INRAe, Avignon, France*; [3] *CNRGV, INRAe, Toulouse, France*; [4] *MGX, CNRS, Montpellier, France*

Root-knot nematodes (RKNs), *Meloidogyne* spp., severely challenge plants worldwide and especially perennials. In *Prunus* spp., the genetic resistance to RKNs mainly relies on NBS-LRR genes [1] that are key factors for pathogens control. The *Ma*plum, *RMja* almond and *RMia* peach genes display different spectra, which are now well defined, for resistance (*R*) to RKNs. The *Ma* gene, cloned in 2011 [2], is characterized by a complete-spectrum and heat-stable resistance that has not been overcome under field and controlled conditions up-to-now. A functional annotation of the TIR-NBS-LRR (TNL) gene family in the peach genome, the model genome for *Prunus*, and in other plant genomes showed that Ma has a peculiar TNL structure which is unique among plants [3]. A previous work had shown that the *RMja* gene mapped on the same chromosome as *Ma* and a high-resolution mapping, from a large almond segregating progeny completed by the construction of a BAC library, allowed to locate the *RMja* candidate region within a 148-kb sequence. We showed that *RMja* maps in the *Ma* resistance cluster and that the *Ma* orthologue is by far the best candidate for *RMja* [4]. This co-localization opens the way to unravel the molecular determinants involved in the resistance to RKNs at this locus. Future research will exploit the sequence polymorphism of three *Ma* orthologous regions, respectively, within plum (complete *R* spectrum), almond (incomplete *R* spectrum) and peach (null *R* spectrum from the RKN-susceptible peach genome sequence). Analysis of the *Ma* TNL cluster revealed that it has evolved orthologues with a unique conserved structure comprised of five repeated post-LRR (PL) domains. In model-plants TNLs with single PL domains, it has been shown that the integrity of this domain is required for function [5]. Besides, recent structural studies revealed the crucial role of PL (also nammed C-JID) domains in effector recognition. In woody perennials such as *Prunus* spp., the polymorphism contained in those PL domains might underlie differential resistance interactions with RKNs and an original immune mechanism.

Keywords: Resistance - Root-knot nematode - TIR-NBS-LRR - Prunus - PL / C-JID domain.

References:

- [1] Saucet et al. 2016. New phytologist. 211: 41-56.
- [2] Claverie et al. 2011. Plant Physiology. 156: 779-92.
- [3] Van Ghelder and Esmenjaud. 2016. BMC Genomics 17: 317.
- [4] Van Ghelder et al. 2018. Frontiers in Plant Science 9: 1269.
- [5] Saucet et al. 2020. Molecular Plant-Microbe Interactions 34:286–296.

S1-P46

Identification of promising genes for resistance to potato cyst nematodes.

Raquel Varandas[1] (raquelrv@gmail.com), Isabel Luci Conceição[1], Conceição Egas[2, 3]

[1] *CFE-Centre for Functional Ecology-Science for People & the Planet, Department of Life Sciences, University of Coimbra, Coimbra, Portugal*; [2] *CNC-Centre for Neuroscience and Cell Biology, University of Coimbra, Coimbra, Portugal*; [3] *Biocant – Transfer Technology Association, BiocantPark, Cantanhede, Portugal*

Potato cyst nematodes (PCN), *Globodera pallida* and *G. rostochiensis*, are a serious threat to potato production globally and are identified by the European and Mediterranean Plant Protection Organization (EPPO) as quarantine organisms (A2 list). The lack of effective control measures and the limitation on the use of chemical pesticides makes it imperative to develop alternative methods for the control of these nematodes. *Solanum sisymbriifolium*, is a plant resistant to PCN and capable of inducing juveniles hatching, being used as a trap crop. However, the molecular mechanisms underlying this resistance are still largely unknown. This work aimed to elucidate some of the genes that may be involved in the resistance of this plant.
Gene expression profiles were characterized in roots of two cultivars of *S. sisymbriifolium*, uninfected and infected by *G. pallida* and several gene expression changes were validated by quantitative RT-PCR (qPCR). The transcriptome sequencing data identified 839 and 699 downregulated genes in cvs. Melody and Sis 6001, respectively, with several genes encoding proteins with transmembrane transporter activity. A larger number of upregulated genes were identified (3248 in cv. Melody and 1344 in cv. Sis 6001), and many genes were associated with cell growth, oxidative stress, and resistance processes. One promising upregulated gene detected was the *SAR8.2* gene, associated with the salicylic acid (SA) pathway and the systemic acquired resistance. These findings are an important step to elucidate the molecular mechanisms of the resistance of plants to nematodes and help develop more effective control strategies.

Keywords: Potato Cyst Nematodes - Molecular mechanisms of resistance - Solanum sisymbriifolium - SAR8.2 gene.

S1-P47

Study of the pathogenicity of *Bursaphelenchus xylophilus* to seedlings of *Larix olgensis*.

Xizhuo Wang (ladydal@163.com), Laifa Wang, Yefan Cao

RIFEEP, Chinese Academy of Forestry, Beijing, China

Pine wilt disease (PWD) is a systemic disease with pinewood nematode (PWN) *Bursaphelenchus xylophilus* as the pathogen and *Monochamus* insects as the vector [1, 2]. The pathogenicity of PWNs to *Larix olgensis* has not been reported so far. To provide theoretical basis on pathogenicity mechanism of *L. olgensis* with PWD, we investigate the pathogenicity and migration of PWN and the activity changes of superoxide (SOD) in *L. olgensis* after inoculated with PWNs. Two-year-old *L. olgensis* seedlings were inoculated separately with three different isolates PWN(QH-1,NM-1, and CM-1), These isolates come from different provinces in China, and the geographical distance is more than 1400 kilometers. After inoculation of 2,000 PWNs per seedling, the pathogenicity, the external symptom and the nematode migration were observed daily. Leaves were collected every day till 7days after inoculation, to analysis the activity change of SOD in *L. olgensis*. In 35 days after the inoculation, the morbidity was recorded and PWNs were extracted separately from wilt seedlings. The pathogenicity and migration of PWNs in *L. olgensis* seedlings and the activity change of SOD were analyzed. *L. olgensis* seedlings were wilt after inoculated QH-1, NM-1 and CM-1 PWNs, but QH-1 PWNs caused seedlings to wilt 5days earlier than other isolates. In 35 days after the inoculation, the morbidity rates of seedlings inoculated were 100%, 70% and 60%, respectively, and the disease-infected indices were 100, 43.3 and 40, respectively. The number of PWNs of QH-1 treatment was significantly greater than NM-1 and CM-1 treatments ($P<0.01$). The migration ability of PWNs after inoculated showed the differences between QH-1, NM-1, and CM-1, the migration ability of QH-1 was higher than other isolates. The activity change of SOD in *L. olgensis* showed the differences between QH-1, NM-1, and CM-1 PWNs treatments. The observation of cross-section showed browning of pith and cavitation of xylem occurred in the early stage, and the QH-1 PWNs treatment caused more serious damage than other PWNs treatments. The pathogenesis of *L. olgensis* and the fertility in different strains of PWNs were significantly different, which may be related to environmental differences leading to changes in the adaptation of PWNs. Fewer PWNs were also collected from the healthy appearance seedlings after inoculated, which means that the wilt of *L. olgensis* is relevant to the migration ability and the fertility of the PWNs.

Keywords: *Bursaphelenchus xylophilus* - *Larix olgensis* - Pathogenicity - Inoculation - Morbidity.

References:

- [1] Futai et al., 2013. Annu Rev Phytol., 51(51): 61-83.
- [2] Ye et al., 2010. Beijing: China For Publishing House, 7-11.

S1-P48

miRcn1 targets ethylene responsive transcription factor 4 (ERF4) to enhance resistance to root-knot nematodes in tomato.

Lihui Wei (weilihui@jaas.ac.cn), Chunting Wang, Dongmei Zhou

Institute of Plant Protection, Jiangsu Academy of Agricultural Sciences, Nanjing, China

Root-knot nematodes (RKN; *Meloidogyne* spp) are plant endoparasites with a broad host range causing huge losses in the world. MicroRNAs play important roles in plant responses to abiotic and biotic stresses by regulating their target gene expressions. There is limited information on the roles of miRNAs play in plant-RKN interaction at early infection stage.To address this, miRNAs of RKN infected and uninfected roots from tomato (*Solanum lycopersicum*) were sequenced by high throughput sequencing. The miRNA- target gene interaction was confirmed by western-blot and real-time PCR. Over-expression transgenic lines of miRNA and target gene are generated to investigate the RKN infection.A novel miRNA (named as miRcn1) was down-regulated upon *M. incognita*infection at 24 hpi, while its target gene ethylene responsive transcription factor 4 (ERF4) was up-regulated. The interaction of miRcn1-ERF4 was further validated by Western-blot and real-time PCR. The miRcn1 over-expressed transgenic line showed an enhanced resistance to *M. incognita* and a lower expression of ERF4. On the contrast, the ERF4 over-expressed transgenic plants were more susceptible to *M. incognita* and led to defect in growth. Altogether, miRcn1 positively regulates tomato resistance to *M. incognita* infection by suppressing ERF4 expression.

Keywords: Root-knot nematodes - miRNAs - ERF4 - miRcn1 - Resistance.

S1-P49

The Arabidopsis transcription factor TCP9 regulates root growth response during cyst nematode infections via ROS-mediated signalling.

Jaap-Jan Willig (jaap-jan.willig@wur.nl), Willem de Jong, Mark Sterken, Joris van Steenbrugge, Jose Lozano Torres, Aska Goverse, Geert Smant

Laboratory of Nematology, Wageningen University and Research, Wageningen, The Netherlands

Low levels of infection by cyst nematodes on some resistant crop varieties result in significant loss in yield, while other heavily infected susceptible varieties show hardly any symptoms at all. This suggests that some plants tolerate biotic stress by plant parasitic nematodes better than others. We hypothesize that tolerant plants accommodate nematode infections by altering the architecture of their root system. Our current objective is to investigate whether the transcription factors TCP9 and TCP20, which are known regulators of plant root architecture, are involved in tolerance to cyst nematodes in Arabidopsis. To this end, we first tested whether TCP9 and TCP20 regulate susceptibility of Arabidopsis to the beet cyst nematode *Heterodera schachtii*. Next, we analysed the root architecture of nematode-infected roots of Arabidopsis *tcp9* and *tcp20* knock-out mutants to assess whether these genes are involved in alterations in primary and secondary roots associated with nematode infections. To better understand why *tcp9* shows a tolerant root phenotype, we performed an RNAseq experiment which shows that there is an increase in peroxidase activity during nematode infection. Finally, we analysed reactive oxygen species biosynthesis and accumulation.

Keywords: Phenotypic Plasticity - Tolerance - TCP transcription factor - Plant-nematode interaction - *Heterodera schachtii.*

S1-P50

Manipulation of host proteins and processes by potato cyst nematode effector RHA1B.

Fangming Xiao (fxiao@uidaho.edu), Li Huang, Yulin Yuan, Joanna Kud, Louise-Marie Dandurand

University of Idaho, Moscow, Idaho, United States

Objective: In order to colonize host plants successfully, the potato cyst nematode *Globodera pallida* secrets effectors to suppress plant defense responses and manipulate host cells into the feeding site syncytium. We have identified a *G. pallida* effector RHA1B that is a ubiquitin ligase and plays a significant role in the parasitism. Significantly, RHA1B can suppress defense signaling and function in concert with multiple host ubiquitin conjugating enzymes to catalyze ubiquitination involved in both proteolytic and non-proteolytic protein modifications. Our goal is to understand the mechanistic basis by which *G. pallida* uses RHA1B to manipulate host physiological processes for successful colonization.
Methods: 1) Immunoprecipitation-based tandem mass spectrometry technique to identify targets of RHA1B in potato; 2) Transcriptome profiling on transgenic potato roots expressing RHA1B to determine potato genes manipulated by RHA1B; 3) Highly sensitive immune affinity purification and liquid chromatography tandem mass spectrometry (LC–MS/MS) analysis to determine host proteins ubiquitinated by RHA1B directly and indirectly.
Results: 1) Multiple putative targets of RHA1B have been identified and determined, including a nematode-specific immune receptor NILR1, a highly conserved CcR4-Not complex involved in regulation of gene expression, a receptor-like protein kinase FERONIA involved in plant defense and root growth and development, and a kinesin-like protein KIN12B related to cell division and cargo transport. Further biochemical analyses indicate RHA1B ubiquitinates Ccr4-NOT and NILR for degradation but, to our surprise, stabilizes KIN12B. 2) RNA-Seq analysis indicates more than 2000 genes are either up- or down-regulated in the RHA1B-expression transgenic potato roots, including large number of genes involved in hormone-based defense response, cell wall modification and cell division/growth. We hypothesize RHA1B indirectly manipulates these genes by ubiquitination-mediated modification of intermediate regulators, of which, CcR4-NOT is thought to be a key factor for such gene regulation. 3) Analysis of potato RHA1B-expression root ubiquitome is in progress and the newly generated data will be presented in the conference.
Conclusion: Our results suggest that *G. pallida* uses RHA1B as a molecular weapon to affect a wider range of physiological processes of host plants via manipulation of both proteolytic and non-proteolytic protein process in infected hosts.

S1-P51

Activation of Gpa2 by *Globodera pallida* RBP(P) variants is suppressed by RBP (S) effector variants: a molecular interplay between different RBP family members.

Vera Putker (vera.putker@wur.nl), Afke Zijlstra, Hein Overmars, Qi Zheng, Joris van Steenbrugge, Amalia Diaz Granados Munoz, Mark Sterken, Geert Smant, Aska Goverse

[1] *Laboratory of Nematology, Wageningen University & Research, Wageningen, Netherlands*

Plant-parasitic nematodes effectors are defined as proteins and small peptides with a wide range of molecular functions that either assist in host invasion, modulation or plant immune responses, or initiation and maintenance of the permanent feeding site. The potato cyst nematode *Globodera pallida* secretes, amongst others, the effector protein RBP which is in case of plant resistance, recognised by its matching immune receptor encoded by a specialised resistance (R) gene: Gpa2. Individuals of the avirulent *G. pallida* population D383 solely produces Gpa2-mediated cell death eliciting RBP effector proteins, while the virulent population Rookmaker produces both Gpa2-mediated cell death eliciting and non-eliciting RBP effector proteins. One determinant for specific nematode recognition of RBP is a polymorphism on position 187 in its amino acid sequence: while a Proline variant (RBP-P) does elicit a Gpa2-mediated response, Serine variants (RBP-S) do not. Based on the co-existence of RBP-P and RBP-S variants in the virulent population Rookmaker we hypothesize an interplay between these two effector variants in which we think that RBP-S variants are dominant over RBP-P variants and somehow interfere in the detection or activation by Gpa2. To test this, we combined RBP-P and RBP-S variants in cell death assays, both in the model plant *N. benthamiana* as well as in the host plant *Solanum tuberosum*. Here we show for the first time that RBP-S is able to suppress the cell death eliciting effect of RBP-P variants.

Keywords: *Globodera pallida* - Potato cyst nematode - Effector biology - Plant immunity.

S1-P52

Population dynamics and diagnosis of *Hoplolaimus galeatus* on hybrid bermudagrass.

William Crow (wtcr@ufl.edu)

Entomology and Nematology, University of Florida, Gainesville, Florida, United States

Hoplolaimus galeatus has long been recognized as causing damage to turfgrasses in the United States. However, due to a lack of effective control measures, the importance of this nematode for turfgrass managers has increased greatly in recent years. On turfgrasses this nematode feeds as both an ectoparasite and as a migratory endoparasite. Bermudagrass (*Cynodon dactylon* and *Cynodon* hybrids) is the predominant grass used on golf courses and athletic fields in the southeastern United States. Bermudagrass is perennial, so in most cases nematode diagnosis is conducted on actively growing turf in the field, and all life stages of *H. galeatus* can be found in the rhizosphere soil and within the roots at any given time. A two year study was conducted in two locations in Florida, one in northern Florida and one in southern Florida. Both locations were planted to hybrid bermudagrass that was naturally infested with *H. galeatus*. At each site there were 120 small plots, with 60 plots receiving no nematicides and 60 receiving applications of fluopyram and abamectin. Turf health and root health were monitored throughout the trial. Nematode samples were collected 4 times per year, and nematodes were extracted from soil using centrifugal-flotation and from roots using mist incubation. Regression was used to quantify the relationships of *H. galeatus* with turf and root health at different sampling and evaluation times. Population densities of *H. galeatus* increased in nematicide treated plots compared to non-treated plots, and correlations between *H. galeatus* and turf health were greater in treated than untreated plots. Both soil and root counts of *H. galeatus* were often correlated with turf health, but correlations were typically greater using counts from soil than from roots. Correlations between counts of *H. galeatus* and turf health were usually greater for turf measurements taken several months after collection of nematode samples than for measurements occurring at the same time as sampling. These results indicate that extraction of *H. galeatus* from soil is generally superior to extraction from roots for diagnostic purposes. Also, counts of *H. galeatus* are better at predicting future nematode damage than at estimating current damage. Finally, the most effective turfgrass nematicides currently used on golf and sports turf are not effective against *H. galeatus*, and damage from this species will likely continue to increase without new tools for their management.

Keywords: Lance nematode - Turfgrass - Diagnostics - *Hoplolaimus galeatus*.

S1-P53

A glycolytic enzyme from plant parasitic nematode induce defence responses in plants.

Badou Mendy[1] (bmendy@uni-bonn.de), Neil Pep Dave Sumaya[1], Maximilian Euler[1], Muhammad Ilyas[1, 2], Divykriti Chopra[1], Alexander Graf[3], Sylvia Schleker[1], Sebastian Eves-van den Akker[4], Shahid Siddique[5], Florian M.W. Grundler[1]

[1] *Molecular Phytomedicine, Rheinische Friedrich-Wilhelms-University of Bonn, INRES, Bonn, North Rhine-Westphalia, Germany*; [2] *Section of Population Genetics, School of Life Sciences, Technical University of Munich, Munich, Bayern, Germany*; [3] *Wissenschaftspark Golm, Max-Planck-Institut für Molekulare Pflanzenphysiologie, Potsdam, Brandenburg, Germany*; [4] *Department of Plant Sciences, The Crop Science Centre, University of Cambridge, CB2, 3EA, United Kingdom*; [5] *Department of Entomology and Nematology, University of California, One Shields Ave, Davis, CA 95616, United States*

Plants adapt to adverse environmental conditions and pathogens attacks with the help of a complex defense network. For the past years, research works has focused on understanding these defense system and identifying molecular players including pathogen associated molecular patterns (PAMPs) that are recognized by plants cell-surface localized receptors leading to PAMP-triggered immunity (PTI). Although numerous PAMPs have already been identified from different plant pathogens yet very little information remains to date regarding the molecular signature derived specifically from plant-parasitic nematodes (PPNs). Previous work with the nematode aqueous diffusate known as NemaWater revealed that a proteinaceous molecule from plant-parasitic nematode is/ are able to activate PTI responses in plants. Here, we characterized enolase, one of the abundant proteins in NemaWater in its ability to activate plant defense. We observed that yeast enolase, a commercially available protein with similar molecular weight as PPN enolase is able to induce oxidative burst and growth inhibiting properties analogous to NemaWater in Arabidopsis. Moreover, plants treated with 10 µg/ml yeast enolase showed an increased resistance against *Heterodera schachtii* and *Meloidogyne incognita.* Using glucuronidase expression lines, we show that yeast enolase can trigger induction of camalexin, an essential phytoalexins against pathogen attack. Furthermore, recombinant enolase from *H. Schachtii* is able to induce ROS-burst in Arabidopsis, tomato and tobacco plants. These results indicate the potential of enolase as one of the specific immune elicitors contained in NemaWater. The capacity of nematode enolase to induce plant defense responses will be discussed.

Keywords: Elicitor - NemaWater - PAMPs - Phytoalexin - PTI.

References:

- Mendy et al., 2017. PLOS Pathogens 13, e1006284.
- Zhang et al., 2018. Acta Parasitologica 63, 252–260.
- Pancholi, 2001. Cellular and Molecular Life Sciences 58, 902–920.
- Chen et al,. 2012. Vaccine 30, 3478–3482.
- Didiasova et al,. 2019. Frontiers in Cell and Developmental Biology 7, 1–11.

S1-P54

An investigation of organophosphate nematicides' effects on *Pratylenchus loosi,* a root lesion nematode in tea plantations

Reza Nafari[1] (rezanafari1987@gmail.com), Zeynab Rezayat[2]

[1] *Shiraz university, Shraz, Fars, Iran*; [2] *Tehran University, Tehran, Iran*

Root lesion nematode, *Pratylenchus loosi*, is one of the most important pests of tea plantations that causes severe damage to the mature tea plants. In order to reduce damage and nematode population on tea plants, organophosphate nematicides Rugby 10G (Cadusafos) and Nemacure 10G and EC400 (Fenamiphos) were tested each at two rates, under a complete Randomized Block Design in a time span of two years. The experiments were carried out in two different areas of tea plantation, Lashidan and Zomeidan in the north of Iran. Root and soilsamples were taken prior to application and two, fiveand nine months after applicationin the first year and two, six and thirteen months after application in the second year. The nematicides caused significant reduction of nematode populationsin the roots (in Farsi text) at all of the dosagesand all sampling intervals, the maximum reduction was obtained in the second year. The green leaf tea yield was significantly increased in plants treated with nematicides.

Keywords: Nematicides - Root lesion nematode - *Pratylenchus loosi* - Tea.

POSTERS

S2. Systematics, phylogeny and phylogeography

S2-P01

Occurrence and population dynamics of *Ditylenchus dipsaci* (stem and bulb nematode) on chickpea fields in Turkey.

Tohid Behmand[1] (tohid.behmand63@gmail.com), Jens berger[2], Abdullah kahraman[3], Ibrahim halil elekcioğlu[1]

[1] *Plant protection department, cukurova university, Adana, Turkey*; [2] *CSIRO Agriculture Flagship, Centre for Environment and Life Sciences, Canberra, Australia*; [3] *Harran University, Faculty of Agriculture, Department of Crop Science, Şanlıurfa, Turkey*

Ditylenchus dipsaci is one of the most damaging plant parasitic nematodes and is widely distributed mainly in temperate areas and attack almost crops (especially chickpea), causing significant damage in Turkey. It is of major economic significance worldwide. This study was conducted to evaluate the population dynamics of the *Ditylenchus dipsaci* in the twelve accessions of wild *Cicer* spp. (*C. reticulatum* and *C. echinospermum*) and domesticated *C. arietinum* in two regions (Ankara and Şanliurfa) during 2015 and 2016 chickpea growing season. A survey was conducted from 36 cicer produced provinces and 211 root and soil samples were collected. *Ditylenchus dipsaci* was detected in 95 soil samples (45% of the total samples). Density of *Ditylenchus dipsaci* populations were highest during June to mid-summer (July) and was lowest during cold months winter (February-March). Moreover, the results showed a positive correlation between soil temperature and population development of nematodes in both regions. Also, there was a significant difference in the reproductive rate of *D. dipsaci* among twelve accessions of chickpea ($P<0.001$). Development of resistant chickpea remains a principal goal in the management of this nematode. The output of this study could be used to advantage in fields infested with this nematode by helping select cultivars with the best levels of partial resistance and providing information that will be useful for suppressing the nematode population below the threshold level.

Keywords: Population dynamics - Ditylenchus dipsaci - Resistance - Chickpea varieties.

S2-P02

A catalogue of nematode karyotypes.

Mark Blaxter[1] (mb35@sanger.ac.uk), Chloe Leech[1], David Lunt[2]

[1] *Tree of Life, Wellcome Sanger Institute, Cambridge, United Kingdom*; [2] *Evolution Group, University of Hull, Hull, United Kingdom*

Observation of the behaviour of nematode chromosomes during meiosis by Boveri was critical to the development of the germline theory of inheritance in the late 19th century. In the current era of genomics, knowledge of the karyotype of a species is very useful in conditioning expectations of a genome assembly. *Caenorhabditis elegans* has six chromosomes, comprising five autosomes and an X. Other *Caenorhabditis* species also have six chromosomes, but in the sister group to *Caenorhabditis*, the *Protorhabditis-Diploscapter* clade, chromosome numbers range from 6 to 1. These changes in chromosome number imply changes in gene linkage, and also therefore in gene coregulation. Previous catalogues of nematode chromosome numbers were limited in scope (for example including data from only plant parasites or only animal parasites) or contain species names that are no longer current. We have collated a catalogue of chromosome numbers, and sex determination karyotypes, from literature from 1883 to the present day. We identified the current names for species described in older literature, and placed these data in a modern taxonomy. We identified chromosome counts for 218 species from 95 genera in 44 families. As is often the case in Nematoda, free-living species from non-rhabditid Chromadoria, Enoplia and Dorylaimia are poorly represented, Haploid chromosome numbers range from 1 to over 50, with variability especially in the plant parasitic Meloidogyninae. The modal reported haploid chromosome number is 6. Sex determination mechanisms, derived from karyotypic differences between males and females, and between different spermatid nuclei, are commonly XX-X0 with several independent origins of XX-XY systems. Ascaridomorpha includes many species with multiple X chromosomes. Many nematodes are parthenogens, and parthenogenesis is associated in some lineages with polyploidy (triploidy and tetraploidy). The catalogue is available via the Genomes on a Tree datasystem (https://goat.genomehubs.org/).

Keywords: Karyotype - Sex determination - Genomics - Catalogue.

S2-P03

Epidemiological approach to potato cyst nematodes in Portugal, with special reference to *Globodera pallida*.

Maria João Camacho[1, 2] (mjoao.camacho@iniav.pt), Eugénia de Andrade[1, 3], Manuel Mota[2], Maria L. Inácio[1, 3]

[1] *Instituto Nacional de Investigação Agrária e Veterinária (INIAV, I.P.), Oeiras, Portugal*; [2] *Mediterranean Institute for Agriculture, Environment and Development (MED), University of Évora, Portugal*; [3] *3GREEN-IT Bioresources for Sustainability, ITQB NOVA, Oeiras, Portugal*

The occurrence of the potato cyst nematodes (PCN) in Portugal has been recorded for decades. *Globodera rostochiensis* was first reported in 1956 [1] and *Globodera pallida* in 1988 [2], both in the north of Portugal. To establish the status of PCN in the country, a field survey was conducted in 2010. This work aims to identify where PCN infestations occur in Portugal and which species are present in these regions. This information is essencial to plan and implement strategies for managing this pest. This research focused on PCN species, *Globodera pallida* and *Globodera rostochiensis*. From 2013 until 2019, 748 soil samples from the rhizosphere of different potato cultivars were surveyed in the Portuguese mainland to detect and identify both species and track their location. PCN are widespread invasive species throughout Portugal. In fact, during the survey period an incidence of 22.5% was estimated for the tested samples. The patterns of infestation vary among regions, increasing from south to north, where PCN were first detected. Currently, both species are present in all potato producing regions of the country, with a greater incidence of *G. pallida*. Phytosanitary control measures are influencing to the observed results. The use of potato cultivars resistant to *G. rostochiensis* led to a decrease of this species but had no influence on *G. pallida* detections, which continues its reproduction freely since there are no effective resistant cultivars for this species. The relationship between the presence, infestation rate, spread and geographical distribution of PCN is discussed in terms of behavioral responses of the potato cultivars and the implications for developing new integrated crop protection measures.

This work is funded by National Funds through FCT - Foundation for Science and Technology under the Project SFRH/BD/138724/2018

Keywords: Survey - Identifications - Management.

References:

- Camacho MJ, Andrade E, Mota M, Nóbrega F, Vicente C, Rusinque L, Inácio ML. 2020. Frontiers in Plant Science 11:606178.

S2-P04

Determination of cyst nematods affecting commercial fields of potato in Julcan, La Libertad, Peru.

Carolina Cedano[1] (carolina_cedano@hotmail.com), Martín Delgado[2], Zinthia Neira[1], Génesis Becerra[1]

[1] *Universidad Nacional de Trujillo, Trujillo, La Libertad, Perú*; [2] *Universidad Privada Antenor Orrego-UPAO, Trujillo, La Libertad, Perú*

In the Andean area of Peru, potato production is limited by the presence of different species of cyst-forming nematodes, which have wide geographic distribution due the easy dissemination and few effective control measures [1]. The objective of this research was to identify the species of cyst-forming nematodes predominant in the commercial potato fields in Julcán, La Libertad, Peru. For that purpose soil samples were taken in different commercial fields of the study area. The samples were screened to remove impurities and dried at room temperature. Subsequently, from each sample, 100 grams of soil were taken in triplicate, the cysts were extracted and the average number of cysts per sample was determined. For the morphometric and anatomical characterization of the cysts and juveniles of the second stage, 25 cysts were taken from each sample, dissections were made to determine shape, color and size [2]. Likewise, cuts of the perineal region were made, to observe the characteristics of the vulvar fenestra, the distance between anus and vulva was measured, the number of stretch marks between fenestra and anus, diameter and shape of the fenestra was counted and with the data determined the Graneck Radio. In addition, the shape and size of the eggs, length of the juveniles, length of the stylet and shape of the basal nodules as well as the size and shape of the tail were determined. The results of morphometric and anatomical studies indicated that *Globodera rostochiensis*, *Globodera pallida* and *Heterodera* spp. are affecting potato fields in Julcán, La Libertad.

Keywords: "Cyst Nematode", fenestra, morphometry, Graneck Radio

References: • [1] Franco J. (1994). Nematode problems in potato production in temperate climates in the Andean region. Nematropica 24:179-195.
[2] Oepp, B. et al. (2013). PM 7/40 (3) *Globodera rostochiensis* and *Globodera pallida,* 43, 119–138.

Keywords: Cyst Nematode - Fenestra - Morphometry - Juveniles - Graneck Radio.

References:

- [1] Franco J. (1994). Nematode problems in potato production in temperate climates in the Andean region. Nematropica 24:179-195.
- [2] Oepp, B. et al. (2013). PM 7/40 (3) Globodera rostochiensis and Globodera pallida, 43, 119–138.

S2-P05

Integrative taxonomy of *Meloidogye ottersoni* (Thorne,1969) Franklin, 1971 parasitizing flooded rice in Bazil.

Vanessa Mattos[1] (nessinha.agro@gmail.com), Raycenne Rosa Leite[2], Ana Cristina M.m. Gomes[1], Leandro Grimaldi Py[3], Daniela A. Souza[1], Philippe Castagnone-Sereno[4], Juvenil E. Cares[2], Regina M.d.g. Carneiro[1]

[1] *Embrapa, Brasilia, Brazil*; [2] *University of Brasilia, Brasilia, Brazil*; [3] *Rua Frei Humberto Weil, 29, Bairro Pereq, Santa Catarina, Brazil*; [4] *INRAe, Sophia Antipolis, France*

A root-knot nematode (RKN) parasitizing rice (*Oryza sativa* L.) and causing damage in flooded field conditions in Santa Catarina (SC), Rio Grande do Sul (RS) and Paraná (PR) states (Brazil) was identified as *Meloidogyne ottersoni*. The species was redescribed from the Brazilian population from Meleiro (SC) and compared with the type description of *M. ottersoni* (Wisconsin, USA) with additional morphological, biochemical and molecular characterization. The female and male presented smaller stylets: 10-12 µm, 14-16 µm, respectively, when compared with *M. graminicola*: 12-14 µm, 16-18 µm, and *M. oryzae*: 14-16 µm, 28-20 µm. *M. ottersoni* presents perineal patterns located on the contour of a slight protuberance. Striae mostly continuous, never raised by transverse irregular striae, which are frequently observed in *M. graminicola* and *M. oryzae*. *Meloidogyne ottersoni* belongs to the RKN group 11 (Jepson, 1987) and reproduction is by meiotic parthenogenesis and the somatic chromosome number is 18. The tails of second-stage juveniles are very long and thin and tapers to a long, narrow, irregular hyaline terminus. The ability of Brazilian *M. ottersoni* population to parasitize the type host: reed canary grass, *Phalaris arundinacea* L., was confirmed. Biochemically, *M. ottersoni* presents a distinct esterase profile (Est) without bands (Est Ot0, Rm=0) for differentiating it from other species: *M. graminicola* Est VS1 (Rm: 0.70) and *M. oryzae* Est O1, (Rm: 1.02). In Maximum Likelihood analysis of ITS, D2D3 rRNA and coII-16S rRNA sequences, populations of *M. ottersoni* n. sp. from different states of Brazil clustered together and were separated from other *Meloidogyne* spp., thus confirming that all four populations are very similar and conspecific.

Keywords: Root knot nematode - *Oryza sativa* - Species complex.

S2-P06

Molecular and biochemical identification of *Meloidogyne* spp. in East-Mediterranean Region of Turkey.

Ibrahim Halil Elekcioglu (halile@cu.edu.tr), Aysenur Aslan

Faculty of Agriculture, Plant Protection Department, Cukurova University, Adana, Turkey

This study which covers the East-Mediterranean region, was carried out in Adana, Mersin and Hatay provinces and towns with dense greenhouse agriculture. The field survey and laboratory study were conducted in order to determine the presence and distribution of *Meloidogyne* species in three provinces. For this purpose, a survey was carried out from April to July 2018, 100 randomly selected root samples were collected in order to isolate nematode juveniles. After soil analysis *Meloidogyne incognita* and *M. javanica* were detected and characterized by molecular PCR (Polymerase Chain Reaction) with specific primers and biochemical PAGE (Polyacrylamide Gel Electrophoresis) techniques. Among all identified species *M. incognita* was detected in 39 samples from 7 locations while M. javanica was detected in 7 samples from 4 locations. After PCR, the amplified product was sequenced and was compared with BLAST sequences from NCBI and in phylogenetic analysis *M. incognita* isolates showed 100 % similarity with each other. As our isolates showed highest similarity with India and China, they were found distant from Thailand and Indonesia. Despite 99% similarity of *M. javanica* isolates with India and China isolates, low similarity with Iraq was obtained. Young mature females of all samples were analyzed by PAGE method of biochemical and esterase enzyme profile in order to confirm the molecular identification results of *M. javanica* and *M. incognita* species detected in 46 examples. At the end of the research, the results of PCR and PAGE method have supported each other and the detection and characterization were performed properly.

Keywords: Vegetable - Root-knot nematode - Identification.

S2-P07

Comparative analysis and evolutionary implications of exon-intron-structure of the *hsp*-90 gene in nematodes.

Elena Fanelli[1] (elena.fanelli@ipsp.cnr.it), Francesco Capriglia[1], Alberto Troccoli[1], Eustachio Tarasco[2], Francesca De Luca[1]

[1] *Nematology, Institute Sustainable Plant Protection, Bari, Italy*; [2] *Entomology, University of Bari, Bari, Italy*

Heat shock proteins (HSP) belong to a protein family that are rapidly synthesised in response to a series of environmental stressors. Among all HSPs, HSP90 is the most conserved and abundant in cells and is involved in response to temperature stress. Temperature affects life stage development and behaviour of nematodes, with different species and strains having optimum temperatures for feeding, survival and reproduction [1, 2]. A portion of the *hsp*-90 gene was amplified and sequenced in free-living, entomopathogenic and plant parasitic nematodes. Sequence analyses of the *hsp*-90 products showed the existence of two isoforms characterized by presence/absence of introns in nematodes. Furthermore, the exon-intron structure of *hsp*-90 genes varied among nematodes including the number of introns, intron position and length. In nematodes *hsp*-90 introns are relatively short compared to vertebrates suggesting that introns are under strong selective pressures. Interestingly, a higher number of introns in *hsp*-90 genes of parasitic nematodes compared to free-living nematodes is observed, and few of them are inserted at the same conserved position. These findings allow us to speculate that introns are gained and loss in the evolution of nematode lineages and may also contribute to adaptation to environmental changes. The duplication of *hsp*-90 gene in parasitic nematodes may indicate its predisposition to exploit climate changes. Further investigations are ongoing to evaluate their evolutionary implications among different groups of nematodes.

Keywords: Heat shock proteins - Sequencing - Gene structure - Nematode - Evolution.

References:

- [1] De Luca et al., 2009. Gene 440: 16-22
- [2] Fanelli et al., 2018. Eur J Plant Pathol.152: 355–365

S2-P08

Variability of *Mesocriconema* populations associated with grapevine decline disease in South Brazil.

Welllington Silva[1] (cbauergbr@gmail.com), Margareth Divers[1], Jerônimo Araújo-Filho[1], Alexandre Menezes-Netto[2], Cesar Gomes[3]

[1] *Plant Protection Post Graduation Program, Universidade Federal de Pelotas, Pelotas, Rio Grande do Sul, Brazil*; [2] *Laboratory Entomology, Epagri, Videira, Santa Catarina, Brazil*; [3] *Lab Phytopathology and Nematology, Embrapa Clima Temperado, Pelotas, Rio Grande do Sul, Brazil*

The Grapevine Decline Disease (GDD) is one of the main factors limiting crop production in Southern Brazil. Data of recent field surveys has revealed a worrying increase in the incidence of this complex disease. Abiotic and biotic factors are involved with this syndrome and the presence of ring nematode (*Mesocriconema* spp.) seems to be an important cause. The taxonomic status of the genus *Mesocriconema* is controversial and a reason for discussion among taxonomists about the validity and composition of species of this genus due to closely-related taxa. Therefore, this study aimed to identify the diversity of *Mesocriconema* species associated with vineyards with GDD symptoms in Southern Brazilian states. Soil samples were collected in vineyards with symptomatic plants from four municipalities of Rio Grande do Sul (Caxias do Sul, Flores da Cunha, Garibaldi, Nova Pádua) and in three municipalities of Santa Catarina (Pinheiro Preto, Tangará and Videira). In order to identify *Mesocriconema* species, temporary slides were used for morphological and morphometric examination according to Raski and Golden (1965) and Andrassy (1965) keys. Adult female specimens of *Mesocriconema* obtained from processed soil were observed, and the micrographs were performed with the 10, 20 and 40, 60 and 100X. The measurements of the images were performed using the LAS Core Software. According to morphological and morphometric data, five species of *Mesocriconema* were identified: *M. xenoplax*, *M. curvatum*, *M. rusticum*, *M. sphaerocephala* and *Mesocriconema* sp. *(≈M. mutabile*). The Principal Component Analysis (PCA), based on five characters (L, St, Ran, VL VB and VL), indicated variation among the populations of Southern Brazil and also among those populations and distant geographical areas from data in the literature (all identified by DNA sequences). The PCA results for *Mesocriconema* were reasonably aligned with those obtained previously. The first two main components (CP) explained 75,82% of the variation. The *Mesocriconema* populations from this study were all grouped in the same cluster and grouped with other *M. xenoplax* isolates described in the literature, confirming our preliminary identification. In our research we detected a complex of *Mesocriconema* species associated with plants with GDD symptoms, suggesting the need for new studies to the characterize the host status of the main rootstocks in Brazil.

Keywords: Mesocriconema - Variability - Grapewine - Decline - Brazil.

S2-P09

Reproductive fitness, pathogenicity, morphometric and genetic variability among geographic isolates of *Pratylenchus penetrans.*

Betre Estifanos[1] (betretadesse@yahoo.com), Peeten Ank Theelen[2], Philippe Packbier[2], Honermeier Bernd[3], Johannes Hallmann[1]

[1] *Epidemiologie und Pathogendiagnostik, Julius Kühn-Institut (JKI), Bundesforschungsinstitut für Kulturpflanzen, Münster, North Rhine-Westphalia, Germany*; [2] *Joordens Zaden, Kessel, Netherlands*; [3] *Agronomy and Plant Breeding I, Justus Liebig University, Gießen, Hessen, Germany*

The root lesion nematode *Pratylenchus penetrans* is known to damage major crops of economic importance. Pure cultures of seven isolates of *P. penetrans* were established from single females recovered from soil samples of different origins in Europe, namely Belgium, Germany, France, United Kingdom and The Netherlands using axenic carrot disc cultures. The diversity among the seven isolates of *P. penetrans* was characterized based on their level of reproduction and pathogenicity on selected hosts, as well as their morphometric and genetic characteristics. The reproductive fitness of the seven *P. penetrans* isolates was tested *in vitro* on carrot discs and *in vivo* on lentil and common vetch. Pathogenicity of those *P. penetrans* isolates was studied on alfalfa, carrot, fodder radish and French marigold. The seven isolates of *P. penetrans* were compared for their variability in genetic characteristics using Random Amplified Polymorphic DNA (RAPD-PCR) method. Differences in reproductive fitness and pathogenicity were observed among *P. penetrans* isolates. In general, reproductive fitness correlated positively with plant damage. Morphometrics of *P. penetrans* isolates were within the range of the original descriptions, but showed differences in some features to populations reported from other countries, such as body length, a, b and c ratio, maximum body width, tail length and length of the post-vulval uterine sac. According to RAPD analysis, the seven *P. penetrans* isolates showed a high level of intraspecific genetic variability. Furthermore, genetic similarities between some *P. penetrans* isolates were reflected in similarities in reproductive fitness, pathogenicity and morphometrics. Results of this study suggest that isolates of *P. penetrans* could be distinguished based on reproductive fitness and pathogenicity, as well as morphological and genetic characterization.

S2-P10

Ektaphelenchus sp. (Rhabditida: Ektaphelenchinae), a tentative new member of the genus, from dead wood in north Iran.

Fariba Heydari (fariba_heidari70@yahoo.com), Mohammad Reza Atighi, Majid Pedram

Department of Plant Pathology, Faculty of Agriculture, Tarbiat Modares Universit, Tehran, Iran

There are currently 32 species under the genus *Ektaphelenchus* [1&2]. In recent years, five species have been described from Iran. During the present study,*Ektaphelenchus* sp. was described and illustrated based upon morphological and morphometric characters. The new species was isolated from dead/rotten wood samples collected from natural forests in north Iran, Golestan province. It is characterized by 347-378 µm long females (shortest representative of the genus so far), having three lines in lateral field, offset cephalic region flat in anterior end, stylet well-developed, 15.0-16.5 µm long, having a wide lumen and lacking basal knobs or swellings, excretory pore slightly more than one metacorpus length posterior to it, vulva at 76.0-79.5%, rectum and anus absent, posterior body (tail) conical with narrower distal end, males common in population with the single precloacal papilla, and paired second and third caudal papillae, arcuate and separate spicules 7-10 µm long with well-developed wide condylus, and tail with a small mucron at terminus. The new species was morphologically compared with typologically similar small-sized species of *Ektaphelenchus* having three lines in the lateral field, conical posterior body end and stylet lacking basal knobs namely *E. joyceae*, *E. taiwanensis* and *E. apophysatus*. It was further compared with *E. olitorius* and *E. goffarti* having a similar female posterior body end shape and unknown number of incisures in the lateral field. It was further also compared with two species currently placed under genus *Seinura*, because of having obscure rectum namely *S. informis* and *S. paratenuicaudata*.

Keywords: Golestan province - Morphometric data - New species - Predatory nematode - Taxonomy.

References:

- [1] Gu, J. et al., 2021. New alien and native Ektaphelenchid nematodes (Tylenchomorpha: Ektaphelenchinae) from China with details on host association and geographical distribution. European Journal of Plant Pathology. 161: 123-145.
- [2] Hunt D. J. 2008. A checklist of the Aphelenchoidea (Nematoda: Tylenchina). Journal of Nematode Morphology and Systematics. 10: 99-135.

S2-P11

Plant-parasitic nematodes associated with the Solanaceae family in Costa Rica.

Danny Humphreys-Pereira (danny.humphreys@ucr.ac.cr), Rebeca Sandoval-Ruiz, Fernando Badilla-Jiménez, Lorena Flores-Chaves

Laboratory of Nematology/CIPROC, Agronomy School,University of Costa Rica, Montes de Oca, San José, Costa Rica

In Costa Rica, crops of the Solanaceae family (potato, sweet pepper and tomato) are highly valuable, and represent an important fraction of the daily diet in the country. Tomatoes, potatoes and sweet pepper are mainly grown in the Central Valley of Costa Rica. Nematodes are one of the most important pests on these crops. However, little is known about the main nematodes affecting Solanaceae, their distribution, the nematode species, the population variability and management. In potato, 67% of the soil samples (46 fields) had *G. pallida* cysts with densities up to 1600 cysts/200cc of soil. In roots, the most frequent nematode was *Pratylenchus* (68% of the samples) followed by *Meloidogyne* and *Globodera* (both with 30%). The highest population densities were observed in *Meloidogyne* with 44,000 J2/100g of roots, *Pratylenchus* with 24,110 nematodes and *Globodera* with 13,280 nematodes. Molecular techniques allowed the identification of *G. pallida*, *M. hapla*, *M. incognita* and *P. penetrans* associated with potatoes. Results from 43 fields of sweet pepper have shown that *Meloidogyne* is the most frequent nematode in this crop (frequency of 84% in roots), followed by *Helicotylenchus* with a frequency of 33% in roots, respectively. Twenty-five populations of *Meloidogyne* extracted from sweet pepper were characterized using molecular markers. The species associated with sweet pepper were *M. incognita*, *M. hapla* and *M. exigua.* This is the first report of *M. exigua* on sweet pepper in the country. Five populations were mixed with two concomitant species, three with the combination *M. incognita*/*M. exigua*, one with *M. incognita*/*M. hapla* and one with *M. hapla*/*M. exigua*. In a total of 30 tomato samples processed at the Laboratory of Nematology, *Helicotylenchus* was the most frequent nematode with 83% followed by *Meloidogyne* with 53% and *Pratylenchus* with 33%. *Meloidogyne* showed the highest population density with 468,000 J2/100g followed by *Helicotylenchus* with 91,800 nematodes and *Pratylenchus* with 5200 nematodes. With the new knowledge about the nematode species that affects Solanaceae crops and their distribution in the country, the future projects will be focused on management, including assays with new varieties or rootstocks with resistance against nematodes.

Keywords: *Meloidogyne* - Tomato - Sweet pepper - Potato - Cysts.

S2-P12

New records of *Globodera pallida* in Costa Rica.

Danny Humphreys-Pereira (danny.humphreys@ucr.ac.cr), Rebeca Sandoval-Ruiz, Lorena Flores-Chaves

Laboratory of Nematology/CIPROC, Agronomy School,University of Costa Rica, Montes de Oca, San José, Costa Rica

In Costa Rica, potatoes are grown in three provinces, mainly in Cartago with 72 % of the total potato area, followed by Alajuela (Zarcero, Alajuela, Poás and Naranjo) with 24% and San José (Dota, Coronado and Goicochea) with 4%. The potato cyst nematode, *Globodera pallida* was detected in the country in 2005, near the Irazú Volcano, Cartago. Currently, the nematode is widely spread around the Irazú volcano, which is the main potato seed production area in the country (area with an altitude up to 3100 m.a.s.l). Since 2017, a nematode survey has been performed in Zarcero, the second most important area that grows vegetables, including potatoes. Soil (100cc) and root (10g) samples were processed through the floatation-centrifugation method. In addition, 200cc of soil was processed with the Fenwick method. Several potato fields in Zarcero showed the presence of *Globodera* sp. on roots and soil samples. Cysts were found in densities up to 100 cysts/200 cc of soil. The population density on roots ranged from 5 to 2350 nematodes/100g and in soil ranged from 8 to 700 J2/100cc of soil. The species was identified by sequencing the *cob* gene and the *ITS* region. The *G. pallida* population sequence from Zarcero was identical to those from Cartago, and a unique haplotype was found in this area. A large survey is being performed and a distribution map of the species prepared for the different localities of Zarcero.

Keywords: *Globodera* - Potato - Cysts - Zarcero - ITS.

S2-P13

Ten populations of the genus *Hoplolaimus* Daday, 1905 (Hoplolaimidae), associated with date palm in south of Kerman province, Iran.

Farahnaz Jahanshahi Afshar[1] (afshar_fj@yahoo.com), Mohammad Afzali[2], Azam Zeynaddini Riseh[2]

[1] *Iranian Res. Ins. of Plant Protection, Agri. Res., Edu. & Ext. Org. (AREEO), Tehran, Iran*; [2] *Department of Plant Protection, Faculty of Agriculture, Vali-e-Asr University of Rafsanjan, Rafsanjan, Iran*

Date palm (*Phoenix dactylifera* L.) is one of the most important economic products in the south of Kerman province. In order to identify plant parasitic nematodes with a tentative role in recently observed comprehensive decline syndrome in southern Iran, several soil and root samples were collected from the rhizosphere of date palm trees in gardens and transferred to the laboratory in 2020 and 2021. The extracted nematodes using sieving and centrifugation methods, were fixed and permanent slides were prepared. As the result, ten populations of four species of *Hoplolaimus* including *H. columbus, H. dubius, H. indicus* (three populations) and *H. seinhorsti,* were identified and reported based upon morphological and morphometric characters. Four populations did not fit the characteristics of currently known species and were reported as *Hoplolaimus* sp.1, *Hoplolaimus* sp.2, *Hoplolaimus* sp.3 and *Hoplolaimus* sp.4. *H. columbus* was reported for the first time from Iran.

Keywords: First report - Hoplolaimidae - Plant parasite nematodes - Taxonomy.

S2-P14

Characterization of nematode pests of enset (*Ensete ventricosum* welw. Cheesman) and their management.

Selamawit Kidane[1] (selamawit.araya.kidane@nmbu.no), Danny Coyne[2], Beira Meressa[3], Solveig Haukeland[4], Trine Hvoslef-Eide[1]

[1] *Plant Science, Norwegian University of Life Sciences, Ås, Norway*; [2] *Nematology, IITA, Nairobi, Kenya*; [3] *Nematology, Jimma University, Jimma, Ethiopia*; [4] *Nematology, Icipe, Nairobi, Kenya*

Enset (*Ensete ventricosum* Welw. Cheesman) is an important starch staple crop, cultivated primarily in southern Ethiopia. Enset has an underground corm, a pseudostem formed from a bundle of leaf sheaths and large leaves, reaching up to 10 m in length with a pseudostem diameter up to 2 m (1). Enset is the main crop of a sustainable indigenous African system that ensures food security in a country that is food deficient. Related to the banana family, enset is similarly infected by plant parasitic nematodes. From previous studies, *Pratylenchus goodeyi* appears to be the dominant nematode pest, which is believed to contribute to reduced productivity of enset (2). However, while surveys have demonstrated high *P. goodeyi* infection levels, there is relatively scant information on how damaging the nematode is to enset production. There is also little information on the variability of the nematode pest in terms of levels of pathogenicity on enset and if so, how this may relate to variability in climate and temperature zones under which enset is grown. The objective of our study is to identify nematode pests of enset, assess the possible damage of nematode pests, with emphasis on *P. goodeyi*, and in relation to the presence of other diseases, how climate and agro ecology may affect this, assess cultivation and management options and to screen the resistance of enset.

Keywords: Enset - Nematode - Pratylenchus goodeyi - Resistance - Management.

References:

- (1) Westphal, E. et al 1975. Agricultural systems in Ethiopia, vol. 826: Centre for Agricultural Publishing and Documentation Wageningen.
- (2) Addis, T. et al 2006. Nematodes associated with enset and banana in the highlands of Ethiopia. International Journal of Nematology, 16, pp.118-125.

S2-P15

Occurrence of plant-parasitic nematodes on *Ensete ventricosum* in Ethiopia with focus on *Pratylenchus goodeyi.*

Selamawit Kidane[1] (Sa.Kidane@cgiar.org), Beira H. Meressa[2], Solveig Haukeland[3, 4], Trine Hvoslef-eide[5], Christer Magnusson[3], Marjolein Couvreur[6], Wim Bert[6], Danny L. Coyne[1]

[1] *International Institute of Tropical Agriculture, IITA, Nairobi, Kenya*; [2] *Jimma University, Jimma, Ethiopia*; [3] *The Norwegian Institute of Bioeconomy Research, NIBIO, Ås, Norway*; [4] *International Centre of Insect Physiology and Ecology, Icipe, Nairobi, Kenya*; [5] *Norwegian University of Environmental and Life Sciences, NMBU, Ås, Norway*; [6] *Nematology Research Unit, Department of Biology, Ghent University, Ghent, Belgium*

Enset (*Ensete ventricosum*) is an important starch staple crop, cultivated primarily in south and southwestern Ethiopia. Enset is the main crop of a sustainable indigenous African system that ensures food security in a country that is food deficient. Related to the banana family, enset is similarly affected by plant-parasitic nematodes. Plant-parasitic nematodes impose a huge constraint on agriculture. The distribution, population density and incidence of plant-parasitic nematodes of enset was determined during August 2018. A total of 308 fields were sampled from major enset-growing zones of Ethiopia. Eleven plant-parasitic nematode taxa were identified, with *Pratylenchus* (lesion nematode) being the most prominent genus present with a prominence value of 1460. It was present in each sample, with a highest mean population density per growing zone of 16, 050 (10 g root)−1, although densities as high as 25,000 were observed in fields at higher altitudes in Guraghe (2200-3000 m a.s.l.). This lesion nematode is found in abundance in the cooler mountainous regions. Visible damage on the roots and corms was manifested as dark purple lesions. Using a combination of morphometric and molecular data, all populations were identified as *P. goodeyi* and similar to populations from Kenya, Uganda and Spain (Tenerife). Differences in population densities amongst cultivars indicate possible resistance of enset to *P. goodeyi*.

Keywords: Food security - Altitude - Lesion nematode - Molecular data - Morphology.

S2-P16

Identification of *Meloidogyne floridensis* populations and their virulence on vegetables in Georgia, USA.

Josiah Marquez[1] (josiah.marquez@uga.edu), Abolfazl Hajihassani[1, 2]

[1] *Department of Plant Pathology, University of Georgia, Tifton, GA, United States*; [2] *Department of Entomology and Nematology, Institute of Food and Agricultural Sciences, University of Florida, Davie, FL, United States*

Meloidogyne floridensis is of particular concern, reproducing on tomato, pepper, corn, and tobacco cultivars with resistance to common root-knot nematode species [1]. During a 2018 survey in south Georgia, USA, *Meloidogyne* populations were collected from multiple fields with collard, cowpea, cucumber, watermelon, and tomato and cultured on tomato cv. Rutgers. Individual females (*n*= 3) were recovered from galled roots and subjected to PCR using three primer sets targeting mitochondrial genes [C2F3/1108 for the fragment of cytochrome c oxidase subunit II and the large subunit rRNA (l-rRNA) gene; TRNAH/MRH106 for the l-rRNA gene; and NAD5F2/NAD5R1 for the NADH dehydrogenase subunit 5 gene]. DNA sequences from C2F3/1108 and TRNAH/MRH106 primer sets had 100-98.6% identity and 100-84.9% query coverage to *M. floridensis* isolates in Florida (accession no. DQ228697, KP732360, and KP732361), whereas NAD5F2/NAD5R1 primer set had 100-99% identity and 100-97.1% query coverage to *M. floridensis* isolates in California (MH729181) and South Carolina (MN072363). Two greenhouse trials in a completely randomized design used a susceptible tomato (cv. Rutgers), a resistant tomato (cv. Skyway; *Mi*-1.2 gene), and vegetable crops associated with the origin of *M. floridensis*isolates in Georgia to determine variability in host preference and reproduction level. All crops other than collard were susceptible to isolates with a reproduction factor (final population/initial population; RF) > 1 ($P < 0.05$). This is the first report of watermelon and cowpea as a host to *M. floridensis.* While cucumber was reported as a non-host to Florida isolates [2], isolate Wi21 from Georgia reproduced well on cucumber with a RF comparable to susceptible tomato. All other isolates had a higher RF on the susceptible tomato than other vegetables ($P < 0.05$). When comparing isolates between tomato cultivars, isolate Do14 produced more eggs ($P < 0.05$) than isolate Wi21 on susceptible tomato, whereas egg mass index for isolates Do14 and Wa8 were greater ($P < 0.05$) than isolate C8 on resistant tomato. Overall, this study suggests that there are variations among *M. floridensis* isolates in pathogenicity and reproduction levels on vegetable crops that need to be considered in developing host resistance.

Keywords: Mitochondrial haplotyping - COX2 - NADH5 - Aggressiveness - Virulence.

References:

- [1] Stanley et al., 2009. Nematropica. 39:255-271.
- [2] Burelle, N. & Nyczepir, A., 2004. J. Nematol. 36: 328.

S2-P17

Detection of Root-Knot Nematodes in Northern Iraq.

Husheng Hamad Kanabi[1] (hoshang.kanabi@gmail.com), Gökhan Aydınlı[2], Sevilhan Mennan[1]

[1] *Plant protection dep., Ondokuz Mayıs University, Agr. Fac., Samsun, Turkey*; [2] *Animal and plant production, Ondokuz Mayıs University, Bafra Vocational School, Bafra, Samsun, Turkey*

Root-knot nematodes, *Meloidogyne* spp., are widely distributed all over the world, infecting nearly all cultivated plants including vegetables, field crops and ornamental plants. But, there are few reports about root-knot nematodes (RKNs) in northern Iraq. Even though the occurrence of RKN in greenhouses was documented, the research comprised small-scale surveys. Moreover, RKNs in Northern Iraq have only been identified based on perineal pattern [1, 2, 3]. Therefore, the objectives of this study was to determine the distribution and identification of RKNs, using both perineal pattern and esterase phenotype, collected from different greenhouses in Sulaymaniyah, Erbil and Duhok provinces, the largest vegetable-producing areas in Northern Iraq. A total of 187 greenhouses were surveyed during October - November 2018. At least 20 females representing each population were obtained from naturally infested plants and used for identification studies. RKN were detected in 70 greenhouses (37.4%) located in Sulaymaniyah (35), Erbil (20) and Duhok (15). The most infected plants were cucumber and tomatoes respectively. Out of 70 populations obtained from greenhouses infested with RKN, 45 *Meloidogyne javanica* (64.3%) and 25 *M. incognita* (35.7%) were identified. The existence of mixed root-knot nematode populations was not detected in this study.

Keywords: Greenhouses - Identificaiton - Northern Iraq - Root-knot nematodes - Esterase phenotype.

References:

- [1] Al Adhami. 1955. Baghdad Suhor Press, Technical Bull,p.10.
- [2] Stephan. 1988. Int. Nematol. Network Newsl., 5:21.
- [3] Stephan and Anton.1991. Basrah J. Agric. Sci., 4: 95-99.

S2-P18

Morphological characterization of Race A and Race B of *Meloidogyne hapla*.

Branimir Nježić (branimir.njezic@agro.unibl.org), Petar Nikolić

Faculty of Agriculture, University of Banja Luka, Banja Luka, Bosnia & Herzegovina

Northern root-knot nematode – *Meloidogyne hapla* is one of four major root-knot nematode species with a cosmopolitan distribution. It is considered as the most cytologicaly and reproductively diverse root-knot nematode. It has two ways of reproduction; automixis (facultative meiotic parthenogenesis) present in Race A with diploid chromosome number and apomixis (obligatory mitotic parthenogenesis) present in Race B with polyploid chromosome number. Morphological characters of perineal pattern and infective juveniles (J2) of 44 populations from Bosnia and Herzegovina sampled from potato fields were compared by light microscopy. Race B was found in two localities on altitude above 800 m and eight populations were characterized. The lowest variation of morphological characters was among J2 originating from the same egg mass, while the highest morphological variation was among two races. There was no significant difference among localities. In the fields where Race B was found on potato tubers with symptoms similar to quarantine species, *M. chitwoodi* and *M. fallax*.

Keywords: Northern Root-knot nematode - Potato nematodes - Taxonomy - Bosnia and Herzegovina.

S2-P19

Morphological and molecular characterization of *Heterodera trifolii* in Costa Rica, and pathogenicity on three plants species.

Lester A. Núñez-Rodríguez (lesnr92@gmail.com), Lorena Flores-Chaves, Danny A. Humphreys-Pereira

Laboratory of nematology,CIPROC, Agronomy School,University of Costa Rica, San José, San Pedro, Costa Rica

Globodera pallida was the only cyst nematode species reported in Costa Rica, and no information was published regarding the morphology, intraspecific variability, biology and the distribution of other cyst nematodes in the country. Two populations of *H. trifolii* were found in the Central Valley of Costa Rica, one extracted from *Trifolium repens* and the other from *Rumex obtusifolius*. Cysts were extracted from soil samples using the Fenwick method[1] and the second-stage juveniles (J2s) were obtained from the cysts. Cysts were lemon shaped with a prominent vulval cone, light to dark brown coloring, and ambifenestrate with strongly developed underbridge. The *H. trifolii* J2 from Costa Rica were distinct to *H. betae* by the shorter average body length (531.1-551.2 vs 584-607 μm), a shorter average stylet length (27.4-28.4 vs 29.6-31.0 μm) and a shorter average tail length (66.3-69.6 vs 71-74 μm)[2]. No *H. trifolii* population from Costa Rica presented males, contrary to their closely species, such as *H. daverti, H. glycines* and *H. schachtii*, that have males. Three molecular markers were sequenced, the D2-D3 expansion segments of the *28S* rDNA, the nuclear region ITS1-5.8S-ITS2 and the mitochondrial gene *cox1*, and compared with sequences retrieved from GenBank. The D2-D3 region had divergence levels of 0.14-0.78% (1-5 nt), 0.42-1.6% (3-11 nt) and 0.16-0.34% (1-2 nt) compared with *H. schachtii*, *H. glycines* and *H. daverti*, respectively. The *ITS* region showed divergence levels of 0.1-1.6% (1-15 nt), 1.6-2.3% (15-22 nt) and 0.1-1.6% (1-16 nt) compared with *H. schachtii*, *H. glycines* and *H. daverti*, respectively. Both the Bayesian phylogenetic analysis based on the *ITS* region and the segment D2-D3 of the *28S* grouped the sequences generated in this study within the *Schachtii* group, with a high support value (for both markers, PP = 100). The phylogenetic analysis based on the partial *cox1* gene resulted in a clade composed with the sequences obtained in this study and sequences of *H. trifolii* (PP = 98). The pathogenicity of *H. trifolii* on *R. obtusifolius*, *T. repens* and table beet was evaluated under greenhouse conditions. A total of 1000 infective J2 (eggs + J2) per plot were inoculated. Results showed that beet was a non-host for *H. trifolii* (reproduction factor = 0), while being susceptible to *T. repens* and *R. obtusifolius* (RF = 3.4 and 17.4, respectively). Importantly, the host status for *H. trifolii* of other crop plant species will be tested.

Keywords: *H. trifolii* - Morphological - Molecular - Beets - Costa Rica.

References:

- [1] Fenwick, D. 1940. Journal of Helminthology 18:155–172
- [2] Subbotin et al. 2010. Nematology Monographs and Perspectives 8B.

S2-P20

Synonymization of *Rotylenchulus macrosoma* Dasgupta et al., 1968 with *R. borealis* Loof & Oostenbrink, 1962 and its ecology and phylogeography.

Juan Emilio Palomares-Rius[1] (palomaresje@ias.csic.es), Ilenia Clavero-Camacho[1], Antonio Archidona-Yuste[2], Carolina Cantalapiedra-Navarrete[1], Emmanuel A. Tzortzakakis[3], Andrea S. Azpilicueta[4], Antonio Saborido[5], P. Castillo[1]

[1] *Plant Protection, Instituto de Agricultura Sostenible-CSIC, Córdoba, Spain*; [2] *Andalusian Institute of Agricultural and Fisheries Research and Training (IFAPA), Córdoba, Spain*; [3] *Department of Viticulture, Vegetable Crops, Floriculture and Plant Protection, Institute of Olive Tree, Subtropical Crops and Viticulture, N.AG.RE.F., Heraklion, Greece*; [4] *Cambrico Biotech S. L., Sevilla, Spain*; [5] *Bayer Crop Science, Barcelona, Spain*

Reniform nematodes of the genus *Rotylenchulus* Linford & Oliveira, 1940 comprise ten valid species. Some species are important plant-parasites such as *R. reniformis* Linford & Oliveira, 1940 in USA and other parts of the world. However, the importance of other species such as *R. macrosoma* Dasgupta et al., 1968 has been neglected and their expansion through European countries has been underestimated in the recent years as well as their genetic diversity. In this study, we compared 12 populations of *R. borealis* and 16 populations of *R. macrosoma*, including paratypes deposited in nematode collections, confirming that morphological characters between both nematode species do not support their separation. In addition, analysis of molecular markers using nuclear ribosomal DNA (28S, ITS1) and mitochondrial DNA (*coxI*) genes, as well as phylogenetic approaches, confirmed the synonymy of *R. macrosoma* with *R. borealis*. Molecular analyses based on *coxI* and D2-D3 segments of 28S RNA markers showed high diversity and pronounced genetic structure among populations of *R. borealis*. The present study confirms the extraordinary morphological and molecular diversity of *R. borealis* in Europe, Africa, and the Middle East and comprises a paradigmatic example of remarkable flexibility of ecological requirements within reniform nematodes. Levels of infestation are in the range from 5 to 1760 individuals / 100 cm3 of soil, indicating that in some fields the infestation levels could cause yield losses. Knowledge of the molecular diversity and prevalence of *R. borealis*, as well as the major environmental and agronomical cues are of essential importance for designing useful control measures to reduce significant damage of this plant-parasitic nematode.

Keywords: Phylogeny - Reniform nematodes - Molecular diversity.

S2-P21

Proposal for a new *Xiphinema* Cobb, 1913 (Longidoridae) species from Iran belonging to the *X. americanum*-group.

Maid Pedram[1] (majid.pedram@modares.ac.ir), Mahyar Mobasseri[1], Farahnaz Jahanshai Afshar[2]

[1] *Faculty of Agriculture, Tarbiat Modares University, Tehran, Iran*; [2] *Iranian Research Institute of Plant Protection, AREEO, Tehran, Iran*

Two populations of a new species of the dagger nematode genus *Xiphinema* belonging to the *X. americanum* group were recovered from natural forests of Golestan province and a pistachio garden in city of Damghan (Semnan province) in association with forest and pistachio trees respectively. Both populations belong to the same species, which is undescribed. This species is mainly characterized by having an offset lip region, guiding ring at 61-82 µm distance from the anterior end, 82-91 and 43-49 µm long odontostyle and odontophore, respectively, 72-92 µm long pharyngeal bulb, visible endosymbiont bacteria occupying ovaries under light microscopy, vulva at 52-56 %, tail conical, dorsally convex, ventrally flat, its tip blunt, 26-32 µm long, and rare male. The alpha-numeric identification codes of the new species are: A3, B23, C3, D3, E3, F-, G2, H2, I2. The molecular phylogenetic studies using two markers revealed the new species stands as a unique lineage in two LSU and ITS phylogenies. The sequencing of 16S rDNA of the endosymbiont bacterium of the new species and the BLAST search using this sequence revealed it belongs to the genus *Candidatus* Xiphinematobacter, having 93% identity with five previously submitted 16S rDNA sequences into the GenBank. The new species was morphologically compared with *X. pachtaicum*, *X. incertum*, *X. californicum*, *X. bricolensis* and *X. santos*.

Keywords: Dagger nematode - Phylogeny - Molecular - Taxonomy.

S2-P22

Proposal for a new species of the genus *Sphaerularia* from Iran.

Majid Pedram (majid.pedram@modares.ac.ir), Mojtaba Keramat, Zeinab Mahboubi

Department of Plant Pathology, Tarbiat Modares University, Tehran, Iran

The genus *Sphaerularia* (Sphaerularioidea, Sphaerulariidae), a nematode with entomoparasitic and free-living forms, currently includes two nominal species viz. *S. bombi*, the type species, described by Dufour [1] and distributed in Canada, Europe and North America; and *S. vespae*, described by Kanzaki *et al*. [2] from Japan. It is mainly characterized by having a uterium, an abnormal large egg-producing structure in entomoparasitic stage. Molecular data are available for both aforementioned species in GeneBank database. During the present study, a population of juveniles of this genus was recovered from decaying wood of a dead *Cypress* sp. tree in Tehran, Iran. It was sequenced for three 18S, ITS and 28S ribosomal DNA loci (partial sequences, accession numbers MT002878, MT002877 and MT002910). The BLAST search using these sequences revealed the 18S sequence has 99.58% identity with *S. vespae* (AB300595) and 99.30 and 99.18 % identity with *S. bombi* (AB250213 and AB250212), the ITS sequence has 91.21% identity with *S. vespae* (AB300595) and the 28S rDNA D2-D3 sequence has 93.21% identity with *S. vespae* (AB300596) and 91.10 and 91.15% identity with *S.* cf. *bombi* and *S. bombi* (AB733664 and DQ328726) and 88.99% identity with *S*. cf. *bombi* (AB733665). By remarkable sequence differences between the Iranian population and those of two aforementioned species, it was proposed as a new species to the genus in a reverse taxonomic approach. Studies to recover and illustrate the adults are in progress. The genus *Bealius* (Sphaerularioidea, Paurodontide) is known in Iran with *B. pinus* described by Esmaeili et al. [3] and was molecularly characterized using 18S data. During the present study, it was re-isolated in a high population number from the same tree from which *Sphaerularia* n. sp. was recovered, sequenced for its 28S rDNA (MT002879) and was included in the aforementioned 28S phylogeny. The 18S, ITS and 28S phylogenies of *Sphaerularia* n. sp. are discussed.

Keywords: New species - Reverse taxonomy - Tehran.

References:

- [1] Dufor, 1937. Ann Sci Natur Zool. 7: 5-20.
- [2] Kanzaki et al. Zool Sci. 24: 1134-1142.
- [3] Esmaeili et al. Nematology. 21: 435-444.

S2-P23

An unknown species of the genus *Longidorus* Mikoletzky, 1922, associated with beech trees in natural forests of northern Iran.

Niloofar Pour Ehtesham (niloofarpourehtesham@gmail.com), Majid Pedram

Department of Plant Pathology, Faculty of Agriculture, Tarbiat Modares University, Tehran, Iran

Following our programs to monitor longidorids occurring in natural forests of northern Iran, a population of the genus *Longidorus* Mikoletzky, 1922 was recovered from the rhizosphere of *Fagus* sp. in natural forests of Golestan province, northern Iran. The polytomous key of the genus by Chen *et al*. [1] and original descriptions of the species described after the key, or transferred to the genus, were used to clarify the taxonomic status of the recovered population. The recovered species is characterized by its narrower anterior body region, giving a bottle-neck appearance, 5.2-7.5 mm long females having anteriorly almost flat lip region, pouch-like amphidial fovea, guiding ring at 30-40 µm distance from anterior end, odontostyle 92-103 µm long, odontophore 60-70 µm long, tail short (c′=0.8-1.0), rounded to short-conical, dorsally more convex with widely rounded end, common males with 50-64 µm long spicules and 9-13 ventromedian supplements. Only the last juvenile developmental stage was recovered. Based on its morphological characteristics, the recovered population belongs to an unknown species of the genus, and the detailed morphological comparisons of the new species with other closely related species were discussed. The partial LSU rDNA D2-D3 and ITS rDNA sequences were made available for the species, and their BLAST search revealed they are unique, compared to currently available sequences deposited in the GenBank database.

Keywords: Longidoridae - Fagus sp. - New species - Taxonomy.

References:

- [1] Chen et al., 2018. Fundamental and Applied Nematology. 20:15-28.

S2-P24

Proposal for a new species of the genus *Sphaerularia* Dufour, 1837 (Sphaerulariidae) from Iran.

Mohammad Reza Atighi (mj_pedram@yahoo.com), Zeinab Mahboubi, Mojtaba Keramat, Majid Pedram

Department of Plant Pathology, Faculty of Agriculture, Tarbiat Modares Universit, Tehran, Iran

The genus *Sphaerularia*, a nematode with entomoparasitic and free-living forms, currently includes two nominal species viz. *S. bombi*, the type species, described by Dufour [1] and distributed in Canada, Europe and North America; and *S. vespae*, described by Kanzaki *et al*. [2] from Japan. It is mainly characterized by having a uterium, an abnormally large egg-producing structure in the entomoparasitic stage. Molecular data are available for both aforementioned species in GeneBank database. During the present study, a population of juveniles of this genus was recovered from decaying wood of a dead *Cypress* sp. tree in Tehran, Iran. It was sequenced for three 18S, ITS and 28S ribosomal DNA loci (partial sequences, accession numbers MT002878, MT002877 and MT002910, respectively). The BLAST search using these sequences revealed the 18S sequence has 99.58% identity with *S. vespae*(AB300595) and 99.30 and 99.18 % identity with *S. bombi* (AB250213 and AB250212), the ITS sequence has 91.21% identity with *S. vespae*(AB300595) and the 28S rDNA D2-D3 sequence has 93.21% identity with *S. vespae* (AB300596) and 91.10 and 91.15% identity with *S.* cf. *bombi*and *S. bombi* (AB733664 and DQ328726) and 88.99% identity with *S*. cf. *bombi* (AB733665). Due to the remarkable DNA sequence differences of Iranian population with both species, it was proposed as a new species in the genus by a reverse taxonomic approach. Studies to recover and illustrate the adults are in progress. The genus *Bealius* (Sphaerularioidea, Paurodontide) is known in Iran with *B. pinus* described by Esmaeili et al. [3] and was molecularly characterized using 18S data. During the present study, it was re-isolated in large numbers from the same tree in which the new species of *Sphaerularia* was recovered, sequenced for its 28S rDNA (MT002879) and included in aforementioned 28S phylogeny.

Keywords: New species - Reverse taxonomy - Tehran.

References:

- [1] Dufor, 1937. Ann Sci Natur Zool. 7: 5-20.
- [2] Kanzaki et al. Zool Sci. 24: 1134-1142.
- [3] Esmaeili et al. Nematology. 21: 435-444.

S2-P25

Morphological and molecular characterization of *Acrobeloides saeedi* Siddiqi, De Ley and Khan, 1992 (Rhabditida, Cephalobidae) from India.

Aasha Rana[1] (aasha.aasharana@ymail.com), Aashaq Hussain Bhat[1], Ashok Kumar Chaubey[1], Joaquín Abolafia[2]

[1] *Nematology Laboratory/Deptt. of Zoology, Chaudhary Charan Singh University, Meerut, Uttar Pradesh, India*; [2] *Department of Animal Biology, Universidad de Jaén, Campus, Las Lagunillas, Jaén, Spain*

Two cultured populations of *Acrobeloides saeedi* labelled DH1 and KMW are described from the Indian subcontinent. Morphologically and morphometrically this material agrees well with other species of the "*maximus"* group (*A. bodenheimeri*, *A. longiuterus* and *A. maximus*), especially with *A. longiuterus* [1]. However, molecularly the Indian material is well differentiated from all of these species. According to this, *A. saeedi* is considered a valid taxon distinguished mainly from *A. bodenheimeri* by having a dextral female reproductive system (*vs* sinistral), from *A. longiuterus* by having larger females (1.03–1.57 *vs* 0.57–0.88 mm) and from *A. maximus* by having seta–like labial processes (*vs* absent) and males as frequent as females (*vs* males very infrequent). Molecular data based on 18S and D2D3 rDNA showed that the sequence alignment of present strains DH1 and KMW showed 21 and 51 bp differences, respectively with *A. maximus.* With *A. longiuterus* (formerly *Acrobeloides camberenensis*), it showed 38 bp differences based on D2D3 regions and on ITS rDNA, it showed 73 bp differences and 23 gaps. With *A. bodenheimeri,* the sequence alignment of 18S genes and D2D3 expansion fragment of 28S genes of present strains showed 22 and 54 bp differences, respectively. Thus molecular data confirm the present strains to be different from species of the "*maximus"* group. In addition, other similar species are revised: *Acrobeloides ishraqi* [2] is considered a new junior synonym of *A. saeedi, Acrobeloides mushtaqi* [2] is considered a new junior synonym of *A. bodenheimeri* and *Acrobeloides gossypii* [3] is also considered a new junior synonym of *A. saeedi*.

Keywords: A.bodenheimeri - A. longiuterus - A. maximus - 18S rDNA - 28S rDNA.

References:

- [1] Rashid and Heyns, 1990. Phytophylactica. 22:189–199.
- [2] Pervez, 2011. Arch Phytopathology Plant Protect. 44:1438–1446.
- [3] Nahiyoon et al., 2019. Pakistan J. Zool. 51(4):1309-1314.

S2-P26

Plant-parasitic nematodes associated with *Musa* spp. crops in Colombia.

Donald Heberth Riascos Ortiz[1, 2] (dhriascoso@unal.edu.co), Carlos Arboleda[1], Ana Teresa Mosquera Espinosa[3], Francia Varón de Agudelo[4], Juliana Magrinelli Osório Rosa[5], Claudio Marcelo G. Oliveira[5], Jaime Eduardo Muñoz Florez[6]

[1] *Molecular Laboratory, Universidad Nacional de Colombia, Palmira, Valle del Cauca, Colombia*; [2] *Laboratorio de Fitopatología, Universidad del Pacífico, Buenaventura, Valle del Cauca, Colombia*; [3] *Pontificia Universidad Javeriana, Cali, Valle del Cauca, Colombia*; [4] *Centro de Investigación en Palma, Bogotá, Cundinamarca, Colombia*; [5] *Instituto Biológico, Campinas, São Paulo, Brazil*; [6] *Universidad Nacional de Colombia, Palmira, Valle del Cauca, Colombia*

Plant-parasitic nematodes are a limiting factor of the production of *Musa* spp. in Colombia. In order to determine the relative importance of plant-parasitic nematodes associated with *Musa* spp. and to identify the species of the genus with wider prominence value - VP, thirty compound samples of roots and soil were taken in plantain and banana crops in the departments of Valle del Cauca, Quindío, Risaralda and Caldas in Colombia, during the period of 2015-2018. For the genus with major VP, identification of species was done through morphological characters, morphometrics, biochemical (esterase electrophoretic profile), molecular (sequencing of D2-D3 expansion segment of ribosomal RNA and Cytochrome oxidase subunit I and Nad5 of mitochondrial DNA), and phylogenetic analysis. The plant-parasitic nematodes with major and intermediate relative importance in roots of *Musa* spp. were *Meloidogyne, Helicotylenchus, Radopholus, Pratylenchus* and *Rotylenchulus*, with a prominence value of 3868, 2396, 876, 65 and 10, while in soil they were *Helicotylenchus, Meloidogyne* and *Rotylenchulus*, with prominence values of 600, 372 and 70, respectively. Through integrative taxonomy we identified in plantain and banana crops *M. incognita, M. arenaria, M. hispanica, M. acrita, H. multicinctus, H. dihystera, H. erythrinae, H. californicus, R. similis, P. araucensis* and *R. reniformis*. This is the first report of *M. acrita, M. hispanica, H. multicinctus, H. dihystera, H. erythrinae, H. californicus* and *R. reniformis* in plantain and banana for Colombia through integrative taxonomy. In conclusion, an ample diversity of plant-parasitic nematodes species are associated with plantain and banana crops in Colombia. This study contributes to the knowledge of the parasitic nematode community of this country, and is essential information for the future design of integrated management programs.

Keywords: Plantain - Banana - Meloidogyne - Helicotylenchus - Rotylenchulus.

References:

- [1] Riascos-Ortiz et al., 2019. Jorunal of Nematology. 51:1-13

S2-P27

Uncovering diversity in plant-parasitic nematodes using morphological, molecular and phylogenetic approaches.

Phougeishangbam Rolish Singh[1] (phougeishangbamrolish.singh@ugent.be), Gerrit Karssen[1, 2], Alliance Nyiragatare[1, 3], Beatrice Kashando[1, 4], Catherine Malike Etongwe[1], Abebe Woldesenbet[1, 5], Marjolein Couvreur[1], Wim Bert[1]

[1] *Nematology Research Unit, Ghent University, Ghent, Belgium*; [2] *National Plant Protection Organizations, Wageningen, Netherlands*; [3] *Biological Science Department, University of Rwanda, Kigali, Rwanda*; [4] *Tanzania Agricultural Research Institute, Kibaha Coast region, Tanzania*; [5] *College of Agriculture and Veterinary Medicine, Jimma University, Jimma, Ethiopia*

Plant-parasitic nematodes (PPN) surveys have been conducted in both agricultural and natural habitats, including samples from sugarcane (Kilimanjaro region of Tanzania) [1], coffee (Jimma region of Ethiopia), finger millet (Eldoret region of Kenya), corn (eastern province of Rwanda), sand dunes (coastal beach of Gran Canaria) and other selected places in Belgium and Netherlands. This revealed an enormous diversity of PPN and allowed us to link more than 50 morphologically characterised PPN species with ribosomal and mitochondrial molecular barcodes (18S, D2-D3 region of 28S and ITS and *cox1* sequences). Major results include discovery of four new PPN species, namely *Heterodera dunensis* from Gran Canaria [2]; *Paratylenchus* n. sp. from the Netherlands, *Pratylenchus rwandae* from Rwanda [3] and *Rotylenchus wimbii* from Kenya [4]; first reports of four *Paratylenchus* species in Belgium, one *Paratylenchus* species in Europe [5] and *Cryphodera* from Africa; synonymisation of *Tylenchorhynchus agri* with *T. crassicaudata* [1], *Rotylenchus rhomboides* with *R. goodeyi* and *Cacopaurus* with *Paratylenchus*; and corrective reassignment of multiple barcodes on public database that were previously unidentified or incorrectly classified [5]. Furthermore, our study also provides, for the first time, full length mitogenome and ribosomal DNA sequences of *R. goodeyi*, revealing the presence of two mitogenome variants within a single population. This new method made it possible to explore the use of full mitogenome sequences, originating from individual nematodes, in PPN diagnostics.

Keywords: Diversity - DNA-barcodes - Plant-parasitic nematodes - Systematics - Taxonomy.

References:

- [1] Singh et al., 2020. J Nematol. 52: e2020-59.
- [2] Singh et al., 2020. J Nematol. 52: e2020-98.
- [3] Singh et al., 2018. Nematology. 20(8): 781-794.
- [4] Singh et al., 2021. J Nematol. 53: e2021-16.
- [5] Singh et al., 2021. Plants 10(2): 408

S2-P28

Morphological, morphometrical and oogenesis aspects of *Tubixaba tuxaua.*

Santino Aleandro Silva[1] (santinoaleandro@gmail.com), Renata da Rosa[2], Ailton Rocha Monteiro[3], Andressa Cristina Zamboni Machado[4]

[1] *Plant Protection, IAPAR, Londrina, Paraná, Brasil*; [2] *Biology, UEL, Londrina, Paraná, Brasil*; [3] *Phytopathology and Nematology, ESALQ, Piracicaba, São Paulo, Brasil*; [4] *Plant Protection, Instituto Agronômico do Paraná, Londrina, Paraná, Brasil*

The giant nematode, *Tubixaba tuxaua*, was described in Brazil in 1980´s associated with soybean plants in Marechal Cândido Rondon, Paraná State. Since then, some studies were published in which its relationships with other crop species were studied, but little progress has been made in the study of the genetic characteristics and behavior of this nematode oogenesis. In order to increase knowledge about this species, samples were taken in Toledo, Paraná State, aiming to obtain exemplars of *T. tuxaua*. Eighteen samples were collected in four municipalities surrounding Toledo, at a depth of 20 to 50 cm, but in only one sample collected in Mercedes, Paraná State, did we recover enough specimens of *T. tuxaua* for our studies. After the initial morphological identification, we took measurements of adult females and males and DNA was extracted and sequenced using the universal primers of the D2/D3 rDNA region; also, the cytogenetic analysis of males and females was performed to observe the oogenesis. Morphometric data and the de Man indices were grouped by the Ward method, together with available morphometric data from literature, sequences were analyzed by a phylogeny bootstrap method and aligned with sequences from Genebank for our specimens and other Dorylaimida species, and the slides with treated smears were observed and photographed under a light microscope. The dendrogram and phylogenetic analyzes adequately separated *T. tuxaua* from another Dorylaimida species, and allowed inferences about the phylogenetic position of this nematode within its group. The cytogenetical analysis allowed the observation of the process of oocyte and spermatozoan formation, as well as the behavior of the oogonial cells. Thus, important information was obtained, either as tools to identify this species or for knowledge about the reproduction of its germ cells.

Keywords: Giant nematode - Phylogeny - Cytogenetic - Taxonomy.

S2-P29

Recombinase polymerase amplification assay for rapid detection of the root-knot nematodes.

Sergei Subbotin (sergei.a.subbotin@gmal.com)

Plant Pest Diagnostic Center, CDFA, Sacramento, CA, USA

Rapid diagnosis tools for detection of root-knot nematodes play an important role in disease control and eradication programs. Recombinase polymerase amplification (RPA) assays were developed targeting the different genes of several species of root-knot nematodes, *Meloidogyne enterolobii, M. javanica, M. incognita* and *M. arenaria*. The RPA assays using TwistAmp® Basic and TwistAmp® exo kits (TwistDx, UK) allowed detection of Meloidogyne species from gall tissues and crude nematode extracts of all stages of target species without a DNA extraction step. The study included three stages: i) testing and selection of RPA specific primer combinations; ii) validation of sensitivity and specificity for real-time RPA assay; and iii) practical evaluation of real-time RPA assay with field samples. The results of real-time RPA assays with Applied Biosystems QuantStudio 6 Flex Real-Time PCR System using a real-time fluorescent detection of a series of crude nematode extracts showed reliable detection with sensitivity of 1/10 of a second-stage juvenile in a RPA reaction tube after 15-20 min. The RPA assay provides affordable, simple, fast and sensitive detection of root-knot-nematodes. The RPA assays have great potential for being applied and implemented in testing programs on root-knot nematodes in diagnostics laboratories.

Keywords: Diagnostics - Meloidogyne - Real-time RPA assay.

S2-P30

Occurrence of *Pratylenchus* species on raspberries in North Italy with morpho-molecular characterization of a new species.

Alberto Troccoli[1] (alberto.troccoli@ipsp.cnr.it), Elena Fanelli[1], Pablo Castillo[2], Grazia Liébanas[2], Alba Cotroneo[3], Francesca De Luca[1]

[1] *CNR, Istituto per la Protezione Sostenibile delle Piante (IPSP), Bari, Italy*; [2] *CSIC, Institute for Sustainable Agriculture (IAS), Cordoba, Spain*; [3] *Regione Piemonte, Settore Fitosanitario e Servizi tecnico-scientifici, Torino, Italy*

Root-lesion nematode species rank below only root-knot and cyst nematodes as having greatest economic impact on crops worldwide. A survey of plant-parasitic nematodes associated with decaying raspberries (*Rubus idaeus*) in northern Italy, revealed that root-lesion nematodes were the most frequently occurring species among other phytonematodes. *Pratylenchus* species, including mainly *P. penetrans* and *P. crenatus,* have been recorded associated to *Rubus* sp. in Canada (Quebec, Colombia Britannica) and USA (North Carolina, Maryland, New Jersey). In the rhizosphere of symptomatic raspberries plants, specimens of at least two *Pratylenchus* species were detected, one of which being a putative new species. Extracted nematodes were studied as live and fixed material, and subsequently identified by integrative taxonomy (morphological and molecular). Detailed morphometrics of one of the two root-lesion nematode populations was consistent with *P. crenatus*. The second root lesion population was characterized by having a conoid, flattened lip region with 3 annuli, short stylet 14-15 mm long with rounded knobs, relatively high vulva position (76-79%), round and full spermatheca, tail sub-cylindrical with bluntly-rounded and smooth tip, and males frequent. This second species is morphometrically similar (cryptic species) to *P. mediterraneus*, differing just by a shorter pharyngeal overlap. Its molecular identification was carried out by sequencing the D2-D3 expansion domains of the 28S rRNA gene and ITS region. ITS-RFLP and sequence analyses revealed that the new *Pratylenchus* sp. from raspberries show species-specific restriction profiles and no corresponding sequences are present in the database. Phylogenetic relationships, by using D2-D3 sequences, placed the new species in a clade with *P. pseudopratensis* and *P. vulnus*, whereas by using ITS sequences the new species clustered with *P. fallax* and *P. pratensis*. This research confirms the high cryptic biodiversity within the genus, and the need for an integrative approach to the identification of *Pratylenchus* species.

Keywords: Integrative taxonomy - New species - Raspberries - Root lesion nematodes.

References:

- [1] McElroy, 1977. Canadian Plant Disease Survey, 57: 3-8.
- [2] Zasada et al. 2015. Plant Dis., 99(7): 939-946.

S2-P31

Plant parasitic nematodes associated with anise in Turkey.

İbrahim Mıstanoğlu[1] (i_mistanoglu@hotmail.com), Gulsum Uysal[2, 3], Zübeyir Devran[3]

[1] *Bolvadin District Directorate of Ministry of Agriculture and Forestry, Afyonkarahisar, Turkey*; [2] *Bati Akdeniz Agricultural Research Institute, Antalya, Turkey*; [3] *Faculty of Agriculture Department of Plant Protection, Akdeniz University, Antalya, Turkey*

Anise (*Pimpinella anisum* L.) is known as an important medicinal aromatic plant. Plant parasitic nematodes cause economic damage in anise. However, the symptoms of nematodes on anise can be confused with nutrient deficiency or symptoms of other soilborne pests and diseases. To support integrated nematode management programs including crop rotation, accurate and rapid identification of plant parasitic nematodes is quite important. The aim of the study was to determine plant parasitic nematodes in anise growing areas in Bolvadin district of Afyonkarahisar using molecular techniques. A survey was conducted in Bolvadin, which is one of the most important anise production areas of Turkey. A total of 42 soil samples were taken from the region and DNA was extracted from larvae by using a kit. All samples were scanned by species-specific primers belong to root-knot nematodes (*Meloidogyne* spp.), lesion nematodes (*Pratylenchus* spp.), cyst nematodes (*Heterodera* spp.) and foliar nematodes (*Aphelenchoides* spp.). As a result of the study, *Meloidogyne hapla*, *Pratylenchus neglectus*, *P. thornei* and *Aphelenchoides besseyi*, were found in the samples as single or mixed populations.

Keywords: Anise - Identification - Plant parasitic nematodes - PCR.

S2-P32

A new cyst nematode, *Heterodera Luodingensis* n. sp. (Nematoda: Heteroderidae) on Rice from Guangdong province, China.

Yanhua Wen[1] (yhwen@scau.edu.cn), Haoyang Zhen[1], Huan Peng[2], Fuxiang Liu[1], Deliang* Peng[2]

[1] *College of Agr./Nematology Lab, South China Agricultural University, Guangzhou, Guangdong Province, China*; [2] *Chinese Academy of Agricultural Sciences, Institute of Plant Protection, Beijing, China*
** Correspondence to: Deliang Peng, E-mail: pengdeliang@caas.cn*

During the survey of cyst nematode on agricultural crops including rice, sugarcane and corn in Luoding and Jiangmen of Guangdong Province, China from March to July 2017, a new cyst forming nematode population was isolated from rice field soil and rice roots. This new species is described and illustrated as *Heterodera Luodingensis* n. sp. by morphology and molecular analyses of rRNA large subunit (LSU) D2–D3 segments and ITS gene sequence. The Cyst is characterized by light to dark brown, spherical to lemon-shaped with prominent vulval cone (usually mastoid shaped), Neck prominent, vulval cone ambifenestrate, abundant bullae, and strong underbridge, anus distinct. Egg-sac present. The second-stage juvenile (J2) is 435.3±16.1µm (408.3-456.3) long, with three lip annuli and stylet is 18.3±1.4 µm (16.6-22.3) long, with rounded knobs. Lateral fields with three incisures. Tail conoid, 61.3µm (53.9-69.1) long, hyaline terminal section distinct, 31.7µm (27.4–36.2) long, occupying 47.5%-58.3% of tail length. SEM photographs of labial region in *en face* view show dumbbell-shaped fusion of submedian lips. Phylogenetic relationships of *Heterodera Luodingensis* n. sp. indicate it belongs to the Cyperi Group. It is distinguished from *Heterodera elachista* by shorter stylet length of second-stage juvenile (J2) (18.3±1.4 µm vs. 22.5±1.1), lighter color of cysts and larger number of bullae. The restriction enzyme fragment diversity analysis (RFLP) of the ITS fragment PCR product of the *H. luodingensis* n. sp. showed that the enzyme digestion map of the *H. luodingensis* n. sp. could be clearly distinguished from the similar species of the cyst nematodes *H. elachista*, *H. oryzicola*, *H. mothi*, *H. guangdongensis*, and *H. cyperi*. This research was supported by Chinese Special R & D Fund for Public Benefit Agriculture (201503114).

Keywords: Cyst nematode - *Heterodera Luodingensis* n. sp. - Rice.

References:

- [1] Zhen, H.Y., et al., 2018, Scientia Agricultura Sinica, 2018, 51(15):2913-2924

S2-P33

Comparative genomics reveals genome architecture and evolutionary adaptation in *Meloidogyne*.

Chuanshuai Xie (xiesc@webmail.hzau.edu.cn), Dadong Dai, Shengqiang Mao, Jinshui Zheng, Donghai Peng, Ming Sun

Huazhong Agricultural University, Wuhan, China

Root-knot nematodes (RKNs) are obligate endoparasitic pathogens in plants, with huge economic damage in agriculture worldwide. Members of the *Meloidogyne incognita* group (MIG), which includes *Meloidogyne incognita*, *Meloidogyne arenaria* and *Meloidogyne javanica*, are notorious root-knot nematodes due to wide host ranges and particular reproductive mode (mitotic parthenogens). Previous studies had shown that MIG originated from hybridization [1,2], but limited by the low-quality genome, the details of genome architecture and evolution are unclear. Here we obtained the high-quality genome of four MIG species (*M. incognita*, *M. arenaria, M. arenaria_psd and M. javanica*) and one diploid lineage *M. graminicola* by using a combination of single-molecule real-time sequencing, Bionano optical mapping and Hi-C. Using *M. graminicola* genome as outgroup, we found a large number of conserved synteny blocks with 1:3 or 1:4, speculating that *M. incognita* and *M. arenaria_psd* may be triploid, *M. arenaria* and *M. javanica* were tetraploid. Phylogenetic analysis proved that the triploid species was likely AAB and the tetraploid species was AABB, suggested that MIG may come from two common ancestral species. To detect genomic changes after hybridization, we divided the Mi genome into two subgenomes, and further identified chromosomal rearrangements, gene loss, and transposon outbreaks in the different subgenomes. Combining RNA-seq and Hi-C data, we found that subgenome A had more dominant in homoeologous expression and more active three-dimensional conformation, while subgenome B had more heterochromatin regions. The further analysis showed genes with homeolog expression bias (HEB) had different Pfam enrichment, suggesting that the root-knot nematode may increase its adaptive capacity by increasing the complexity of gene expression pattern. We are working on *Meloidogyn spp.* genomics to systematically reveal the evolutionary history and adaptive mechanism of the MIG species.

Keywords: Root-knot nematodes - Hybridization - Evolution.

References:

- [1] Blanc-Mathieu R et al.,2017.PLoS genetics.13(6): e1006777
- [2] Szitenberg A et al.,2017.Genome biology and evolution.9(10): 2844-2861

POSTERS

S3. Biodiversity and ecology

S3-P01

Interaction of the bacteria *Photorhabdus luminescens* with life history traits and virulence of *Heterorhabditis bacteriophora.*

Temesgen Addis[1] (t.addis@e-nema.de), Elsie Enow[2], Augustina Tetteh[2], Carlos Molina[1], Ralf-Udo Ehlers[1]

[1] *e-nema GmbH, Schwentinental, Germany*; [2] *University Gent, Gent, Belgium*

Life history traits (LHTs) of *Heterorhabditis bacteriophora* were assessed at 25°C in a hanging drop technique using *Photorhabdus luminescens* in a semi-solid nematode growth medium at cell densities ranging from 2.5-20 × 10^9 cells ml^{-1}. Increasing food density had a significant positive influence on the body volume of hermaphrodites. The offspring production ranged from 50-269 juveniles/hermaphrodite. The lifespan of hermaphrodites, which was predetermined by the beginning of the *endotokia matricida,* remained similar (8 days). In another investigation six different *P. luminescens* strains/species were combined with two *H. bacteriophora* strains in liquid culture and the effect on dauer juvenile (DJ) recovery, yield and virulence was recorded. Exchange of symbiotic bacteria affected DJ recovery that ranged from 20-88%. Similarly, DJ yield was significantly influenced by the type of bacterial strain. It was found that native bacteria are not necessarily more virulent or provide the highest yield. However, variation between the two nematode strains in DJ recovery and yield was higher compared with variations obtained with exchanging the bacterial strains. The virulence of a nematode-bacterium combination seems to be governed more by the nematode strain than the associated bacterial strain.

Keywords: LHT - Symbiotic bacterium - Entomopathogenic nematode - Endotokia matricida - Mutualism.

S3-P02

Anatomical alterations in Aleppo pine roots induced by the dagger nematode *Xiphinema vuittenezi.*

Leena Irshaid[1] (lalbanna@ju.edu.jo), Monther Sadder[2], Luma Al Banna[3]

[1] *Department of Agricultural Biotechnology, Al Balqa'a Applied University, Al salt, Jordan*; [2] *Department of Horticulture and Crop Scienc, The University of Jordan, Amman, Jordan*; [3] *Department of Plant Protection, The University of Jordan, Amman, Jordan*

The dagger nematode *Xiphinema vuittenezi* was recovered from rhizosoil of declining Aleppo pine trees (*Pinus halepensis*) grown in AL-Jubiha area in the Northern Mediterranean phytogeographical region of Jordan. We aimed in this study to understand the impact of *X. vuittenezi* on pine roots. Inoculation of pine seedlings with *X. vuittenezi* resulted in pale light green upper needles and lower brown ones, with swellings and dichotomous branching of the root tips. Our observations revealed that the dagger nematode *X. vuittenezi* fed on root tip and the parts just above the root tip. Histopathological studies of pine roots inoculated with *X. vuittenezi* after one hour, one day, and one week of feeding were conducted. After one hour of nematode feeding on root tip, the root cap was detached with empty spaces in the infected root tip. Nuclei of infected cortex were relatively larger than healthy (non-inoculated) roots. When the nematode attacked the roots beyond root tip, starch accumulated in the cortex with no other obvious changes on cortical cells or nuclei compared to non-inoculated ones. One day after inoculation, cortical cells of the root tip increased in size with enlarged nuclei and swelled root tips. The lignin contents increased in nematode-fed tips compared to the non-inoculated ones. In the nematode-fed region beyond the root tip, cortical cells and nuclei were larger than those of non-inoculated roots. Additionally, starch increased in infected roots. After one week of nematode attack, the root cap was absent in infected root tip while it was still present in the healthy one. More cellulose and starch were present in infected root tips. Swellings were noticed in infected tips with more expanded cavities. The cells and the nuclei of the cortex in the nematode-fed region beyond root tip enlarged with exception of the presence of one multinucleate cell that might be a giant cell. Accumulation of tannins was greater in the infected roots than in the healthy roots. Interestingly, no necrosis was noticed after one week of attack. Further histochemical studies are needed to understand the changes in root architecture.

S3-P03

Community composition and metabolic footprints of soil nematodes in fruit systems in Mediterranean areas.

Brynelly Bastidas[1, 2] (bebastidas@uc.cl), Eduardo Arellano[1, 2], Aurora Gaxiola[3, 4], Erwin Aballay[5], Sara Sánchez-Moreno[6]

[1] *Pontificia Universidad Catolica de Chile, Santiago, Chile*; [2] *Center of Applied Ecology and Sustainability (CAPES), Santiago, Chile*; [3] *Departamento de Ecología, Facultad de Ciencias Biològicas, PUC, Pontificia Universidad Catolica de Chile, Santiago, Chile*; [4] *Instituto de Ecología y Biodiversidad., Santiago, Chile*; [5] *Departamento de Sanidad Vegetal, Universidad de Chile, Santiago, Chile*; [6] *Departamento de Medio Ambiente y Agronomía, Centro Nacional Instituto de Investigación y Tecnología AgrariayAlimentaria INIA, Madrid, Spain*

Soil nematode communities play an essential role in ecosystem energy flow and provide information on ecosystem services through metabolic footprints. However, there are still no clear patterns of the composition and metabolic footprints of the nematode community in some biomes as occurs in Mediterranean climates. The central zone of Chile presents this type of climate and is where the most significant extension of the country's fruit sector is located, which leads us to consider that the ecosystem services of that area must be affected by the high agricultural activity. The objective of this study was to evaluate the composition and metabolic footprints of the soil nematode community in the most extensive fruit systems in the Mediterranean region, such as grape and cherry crops. Soil samples were taken at a depth of 30 cm in 5 different geographical areas; each area had cherry, grape, and non-cultivated areas. Physicochemical properties of each soil were also analyzed, and the biomass and metabolic activity of each guild of the nematode community were quantified to obtain the metabolic footprint. The results showed that the greatest diversity and abundance of total nematodes was generally found in non-agricultural areas, contrary to the agricultural soils of grape and cherry crops, where the abundance and diversity were lower and without significant differences. A high enrichment footprint indicates an increase in resource input into the soil food web, while a low enrichment footprint suggests a decrease in available prey. Therefore, a decrease in ecosystem services is observed in both soils of fruit crops, losing the pest suppression service, which increased the guilds of phytoparasitic nematodes that cause severe damage to crops, also increasing mineralization services mediated by bacterivorous nematodes. Significant differences were observed between the crops and the physicochemical properties of the soil: bulk density (DA), carbon (C), phosphorus (P), electrical conductivity (EC), aggregates, except in organic matter (OM), pH, and Nitrogen (N). Our study shows that agricultural activities associated with high fruit extension in the central zone of Chile have affected the composition of the nematode community and the proportion of ecosystem services.

Keywords: Nematodes - Soil - Fruit systems - Metabolic footprints - Mediterranean.

References:

- Sánchez-Moreno, S., & Ferris, H. (2018). Plant-Parasitic Nematodes in Subtropical and Tropical Agriculture. CAB International, Wallingford, 62-83.
- Guerra, C. A., et al., (2020). Nature communications, 11(1), 1-13.

S3-P04

Nematode parasite loads on ruminant grazing sites at Allen Island, Maine, USA.

Ellen Batchelder (ebatchelder@unity.edu)

School of Environmental Conservation, Unity College, Unity, ME, United States

The nematode *Haemonchus contortus* is an economically important livestock parasite. Eggs are deposited on pastures in feces from infected animals, hatch, and develop in a few days into larvae that can infect new grazing hosts. Ingested *H. contortus* make their way into the abomasum, or true stomach, of the host where they feed, mature, and reproduce. As they suck their host's blood, they cause symptoms including weight loss, anemia, and sometimes death. They are especially problematic because they can overwinter in a hypobiotic state in host tissues, they are exceptionally fecund, and have rapidly developed resistance to drug treatments. It is clear that multiple strategies including pasture rotations are necessary to reduce parasite loads and that appropriate strategies will depend on many specific factors such as local climate, herd density and pasture size, etc. The objective of this study is to determine the temporal distribution of larval stages on pastures used by a small hobby flock of sheep located on a coastal island in the Gulf of Maine. Specifically, to understand how long infective larvae persist on pastures in this particular environment, if persistence differs among the local microhabitats, and if efforts to remove sheep feces from pastures have an effect on larval numbers.
Pasture samples were collected in several different areas monthly between March (end of snow pack) and September. Nematodes were separated from the herbage by decanting, sieving, and sedimentation, then fixed in formaldehyde. Enumeration and identification of parasitic and free-living nematodes using mass slide preparations is ongoing. Ultimately, the results should suggest specific pasture management practices to complement animal management practices and anthelminthic treatments in controlling parasite loads in sheep, particularly on small scale farms. In addition, the free-living nematodes also present in samples are being described to provide insights into nematode biodiversity in the mid-coast Maine region.

Keywords: Ruminant - Parasitic - Management - Biodiversity.

S3-P05

Free-living nematode assemblages in the rhizosphere of watermelon plants in Nigeria: a baseline study.

Tesleem Bello[1] (tesleembello@yahoo.com), Danny Coyne[2], Hendrika Fourie[1]

[1] *Unit for Environmental Sciences, North-West University, Potchefstroom, North-West, South Africa*; [2] *Nematology, IITA, Ibadan, Oyo, Nigeria*

Watermelon is increasingly produced and consumed in Nigeria and sub-Saharan Africa. However, limited information exists regarding nematode pests and beneficial/free-living nematodes associated with the crop. The present study recorded the abundance and diversity of free-living nematodes from 50 watermelon fields across south-west Nigeria during 2016/2017. Of the 30 genera identified from soil samples, *Cephalobus*, followed by *Rhabditis*, *Aphelenchus* and *Aporcelaimus*, were predominant. Variation in nematode community structures across the 50 fields was apparent for mean maturity indices, metabolic footprints, feeding-type composition and coloniser-persister (c-p) structure. Faunal analyses characterised 52% of the fields as having stable and enriched soil food webs, which is beneficial for crop production. Significant correlations were apparent between some nematode genera and selected soil properties, and rainfall. This study provides the first information of free-living nematodes associated with watermelon from sub-Saharan Africa, offering novel and baseline information on their abundance and diversity in south-west Nigeria.

Keywords: Beneficial nematodes - *Citrullus lanatus* - Faunal analysis - Prominence values - Soil health.

S3-P06

Meloidogyne species and other plant-parasitic nematodes associated with watermelon in Nigeria.

Tesleem Bello[1] (tesleembello@yahoo.com), Milad Rashidifard[1], Danny Coyne[2], Hendrika Fourie[1]

[1] *Unit for Environmental Sciences, North-West University, Potchefstroom, North-West, South Africa*; [2] *Nematology Unit, IITA, Ibadan, Oyo, Nigeria*

Plant-parasitic nematodes have been documented from various parts of the world as being economically damaging on watermelon. Little information is available, however, on nematode pests associated with the crop in sub-Saharan Africa. The present study recorded the abundance and prevalence of plant parasitic nematodes from 25 localities across south-west Nigeria during 2016/2017. Of twelve nematode species identified, those of *Meloidogyne* spp. were predominant, followed by *Helicotylenchus dihystera*, *Pratylenchus zeae* and *Scutellonema bradys*. Morphology and molecular techniques were used to identify four *Meloidogyne* spp. *Meloidogyne enterolobii* was the most prevalent followed by *M. incognita*, *M. javanica* and *M. arenaria*. *Meloidogyne arenaria* is here reported for the first time from south-west Nigerian cropping systems. Significant associations were observed between the frequency of occurrence of the predominant nematode species and soil properties as well as rainfall. Results provide baseline information for watermelon in Nigeria and in a wider context for sub-Saharan Africa.

Keywords: Mean population density - Molecular techniques - Morphology - Perineal-pattern - Prominence values.

S3-P07

Diversity of plant-parasitic nematodes associated with *Crocus sativus* L. in Morocco: relationships with edaphic factors.

Salah-Eddine Laasli[1] (laaslisalaheddine@gmail.com), Fouad Mokrini[2], Abdelfattah Dababat[3], Silas Kipsang Bett[1]

[1] *Nematology Research Unit, Ghent University, Gent, Belgium*; [2] *Biotechnology unit, INRAe, Rabat, Morocco*; [3] *CIMMYT Turkey, Ankara, Turkey*

Saffron (*Crocus sativus* L.) fields in Morocco's Taliouine and Taznakht regions were surveyed between January and April 2018 to study the diversity and incidence of plant-parasitic nematodes and assess the effects of soil physico-chemical properties on the nematodes. Fourteen nematode genera were identified in soil and root samples collected from 66 saffron fields. The most common plant-parasitic nematodes in Taliouine region were *Pratylenchus* spp. and *Helicotylenchus* spp. In Taznakht region, the most common nematodes were *Pratylenchus* spp., *Tylenchorhynchus* spp. and *Ditylenchus dipsaci*. Nematodes, particularly *Pratylenchus* spp. and *Ditylenchus* spp., were abundant and frequent throughout the region. Several nematode genera were significantly associated with soil texture and mineral content, indicating that soil properties play an important role in plant-parasitic nematode communities. This description of plant-parasitic nematode assemblages associated with saffron fields in Morocco and their relationships with soil physico-chemical properties provides a starting point from which appropriate nematode management strategies can be implemented.

Keywords: *Ditylenchus* spp. - Ecology - Nematode survey - *Pratylenchus* spp - Soil texture.

References:

- Mokrini et al., 2019. Nematol 22(1): 87-102.

S3-P08

MaisSolo: nematodes as bioindicators of soil status.

Maria João Camacho[1, 2] (mjoao.camacho@iniav.pt), Leidy Rusinque[3], Filomena Nobrega[4], Maria L. Inácio[3, 5]

[1] *NemaINIAV, INIAV, I.P. - Instituto Nacional de Investigação Agrária e Veterinária, Oeiras, Évora, Portugal*; [2] *U. Évora/NemaLab, MED – Mediterranean Institute for Agriculture, Environment and Development, Ins, Évora, Portugal*; [3] *NemaINIAV, INIAV, I.P. - Instituto Nacional de Investigação Agrária e Veterinária, Oeiras, Portugal*; [4] *Bioquímica, INIAV, I.P. - Instituto Nacional de Investigação Agrária e Veterinária, Oeiras, Portugal*; [5] *ITQB NOVA, GREEN-IT Bioresources for Sustainability, Oeiras, Portugal*

The predominant horticultural production systems for industry in Ribatejo (Portugal) are based on crops with high phytotechnic intervention, often in monoculture and with a high degree of intensification. At the soil nematofauna level, these practices result in biodiversity imbalances and lead to the emergence of serious phytosanitary problems for which there is a growing lack of control methods. To counteract this trend, the project MaisSolo aims to demonstrate the advantages of changing current monoculture systems to practices that include, for example, the installation of cover crops during the fall-winter period, preceding the main crop of the agricultural year. This work evaluated nematodes as bioindicators of soil health in experimental plots, where different cover crops were installed prior to main crop cultivation: 1) biodiverse mixture of grasses and legumes, including *Trifolium resupinatum* inoculated with nitrogen-fixing rhizobia; 2) *Lolium multiflorum* (annual ryegrass), a mycotrophic grass that favours soil enrichment in endemic mycorrhizal fungi; and 3) *Raphanus sativus* (forage turnip), a biofumigant species which incorporated into soil contributes to the elimination of pathogens. A control plot was maintained without any cover crop. The proportion of the different trophic groups in the soil nematofauna can be converted into relevant information regarding its biological status. Samples from the soil rhizosphere were collected and the abundance of different nematode trophic groups determined. A high proportion of plant parasitic nematodes (belonging to genera which do not cause damage to the established cultures), bacterial feeders and fungal feeders were detected. Omnivorous and predatory nematodes were found in low numbers. The higher prevalence of plant parasitic nematodes was due to the vegetation cover, since there was a greater production of root biomass that support their multiplication. The reduced number of omnivorous and predatory nematodes confirms the type of farming with large cultural intensification (annual crop), in which fallow period of the plots is short and not enough for nematode populations to recover after imbalance factors (tillage, application of pesticides, etc.).

Financial support was provided by Project PDR 2020-101-030821 (Grupo Operacional MaisSolo).

Keywords: Nematofauna - Biological indicators - Soil - Agrobiodiversity.

S3-P09

Nematode communities as bio-indicators of constructed soil: a case of study in a Mediterranean city, Montpellier.

Camille Chauvin[1] (camille.chauvin@elisol-environnement.fr), Olivier Taugourdeau[2], Jennifer Hellal[3], Cécile Villenave[1]

[1] *ELISOL environnement, Congénies, France*; [2] *Valorhiz, Montpellier, France*; [3] *BRGM, Orléans, France*

In urban and industrial area, soil restoration has become a major issue to promote park and green space [1]. Soil restoration can be based on practices such as soil decompaction and cover-crops but also on soil construction through the assemblage of treated soil and recycled wastes [2, 3]. In this study, we assessed how nematode communities can be use as soil bioindicators to monitor soil refunctionalization on an old vehicle passing area in Montpellier (France). Three situations were followed during an 18-months period: TE, unrearranged area as control; GE, soil decompaction at 40 cm and seeding; TG, stripping of 40 cm, then backfilling the pit with "Terragenèse®" a material developed by the Valorhiz society. At the beginning of the trial, all situations showed low nematode abundance and low diversity. Unpacking and seeding on the GE soil improved free-living nematodes abundance and structure index value significantly more than in TE soil. Eighteen months later, although the biological activity is still moderate in this soil, the functional diversity and taxonomic composition of the nematode communities are approaching those of a meadow land. The constructed soil TG enabled an abundant microbivore nematode population during the first 12 months of the trial, but the nematode communities remained poorly diversified with low structure index (SI) and high variation of the enrichment index (EI). In this constructed soil, the biological activity was improved but specialized on organic matter degradation. The effect was of short duration as free-living nematode abundance decreased after 18 months. Results on TG modality highlight that nematodes can serve as bioindicators for monitoring the land refunctionalization in the first few months and for evaluating soil engineering methods. Longer-term studies are needed to evaluate the sustainability of functioning of the new soils.

Keywords: Soil food web - Soil restoration - Constructed soils - Nematode communities - Urban area.

References:

- [1] Geisen, S et al., (2019). Current Biology, 29(19), 1036-1044.
- [2] Séré, G et al., (2008). Journal of soils and sediments, 8(2), 130-136
- [3] Villenave, C. et al., (2018). Eurasian soil science, 51(10), 1266-1273.

S3-P10

Rice-root nematodes (*Hirschmanniella* spp.) and rice root-knot nematodes (*Meloidogyne graminicola*) in Takeo and Prey Veng Provinces, Cambodia.

Buncha Chinnasri[1] (agrbcc@ku.ac.th), Buyung Hadi[2], Rathmuny Then[2], Natthidech Beesa[1], Kansiree Jindapunnapat[3], Rica Flor[2], Kim Eang Tho[4]

[1] *Department of Plant Pathology, Faculty of Agriculture, Kasetsart University, Bangkok, Thailand*; [2] *Cambodia Office, The International Rice Research Institute (IRRI), Phnom Penh, Cambodia*; [3] *Department of Entomology and Plant Pathology, Faculty of Agriculture, Khon Kaen University, Khon Kaen, Thailand*; [4] *Division of Research and Extension, Royal University of Agriculture (RUA), Phnom Penh, Cambodia*

The population dynamics of rice-root nematodes (*Hirschmanniella* spp.) and rice root-knot nematodes (*Meloidogyne graminicola*) were measured during the annual rice growing period in Takeo and Prey Veng provinces, Cambodia. Nematode samplings were conducted four times: September, 2018, November, 2018, March, 2019, and June, 2019. Details of rice varieties and rice ages in each survey were recorded. Thirty rice plots (grown with various rice varieties) in each province were selected for soil and rice root samplings. In each plot, five rice plants were randomly uprooted (at the radius of three meters from the center of the plot). The result showed that *Hirschmanniella* existed more predominantly than *M. graminicola*, especially in rice roots. Additionally, the survey demonstrated that flooding conditions suppressed populations of both nematodes as shown by greater numbers of nematodes in September (early flooding season) than November (late flooding season). *Hirschmanniella* survived flooding better than did *M. graminicola.* During the off-season, *Hirshmanniella* and *M. graminicola* survived in high numbers in volunteer rice, stumps and soil. *Hirschmenniella* collected from Takeo and Prey Veng provinces had morphological characteristics and morphometrics somewhat similar to *H. mucronata*. Molecular identification based on the PCR technique using the universal primers (D2A/D3B) and sequence comparison with the GenBank database affirmed that the nematodes were *H. mucronata.* Resistance and susceptibility of different rice varieties to nematodes were also determined in naturally-grown rice plants in Takeo and Prey Veng provinces. The results demonstrated low number of *Hirschmanniella* in the roots of PHK, REC, and IR54 rice varieties.

Keywords: *Hirschmanniella mucronata* - *Meloidogyne graminicola* - Rice.

S3-P11

Pin nematodes (*Paratylenchus* spp.) parasitizing *Prunus* in Spain: distribution, ecological factors and specific PCR for major species identification.

Ilenia Clavero-Camacho[1] (iclavero@ias.csic.es), Carolina Cantalapiedra-Navarrete[1], Antonio Archidona-Yuste[2], Pablo Castillo[1], Juan Emilio Palomares-Rius[1]

[1] *Institute for Sustainable Agriculture, Córdoba, Spain*; [2] *Andalusian Institute of Agricultural and Fisheries Research and Training, Córdoba, Spain*

Pin nematodes of the genus *Paratylenchus* are obligate plant-ectoparasitic nematodes of small body length (<600 µm) with a wide host range and global distribution. Some species are considered pathogenic in crops; therefore, understanding the environmental and agronomic factors involved in their distribution is critical to design management strategies to reduce plant damage. This study aimed to evaluate the prevalence and distribution of *Paratylenchus* species in the main areas of *Prunus* spp. production in Spain, their ecological constraints and new molecular tools for the specific identification of major species. A total of 219 sampling sites were surveyed and twelve *Paratylenchus* species were identified based on integrative taxonomic approach (*P. baldaccii*, *P. enigmaticus*, *P. goodeyi*, *P. hamatus*, *P. holdemani*, *P. indalus*, *P. israelensis*, *P. pedrami*, *P. tateae*, *P. tenuicaudatus*, *P. veruculatus* and *P. zurgenerus*). The most common pin nematode was *P. hamatus*, followed by *P. tenuicaudatus*. Nematode abundance was influenced by climatic characteristics, soil chemical properties and agronomic management practices. Nine explanatory variables were selected as the most strongly associated with *Paratylenchus* distribution. Specifically, *P. tenuicaudatus* was significantly correlated with soil chemical characteristics, such as, pH, carbon, sulphur and sodium content. Whereas *P. goodeyi* was closely related to fields with less than 10 years of almond cultivation. Species-specific PCRs were developed for *P. hamatus* and *P. tenuicaudatus* and their validity was evaluated studying the molecular variability of these species and against other *Paratylenchus* species.

Keywords: Distribution - Paratylenchus hamatus - Paratylenchus tenuicaudatus - PCR.

S3-P12

Potato-cyst nematodes, *Globodera* spp., and root-knot nematodes, *Meloidogyne* spp., on potato in Portugal.

Isabel Conceição (luci@zoo.uc.pt), Rita Gonçalves[1], Ana Martins[2], Sofia R. Costa[2], M. Teresa M. Almeida[2], Raquel Varandas[1]

[1] *CFE, Department of Life Sciences, University of Coimbra, Coimbra, Portugal;* [2] *CBMA, University of Minho, Campus de Gualtar, Braga, Portugal*

Potato, *Solanum tuberosum*, is a highly relevant crop in Portugal, where it is mostly cultivated in the Centre and North regions. Potato plants are susceptible to pests and diseases that compromise its yield quantity and quality. Many plant-parasitic nematode genera can feed on potato roots, but potato cyst nematodes (PCN), *Globodera* spp., and the root-knot nematodes (RKN), *Meloidogyne* spp., can be important pathogens of potato. The aim of this work was to identify PCN and RKN species in potato growing regions of Portugal as part of the TREND project (POCI-01-0145-FEDER-029289; PTDC/ASP-PLA/29282/2017). Soil and root samples were collected in 54 fields in 2018 and 2019. Nematode cysts were extracted from soil, with a modified Fenwick can. *Meloidogyne* spp. females were extracted from roots using a stereomicroscope. Identification of *Meloidogyne* isolates was made by morphological characterization of perineal patterns and confirmed by esterase phenotype of females [1] obtained through enzyme gel electrophoresis. In 54 samples, RKN were detected in 6 samples and identified as *M. hapla, M. hispanica*, *M. incognita* (2), *M. javanica*, and 1 as a mixture of *M. hapla*, *M. hispanica* and *M. javanica.* Identification of the *Globodera* isolates was performed by PCR-RFLPs, with specific primers and restriction enzymes *Alu*I and *Hinf*I [2, 3]. PCN isolates were detected in 7 samples and identified as *G. pallida* (6) and *G. rostochiensis* (1). In the North of Portugal, only 3 PCN isolates (2 *G. pallida* and 1 *G. rostochiensis*) and 1 RKN isolate (*M. hapla*) were detected. The other isolates were found in the Centre of Portugal. These differences may be due to the warmer climate of the centre of the country. Accurate identification of *Meloidogyne* spp. and *Globodera* spp. isolates present in potato fields is important for quarantine purposes and to implement effective integrated management programs. Global climatic changes may lead to a replacement of PCN populations by RKN populations, which are better adapted to warmer temperatures and may become an even more serious threat to potato crops in the future.

Keywords: Biochemical identification - Molecular identification - Potato cyst nematodes - Root-knot nematodes.

References:

- [1] Pais et al., 1986. Ciência Biológica 6: 19-34.
- [2] Ferris et al., 1993. Fundam Appl Nematol 16: 177-184.
- [3] Širca et al., 2010. Phytopathol Mediterr 49(3): 361-69.

S3-P13

First report of *Caenorhabditis brenneri* (Nematoda: Rhabditida) association with the terrestrial slug *Philippinella moellendorffi* from the Philippines.

Loel Dalan[1] (loel.dalan@g.msuiit.edu.ph), Michelle Anne Diano[1], Irma Tandingan-De Ley[2], Nanette Hope Sumaya[1]

[1] *Flora Biodiversity Laboratory-PRISM, Department of Biological Sciences, Mindanao State University-Iligan Institute of Technology, Iligan, Zamboanga del Norte, Philippines*; [2] *Department of Nematology, University of California-Riverside, Riverside, California, United States of America*

Gastropod-associated nematodes have been previously studied and documented worldwide, with some species forming host-specific association as obligate parasites of molluscs while others form intermediate and temporary association. *Philippinella moellendorffi* recently identified from Little Baguio, Imelda, Zamboanga Sibugay were collected and maintained aseptically in the laboratory until mortality was observed. Cadavers were washed and cultured in nutrient agar (NA) media to allow nematode proliferation. Pure cultures were obtained through a series of subcultures using one gravid female. Morphology, morphometrics, and molecular analyses (18S rDNA and D2-D3 expansion segments of 28S rDNA) were employed as diagnostic tools in identifying the isolated nematode species associated with *P. moellendorffi.* The *P. moellendorffi*-associated nematode was identified as *Caenorhabditis brenneri,* thus designated as *C. brenneri* strain IZSP from the Philippines. This study showed that *C. brenneri* also formed association with terrestrial slugs and furthermore contributed to the host potential of *Philippinella* species to be a vector of non-parasitic nematode species. This finding sets the first association record of *C. brenneri* with terrestrial slug *P. moellendorffi,* a new slug record in Mindanao, Philippines.

Keywords: *Caenorhabditis* - Gastropods - Philippinella - 18s rDNA - D2-D3 expansion segments of 28S rDNA.

S3-P14

Selective feeding and reproductive activities of a facultative plant-parasitic nematode *(Aphelenchoides besseyi)* and a fungal feeder *(A. pseudogoodeyi)* on isolates of fungi pathogenic and non-pathogenic to strawberry.

Clemen De Oliveira[1] (clemendeoliveira@ufl.edu), Johan Desaeger[1], Janete Brito[2], Natalia Peres[3], Teresa Seijo[3]

[1] *Entomology and Nematology Department, University of Florida, Wimauma, FL, United States*; [2] *Nematology Section, Florida Department of Agriculture, Wimauma, FL, United States*; [3] *Plant pathology Department, University of Florida, Wimauma, FL, United States*

Aphelenchoides besseyi and *A. pseudogoodeyi* are foliar nematodes associated with strawberry in Florida. In 2016, *A. besseyi* was found on several strawberry farms in Florida, causing severe stunting and considerable damage. Recent surveys in Florida strawberry fields have revealed the presence of many other *Aphelenchoides* spp. One of those is a newly discovered species, *A. pseudogoodeyi*, which appears to be a strictly fungal feeding nematode (Oliveira et al., 2019). It is hypothesized that both nematode species could feed and reproduce on fungi present in the soil. Feeding and reproductive abilities of these two nematodes were assessed on Florida isolates of four fungi pathogenic to strawberry; *Colletotrichum gloeosporioides Macrophomina phaseolina, Pestalotiopsis clavispora*, and *Botrytis cinerea*, one non-pathogenic; *Fusarium oxysporum* and one fungus pathogenic to peach; *Monilinia fructicola*. *Aphelenchoides besseyi* reproduction was significantly higher (p-value<2e-16) on strawberry pathogenic isolates of *B. cinerea*, *C. gloesporioides,* and *P. clarispora* than on the non-pathogenic isolates of *F. oxysporum*, and *M. fructicola*. On the contrary, *A. pseudogoodeyi*'s reproductive rates did not differ across the fungus cultures. *M. phaseolina* was an exception because it did not produce mycelium in the medium used, thus being a poor host for both nematodes. Our findings indicate that *A. besseyi* is more selective in its feeding preference on fungi than *A. pseudogoodeyi*. Fungi play an important role in maintaining the populations of the two nematodes in the soil. The removal of strawberry plant residues infected by fungi is a desirable and effective management practice to suppress *A. besseyi* in commercial strawberry fields in Florida.

Keywords: Foliar nematodes - Strawberry pathogens - Feeding habits - Nematode-fungi interaction.

References:

- Oliveira et al., 2019. J. Plant Diseases. 103 (11): 2825-2842.

S3-P15

Nematode survey of grassland habitats in two nature reserves of the Free State Province, South Africa.

Anke de Smidt (ankedesmidt@gmail.com), Candice Jansen van Rensburg, Liesl L Van As

Zoology and Entomolgy, University of the Free State, Bloemfontein, Free State Province, South Africa

The Phylum Nematoda Potts, 1932 is highly diverse in terms of species richness and inhabit almost every known ecological niche on earth [1]. Even though these organisms are ubiquitous, many species are still being described [2]. Records from the South African Plant-Parasitic Nematode Survey (SAPPNS) and National Collection of Nematodes (NCN) showed little information of free-living and plant-parasitic nematodes in grassland habitats from the Free State Province, especially in nature reserves. Soil samples were collected at different times of the year between 2017 and 2018 and January 2019 from grassland areas within Soetdoring and Willem Pretorius Nature Reserves, respectively. Nematodes were extracted and processed in the laboratory using standard techniques. Twenty-eight genera belonging to 18 families were identified from Soetdoring Nature Reserve, while twenty-nine genera belonging to 17 families were identified from Willem Pretorius Nature Reserve. The family Cephalobidae were most commonly observed throughout the survey. Plant feeders were the most abundant trophic group, followed by fungivores and omnivores, that were identified from the grassland samples per survey, in both reserves. The highest Maturity Index (MI) value was recorded from the grassland sample of Soetdoring Nature Reserve in June 2018, while the lowest MI was observed in the grassland sample of Willem Pretorius Nature Reserve in January 2019. The nematode diversity was the highest in the grassland sample in Soetdoring Nature Reserve in June 2018, while the lowest nematode diversity was recorded in July 2017 from the grassland sample in Willem Pretorius Nature Reserve. The nematode faunal profile analysis showed a majority of samples clustered into Quadrat C, demonstrating that both grassland habitat food webs were stable with multiple trophic linkages. The information gathered from this survey has already contributed to the knowledge of free-living and plant-parasitic nematodes from grassland habitats in South Africa and lays the foundation for similar studies in the future.

Keywords: Free-living nematodes - Free-State - Grasslands - Nature reserves.

References:

- [1] Mulder et al., 2005. Ecotoxicology and Environmental Safety 62: 278-289.
- [2] Du Preez et al., 2013. Czech Speleological Society pp 386-390.

S3-P16

Isolation and identification of bacteria associated with nematodes from *Achatina fulica*: an unexpected occurrence of anaerobic *Clostridium lundense.*

Michelle Anne Diano (michelleanne.diano@msuiit.edu.ph), Nanette Hope Sumaya

Flora Biodiversity Laboratory - PRISM, Department of Biological Sciences, Mindanao State University-Iligan Institute of Technology, Iligan City, Philippines

The interplay between host-microbe interactions ranging from mutualistic to pathogenic relationships are quite interesting. Such associations between nematode-gastropod complexes and bacteria, if fully unraveled, may provide beneficial impacts on human interest and health. It is perceived that bacterivores in soil are important for many ecosystem functions, and out of a vast range of microorganisms found in soil only a few to date are documented to make a physical interaction with nematodes. To study this, bacteria associated with nematodes collected from a cosmopolitan invasive gastropod species, the giant African land snail, *Achatina fulica* were isolated and identified. Morphological, biochemical, and molecular analysis using 16S rRNA gene marker identified a total of seven bacterial strains. Two bacterial strains of *Bacillus cereus* are associated with *Caenorhabditis brenneri*; three bacterial strains of *Proteus mirabilis* with *Caenorhabditis briggsae*; one bacterium, *Providencia vermicola* is associated with a *Panagrolaimus* sp. and also one bacterium, *Clostridium lundense* with *Rhabditis* sp. Whereas the other bacteria are commensals in many different environments, the prevalence of *C. lundense*, a strict anaerobic, mesophilic, endospore-forming, lipolytic bacterium isolated originally from bovine rumen fluid, is rather unexpected. This is the first ever report of *C. lundense* associated with nematodes. This study mainly provides an additional account and collection of naturally-isolated nematode-bacteria, contributing to limited knowledge on its diversity, ecology and associations. Additionally, the associated bacteria promoted growth and development to nematodes, and did not cause nematode mortality on in vitro cultures. Further studies on the life history trait of nematodes (i.e reproduction rate, etc.) using their associated bacteria can be conducted for deeper understanding on their interactions. A culture-independent method using high throughput sequencing to examine the metagenome in the nematode-bacteria associations is also a possible research outlook.

Keywords: Host-microbe - 16S rRNA - Biochemical - Nematode-gastropod complex - Associations.

S3-P17

Survival of *Meloidogyne graminicola* in soil under different moisture conditions.

Claudia Dias-Arieira[1] (crdarieira@uem.br), Mayra Soares[2]

[1] *Agronomy, Universidade Estadual de Maringá, Umuarama, Parana, Brasil*; [2] *Agronomy, Universidade Estadual de Maringá, Maringá, Parana, Brazil*

Plant-parasitic nematodes may persist in the soil, even under adverse conditions, such as drought or flood. This study aimed to determine the ability of *Meloidogyne graminicola* to survive for 1 year in soils under different moisture conditions. The experiment was conducted in a greenhouse using a completely randomized 3 × 12 factorial design, with three moisture levels and 12 incubation periods. Rice plants cv. IRGA were transferred to pots containing 700 mL of a mixture of soil and sand (1:1) (previously autoclaved at 120 °C for 2 h) and inoculated with 1500 eggs and second-stage juveniles of *M. graminicola*, obtained from a single-species population. At 60 days after inoculation, shoots were cut off at ground level, the soil was lightly tilled, and irrigation regimes were applied. The following moisture treatments were used: (a) continuously flooded to 1 cm above the soil surface, (b) irrigated to about 60% of the field capacity, and (c) dry (without irrigation). Rice seeds were sown monthly for 1 year, with seven replications for each treatment. Plants were harvested at 60 days after each sowing date, and the roots were evaluated for nematode count. Nematode total differed significantly between moisture conditions only at time zero (60 days after inoculation), when dry soils had more nematodes than flooded soils. Nematodes survived up to 12 months in dry and flooded soils, but populations decreased linearly with time. In total, 37355, 20204, 7229, 3341, and 536 nematodes were recovered from dry soil in months 1 to 5, respectively. Thereafter, less than 300 nematodes were collected, except for month 12, when 480 nematodes were recovered. In flooded soil, nematode total decreased from 19699 in the first month to 317 in the seventh month and less than 60 thereafter. Under moist soil conditions, nematode populations varied from 2822 in month 1 to 360 in month 6, and later, the number was zero. The results show that *M. graminicola* can survive for 1 year in dry and flooded soils.

Keywords: Rice - Nematode - Ecology.

S3-P18

Should I stay, or should I go? Triggers of nematode dispersal.

Birgit Gansfort (birgit.gansfort@uni-bielefeld.de), Bianca Kreuzinger-Janik, Walter Traunspurger, Christoph Ptatscheck

Bielefeld University, Bielefeld, Germany

The movement of organisms from one site to another (i.e. dispersal) appears as a simple process on scale of individuals but has ecological and evolutional consequences on population, community and metacommunity scale. Therefore, researchers have aimed to disentangle the determinants of dispersal in ecological systems. It has generally been concluded that dispersal is rather than simple and random, a complex and informed process which depends on environmental condition such as bottom-up and top-down control as well as population density. However, in this context the role of interspecific competition albeit it is an omnipresent major ecological interaction has largely been ignored. Nematodes are mostly suspected to be prone to passive and random dispersal, but active dispersal also occurs, and it may be asked if and how it relates to the biotic and abiotic environment. Therefore, here we tested the effect of food availability, predation as well as intra- and interspecific competition and the combinations thereof on the active dispersal of three free- living nematode species (*Caenorhabditis elegans, Acrobeloides nanus* and *Panagrolaimus thienemanni*) by measuring their dispersal rates in experiments with two-patched systems. It was clearly shown that the availability of food inhibited nematode dispersal and that flatworm as potential predator negatively affected nematode dispersal rates. The presence of a competitive species significantly altered dispersal rates. However, the effects varied regarding the three tested species: *C. elegans*and *P. thienemanni* showed up to 129% and 43% higher dispersal rates when another species was present compared to single-species treatments. In contrast, the dispersal of *A. nanus* was reduced by 92% under the presence of a competitor. Further, it was shown that the reactions of nematodes to heterospecifics were fundamentally different to the effect of intraspecific competition. Thus, our experimental work implies that insights in dispersal pattern based on single-species experiments cannot directly be scaled up to community systems. Therefore, we state that more experiments including interspecific interactions are needed if the objective is to predict natural dispersal pattern and that nematodes are useful model organisms for this purpose.

Keywords: Interspecific interaction - Emigration - Metacommunity - Context-dependent dispersal.

S3-P20

Phytoparasitic nematodes of strawberry and their management in Egypt.

Mostafa Hammam (mmahammam@yahoo.com), Wafaa El-Nagdi, Moawad Mohamed, Mahfouz Abd-Elgawad

Plant Pathology Department, National Research Centre, El-Behooth St., Dokki 12622, Giza, Egypt

Egypt is progressing steadily to higher global rank in the cultivation and exportation of strawberries backed by its fertile soils, Mediterranean climate, inexpensive labor, and geographic location. Egyptian strawberry has early fruiting and long harvest season, good quality, low production costs, and close export markets [1]. Therefore, the likely development of strawberry pests in parallel to more cultivated areas should be tracked to optimize their control measures. Plant-parasitic nematodes rank high among pests and diseases that can cause considerable yield losses of strawberry. The nematode losses vary according to nematode species, population density, and biotic and abiotic factors. Therefore, we have recently surveyed the two largest governorates of growing strawberries in Egypt. Population density, frequency of occurrence, and prominence values of the encountered nematode species were recorded. For root-knot nematodes (RKNs), *Meloidogyne* spp., these nematode parameters were higher in El-Beheira than in El-Qalioubia governorate. At El-Salam village, the average of RKN densities were 1645 per 200g soil and 270 females per g fibrous roots. Lesion nematodes were second in order of economically important nematodes on strawberry plants. They were detected from two of three counties at El-Qalioubia governorate only at population densities of 80 and 130 nematodes per 200g soil and 5g roots, respectively. Integrated nematode management including adequate and safe fumigation as well as other agricultural practices can offer sustainable cultivation of strawberry [2]. For strawberry nurseries in Egypt, there is an urgent need to produce enough national certified seedlings. This research is funded in part by Science and Technology Development Fund (EG-US project ID 172).

Keywords: Nematode management - Sampling - Strawberry - Survey - Yield loss.

References:

- Abd-Elgawad, 2019. Bull. NRC 43:7 https://doi.org/10.1186/s42269-019-0049-2.
- Hammam et al., 2019. Egypt J Agronematol. 18(1): 1-17.

S3-P21

Linking barcodes, images and metadata to construct nematode reference libraries in BOLD, the Barcode of Life Database.

Rebecca Higgins (rhiggins3@unl.edu), Blanche Butera, Timothy Harris, Peter Mullin, Kirsten Powers, Thomas Powers

Department of Plant Pathology, University of Nebraska, Lincoln, NE, United States

Historically nematodes have been described and diagnosed using morphological characters. Typically voucher material in the form of fixed specimens on slides and bulk specimens in formalin have been deposited in curated museums. Now nucleotide characters and image vouchers are common, and often required, components of nematode species descriptions. Ironically, there are relatively few database options that serve as a sequence and image repository. BOLD, The Barcode of Life Database is one such repository that functions as a primary data storage and analytics platform for DNA barcode data [1]. Currently (Jan. 15, 2020) the nematode database includes 22,894 specimens with sequences, representing 2,624 species. We have been populating BOLD reference libraries with nematode specimens from two projects with different approaches to adding detailed nematode content. The first project is taxonomically focused on the suborder, Criconematina, with efforts to collect all taxa on the North American continent [2]. The second project has a community focus with efforts to database all nematode taxa from agricultural and native forest sites near and in Volcanoes National Park, Rwanda [3]. BOLD reference libraries allows us, in one database, to readily deposit specimen information and all associated metadata. It is a repository for image vouchers, morphometrics, and nucleotide sequences. Image and text components of habitat, host and collection information are linked to each specimen. GPS coordinates of collection sites can be mapped and displayed at multiple geographic scales. The “workbench”, BOLD’s analytics tool, allows an investigator to compare and generate sequence-based metrics. Specimen morphology can be compared by selecting images of specific regions to develop an array of heads, tails, or entire bodies. BOLD uses a common genetic marker, COI, as a metric to assess animal biodiversity. Our goal in populating BOLD is to ensure that verifiable specimen information is accurately presented for assessments of global nematode biodiversity.

Keywords: Biodiversity - COI.

References:

- [1] Ratnasingham and Hebert 2007. Mol. Ecol. Notes 7(3), pp.355-364.
- [2] Powers et al. 2016. Zootaxa. Mar 3; 4085(3):301-44
- [3] Butera et al. 2019. UNL Integrated Studies senior thesis.

S3-P22

Tracing patterns of glacial refugia with *Scottnema lindsayae* nematode.

Abigail Jackson[1] (ajackson260@gmail.com), Thomas Powers[2], Kristen Powers[2], Timothy Harris[2], Diana Wall[3], Byron Adams[4]

[1] *Biology Department, Brigham Young University, Provo, Utah, United States*; [2] *University of Nebraska, Lincoln, Lincoln, NE, United States*; [3] *Colorado State University, Fort Collins, CO, United States*; [4] *Biology Department, Brigham Young University, Provo, UT, United States*

As part of a Long-Term Ecological Research (LTER) site, the McMurdo Dry Valleys (MDV) of Antarctica boast thirty plus years of multidisciplinary data from an infamously harsh environment that fosters neither high terrestrial biodiversity nor species recruitment. Freezing temperatures and saline soils limit terrestrial life to simple, dispersed microbial communities and microscopic soil invertebrates providing a unique opportunity to study nematodes in a naturally simplified environment. Taking advantage of these prerequisites, we aimed to test phylogeographic hypotheses regarding dispersal patterns of terrestrial free-living nematodes after large-scale environmental disruptions like ice-sheet dynamics, glacial advance and recession, and climate-driven changes to the soil environment. Nematodes in this system are sensitive to slight changes in environmental conditions and require refugia during periods of environmental disruption. Refugia are habitable, high elevation soil ecosystems that avoided inundation by expanding glaciers and have continually experienced periodic wetting, reducing inhospitable saline conditions of the MDV. We tested putative refugia on a fine-scale utilizing the mitochondrial COI gene of the most ubiquitous terrestrial animal in Antarctica – *Scottnema lindsayae* – by determining phylogeographic patterns shaped by historical glacial cycles and subsequent dispersal. We sequenced 250 bp COI haplotypes from 24 sites across eight valley systems in the MDV. These sites represent high and low elevations corresponding to putative refugia and recolonized glacial forefields respectively. We analyzed phylogeographic patterns of haplotype diversity in the context of hypothesis testing consistent with persistent refugia. We predict that putative refugia populations will have signatures of high genetic diversity, while subsequent glacial retreats allowing range expansion will generate patterns of decreasing variation from refugia to recolonized glacial forefields. Utilizing phylogeographic approaches on the most ubiquitous microfauna species, *S. lindsayae*, refugia hypothesis testing can be applied on a fine, universally applicable scale. This study demonstrates phylogeographic patterns of dispersal from refugia where terrestrial biota survived periods of extreme climate-driven environmental changes. These results may be used to inform predictions about the community's response to future climate changes and the process of speciation in the MDV.

Keywords: Antarctica - Phylogeography - Refugia - Glaciations - Speciation.

S3-P23

Morphological and molecular identification of species of *Meloidogyne* and distribution in pomegranate orchards of Iran.

Nafiseh Katooli[1] (n.katooli@gmail.com), Esmat Mahdikhani Moghadam[2], Reza Aghnoum[3]

[1] *Plant Protection Department, PhD student in plant protection. Faculty of Agricultural Sciences Department of, Mashhad, Iran*; [2] *Plant Protection Department, Professor in Department of plant protection, College of Agriculture, Ferdowsi Un, Mashhad, Iran*; [3] *Seed and Plant Improvement Research Department, Khorasan Razavi Agricultural and Natural Resources Research and Education Center, AREEO,Mashhad, Iran*

Pomegranate is one of the most popular fruit trees and one of the most important export products in Iran. Root-knot nematodes (*Meloidogyne* spp.) are the most damaging plant-parasitic nematodes of pomegranate, which cause root galls and disrupt the absorption and transfer of water and food in the plant and eventually decrease the growth of pomegranate fruit. To investigate the infection of pomegranate orchards to *Meloidogyne* species, 195 soil and root samples were collected from major areas of pomegranate cultivation from Razavi, Northern and Southern Khorasan provinces, during 2015-2016. The highest area of pomegranate cultivation is located in Torbat Heydarieh, Bardaskan, Kashmar, Khalil Abad, Bajestan, and Ferdows cities. After extraction of nematodes from the root and surrounding soil, species identification was carried out according to the molecular method by sequences of D2a-D3b expansion domains of 28S rDNA, sequence characterized amplified regions (SCAR). The morphological diagnostic was conducted on the perineal pattern of females. Several species of *Meloidogyne* were identified and showed band399 bp in *M. incognita* by Inc-14 primer, 670 bp in *M. javanica* by Jav primer, and 420 bp in *M. arenaria* by ar primer. *M. cruciani* showed 820 bp band by D2a-D3b primer but the sequence was not distinct from other species and use perineal pattern morphology. Subcuticular punctuation around the anus on lateral posterior sides has characterized the perineal pattern in *M. cruciani* , *M. cruciani* was reported of cucumber, tomato, pumpkin and pistachio in Iran, lately. In this study, the above species was isolated and identified from the roots of pomegranate trees for the first time in Iran. Infection of root-knot nematode pomegranate was observed in almost cities but the intensity of infection was very variable. The highest percentage of infection on root-knot nematodes pomegranate orchards was observed in cities of Bardaskan (Anabad section of Fatemieh village) with 57.5% and Bajestan (Chah Paliz village) with 32%. *M. incognita* has the highest distribution in pomegranate orchards. The highest percentage of infected orchards was respective estimates in provinces of Razavi Khorasan: in Bardaskan 19.93%, Bajestan 12.3%, Khalil Abad 6.9%, Kashmar 4.3%, Torbat Heidariyeh 3.5%, South Khorasan in Ferdows 5.4% and Nehbandan 1.3%, and North Khorasan in Maneh 1.1% and Jajarm 0.8% of cities.

Keywords: Pomegranate - Root-knot nematode - SCAR Primers - D2a-D3b primer.

References:

- Zijlstra et al. 2000..Nematology, 2(8): 847-853.

S3-P24

Glyphosate-tolerant and conventional soybean cultivars and the plant-parasitic nematodes associated with its rhizosphere.

Akhona Mbatyoti[1] (24088978@nwu.ac.za), Mieke Daneel[2], Antoinette Swart[3], Mariette Marais[3], Dirk De Waele[1], Hendrika Fourie[1], Rinus Knoetze[4]

[1] *Integrated Pest Management, North-West University, Potchefstroom, South Africa*; [2] *Nematology, Agricultural Research Council-Tropical & Subtropical Crops, Mbombela, South Africa*; [3] *Nematology, Agricultural Research Council-Plant Health & Protection, Queenswood, South Africa*; [4] *Plant Protection, Agricultural Research Council, South Africa, Stellenbosch, South Africa*

Information about the non-target effects of glyphosate, a broad-spectrum systemic herbicide, on terrestrial nematode assemblages is limited and not well documented for South Africa. Therefore, the abundance and diversity of plant-parasitic nematodes in roots and rhizosphere soil of commercial glyphosate-tolerant (genetically modified; GM) and conventional (non-glyphosate-tolerant) soybean cultivars from cultivated fields were obtained for the 2011/2012 and 2012/2013 growing seasons. Grass and soil from adjacent natural vegetation were sampled, representing the reference system. Thirty plant-parasitic nematode species, belonging to 13 genera, were identified from the rhizospheres (roots and soil) of soybean cultivars and natural vegetation. *Meloidogyne* (for glyphosate-tolerant and conventional soybean), followed by *Helicotylenchus* (for glyphosate-tolerant soybean) and *Scutellonema* (for conventional soybean) were the predominant genera present in soil samples. Seven species, *viz*. *Pratylenchus flakkensis, Pratylenchus scribneri, Pratylenchus vulnus, Rotylenchus brevicaudatus, Telotylenchus avaricus, Tylenchorhynchus brevicaudatus* and *Quinisulcius capitatus* represent first reports for South African soybean fields. This study suggested that glyphosate had no adverse effects on plant-parasitic nematode assemblages although the impact of each ecosystem on these organisms could not be defined.

S3-P25

Chemotactic response and motility of mollusc parasitic nematode *Phasmarhabditis papillosa* toward mucus from different slug species.

Ziga Laznik[1] (ziga.laznik@bf.uni-lj.si), Stanislav Trdan[1], Ivana Majić[2]

[1] *Agronomy, University of Ljubljana, Biotehnical faculty, Ljubljana, Slovenia*; [2] *Section of Entomology and Nematology, University of Osijek, Faculty of Agrobiotechnical Sciences Osijek, Osijek, Croatia*

Phasmarhabditis papillosa is a promising biocontrol agent of gastropods in crops. To enhance the efficacy of the slug parasitic nematodes a better understanding of the chemical signaling between the host and parasite is needed. The objective of this research was to test the stimulus-response behavior of the slug parasitic nematode *P. papillosa* towards mucus of the following mollusc species: *Arion vulgaris, Limax maximus, Deroceras reticulatum,* and *Helix pomatia*. The nematode chemotactic response was observed as their directed motility toward mucus of the tested molluscs at two temperature regimes under laboratory conditions. The motility of *P. papillosa* was strongly expressed at a higher temperature (20 °C) than at a lower one (15 °C). When comparing mucus of different mollusc species, none of the tested molluscs significantly affected the directed motility of *P. papillosa* at a lower temperature (15 °C). At a higher temperature (20 °C), the mucus of *L. maximus* and *H. pomatia* were more attractive for the nematodes compared to the mucus of *A. vulgaris* or *D. reticulatum*. Our results indicate that *P. papillosa* distinguishes among different molluscs species by exhibiting chemotactic response to a signal emanating from molluscs mucus.

Keywords: *Phasmarhabditis papillosa* - Chemotactic response - Slugs - Snails.

S3-P26

Potato Cyst Nematode Diversity and Adaptation in the Tropics: A case of Kenyan cropping system.

Harrison Mburu[1] (hmburu@cgiar.org), Laura Cortada[1], Solveig Haukeland[2], Danny Coyne[1], Joel Bargul[3], Varypatakis, K Cellular and Molecular Science Group[4], Jamie Orr[5], Vivian Blok[5], John Jones[5]

[1] *Nematology, International Institute of Tropical Agriculture (IITA), Nairobi, Nairobi, Kenya*; [2] *Nematology, International Centre for Insect Physiology and Ecology (ICIPE), Nairobi, Nairobi, Kenya*; [3] *Biochemistry, International Centre for Insect Physiology and Ecology (ICIPE), Nairobi, Nairobi, Kenya*; [4] *James Hutton institute, Dundee, United Kingdom*; [5] *Cellular and Molecular Science Group, James Hutton institute, Dundee, United Kingdom*

Potato cyst nematodes (PCN) are quarantine pests known to cause major yield and economic losses. These invasive pests were recently detected in Kenya and epidemiologic studies have shown that the pest is now widely distributed across all the major potato growing regions in the country and presents high incidence levels. The introductory events that led to the arrival of PCN to Kenya remain unknown. Factors that might have favourably influenced the spread of PCN in Kenya are: warm tropical climate with absence of prolonged frosting periods, year-round production of potato with minimum two cropping seasons per year, limited rotation practices, small farm sizes, replanting of PCN-infested farm-saved seed, and farmers' preference of PCN-susceptible potato cultivars . Molecular tests and bioassays were conducted to further characterize two different PCN populations from Kenya which originated from two different agro-ecological zones. Hatching bioassays indicated that the two *G. rostochiensis* populations studied do not undergo a diapause stage, being able to indistinctively hatch ($p > 0.05$) upon stimulation with potato root exudates after storage at 4˚C, 10˚C, 15 ˚C, 18 ˚C, 20 ˚C and 25 ˚C; the Kenyan isolates also presented two distinct optimal hatching temperature regimes. Barcoding analyses to understand the pest diversity confirms the presence of both *G. pallida* and *G. rostochiensis* and suggests the presence of two distinct genotypes of *G. rostochiensis* in the country that could correlate with to different pathotypes of this species. Furthermore, our analyses confirms the presence of other *Globodera spp.* and *Heterodera* species. These results indicate that the local populations of *G. rostochiensis* in Kenya may have adapted their biology to the local agronomic and climatic conditions; our findings will help researchers to have a better understanding of the ecological behavior of *G. rostochiensis* in Kenya, in order to develop effective management strategies suitable within local cropping systems

Keywords: *Globodera pallida* - *Globodera rostochiensis* - Kenya - Agro-ecological zones - Diapause.

References:

- Mwangi et al., 2015. New Dis. Rep. 31: 18-18.
- Mburu et al., 2018. Plant Dis. 102(8): 1671-1671
- Kaczmareck et al., 2019. Plant Pathol. 68(5): 962-976

S3-P27

In vitro and *in vivo* evaluation of fungal strains with biocontrol characteristics for their effects on the motility and reproduction of *Meloidogyne incognita*.

Marné Pretorius (marne0911pretorius@gmail.com), Gerhard Engelbrecht, Gerhard Du Preez, Hendrika Fourie

Unit for Environmental Sciences and Management, Integrated Pest Management, North-West University, Potchefstroom, South Africa

Root-knot nematode species pose a global threat to grain crop production since they cause major yield losses. Since classical nematicides are continually withdrawn from markets, safer nematode management alternatives are gaining popularity. This includes the use of biological agents, *viz*. fungi and bacteria in particular, for the management of major nematode pests such as *Meloidogyne.* During this study five fungal strains, namely four *Trichoderma* isolates and *Clonostachys rosea*, supplied by Plant Health Products (Pty) Ltd (now trading as Andermatt Group AG), were tested for their adverse effects on *M. incognita*. *In vitro* trials were conducted to determine the effects of the fungal isolates on J2 motility, while *in vivo* trials investigated their effects on female reproduction. The first set of *in vitro* studies, during which the fungal strains were propagated on potato dextrose agar, resulted in non-optimal growth and spore production. Follow-up *in vitro* trials using a bran-soya flour mixture as growth medium for the fungal strains resulted in the highest concentrations of 109 spores/ml being the most effective (80-100%) in reducing J2 motility of all strains. *In vivo* glasshouse trials, conducted for four of the five strains showed that *T. harzianum, T. asperellum, T. virens* and *C. rosea* (applied 108 and 107 spores/ml) as either seed or direct treatments, was able to reduce *M. incognita* egg and J2 densities 48 days after planting and inoculation between 48% - 80%; *C. rosea* had the lowest and *T. virens* had the highest reduction respectively. Data for *T. asperellum* KD was inconclusive and not referred to here. Follow-up glasshouse trials, currently in progress, include a higher concentration of 109 spores/ml as well as two products (*Trichoderma* and *Clonostachys* strains) that were commercialised recently and after this study began. Results of these trials will be presented.

S3-P28

Nematode biodiversity as a soil health indicator in agroforestry ecosystems.

Josipa Puškarić (josipa.puskaric@fazos.hr), Vladimir Ivezić, Brigita Popović, Mirjana Brmež

Faculty of Agrobiotechnical Sciences, Osijek, Croatia

Analyses of the nematode community structure can serve as useful indicators of the agricultural soil health and as indicators of the agroecosystem sustainability. Agroforestry, a combination of woody species and crops, is being offered as one of the solutions for improving soil health and its biodiversity. This combination reduces the negative impacts of climate on agricultural production and creates a new microclimate within the plantations. In this study, the biodiversity of nematodes was compared under 3 different agroecosytems: 1) agricultural crop (C), 2) walnut orchard (W), and 3) consociation of agricultural crop and walnut orchard (C+W); at two sites in Croatia, Dakovo and Ivankovo. The agricultural crop was wheat in 2017/2018, followed by rape seed (green fertilization) in 2018, and buckwheat in 2019. Samples were taken on 11 occasions during that period, in 4 repetitions. The numbers of genera differ statistically significantly under the different treatments in favor of consociation in both sites. At the Đakovo site, statistically lower numbers of genera were found in C treatment (14.77) than in C+W (17.66) and W (16.61). At the Ivankovo site, there was also a statistically lower number of genera in treatment C (14.48) than in C+W (16.91) and W (16.30). Statistically significant differences were found in the percentage of all trophic groups except the predators in the Dakovo site. The highest percentage of bacterivores was found in treatment C (63.3) compared to C+W (51.67) and W (37.15). A higher percentage of fungivores was found in treatment W (29.77) than C (21.41) and C+W (21.11). The percentage of plant parasitic nematodes was higher in treatment W (29.28) than C+W (20.11) and C (10.53). The percentage of omnivores was the highest in treatment C+W (6.23) and W (6.16) compared to C (3.81). Statistically significant differences were found in a percentage of bacterivores and plant parasitic nematodes in the Ivankovo site. The highest percentage of bacterivores in Ivankovo were found in treatment W (51.89) compared to C+W (44.83) and C (34.43). The percentage of plant parasitic nematodes was higher in treatment C (30.6) than C+W (19.41) and W (15.19). Regardless the differences in percentages of trophic groups in the study sites, the results suggest that consociation of woody species and agricultural crops increases soil biodiversity and thus improves soil health, which is a basic prerequisite for succesful agricultural production.

Keywords: Nematode biodiversity - Agroforestry ecosystems - Soil health - Nematode genera - Trophic groups.

S3-P29

Root-Knot Nematode Species Associated with Horticultural Crops in the Island of Azores, Portugal.

Leidy Rusinque[1] (leidy.rusinque@iniav.pt), Filomena Nobrega[2], Laura Cordeiro[3], Arlindo Lima[4, 5], Samuel Andrade[6], Maria Lurdes Inácio[1, 7]

[1] *Laboratory of Nematology, Instituto Nacional de Investigação Agraria e Veterinária, INIAV, Oeiras, Portugal*; [2] *Laboratory of Biochemistry, Instituto Nacional de Investigação Agraria e Veterinária, INIAV, Oeiras, Portugal*; [3] *Direção Regional da Agricultura, DRAG Azores, Angra do heroismo, Portugal*; [4] *LEAF – Linking Environment Agriculture and Food, Instituto Superior de Agronomia, Lisbon, Portugal*; [5] *LPVVA – Laboratório de Patologia Vegetal "Veríssimo de Almeida", Instituto Superior de Agronomia, Lisbon, Portugal*; [6] *LPVVA – Laboratório de Patologia Vegetal "Veríssimo de Almeida", Instituto Superior de Agronomia, Lisboa, Portugal*; [7] *GREEN-IT Bioresources for Sustainability, Universidade Nova de Lisboa (ITQN NOVA), Oeiras, Portugal*

Agriculture is one of the most important economic sectors and a significant component of the island of Azores, responsible for a 46% of the regional economy. According to the latest Land Occupation Report, approximately half of the Azores territory is occupied by agriculture (48.8%), followed with a 42.6% by forestry and natural and semi-natural vegetation. Plant-parasitic nematodes (PPN) represent a significant constraint to agricultural production, as they cause serious losses in quantity and quality worldwide. Among the PPN, root-knot nematodes (RKN), *Meloidogyne* spp., are one of the longest known plant parasitic nematodes and most devastating pests of economically important crops. *Meloidogyne* spp. have an extensive host range that includes nearly every horticultural, fruit and ornamental crop comprising more than 100 species. In Portugal, many species have been reported: *Meloidogyne arenaria* (Neal, 1889) Chitwood, 1949; *M. chitwoodi* Golden et al., 1980; *M. hapla* Chitwood, 1949; *M. hispanica* Hirschman, 1986; *M. incognita* (Kofoid and White, 1919) Chitwood, 1949; *M. javanica* (Trub, 1885) Chitwood, 1949; *M. lusitanica* Abrantes and Santos 1991; *M. luci* Carneiro et al., 2014; *M. enterolobii* Yang et al., 1983 and *M. naasi* Franklin, 1965. Due to the risk various *Meloidogyne* species pose to agricultural production, a national field survey programme that include the islands of Azores and Madeira was established in 2019, and led to the detection of *M. luci* associated with potato in the island of Pico-Azores. Although this programme is in place, only the species included in the A2 and Alert EPPO lists are surveyed in the samples (*M. chitwoodi*, *M. fallax* Karsen, 1996, *M. enterolobii* and *M. luci*). Currently, in the island of Azores, there is a lack of detailed information about the presence of *Meloidogyne* species. So, the aim of the present study was to determine the frequency of occurrence of the RKN and identify biochemically the species present in the fields. A total of 80 samples, comprising 23 species of plants from 13 localities in 4 municipalities of the island were collected. From the 80 samples analysed, *Meloidogyne* spp. was detected in 60 (75%) of them, showing a high prevalence of this plant-parasitic nematode in the Island of São Miguel. The most prevalent esterase phenotype detected was *M. incognita* I2 followed by *M. arenaria* and *M. javanica*. Acknowledgements: PhD fellowship 2020.05541.BD and project PTDC/ASP PLA/31946/2017 (KnowLuci).

Keywords: Identification - EST-Phenotype - *Meloidogyne.*

References:

- Massot, A. 2015. Available online: https://www.europarl.europa.eu/RegData/etudes/ STUD/2015/567667/IPOL_ STU(2015)567667_PT.pdf (accessed on 17 November 2021).
- Jones, J.T.; Haegeman, A.; Danchin, E.G.J.; Gaur, H.S.; Helder, J.; Jones, M.G.K.; Kikuchi, T.; Manzanilla-López, R.; Palomares-Rius, J.E.; Wesemael, W.M.L. 2013. Mol. Plant. Pathol. 14: 946–961.
- Pais, C.S.; de Abrantes, I.M.O.; Fernandes, M.F.M.; de Santos, M.S.N.A. T. 1986. Ciência Biol. Ecol. Syst. 6: 19–34.
- Standard Protocol PM 7/119 (1); Nematode Extraction; EPPO Bulletin 43:471-95. EPPO: Paris, France, 2013.
- Esbenshade, P.R.; Triantaphyllou, A.C. 1985. J. Nematol. 17: 6–20.

S3-P30

Assessment of plant-parasitic nematodes in Portuguese rice agro-systems: preliminary findings.

Leidy Rusinque[1, 2] (leidy.rusinque@iniav.pt), Carla Maleita[2, 3], Filomena Nóbrega[4], Maria Lurdes Inácio[1, 5]

[1] *Laboratory of Nematology, Instituto Nacional de Investigação Agaria e Veterinária, INIAV, Oeiras, Portugal*; [2] *Centre for Functional Ecology - Science for People & the Planet, University of Coimbra, Coimbra, Portugal*; [3] *Chemical Process Engineering and Forest Products Research Centre, University of Coimbra, Coimbra, Portugal*; [4] *Laboratory of Biochemistry, Instituto Nacional de Investigação Agaria e Veterinária, INIAV, Oeiras, Portugal*; [5] *GREEN-IT Bioresources for Sustainability, ITQB NOVA, Oeiras, Portugal*

Rice (*Oryza sativa* L.) is the primary source of food for more than half of the population. Portugal is the largest European consumer and the 4th largest EU producer. Plant-parasitic nematodes (PPN) contribute to severe losses in production worldwide. Among the PPN, root-knot nematodes (RKN, *Meloidogyne* spp.), and particularly *M. graminicola* (*Mg*), are the most serious pests of tropical rice production. Climate changes and the trade activity are promoting the spread of pests northwards, which means that temperate rice agro-systems can also be seriously affected. Moreover, *Meloidogyne graminicola* was reported in Italy in 2016 and included in the European and Mediterranean Plant Protection Organization (EPPO) Alert List. Due to the lack of detailed information about the presence of *Meloidogyne* sp. or any other nematodes of economic importance in portuguese rice fields, this study aims to assess the presence/prevalence of *Mg* and other PPN, including the quarantine aerial rice white-tip nematode *Aphelenchoides besseyi*. Soil, roots and leaves samples were collected from the basins of rivers Mondego and Tejo. Nematodes were extracted using the floating/sieving/centrifugal flotation method and Baermann funnel technique, respectively, and identified at genus level. Roots were observed to detect the presence of galls. Identification is carried out using morphological, biochemical and/or molecular approaches. So far, 50 soil and 50 root samples (rice and weeds) were analysed. Preliminary results from soil samples show that the most predominant nematodes are free-living. Bacteria-feeders were found in 86% of the samples follow by omnivores with a 36%. Predators, PPN and fungal feeders were present in a reduced number of samples, 18%, 24%, 14%, respectively. Baterial-feeders were found in high numbers, around 50-150 nematodes/400 mL soil, follow by omnivores, 5–10 nematodes/400mL. The remaining groups presented very low numbers. No nematodes were found in leave samples. The presence of PPN was only detected in samples from the basins of river Mondego and include nematodes of the genera *Helicotylenchus*, *Aphelenchus*, *Telotylenchus*and *Aphelenchoides,* which are being identified to species level using molecular methods. So far, the presence of *Mg* associated with rice has not been detected in Portugal. However, surveys will continue in the same regions and basins of river sado. Acknowledgements: PhD Fellowship 2020.05541.BD and Projects UIDB/04004/2020, UIDB/00102/2020.

Keywords: Survey - Identification - *Meloidogyne* sp - Rice white-tip nematode.

References:

- Fanelli, E. et al., 2017. European Journal of Plant Pathology 149:467-476.
- Jones, J.T. et al., 2013. Molecular Plant Pathology 14:946-961.
- Mantelin, S., et al., 2017. Molecular Plant Pathology 18(1): 3-15.
- Petitot, A.S., et al., 2017. Annals of Botany 119: 885-899.
- Zhant, L.P., et al., 2018. Journal Integ. Agriculture 17:621-630.

S3-P31

Mitochondrial DNA-based identification of *Meloidogyne* spp. from pineapple roots and cutivated soils in Kenya.

David Kihoro Sirengo[1] (davidkihoro9@gmail.com), Emmanuel Olajide[1], Solveig Haukeland[2], Oliver Chitambo[3], Agnes Kiriga[2], Marjolein Couvreur[1], Laura Cortada[4], Wim Bert[1]

[1] *Nematology Research Unit, Ghent University, Ghent, Belgium*; [2] *ICIPE, Nairobi, Kenya*; [3] *INRES Molecular Phytomedicine, Universität Bonn, Bonn, Germany*; [4] *IITA, Nairobi, Kenya*

The perennial nature of pineapple and continuous monocropping favours the build-up of pests and diseases, including-plant parasitic nematodes. In Kenya, knowledge on the diversity of root-knot nematode (RKN) species infecting pineapple is lacking. The precise identification of the RKN species present is necessary to develop sustainable nematode management strategies. However, tropical RKN, especially the *M. incognita* group (MIG), are very difficult to identify due to their phenotypic plasticity and the presence of closely reticulate-related parthenogenetic lineages. To characterize the species of *Meloidogyne* species infecting pineapple, a purposive sampling technique was applied at one of the largest pineapple plantations in Kenya. A total of 75 galled pineapple plants were collected from different pineapple field blocks. After the plants were uprooted, ~ 50 g of soil within the rhizosphere of the roots (top 15 cm) were collected and bulked. For root-knot nematode (RKNs) detection from the soil, blackjack (*Bidens pilosa*) and amaranth (*Amaranthus spinosus*) were used as bait plants. Females were hand-picked from galled roots and were identified using morphological and mitochondrial DNA-based identification (*Nad5* barcoding). All the females amplified yielded PCR products except one. The *Nad5* mtDNA sequence data indicated the presence of *Meloidogyne javanica*, *M. incognita,* and *M. enterolobii*. To our knowledge, this is the first record of *M. enterolobii* from pineapple soil and this species can constitute a potential threat to pineapple production in Kenya. *M. enterolobii* is known to have a wide host range, high reproduction rate and it is highly damaging on most cultivated crops. This study warrants consideration of several species during resistance screening activities and other management strategies for pineapples. Although this study is based on a limited number of samples, the remarkable different results obtained based on direct sampling, blackjack or amaranth as bait plant underscores the possibility to find undetected species with bait plants and cautions the interpretation of data based on one technique alone. We are currently collecting additional infected galled pineapple roots from pineapple farmer's in Kenya to understand the actual distribution of *Meloidogyne* spp. in pineapple farms in Kenya.

Keywords: Nad5 - Kenya - *M. enterolobii* - Pineapples - Root-knot nematodes.

References:

- Agu 2008. Plant Sciences Research. 1: 36-39.
- Bert et al., 2008. Molecular phylogenetics and evolution. 48: 728-744.
- Chitambo et al., 2016. Plant Disease. 100: 1954.
- Janssen et al., 2016. Scientific Reports. 6: 22591

S3-P32

Multiple-nutrient limitation of soil free-living nematodes in ferralsols from natural grasslands in Madagascar.

Jean Trap[1] (jean.trap@ird.fr), Manoa Raminoarison[2], Blanchart Eric[3], Claude Plassard[4], Bernard Laetitia[1]

[1] *UMR Eco&Sols, Institut de Recherche pour le Développement, Montpellier, FRANCE*; [2] *Laboratoire des Radio-Isotopes, Antananarivo, Madagascar*; [3] *Institut de Recherche pour le Développement, Montpellier, France*; [4] *UMR Eco&Sols, INRAE, Montpellier, France*

Ferralsols from the Highlands of Madagascar are highly weathered nutrient-depleted soils exhibiting high acidity and low organic matter content. In these tropical soils, the microbial activity is assumed to be strongly limited by carbon and phosphorus availability. Free-living nematodes grazing on microbial cells are thus likely to be limited by these same nutrients but there is no evidence at this time. We tested this hypothesis using a nutrient-omission trial in microcosms. In controlled conditions, we manipulated the presence/absence of 8 chemical elements (C, N, P, S, Ca, Mg, K, Si) and a mixture of micro-nutrients (B, Mn, Cu, Co, Na, Mo) in a fresh soil collected from a natural grassland in Madagascar. Microcosms with all elements absent and with all elements present were used as negative and positive controls, respectively (11 treatments with 3 replicates). After 28 days of incubation, soil nematodes were extracted and counted. We also measured nematode biomass using the length and width of 100 individuals for each sample. In parallel, soil microbial parameters were assessed (respiration, molecular biomass, microbial biomass C and P, microbial community structure by DNA pyrosequencing, hydrolytic enzymatic activities). The abundance of bacterial- and fungal-feeding nematodes was strongly co-limited by C, P, Mg and micro-nutrients. Conversely to our expectations, there was no dominant limitation of C and P. We also observed a N, S and Ca co-limitation, but less pronounced. We did not find K limitation. Nematode biomass exhibited the same pattern. We did not find any plant-feeding nematodes while predators-omnivorous nematodes only occurred in two samples. Microbial respiration was strongly related to nematode abundance, suggesting that the abundance of free-living nematodes could be indicators of microbial activity. To conclude, in the attempt to intensify soil ecological functions achieved by nematodes within the agro-ecological framework, the supply of at least C, P, Mg and micro-nutrients is needed to increase the abundance and activity of free-living nematodes. Providing N, Ca and S in these highly weathered soils, when possible, also appear beneficial. Thus, providing a combination of multiple materials (animal manure, compost, vermicompost, dolomite, ashes, etc.) with contrasting chemical compositions may be a suitable way to drive the function of free-living nematodes important for nutrient cycling in soil.

Keywords: Tropical soil - Nutrient limitation - Free-living nematodes - Microcosm - Madagascar.

S3-P33

Nematofauna analysis and ecotoxicological bioassay using *C. elegans* applied to soil toxicity assessment of three polluted sites.

Cécile Villenave[1] (cecile.villenave@elisol.fr), Marina Le Guédard[2], Hélène Cérémonie[1], Olivier Faure[3], Anne Jimenez[1], Manon Trambolho[1]

[1] *ELISOL environnement, Congénies, France*; [2] *LEB Aquitaine Transfert-ADERA, Villenave d'Ornon, France*; [3] *École des Mines de Saint-Étienne, Saint Etienne, France*

Rehabilitation of polluted sites aims to reduce or stabilize soil pollution. As a consequence, the soil rehabilitation process has to be evaluated to check its efficiency and the retentive soil toxicity. Nematofauna are an "in situ" bioindicator, characterizing the biological functioning of the soil by integrating many factors: pedo-climatic characteristics, anthropic situations (management practices, contamination: types, frequency, etc) [1]. Thanks to the great functional diversity of nematodes and their sensitivity to the conditions of their living environment, nematofauna give qualitative and quantitative information on direct or indirect disturbances present in the soil. A direct measurement of the soil toxicity on organisms can also be important information for determining the effects generated by contaminants on soil functioning. The "ex situ" ecotoxicological standardized test (NF ISO 10872 2010) measures the inhibition of the growth, fertility or reproduction of the nematode *Caenorhabditis elegans* in contaminated compared to a non-contaminated soil [2]. As part of the APPOLINE program (Gesipol from ADEME), both tools *in situ* (nematofauna) and *in vitro* (ecotoxicological bioassay) were used on three polluted sites located in France to evaluate: (1) a phytostabilization process carried out in a former gold mine (Aude, France)
(2) a phytostabilisation process in an old metallurgical crass-burner (Loire, France) (3) the impact of accidental PCB contamination on soil (Loire, France). The results of studies conducted on these 3 sites will be presented.

Keywords: Polluted soils - Soil nematofauna - Bioindicator - Ecotoxicological bioassay.

References:

- [1] Salamun et al., 2012. Ecotoxicology. 21, 2319-2330.
- [2] Le Guédard et al., 2017, ADEME. 187 p.

S3-P34

Evaluation of carbofuran metabolism in three different soil types and effect of the metabolites on nematode population

Deborah Olaore[1] (fabiyitoyinike@hotmail.com), Oluwatoyin Fabiyi[2, 3]

[1] *University of Ilorin Faculty of Agriculture, Ilorin, Nigeria*; [2] *Department of Crop Protection, University of Ilorin Faculty of Agriculture, Ilorin, Kwara, Nigeria*; [3] *Department of Crop Protection, University of Ilorin Faculty of Agriculture, Ilorin, Nigeria*

Synthetic nematicides play an important role in the control of plant parasitic nematodes. Their importance in the control of plant parasitic nematodes cannot be overemphasized. However, they are a threat to the environment due to their toxic nature. Some nematicides are degraded readily by certain bacteria in the soil while some may prove recalcitrant. The degradation of carbofuran in the soil is affected by microbial activity which ends in toxic or less toxic metabolites. The biodegradation process of carbofuran was evaluated and effects of the resulting metabolites on nematode population were accessed in sandy, loamy and silt soils. 0.5g each of the soil was serially diluted at 1:10, 1:100 and 1:1000 and spread on Starch Casein Agar, ISP2, Muellar Hinton Agar and Saboroud Dextrose Agar for *Actinomycetes*species and Nutrient Agar and Tryptone soya Agar for *Pseudomonas* species to grow. Molecular characterization was done on the carbofuran degrading bacteria isolated for proper identification. A biodegradation assay was set up while *Meloidogyne incognita* and carbofuran was incorporated into the soil to monitor degradation rate and population of the nematodes for 30 days. The result of the GCMS analysis revealed the presence of metabolites like 3-hydroxy-carbofuran, 3-keto-carbofuran and carbofuran-phenol which implies that there was significant ($p<0.05$) degradation and the population of *M. incognita* in soil was reduced by the metabolites, but not as much as the mother compound carbofuran. Metabolism of carbofuran could be induced by actinomycetes to reduce its toxicity in the environment.

Keywords: Nematicides - Carbofuran - Biodegradation - Actinomycetes - Pseudomonas.

References:

- Mishra et al., [2021]. Chemosphere. 279:
- Rana et al., [2019]. https://doi.org/10.1007/987-981-13-9117-0_11

S3-P35

Gone with the wind: The passive dispersal of nematodes

Christoph Ptatscheck (christoph.ptatscheck@uni-bielefeld.de), Birgit Gansfort, Walter Traunspurger

Animal Ecology, Bielefeld University, Bielefeld, Germany

Dispersal is a vital component of the life- history of nearly all organisms. The ability to disperse determines the distribution and abundance of a species and thus its community dynamic at different sites. While larger animals can cover distances on their own, small (<2 mm) organisms like nematodes are often passively dispersed, resulting in their more ubiquitous occurrence. Next to the transport with larger animals (e.g., flying insects, birds, or mammals) the wind may be an important vector for the overland transport of nematodes. However, concrete dispersal rates have hardly been examined so far. Here, I present the results of a one-year field experiment investigating the composition and dispersal rates of wind drifted nematodes. Aeroplancton was collected in a natural environment, surrounded by potential source habitats and on the roof a 10-storey building (35 m above the ground).

Among the taxa collected in this study, nematodes had the highest dispersal rates (up to >3000 individuals m−2 in 4 weeks) and represented >44% of aeroplankton. They had a higher dispersal potential near to source habitats and only 15 of the 27 collected species were trapped on the roof, dominated by *Chiloplectus andrassyi.* Contrary to the common assumption only living nematodes, and no propagules, were dispersed, mainly represented by individuals < 0.75 mm. Wind speed and humidity were significantly related to nematode dispersal rates. The results indicated that wind dispersal of nematodes which has often been underestimated enables a continues transport of flying worms over long distances.

Keywords: Aeroplakton - Wind dispersal - Dispersal rates.

References:

- Ptatscheck et al. 2018. The extent of wind-mediated dispersal of small metazoans, focusing nematodes. Scientific Reports. 8(1):6814.

S3-P36

Nematode community patterns along an elevational gradient in the Santa Catalina Sky Islands in Arizona

S. Patricia Stock[1, 2] (spstock@email.arizona.edu), Satyendra Pothula[1], Emilie Lefoulon[1], Stephen Pryor[1, 3]

[1] *School of Animal and Comparative Biomedical Sciences, University of Arizona, Tucson, AZ, USA*; [2] *College of Agriculture, California State University, Chico, CA, USA*; [3] *College of Agriculture, California State University, San Luis Obispo, CA, USA*

Elevation gradient of mountains is an excellent “natural experimental system” to predict species adaptation to changing environment and to test the distribution of resources available to plants and soil organisms [1]. Indeed, mountain slopes provide strong variation that can alter abundance and diversity of above and below ground species [2]. Moreover, vegetation, soil, and their associated organisms are naturally interlinked in mountains [3]. Soil organisms play crucial roles in ecosystem functioning such as organic matter turnover and nutrient cycling. Among them, nematodes occupy key roles interacting with a variety of organisms such as bacteria, fungi, plants, and other nematodes in the soil [4]. Several nematode indices have been developed to assess variations in soil ecosystems and as indicators of structure, function, and overall condition of soil ecosystems [5]. To date, very few studies have focused on nematode community patterns along elevation gradients. Thus, in this study, we assessed nematode species composition in the Santa Catalina mountains which are one of the Sky Islands in southeastern Arizona, USA. Soil samples were collected along an elevation gradient that ranged from 730 m to 2,766 m, and transition from Sonoran Desert scrub to subalpine fir forests near the summit of Mt. Lemmon. A multiplex metabarcoding strategy targeting two regions of 18S was used for nematode identification purposes. PCR products were sequenced on an Illumina Miseq platform. Amplicon sequence reads were processed using the DADA2 pipeline. Taxonomy of ASVs were assigned based on the Silva_111 database. Nematode ASVs were assigned to different trophic groups. Diversity indices were calculated and analyzed statistically using ANOVA and significance was tested by Fisher’s least significant difference test. Additionally, nematode communities were correlated with soil properties using the Envfit function of the Vegan package in R. Results from this study will be presented and discussed.

Keywords: Sky Islands - nematodes - diversty - elevation gradient - metabarcoding.

References:

1. [1] Körner, 2007 Trends in ecology & evolution, 22: 569-574.
2. [2] Alexander et al., 2015 Nature,525: 1849-1860.
3. [3] Hodkinson, 2005 Biological reviews, 80: 489-513.
4. [4] Hagedorn et al., 2019 Science, 36: 1119-1123.
5. [5] Yeates et al., 1993 Journal of Nematology, 25: 315-331.

S3-P37

Documentation and characterization of nematode biodiversity in Nilgiri forests of India for their functional role in soil health.

Kavitha Govindasamy (kavitha_nem@yahoo.com)

Department of Nematology, Tamil Nadu Agricultural University, Coimbatore, Tamil Nadu, India

Despite increasing attention given to biodiversity in recent years, relatively little attention is given to forest soil microorganisms. This study, supported by the Tamil Nadu Forest Department, involved ecological research on functional roles of unexplored beneficial organisms that are belowground that maintain health of forest soils. Soil ecosystems support a diversity of microbes where nematodes occupy a central position in detritus food webs that play significant roles in soil functioning. Soil samples collected during the survey in undisturbed forests of Kallar, Burliyar, Conoor, Ooty and Kotagiri of Nilgiris in India were analyzed. Bacterial feeders (*Rhabditis* sp, *Acrobeles* sp, *Diplogaster* sp.) and fungal feeders (*Aphelenchoides* sp., *Aphelenchus* sp. and *Filenchus* sp.) were found predominantly. These beneficial nematode specimens are documented and morphologically and molecularly characterized. Functional roles of the nematodes were studied by soil nutrient analysis. Bacterial feeding nematodes dominated in lower altitudes of Nilgiris which resulted in higher rates of decomposition and in turn increased the organic carbon content (1.91%). They enhanced the decomposition process and nutrient mineralization due to release of excess nitrogen assimilated in the bacteria. Fungal feeding nematodes were more abundant in higher altitudes with soil having acidic pH due to leaching, which favored the growth of fungi and actinomycetes. Nematode activity increased the available phosphorous (34.6kg/ha) with a narrow C:N ratio (12:1) in the forest soil. In contrast the population of the bio-indicators were much less in the agricultural and polluted soils, due to human intervention. Hence, the study has paved a way for defining long-term monitoring and conservation of these bio-resources and forecasting the change in the soil health status in the changing climatic scenario.

Keywords: Bacterial and Fungal feeding Nematode - Undisturbed forest soils - Soil Health status - Decomposition - Nutrient Mobilization.

References:

- Hanel et al., 2010. Helminthologia 47: 123-135.
- Yeates, G. W. 1971. Pedobiologia 11:173-179.

POSTERS

S5. Integrated nematode management

S5-P01

First report of *Meloidogyne arenaria* on calla (*Zantedeschia aethiopica*) in Serbia.

Jasmina Bačić[1] (jasmina.bacic@jasminabacic.rs), Jelena Kušić[2], Polona Strajnar[3], Barbara Gerič Stare[3], Saša Širca[3]

[1] *Plant Nematology Laboratory, Agricultural Extension Service Institute Tamiš, Pančevo, Serbia*; [2] *Institute of molecular genetics and genetic engineering, Belgrade, Serbia*; [3] *Plant Protection Department, Agricultural Institute of Slovenia, Ljubljana, Slovenia*

Meloidogyne arenaria is found worldwide in tropical and subtropical regions and it prefers warm habitats. In temperate regions *M. arenaria* is usually present in glasshouses attacking a large number of host plants, including ornamentals [1]. In Serbia, *M. arenaria* has been identified in glasshouses on tomato, cucumber and pepper [2]. In October 2017, highly damaged calla plants (*Zantedeschia aethiopica*, originated from Kotor, Montenegro) with yellowing and wilting symptoms were observed in a glasshouse in Crepaja (Vojvodina province) in northern Serbia. Galls on roots were not observed but numerous females were extracted from washed thickened portions of roots under a stereomicroscope. Morphological characteristics of the female's perineal patterns were analyzed on freshly isolated females and the sample was identified as *M. arenaria.*

For species identification confirmation young egg-laying females were used for isozyme phenotyping by esterase (EST) and malate dehydrogenase (MDH) activity and revealed the isozyme phenotypes of N1 for MDH and A2 for EST, characteristic for *M. arenaria* [3]. Species identification was further confirmed by molecular analyses using group-specific primers in the rDNA region to determine tropical RKNs and species-specific SCAR primers for *M. arenaria* species identification [4].

This is the first report of *M. arenaria* on calla in Serbia. During 2018 and 2019, serious damage was observed on calla in nearby location of Crepaja (Sefkerin) in northern Serbia. *Meloidogyne* sampled from this glasshouse was identified as *M. arenaria* by morphological and molecular analyses as well. This finding has great importance for the growers who propagate calla by bulbs, so they can take appropriate management steps in order to prevent further pest spread.

This work was performed in frame of the EUPHRESCO project "Global warming and distribution of root-knot nematode species of the tropical group (MeloTrop)" in the period of April 2017-March 2020 [5].

Keywords: Root-knot nematodes - Occurence - Identification - Ornamentals.

References:

- [1] Machado et al., 2013. Australasian Plant. Dis.Notes. 8: 131-132.
- [2] Jovičić & Grujičić, 1986. Plant Protection. Vol. 37 (1) No.175:31-40.
- [3] Strajnar et al., 2009. Russ. J. Nematol. 17:135.
- [4] Adam et al., 2007. Plant Pathology. 56:190-197.
- [5] Gerič Stare et al., 2018. Abstracts 70 th International Symposium on Crop Protection, Ghent, Belgium, 173.

S5-P02

Vertical distribution of nematodes in peanut-cotton cropping systems.

Lesley Schumacher[1] (lesleyschumacher@ufl.edu), Zane Grabau[1], David Wright[2], Sabina Budhathoki[1]

[1] *Entomology and Nematology, University of Florida, Gainesville, FL, United States*; [2] *North Florida Research and Education Center, University of Florida, Quincy, FL, United States*

Cotton (*Gossypium hirsutum*) and peanut (*Arachis hypogaea*) are important field crops grown in the southeastern United States. The plant-parasitic reniform nematode (*Rotylenchulus reniformis*) is a pathogen of cotton capable of reducing lint yields, so crop rotation is commonly used for its management [1]. One specific rotation system, sod-based rotation, uses two years of bahiagrass (*Paspalum notatum*) followed by one year each of peanut and cotton compared with a conventional crop rotation (peanut-cotton-cotton). Peanut and bahiagrass are poor hosts of reniform nematode. Agronomic benefits of a sod-based rotation like improved cotton root growth are well-known, but information about its effects on free-living, non-parasitic nematodes (fungivores, bacterivores, omnivores, predators) is limited [2]. These nematodes contribute to soil nutrient cycling and may be beneficial in crop production. Additionally, reniform nematode is present deep in the soil profile, but not much is known about free-living nematodes at deeper depths. By surveying a long-term experimental site in Quincy, FL, our study aimed to investigate both plant-parasitic and free-living nematodes at different soil depths in sod-based and conventional rotation with or without irrigation. Soil samples were collected to a depth of 120 cm before planting (March 2017, March 2018), after harvest (October 2017, October 2018), and in the winter (January 2018, January 2019) using a hydraulic probe. Free-living and reniform nematode abundances were analyzed in 30 cm-sections. No irrigation effects were observed in any of the sampling dates ($P>0.05$). There were significant crop by depth interactions ($P<0.05$) for free-living nematode abundances for all sampling dates, but only in harvest sampling dates for reniform nematode abundances. For these interactions, effects varied by sampling date for free-living nematode abundances. Yet, sod-based rotation reduced reniform nematode abundances at all depths compared with conventional rotation for post-harvest sampling dates. Overall, both free-living and plant-parasitic nematodes were present up to 120 cm deep in the soil profile and cropping system affected nematode abundances.

Keywords: Reniform nematode - Free-living nematodes - Crop rotation - Vertical distribution - Nematode management.

References:

- [1] Moore and Lawrence, 2012. Nematropica 42:227-236.
- [2] Wright et al., 2010. ASA-CSSA-SSSA International Annual Meetings.

S5-P03

Management of plant parasitic nematodes in California almond production with fluopyram (Velum™ One).

J. Alfonso Cabrera[1] (alfonso.cabrera@bayer.com), Helmut Fuersch[2]

[1] *CropScience, Bayer, Fresno, California, United States*; [2] *CropScience, Bayer, Monheim, NRW, Germany*

Almond production in California currently surpasses $5 billion per year. Plant parasitic nematodes can be a major concern in California almond orchards by reducing yields, decreasing tree vigor, and depressing overall orchard productivity. Among the most damaging nematodes is the ring nematode (*Mesocriconema xenoplax*). Currently, almond growers have only a few alternatives to manage these nematodes after planting. The goal of this investigation was to evaluate the performance of fluopyram (Velum™ One) as a tool to manage these important nematodes. Two multiyear trials were conducted in almond orchards at Fresno and Winton, California, to determine the suppressive activity of fluopyram on ring nematodes and its subsequent effect on almond yield. Additionally, 8 trials were conducted in commercial almond orchards throughout California in growing areas infested with plant parasitic nematodes to evaluate the impact of fluopyram on yield. Fluopyram was applied via chemigation after an initial soil pre-wet period and was subsequently followed with sufficient water to spread/move the product in the soil. Fluopyram was injected once or twice a year at 250 g ai/ha or 237 g ai/ha. Both multiyear trials showed suppressive activity of fluopyram on ring nematodes while increasing almond yield. The average of 8 commercial trials showed that fluopyram increased yield. In four of these trials where almond nut meat was evaluated fluopyram increased nut meat weight when compared to the untreated control. In conclusion, these field tests demonstrated the potential of fluopyram as a parasitic nematode management tool in California almond orchards. Additionally, in these trials fluopyram showed its potential to increase yield in orchards infested with plant parasitic nematodes. Further research is being conducted to determine the potential tree growth benefits following fluopyram applications in recently established young almond orchards.

Keywords: *Mesocriconema xenoplax* - Almond orchards - Yield - Fluopyram - Velum™ One.

S5-P04

Sorghum genotypes reaction to *Meloidogyne javanica.*

Angelica Calandrelli[1] (a.calandrelli@hotmail.com), Monique Thiara Rodrigues e Silva[1], Angelica Miamoto[1], Cláudia Regina Dias-Arieira[2]

[1] *Postgraduate Program in Agronomy, State University of Maringa, Maringá, Paraná, Brazil*; [2] *Postgraduate Program in Agronomy - Umuarama, State University of Maringa, Umuarama, Paraná, Brazil*

Over the past years, the area of sorghum cultivation has increased in Brazil, especially in the succession system with soybean. Since such a plant may be susceptible to nematodes of importance to soybean, such as *Meloidogyne javanica*, caution is needed when choosing genotypes for use in areas infested with the pathogen, to integrate efficient management systems. Thus, the aim of this study was to evaluate the reaction of six sorghum genotypes to *M. javanica*. The experiment was conducted in a greenhouse, in a completely randomized design, with eight replications. Each plot consisted of one plant, sown in a pot containing 1 L of the autoclaved substrate, in a proportion of 1:2 (soil: sand). The genotypes Nucover 100, Nugrain 320, Fox, Nugrain 430, Alvo, and NTX S100 were evaluated. Soy cv. M6410 IPRO was used as the susceptibility standard. At seven days after germination, the plants were inoculated with 2000 eggs + eventual second-stage juveniles (J2) of *M. javanica* and, 60 days after inoculation, they were evaluated for nematode reproduction. The genotypes presented average numbers of *M. javanica* per gram of root ranging from 4,100 to 7,900, while soybean presented an average of 67,700. The reproduction factor observed for soybean was 16.6, while the sorghum genotypes showed averages ranging from 0.2 to 0.5. It was concluded that all sorghum cultivars behaved as resistant to *M. javanica*, suggesting that the introduction of these cultivars can contribute positively to root-knot nematode management systems.

Keywords: Root-knot nematode - Sorghum bicolor - Management.

S5-P05

Effects of *Solanum linnaeanum* and *S. sisymbriifolium* on *Globodera pallida* hatching and mortality.

Laura Soraia Perpétuo[1, 2, 3] (soraiaperpetuo@gmail.com), Maria José Cunha[2], Isabel Luci Conceição[1], Maria Teresa Batista[3]

[1] *Centre for Functional Ecology-Science for People & the Planet (CFE), Department of Life Sciences, University of Coimbra, P-3000 456, Coimbra, Portugal*; [2] *Agronomic Sciences/Research Center CERNAS, Polytechnic Institute of Coimbra, High School of Agriculture Bencanta, 3045-601, Coimbra, Portugal*; [3] *CIEPQPF, Health Sciences Center, Faculty Pharmacy, University of Coimbra, Azinhaga de Santa Comba, 3000548, Coimbra, Portugal*

Potato cyst nematodes, *Globodera* spp., cause damage on potatoes in more than 60 countries. *Globodera pallida* is the most difficult species to control and its management is mainly achieved by cultural practices, crop rotation and resistant cultivars, combined with agrochemicals. However, several nematicides are being removed from the market due to their toxic effects on the environment and human health. New strategies for nematode management, such as less toxic and environmentally acceptable substitutes for commercial nematicides, are needed. This generates a significant market opportunity for the development of alternative and biorational products, namely the botanical nematicides, by screening naturally occurring compounds in plants. The Solanaceae are economically important and many species contain powerful alkaloids, some being highly toxic and known for their pesticidal effect. In this study, the effects of aqueous extracts (roots of plants with 1 and 2 months and immature fruits) and root exudates of *Solanum linnaeanum* and *S. sisymbriifolium* cv. Sis 6001, were evaluated based on *G. pallida* hatching and mortality. All extracts and root exudates affected nematode hatching. In all treatments the cumulative hatching effect (%), after 30 days, was significantly lower than in the control (potato root exudates – cv. Désirée). Considering the mortality, no effects were detected after 10 days of continuous exposure when compared to control (distilled water). This work demonstrate that these *Solanum* species can be used for the development of a more sustainable agriculture and a more ecological plant parasitic nematode management.

Keywords: *Globodera pallida* - Immature fruits - Roots - *Solanum linnaeanum* - *Solanum sisymbriifolium*.

S5-P07

Corteva nematicides' compatibility with soil applied biologicals for nematode, insect and disease management.

Jeanne De Waal[1] (jeanne.dewaal@corteva.com), Yannis Stamatas[2], T.C. Thoden[3], J.A. Wiles[4]

[1] *Corteva Agriscience, Western Cape, South Africa*; [2] *Corteva Agriscience, Halandri, Greece*; [3] *Corteva Agriscience, München, Germany*; [4] *Corteva Agriscience, Cambridge, England*

Today plant-parasitic nematodes remain a key threat to the sustainable production of many important crops, including fruiting vegetables and root crops. They rob producers of both total yield and the quality of their produce through their damage to the function of plant roots or by directly affecting tubers underground. This paper highlights the compatibility of Corteva nematicides (oxamyl, fluazaindolizine) with a broad range of soil-applied biological products that may be used by growers as part of integrated programs to suppress insect, nematode and diseases, and sustain a healthy, functional soil for crop production. Corteva nematicides have shown good compatibility with various commercially available soil biologicals under worst-case laboratory conditions. Due to their selectivity they can be considered compatible with numerous biological organisms that support soil health or suppress soil pests and diseases in cropping systems. In laboratory tests they showed good compatibility with various beneficial soil fungi. The radial growth of fungi on treated agar dishes was not inhibited by them on concentrations of 50 ppm while radial growth was inhibited by an SDHI product at 5 and 50 ppm. Products containing entomoparasitic nematodes are becoming increasingly available to growers and can selectively kill insect pests in the soil. In tests, various commercially available species of entomopathogenic nematodes were shown to be still infective to mealworm larvae (surrogate insect pests) even after 7 days of pre-exposure to both oxamyl and fluazaindolizine at up to 50 ppm. Overall the data presented show that Corteva nematicides are valuable tools for modern integrated nematode management, and are compatible with the increasing use of soil-applied biological products and the focus on sustaining soil health.

S5-P08

Castor bean cake extracts: nematicidal potential and chemical composition.

Claudia Dias-Arieira[1] (crdarieira@uem.br), Guilherme Tarini[1], Lais Fernanda Fontana[1], Beatriz Pereira Moreno[2], Maria Helena Sarragiotto[2], Beatriz Cervejeira Bolanho[1]

[1] *Center of Agrarian Sciences, Universidade Estadual de Maringa, Umuarama, Parana, Brazil*; [2] *Chemical engineering, Universidade Estadual de Maringa, Maringá, Parana, Brazil*

The pressing of castor bean seeds to obtain the oil results in a residue called castor cake, whose accumulation in the environment may be undesirable, due to the toxic compounds present in the seeds, including ricin, ricinin and ricinoleic acid [1]. However, such residue has potential for nematode control. Thus, the aim was to evaluate the nematicidal effect of the aqueous and ethanolic extracts of castor cake on *Meloidogyne javanica*, as well as performing their chemical characterization. Nematode control was evaluated in a greenhouse, with four treatments: aqueous extract, 60% ethanol extract, distilled water and abamectin. For this, soybean was sown in a furrow in the substrate (soil:sand 2:1, autoclaved) and inoculated with 2000 eggs+J2. Then, the furrow was closed, and the treatment was carried out by applying the extracts or the water (control) on the surface. Abamectin was applied by seed treatment. The extracts were prepared in a 1:10 (w:v) ratio, using distilled water or 60% ethanolic solution (99% pure ethanol). After 60 days, the total number of nematodes and nematodes g^{-1} root were evaluated. The extracts were evaluated by high performance liquid chromatography (HPLC) for the concentration of phenolic compounds, and by nuclear magnetic resonance (NMR) of 1H and 13C for the presence of other compounds. Both extracts promoted 43% reduction in the total number of nematodes, and reductions from 60% (aqueous) to 67% (ethanolic) in the number of nematodes g-1 root. The phenolic compounds present were hydroxybenzoic, chlorogenic, syringic, coumaric, ferrulic, trans-cinnamic, gallic and caffeic acids. NMR spectra allowed the identification of a mixture of sucrose and sinapic acid. This research proves the efficiency of castor cake extracts for nematode control and that they are rich in hydroxybenzoic and chlorogenic acid. It also shows the presence of compounds whose nematicidal action needs to be better elucidated.

Keywords: *Ricinus communis* - Phenolic compounds - *Meloidogyne javanica*.

References:

- [1] Yeboah et al., 2021. Food Sci. Technol. 41:399-413.

S5-P09

Design and assessment of innovative Mediterranean vegetable cropping systems to manage root-knot nematodes.

Caroline Djian-caporalino[1] (caroline.caporalino@inra.fr), Mireille Navarrete[2], Ariane Fazari[1], Nathalie Marteu[1], Marc Baily-Bechet[1], Arnaud Dufils[2], Marc Tchamitchian[2], Amélie Lefèvre[3], Laure Pares[3], Thierry Mateille[4], Johannes Tavoillot[4], Alain Palloix[5], Anne-Marie Sage-Palloix[5], Hélène Védie[6], Claire Goillon[7], Philippe Castagnone-Sereno[1]

[1] *Institut Sophia Agrobioetch, INRAe - Université Côte d'Azur - CNRS, Sophia Antipolis, France*; [2] *Ecodeveloppement, INRAE, Avignon, France*; [3] *Domaine expérimental Alénya-Roussillon, INRAe - Université de Montpellier, Alénya, France*; [4] *CBGP, IRD - CIRAD - INRAe, Montpellier, France*; [5] *GAFL, INRAe, Montfavet, France*; [6] *Groupe de Recherche en Agriculture Biologique, Avignon, France*; [7] *Association Provençale de Recherche et d'Expérimentation Légumière, Saint-Rémy-de-Provence, France*

A systems approach based on co-design and experimental field evaluation of cropping systems (CSs) combining technical and varietal innovations has been implemented for sustainable management of root-knot nematodes (RKN) in Mediterranean sheltered vegetable systems [1].
Cropping systems combining genetic resistance and cultural practices (crop rotations including susceptible, resistant, and non-host plants, intercropping management with nematicidal cover crops or soil solarization) were assessed during 4 years (i) to reduce RKN populations and increase the durability of varietal resistances, (ii) to study their impact on soil ecology (plant-parasitic and free-living nematode communities), and (iii) to evaluate their acceptability by farmers. Three CS prototypes resulting from a co-design process with research and development stakeholders have been compared with CS conventionally carried out in the Mediterranean area and evaluated thanks to complementary methods: 1 / system experiments in 3 commercial farms in Southern France; 2 / analytical experiments to decipher the mechanisms of action for some levers; 3 / surveys to evaluate the acceptability of the prototypes by farmers. All three CSs were effective (90% of RKN decrease., protection of partially resistant Solanaceae, no negative effect on non-phytoparasitic nematodes) and sustainable when application conditions and soil biological equilibrium were favorable (global soil nematofauna diversified and abundant) [1,2,3,4,5]. Their acceptability depended on the farming type and the attitude of farmers towards innovation [1,3]. These CSs still need to be improved, in consultation with participation of farmers, in terms of efficiency, by introducing new agroecological levers, as well as innovation' cost. This is currently done in the framework of a project of operational groups of PEI (European Pole of Innovation), program FEADER in PACA (2018-2021): "GONem: Operational Group on the management of root-knot NEMatodes in vegetable crops in the PACA region". Future research will also have to open up to a more comprehensive management of soil health. This work was part of the GEDUNEM project (Varietal and technical innovations for the sustainable and integrated management of RKN in protected vegetable cropping systems) supported by the INRA metaprogram SMaCH (Sustainable Management of Crop Health) and the GIS PicLeg (Scientific interest group on integrated vegetable production).

Keywords: Plant-parasitic nematodes - Integrated pest management - Resistance gene durability - Agronomic practices - system experiment.

References:

- [1] Djian-Caporalino et al., 2019. Biotechnologie, Agronomie, Société et Environnement (BASE) 23 (1) : 7-21.
- [2] Djian-Caporalino et al., 2019. Crop Protection 122 : 142-150.
- [3] Navarrete et al., 2016. Agronomy and Sustainable Development 36 : 68-7.
- [4] Goillon et al., 2016. Phytoma La défense des végétaux 698 : 39-44.
- [5] Djian-Caporalino & Mateille, 2018. Innovations Agronomiques 69: 83-89.

S5-P10

Investigations of mixed infections of root lesion nematodes (*Pratylenchus thornei* and *Pratylenchus neglectus*) and cereal cyst nematodes (*Heterodera avenae* and *Heterodera latipons*) on wheat.

İbrahim Halil Elekcioğlu (halile@cu.edu.tr), Ece Börtecine Kasapoglu Uludamar

Plant Protection Department, Agricultural Faculty, Cukurova University, Adana, Turkey

Reproduction rate and competition status of mixed and single population of *Pratylenchus thornei, P. neglectus, Heterodera avenae* and *H. latipons* were investigated on 20 summer and winter wheat cultivars between 2016 and 2017 under screen house condition. All soil and root samples were extracted by Fenwick and Petri methods. It was determined that cyst nematodes reproduced better than root lesion nematodes in mixed and single infection. In mixed infection, *P. thornei* and *P. neglectus*showed quite low reproduction rates in all experiments. When *H. avenae*, *H. latipons*, *P. thornei*and *P.neglectus* were to be mixed infections in the experiments, cereal cyst nematodes suppressed the reproduction of root lesion nematodes. This study was aimed also to screening of the resistant genes against to cyst nematodes in plants and the changes of root cells after multiple infections of cyst and root lesion nematodes. To investigate of root cell, roots were embedded in paraffin, cut by microtome. The slides of the root cells were examined under light microscope. The root lesion nematodes damaged the cortical cells along the feeding path like cavity. Due to the nematode infection, cortical cells of most wheat cultivars were destroyed by the development of many cavities in the wheat root cells. It was observed that root lesion nematodes are collectively fed in the cell and form fusion by melting the cell wall, cyst nematodes caused to form small syncytium in resistant cultivars. Other results of this study was about resistant genes that the Cre1 and Cre3 genes of the cereal cyst nematode resistance genes did not provide full resistance. It was found that Uzunyayla, Atlı2002, Yayla305, Gerek79, Harmankaya99, Aldane, Silverstar varieties possessed both Cre3 and Cre1 genes. The number of eggs in cysts changed at different conditions. In addition, inter-species competition has substantially changed the reproduction of the population in soil. This project was supported TUBITAK214O419.

Keywords: Intra-species competition - Histopathology - Cereal cyst nematodes - Root lesion nematodes.

S5-P11

Field performance of several maturity group IV and V soybean cultivars in a southern root-knot nematode infested field.

Michael Emerson (memerson@uaex.edu), Travis Faske

Entomology and Plant Pathology, University of Arkansas, Lonoke, AR, United States

The southern root-knot nematode (*Meloidogyne incognita*) is the most important yield-limiting plant-parasitic nematode that affects soybean production in Arkansas. Host plant resistance is an effective management tool; however, there is limited information on the host suitability of the most commonly grown commercially available cultivars grown in the Mid-Southern U.S. The objective of this study was to evaluate the susceptibility of several commonly grown maturity group IV and V cultivars. Fifty-six soybean cultivars were planted in a field with a severe population density of southern root-knot nematode. Eight plant were sampled at R4-R5 stage of growth and the percent root system galled was estimated for each plant. Thirteen percent of the maturity group IV, glyphosate/dicamba cultivars were rated as resistant. These cultivars had an average gall rating of 2.8 and yield of 4,909 kg/ha, whereas the remaining were rated susceptible with an average gall rating of 36 and yield of 3,295 kg/ha. Sixty-seven percent of the maturity group IV, glufosinate cultivars were rated resistant. These cultivars had an average gall rating of 2.1 and an average yield of 4,102 kg/ha, whereas the remaining were rated susceptible with an average gall rating of 22.3 and yield of 4,170 kg/ha. Sixty seven percent of the maturity group V, glyphosate/dicamba cultivars were rated resistant. These cultivars had an average gall rating of 0.9 and yield of 4,909 kg/ha, whereas the remaining were rated susceptible with an average gall rating of 20.1 and yield of 3699 kg/ha. Fifty percent of the maturity group V glufosinate cultivars were rated resistant. These cultivars had an average gall rating of 1.5 and yield of 4,909 kg/ha, whereas the remaining were rated susceptible with an average gall rating of 11.4 and yield of 4,035 kg/ha. Though the majority of these soybean cultivars were susceptible, a few were resistant, which would be a better option in fields with a damaging population density of root-knot nematode.

Keywords: Soybeans - Southern Root-Knot Nematode - *Meloidogyne incognita*.

S5-P12

Ultrasound-assisted extraction of nematicidal compounds from *Ricinus communis* and its potential against *Meloidogyne javanica*.

Lais Fontana[1] (laisffontana@hotmail.com), Guilherme Tarini[2], Bruna O Toniato[2], Angélica A. Melo[2], Adão Júnior Izidoro[2], Cláudia Regina Dias-Arieira[2]

[1] *Fitopatology, UEM, umuarama, PR, Brazil*; [2] *Fitopatology, UEM, Umuarama, Brazil*

Plant compounds are being widely investigated due to lower environmental impact, rapid degradation and high effectiveness for nematode management. In this context, we tested the ultrasound-assisted technique in the extraction of *Ricinus communis* compounds and assessed the content of the total phenolic compounds of the extract obtained. The effectiveness of the extract for *Meloidogyne javanica* suppression was also investigated. The bioactive compounds were extracted in an ultrasonic bath (frequency 40 kHz, power 132 W) in a solution of water and ethanol as solvent, in four different concentrations (0, 20, 40 and 60% ethanol). In each trial 10g of plant tissue (*R. communis*) and 100 ml of solvent were used, and the samples were left in the ultrasound bath for 30 minutes. The same solvent solutions were used in a non-ultrasound treatment for comparison. The crude extract obtained in the different solvent concentrations was diluted at concentrations of 0 (control, distilled water), 5, 10, 15, 20 and 25% and tested for its effect on hatching and mortality of *M. javanica*. To do so a 4x2 factorial (four solutions and 2 extraction methods) completely randomized experimental design with five replicates was performed for each of the 5 diluted extracts. The total phenolic compound content for each solvent concentration used in the extraction was expressed in mg of gallic acid equivalent (GAE) in 100g of the sample. For each replicate 200 eggs and 200 juveniles were placed in 5ml of the extract solution and the effect on *M. javanica* juveniles mortality was assessed 24 hours later, while the effect on hatching was evaluated 7 days later. Hatching reductions of 98% and 95% were observed with the extracts obtained from the ultrasound (WU) technique when compared to 40% and 60% for the control ethanol concentration solvent, at 25% dilution. With the extracts obtained without the ultrasound technique (WOU) the hatching reduction was 69% and 65% using the same solvent concentration and extract dilution. Mortality rates ranged from 0.7% for the control, to 62% and 64% when using 60% concentrate solvent with both WU and WOU, respectively. The total phenolic compounds obtained from the extracts recovered via ultrasound was 905,701 mg GAE (WU) and 475,26 mg GAE (WOU) at 60% solvent. Therefore, the ultrasound assisted extraction technique was efficient in the total phenolic compounds extraction of *Ricinus communis*, and the extract had a positive effect on *M. javanica* suppression.

Keywords: Alternative control - Castor beans - Plant extracts - Root-knot nematode.

S5-P13

Detection of the root-knot nematode *Meloidogyne luci* parasitizing tomatoes in Sacatepéquez Province, Guatemala.

Maria Geane Fontes[1] (geane.fontes@eastwestseed.com), Alvaro Javier Rodas Sierra[1], Sandy Maoly Franco Castañeda[1], Carina Mariani Leite Lopes[2], Cherry Relevante-Belagantol[3]

[1] *East-West Seed Guatemala, Santa Lucia Milpas Altas, Sacatepéquez, Guatemala*; [2] *Nemafito Laboratory, Luis Eduardo Magalhães, Bahia, Brazil*; [3] *East-West Seed Philippines, Bulacan, Philippines*

Root-knot nematodes (RKN) *Meloidogyne* spp. represent one of the most damaging pests of vegetable crops worldwide. The genus *Meloidogyne* includes nearly 100 species infecting a wide range of hosts, including tomato (*Solanum lycopersicum* L.). RKN are considered the most important and widely distributed group of plant-parasitic nematodes, causing serious yield losses in tomatoes. Guatemala represents an important country in Central America in the production of tomatoes. Its production is both for domestic consumption and for export. This work aimed to identify a RKN species present in a tomato field in Sacatepéquez province, Guatemala. In 2021, a survey was conducted in a tomato field in Antigua (Sacatepéquez province) for the presence of RKN. For that, root samples of ten tomato plants exhibiting galls were collected, in order to identify the *Meloidogyne* species in these samples, individual females were handpicked from infected tomato roots and submitted to species characterization by electrophoretic analysis of the esterase (EST) isozyme patterns according to Esbenshade & Triantaphyllou (1990) [1]. *Meloidogyne javanica* (EST J3) was included as a reference. The observation of polyacrylamide gels showed that all *Meloidogyne* populations studied had the three banded esterase pattern recognized as EST L3, specific to the species *Meloidogyne luci* Carneiro *et al*., 2014 (Tylenchida: Meloidogynidae) [2]. The newly described RKN *M. luci* has been detected parasitizing a wide host range and has been associated with several economically important crops in several countries all over the world including Guatemala, however, to the best of our knowledge, this is the first report of the RKN *M. luci* in Sacatepéquez province. This study needs to be expanded to other regions in order to identify the main RKN species in Guatemala to support in the establishment of control measures to prevent the spread of this pathogen.

Keywords: *Meloidogyne luci* - Esterase - EST L3 - *Solanum lycopersicum* L.

References:

- Esbenshade & Triantaphyllou. 1990. Journal of Nematology 22: 10-5.
- Carneiro et al., 2014. Nematology. 16(3): 289–301.

S5-P14

Assessment of the resistance spectrum of the tomato *Mi*-1.2 gene/locus against fifteen *Meloidogyne* species.

Marcia Gabriel[1] (gabriel.marcia@gmail.com), Stela Maris Kulczynski[1], Marlove Fátima Brião Muniz[1], Leonardo Silva Boiteux[2], Regina Maria Dechechi G. Carneiro[3]

[1] *Phytopathology laboratory, University of Santa Maria, Santa Maria, RS, Brazil*; [2] *Genetic breeding laboratory, Embrapa Hortaliças, Brasilia, DF, Brazil*; [3] *nematology laboratory, Embrapa Genetic Resources and Biotechnology, Brasilia, DF, Brazil*

The root-knot nematode (RKN) species, genus *Meloidogyne,* are polyphagous and have cultivated tomatoes (*Solanum lycopersicum* L.) as preferential hosts, leading to significant yield and quality losses. The dominant *Mi*-1.2 gene confers resistance in tomato to the three most important RKN species – *M. incognita, M. javanica,* and *M. arenaria*. However, little information is available about the reaction of tomato accessions carrying this gene to a wide range of tomato-infecting RKN species. The resistance spectrum of the *Mi*-1.2 gene/locus was evaluated against populations of 15 *Meloidogyne* species, employing two contrasting tomato cultivars 'Santa Clara' (homozygous recessive *mi*-1.2/*mi*-1.2 = susceptible) and 'Debora Plus' (heterozygous *Mi*-1.2/*mi*-1.2 = resistant). Seedlings were inoculated with 5,000 eggs of each *Meloidogyne* species separately. Evaluations were performed 90 days after inoculation and the nematode reproduction factors were calculated. The *Mi*-1.2-bearing hybrid 'Debora Plus' was classified as susceptible only to *M. enterolobii* and *M. hapla* and displayed a resistant reaction to the other 13 species viz. *M. javanica*, *M. incognita*, *M. arenaria*, *M. morocciensis*, *M. ethiopica*, *M. inornata*, *M. luci*, *M. konaensis*, *M. paranaensis*, *M. izalcoensis*, *M. hispanica*, *M. petuniae*, and *M. exigua*. The *Mi*-1.2 is located on the tomato chromosome 6 within a cluster of seven homolog genes of the nucleotide-binding site – leucine-rich repeat (NBS–LRR) family. Therefore, additional investigations should be done to confirm if this multiple *Meloidogyne* species resistance phenotype is controlled exclusively by the *Mi*-1.2 gene or by a combined action of other closely linked genes. This comprehensive evaluation of the resistance spectrum of the dominant *Mi*-1.2 gene/locus to multiple *Meloidogyne* species suggested that the employment of this genetic factor has the potential of avoiding losses induced by a wide range of *Meloidogyne* species with natural occurrence across tropical and subtropical soils.

Keywords: *Solanum lycopersicum* L - Resistance - Root-knot nematode.

S5-P15

Can *Arbutus unedo L.* leaves be used as a biological alternative to control *Meloidogyne javanica*?

Cristina Galhano[1] (cicgalhano@esac.pt), Vicente Damian Guevara Jacuinde[2], Inês Seabra[3], Marta Henriques[4], María Virginia Ozcariz-Fermoselle[5], Paula Lorenzo[6]

[1] *Department of Environment, IPC-ESAC/ Centre for Functional Ecology, Coimbra, Portugal*; [2] *Universidad del Estado de Hidalgo, Hidalgo, Mexico*; [3] *Bioengineering Department, Lehigh University, Bethlehem, United States*; [4] *Department of Food Engineering, IPC-ESAC/ CERNAS, Coimbra, Portugal*; [5] *Departamento de Ingeniería Agrícola Y Forestal, Universidad de Valladolid, Campus Palencia, Palencia, Spain*; [6] *Department of Life Sciences, University of Coimbra, Centre for Functional Ecology, Coimbra, Portugal*

Arbutus unedo L., strawberry tree, is a plant species well adapted to the Mediterranean climate. In Portugal, it is naturally spread over 3.2 million ha, corresponding to 35% of the national territory. Previous studies revealed the presence of some interesting chemical compounds in the leaves of this species. However, fruits are the only part widely used to produce alcoholic beverages, so this plant still underused. The root knot nematode, *Meloidogyne javanica*, which causes significant losses in several crops, is mainly controlled by synthetic nematicides. The main goal of this work was to search for potential uses of *A. unedo* leaves, in order to find additional value to a Portuguese endogenous resource and, simultaneously, try to find an environmentallyfriendly alternative to control *M. javanica*. The potential bionematicidal effect of an *A. unedo* leaf extract was evaluated on *M. javanica*. The aqueous extract was obtained using dried powdered leaves at a 1:5 (m/v) for 24 hours, at room temperature. *In vitro* bioassays were carried out in quadruplicate using glass blocks and applying three different extract concentrations: 5, 10, 20 % and distilled water as control. Each glass block contained 0.5 mL of extract and 20 nematodes in the second juvenile stage (J2). The number of immobile and dead nematodes were recorded at 24, 48 72, and 96 hours after exposure. Data were submitted to a Generalized Linear model analysis to test for the effect of extract concentration and time using the Poisson distribution and Log link function. The *A. unedo* extract had both nematicidal and nematostatic effects on J2. All the tested concentrations significantly increased J2 mortality compared to the control. The highest mortality was achieved for an extract concentration of 20%, followed by 10 and 5%, both with the same effect. Concentrations were significantly effective only at 96 hours after exposure, causing mortalities of 41.3, 36.3 and 61.3% in 5, 10, and 20% concentrations, respectively. Immobility was significantly higher in the 20% extract compared to the control. Based on the results, the *A. unedo* leaves aqueous extract can be considered as a potential alternative to synthetic nematicides to control *M. javanica.* Therefore, more detailed studies should be carried out to better understand the potential of this endogenous resource, as a biological solution to control plantnematodes, preserving the environment and employing natural resources.

Keywords: Nematicidal - Nematostatic - Root knot nematodes - Strawberry tree - Portuguese endogenous resources.

S5-P16

Could an agri-food be converted into a valuable environmentallyfriendly nematicide?

Cristina Galhano[1] (cicgalhano@esac.pt), Sara Barbosa[2], Inês Seabra[3], Mara Braga[4], Marta Henriques[5]

[1] *Department of Environment, IPC-ESAC/Centre for Functional Ecology, Coimbra, Portugal*; [2] *IPC-ESAC, Coimbra, Portugal*; [3] *Bioengineering Department, Lehigh University, Bethlehem, United States*; [4] *Department of Chemical Engineering, University of Coimbra/CIEPQPF, Coimbra, Portugal*; [5] *Department of Food Engineering, IPC-ESAC/CERNAS, Coimbra, Portugal*

Root-knot nematodes (RKN), *Meloidogyne* spp., are some of the most important plant-parasitic nematodes, causing significant quality and quantity production losses of a wide range of crops, some of them with relevant economic importance. Society's concerns regarding public health and environment, and the need to achieve planet sustainability, have created the need to find eco-sustainable alternatives for synthetic chemical pesticides, including for nematicides. For example, sustainability can be promoted through a Circular Economy, converting agri-food waste into resources. Thus, the main objective of this study was to evaluate the bionematicidal potential of extracts from faba bean pod (*Vicia faba*), oxidized and fresh, on second juvenile stage (J2) of *Meloidogyne javanica*. *In vitro* studies were carried out using the two types of extracts obtained with different solvents (water, 20% ethanol and 70% ethanol) at three different temperatures (45, 60 and 75°C), using the same solid:liquid concentration of 0.05 g / mL, and distilled water as a control. Four replicates were made for each extract treatment and control. The alive, immobile and dead nematodes were observed at 24, 48, 72 and 96 hours after exposure. All the extracts studied had nematicidal potential on *M*. *javanica*. We observed 100% mortality in the oxidized faba bean pod extracts at 72 hours and in the fresh faba bean pod extracts at 96h. The oxidized faba bean pod extract with 20% ethanol, at 75 °C was the most effective. The results showed that this agri-food waste has high nematicidal potential and may therefore constitute raw material for the development of eco-sustainable alternatives. Nevertheless, further studies envisaging the proposal as a new bionematicide product must be carried out.

Keywords: Faba bean pod extracts - *Meloidogyne javanica* - Plant nematodes - Vicia faba - Waste.

S5-P17

Mapping a new resistance to the potato cyst nematode *Globodera pallida* from the wild potato *Solanum spegazzinii*.

Ulrike Gartner[1, 2] (ulrike.gartner@hutton.ac.uk), Miles Armstrong[3], Sanjeev Kumar Sharma[1], Vivian Blok[1], Ingo Hein[1, 3], Glenn Bryan[1]

[1] *Cell and Molecular Sciences, The James Hutton Institute, Dundee, United Kingdom*; [2] *School of Biology, University of St. Andrews, St. Andrews, United Kingdom*; [3] *Plant Sciences, University of Dundee, Dundee, United Kingdom*

Potato cyst nematodes are economically the most important parasitic nematodes of potato causing at least 9% crop loss worldwide. One way to address this problem, ideally as part of an integrated pest management strategy, is to find new sources of naturally occurring resistance in wild potato species and to introgress them into potato breeding material. The diploid potato wild species *Solanum spegazzinii* Bitter accession 7195, that shows resistance to *Globodera pallida* pathotypes Pa1 and Pa2/3, is investigated here.
A cross and first backcross of *S. spegazzinii* with *S. tuberosum* group Phureja cultivar Mayan Gold was performed, and progeny clones were phenotyped for resistance to *G. pallida* Pa2/3 population Lindley in a root trainer bioassay. Bulked-segregant analysis using generic-mapping enrichment sequencing and genotyping-by-sequencing to identify SNPs that are genetically linked to the resistance were performed. These SNP markers were converted into allele specific PCR assays and the resistance was mapped using a graphical genotyping approach. Tetraploid-diploid interploidy crosses of relevant potato cultivars with the *G. pallida* resistant diploid *S. spegazzinii*, which rely on unreduced male gametes of the diploid parent, were also performed.
The segregation pattern of the resistance/susceptibility in the progeny clones is compatible with either one or two independent genes, with one gene having a much larger effect than the other, conferring the resistance.
By graphical genotyping of 14 informative SNPs the resistance gene could be mapped to less than 150 kb on chromosome VI. There is no *R*-gene cluster reported in this region of the DNA sequence of doubled monoploid *S. tuberosum* group Phureja clone DM 1-3 516 R44 (DM) used as a reference genome.
Initial results found that one cross of the *G. pallida* susceptible tetraploid potato cultivar Alouette with the *S. spegazzinii* backcross provided two progeny plants that had reduced susceptibility to *G. pallida*.
So far there is no conclusive evidence that the resistance to *G. pallida* described here is conferred by an *R*-gene, although *S. spegazzinii* might have undergone translocation of *R*-genes after it separated from the cultivated species. This new resistance is an ideal candidate for introgression into tetraploid potatoes, with the eventual aim to pyramid several different PCN resistance genes to get broad-spectrum and durable resistance to *G. pallida* in potato.

Keywords: Potato cyst nematode - Resistance - Wild potato species - Mapping of resistance.

S5-P18

Genetic diversity of soybean cyst nematode (SCN) *Heterodera glycines* populations in southeastern Goiás state, Brasil.

Fernando Godinho de Araújo[1] (fernando.godinho@ifgoiano.edu.br), Janaina Alves de Almeida Moreira[2], Miran Carvalho Tavares[3], Ivandilson Pessoa Pinto de Menezes[3]

[1] *Campus Urutaí, Instituto Federal Goiano, Urutaí, Goias, Brasil*; [2] *Escola de Agronomia, Universidade Federal de Goiás, Goiânia, Goiás, Brasil*; [3] *Campus Urutaí, Instituto Federal Goiano, Urutaí, Goiás, Brasil*

Heterodera glycines, also known as soybean cyst nematode (SCN), is one of the phytosanitary issues that prevents high soybean yields (*Glycine max*) due to its dissemination capacity and high physiological race variability. The first step towards the proper and safe use of resistant cultivars lies on correctly identifying the physiological race of this nematode in the crop in order to avoid new breed-selection pressure. Thus, the aim of the present study is to genetically characterize the races of *Heterodera glycines* populations from soybean-producer counties belonging to the Railroad (Estrada de Ferro) Region (Goiás State, Brazil), as well as to investigate their molecular characterization based on RAPD markers. RAPD data were evaluated in a binary way, in which values (1) and (0) were attributed to the presence and absence of amplified bands for each primer, respectively. The binary matrix was used to estimate the genetic distance between populations, based on the Jaccard dissimilarity index. The clustering analysis was based on the calculated genetic dissimilarity matrix, according to Ward's method. Eight (8) different physiological *Heterodera glycines* races were found in the investigated counties. Only 10 out of 28 RAPD primers were polymorphic among SCN populations, whereas the remaining primers did not amplify, or presented low amplification of the DNA fragment. Five genetically-different groups were found among *H. glycines* population accessions, which represents a high genetic variability rate.

Keywords: Characterization - HG Type - Polymorphism - Race test - Soybean cyst nematode.

S5-P19

Nematicide and ovicide effect of thiophanate-methyl and fluazinam (Certeza N®) against nematodes.

José Paulo Gonçalves Franco Silva (jose.paulo@ihara.com.br), Eveli Macedo, Thais Souza

Technical Department, IHARA, Sorocaba, SP, Brazil

Meloidogyne incognita and *Pratylenchus brachyurus* are predominant species of nematodes that occur in Brazil. Around 60% of the soybean agriculture these species in their fields, and more than 90% conduct seed treatment in order to manage the incidence of diseases and the population of nematodes. Certeza N® (thiophanate-methyl + fluazinam) is a product developed by IHARA and it has been useful for the management of several species of fungus that occur on row crops, such as *Rhizoctonia solani*, *Pythium* spp., *Aspergillus*spp., and *Fusarium* spp. Based on results from field experiments, potential was shown for the product and its mixture compounds for management of nematodes on important crops, such as soybean and corn. Here, we conducted laboratory studies (*in vitro*) in order to evaluate the possible nematicide effect of the active ingredients thiophanate-methyl and fluazinam, and on the mixture, on juveniles of *P. brachyurus*. The effects of the compounds and the mixture on eggs of *M. incognita* were also evaluated in order to verify the effect on its hatch. For this, juveniles were exposed to the chemical solution for 24h, and moved to a water solution after this period [1, 2, 3]. Both active ingredients, tiofanate and fluazinan, and the mixture, presented nematicidal effects against juveniles of *P. brachyurus*. Related to the ovicidal effects of *M. incognita*, the number of juveniles hatched from eggs were assessed on 3, 7 and 14 days after exposition to the treatments. The active ingredients, isolated or in the mixture, presented 50 to 60% reduction of egg hatch. The study shows the potential of the active ingredients and the product Certeza N® for the management of nematodes on important agriculture crops. New studies might be conducted in order to clarify the mode of action of thiophanate-methyl and fluazinam against on the species of nematodes.

Keywords: Root-knot nematode - Lesion nematode - Chemical control.

References:

- [1] Baermann, G. 1917. Ned. Indie, 57: 131-137.
- [2] Byrd, D.W. et al. 1983. Journal of Nematology, 15(1): 142-143.
- [3] Hussey, R.S. and Barker, R.K. 1973. Plant Disease Reporter, 57: 1025-1028.

S5-P20

Management strategies utilizing fertilizers and nematicides to reduce *Rotylenchulus reniformis* induced damage on cotton.

Kara Gordon[1] (KLG0037@auburn.edu), Kathy Lawrence[1], Brad Meyer[2], Drew Schrimsher[3]

[1] *Auburn University, Auburn University, Alabama, United States*; [2] *Agri-AFC, Decatur, Alabama, United States*; [3] *Agri-AFC, Slocomb, Alabama, United States*

The reniform nematode (*Rotylenchulus reniformis*) causes major yield loss on Upland cotton in the Southern United States. A proper management strategy is crucial to sustaining plant health and preventing financial decline. The objective of this study was to apply a combination of nematicides and fertilizers at different plant growth stages to combat *Rotylenchulus reniformis* induced yield loss. Field trials were conducted in North Alabama, United States of America and were arranged in a randomized complete block design, using Deltapine 1646 B2XF cotton. Treatments consisted of Aeris (Thiodicarb and Imidacloprid), $(NH4)_2SO_4$, 28-0-0-5, Max-In Sulfur (0-0-19) and Vydate (Oxamyl). To mitigate initial reniform density, Aeris (Thiodicarb and Imidacloprid) was applied at planting to select treatments. All chemicals, with the exception of Aeris, were applied at pinhead square (PHS) and/or first bloom (FB). Field trials found that a combination of Aeris, (NH4)2S04, Max-In Sulfur and Vydate, applied at PHS, resulted in the largest root fresh weight and significantly ($P \leq .05$) lowered reniform eggs/g of root when compared to all other treatments. Trails also indicated that a treatment containing Aeris, (NH4)2S04, Max-In Sulfur and Vydate, applied at PHS and FB, increased seed cotton yields by 21% when compared to treatments with no nematicide. This treatment was also a cost-effective management strategy when comparing seed cotton yield to chemical application input costs. Management strategies that focus on nematode suppression and reduced application input expenses are more likely to prevent financial losses from *Rotylenchulus reniformis* induced damage.

S5-P21

Effect of fluopyram, spirotetramat, and oxamyl on the cereal cyst nematodes, *Heterodera avenae* in Idaho.

Saad Hafez[1] (shafez@uidaho.edu), David Kihoro Sirengo[2], Floridelia Garcia[2], Michael Thornton[2], Oliver Neher[3], Sundararaj Palanisamy[4], Esmaeil Fallahi[2], Idris Soliman[2], Kelly Luff[5]

[1] *Plant Pathology, Entomology and Nematology, University of Idaho, Parma, ID, United States*; [2] *Plant Pathology, Entomology and Nematology, University of Idaho, parma, Idaho, United States*; [3] *Sugarbeet quality improvement, Amalgamated Sugarbeet Company, Boise, Idaho, United States*; [4] *Zoology, Bharathiar University, Coimbatore, Tamil Nadu, India*; [5] *Bayer CropScience, Kimberly, Idaho, United States*

Cereal cyst nematode (CCN, *Heterodera avenae*), an important plant-parasitic nematode of wheat, causes yield losses as high as 92% and is widely distributed in the Pacific Northwest regions of United States warranting immediate development of effective methods to protect wheat crops from *H. avenae* damage. Several management practices have been put in place in other parts of the world to control this devastating pathogen but chemical management remains the most effective. Fluopyram, spirotetramat and oxamyl have been registered for controlling root-knot and lesion nematodes on potato and onions in Idaho. However, the potential of these chemistries to control *H. avenae* on wheat remains unknown. Two experiments were conducted in which the three non-fumigant nematicides fluopyram, spirotetramat and oxamyl were examined at three different application timings for their efficacy in reducing all stages of CCN. Experiments were conducted in the greenhouse and microplots. Each was inoculated with 5000 eggs per greenhouse pot or microplot bucket and the effects on plant growth parameters and yield was documented. Nematicides were applied 1week prior to inoculation of eggs, at inoculation of eggs, and eight weeks after seedling emergence. All treatments reduced the number of cysts, viable eggs, juveniles and males at all application timings compared to untreated control ($p \leq 0.01$) under greenhouse conditions. Application of spirotetramat 1 week prior to inoculation, at inoculation, and 8 weeks post seedling emergence increased plant fresh weight, wheat yield ($p \leq 0.01$), and plant height ($p \leq 0.05$) compared to the untreated control, fluopyram and oxamyl under greenhouse conditions. The number of cysts formed after harvest under microplot conditions were significantly lower for all application timings of all treatments compared to untreated control ($p \leq 0.01$). However, fluopyram and oxamyl had a reduction rate of over 70% compared to spirotetramat and the untreated control. No significant treatment effects on plant height, top fresh weight, and yield was observed when compared to the untreated control. These chemicals prove effective and need further evaluation for registration against Heterodera avenae in Idaho.

Keywords: *Heterodera avenae* - Non-fumigant - Wheat.

References:

- Smiley et al., 2011. Plant Dis. 95(8): 983-989
- Smiley et al., 2012. Plant Dis. 96(10): 1537-1547
- Li et al., 2021. Plant Dis. 105(9): 2466-2471

S5-P22

Development of *Heterodera schachtii* in sugar beet genotypes with varying levels of resistance.

Johannes Roeb[1] (johannes.roeb@julius-kuehn.de), Florian Grundler[2], Johannes Hallmann[1]

[1] *Epidemiology and Pathogen Diagnostics, Julius Kühn-Institut, Münster, Germany*; [2] *Molecular Phytomedicine, Bonn University, Bonn, Germany*

The beet cyst nematode (BCN, *Heterodera schachtii*) is a major pest of sugar beet in many production areas around the world. BCN is mainly managed by growing tolerant sugar beet cultivars. These cultivars are not only tolerant, but also show a 30-60% lower nematode reproduction in comparison to susceptible cultivars. To better understand the mechanisms of different levels of resistance, development of *H. schachtii* in susceptible, resistant and tolerant sugar beet cultivars was studied in a series of climate chamber experiments. Seedlings were grown in 100-ml-containers with sand and inoculated in the 2-leaf stage with 500 BCN second-stage juveniles. Nematode development was evaluated at weekly intervals until the first generation was completed. In summary, 1) resistance and partial resistance did not affect root penetration rate of *H. schachtii*, 2) resistance frequently interrupted the development of juveniles and almost completely prevented the development of adult females, 3) partial resistance changed the sex ratio in favour of males and delayed the development of females, 4) resistance and partial resistance reduced the number of developed cysts and the cyst content and 5) sugar beet genotypes with partial resistance could be differentiated according to the average number of eggs and juveniles per cyst. The project was funded by the Federal Ministry of Food and Agriculture through the Fachagentur Nachwachsende Rohstoffe e. V. (FKZ 22022812).

Keywords: *Heterodera schachtii* - Beet cyst nematode - Partial resistance - Tolerance.

S5-P23

'Magic Bullets' for plant parasitic nematodes.

Lindy Holden-Dye (l.m.holden-dye@soton.ac.uk)

University of Southampton, Southampton, United Kingdom

The 21st century demand for optimising crop production combined with an increasing demand from the public and regulatory bodies for environmental protection presents a challenging context for advances in crop protection. New chemicals must be both very efficacious and very selective for the parasite; in the words of the Nobel Prize winning chemist Paul Ehrlich they must be '*magic bullets*'. This is a very high bar to set. The most widely used nematicides miss this by a long mark and the adverse impacts of the organophosphate and carbamates are well recognised. One way to address this challenge is through fundamental research that reveals unique and distinctive features of the plant parasitic nematode e.g. biochemistry, metabolism and cell signalling. In this talk, I will broadly discuss these aspects in relation to new insight into key regulators of the parasitic life cycle and new generation nematicides that have an improved profile for selective toxicity.

S5-P24

Host status of cover crops for root lesion nematode species (*Pratylenchus* spp.) associated with apple in South Africa.

Rinus Knoetze[1, 2] (knoetzer@arc.agric.za), M. Addison[2, 3]

[1] *Infruitec-Nietvoorbij, Agricultural Research Council, Stellenbosch, South Africa*; [2] *Department of Conservation Ecology and Entomology, Matieland, South Africa*; [3] *Hortgro, Paarl, South Africa*

Currently the most preferred method of orchard management is to plant a cover crop in the work row to improve fertility and soil structure, and reduce erosion. However, in choosing a cover crop for use in apple orchards it is important to ensure that it is not a good host for lesion nematodes. Various cover crops were inoculated with lesion nematodes that were cultured on carrot discs. After 12 weeks, the crops were evaluated for their susceptibility to lesion nematodes. To do this, the reproduction factor of the nematodes in each crop as well as the percentage of the nematodes g-1 roots of each crop in relation to the nematodes g-1roots of a known susceptible crop was calculated. Indian buckwheat is considered a moderate to good host for *P. hippeastri, P. penetrans* and *P. vulnus*, and can cause an increase in nematode numbers if planted at high density. Nasturtium is considered a moderate to good host for these nematodes and has the potential to increase nematode numbers in the orchard. Pink serradella (cv. Margarita), rye (cv. Duiker Max) and subterranean clover (Aarbei klawer) are considered good hosts for *P. penetrans* only and should be avoided as cover crops if *P. penetrans* is present in an orchard. Reproduction of *P. hippeastri* is minimal on most cover crops in winter, but has the potential to increase on certain crops (pink serradella, rye, Triticale, subterranean clover, medics) during spring and summer if the cover crops persist in the orchard.

Keywords: Lesion nematodes - Apple - Cover crops - Susceptibility - Management.

S5-P25

Risk analysis, waste disinfestation methods and rotation plants as tools for a management of the risks associated with nematodes.

Anne-Claire Le Roux[1] (anneclaire.leroux@inov3pt.fr), Marie-Sophie Neveux[1], Fabrice Ollivier[2], Emilie Huchet[1], Pauline Dewaegeneire[3], Laure Berton[1], Hoël Hotte[2], Marie-Claire Kerlan[4], Virginie Gobert[3], Yves Le hingrat[1], Laurent Folcher[2]

[1] *FN3PT / inov3PT, Le Rheu, France*; [2] *Plant Health Laboratory, Nematology Unit, ANSES, Le Rheu, France*; [3] *FN3PT / inov3PT, Achicourt, France*; [4] *INRAE, Ploudaniel, France*

Nematools project aimed to reinforce prophylactic methods to prevent the introduction and the dissemination of regulated phytoparasites nematodes *Meloidogyne chitwoodi*, *M. fallax*, *Globodera pallida* and *G. rostochiensis* which are serious threats for potato crops. This project improved control strategies in order to avoid their spread across the French territory. One of the project task was to assess the risks linked to both industrial and laboratory processes and to characterize the efficacy of some decontamination methods. Another task focused on the identification of rotation plants useful for the limitation of quarantine nematodes belonging to *Meloidogyne* genus. In terms of main results, the actions carried out in partnership with industrials and experimental stations allowed the production of a user-friendly risk analysis tool characterizing the critical phases of different waste and effluent treatment processes and providing guidelines on the sanitizing performances of different decontamination processes such as composting, lagooning, anaerobic digestion, chlorination, and heat treatment. This project also allowed screening under controlled conditions of plant species which are not conducive to the development and propagation of root-knot nematodes such as *M. chitwoodi* and *M. fallax* and may be proposed as rotation plants on contaminated sites. Among these plants, specific radish (*Raphanus sativus*) and rocket (*Eruca sativa*) varieties clearly reduced the soil infestation level with variable efficiency between these two *Meloidogyne* species and their tested populations.

Keywords: Pest risk analysis (PRA) - Regulated plant parasitic nematodes - Alternative control methods - Wastes disinfection - Sanitizing plants.

S5-P26

Managing causal pathogens of the potato early die complex with a new chemistry, fluopyram.

Kelly Luff[1] (kelly.luff@bayer.com), Saad Hafez[2]

[1] *Crop Science Division, Bayer, Kimberly, ID, United States*; [2] *University of Idaho, Parma, ID, United States*

The potato early dying complex is problematic in potato growing areas throughout the world. Economic pressure coupled with the lack of alternative cash crops result in narrow crop rotations that amplify soil borne pest populations responsible for this issue. Potato early dying is a complex of both biotic and abiotic factors. The primary causal agent of potato early dying is *Verticillium dahliae*. However, several other pathogens may contribute, including root lesion nematodes (*Pratylenchus* spp.), the early blight fungus (*Alternaria solani* & *A. alternata*), the black dot fungus (*Colletotrichum coccodes*) and the white mold fungus (*Sclerotinia sclerotiorum*). Plant stress resulting from adverse weather conditions and nutrient or water imbalances can predispose potato plants to some of these pathogens. Germplasm offering effective tolerance to all major contributors of potato early dying is not yet available to the potato industry. Current management practices include planting certified seed, crop rotation, fumigation, avoidance of excessive irrigation early in the season and minimizing plant stress by encouraging uniform and continuous plant growth through balanced fertility and optimal irrigation. Pre-plant application of metam sodium has been the standard chemical practice for managing potato early dying. However, high cost, worker safety and increasingly stringent regulations are motivating growers to look for alternative practices. New non-fumigant approaches are being developed by basic manufacturers of crop protection chemistries to manage the various pathogens responsible for this disease complex. Among these new chemicals is fluopyram, developed by Bayer CropScience as Velum® Prime for soil uses. Initial research with this active ingredient demonstrated strong activity on the early blight complex and white mold, and suppressive activity on the black dot fungus. Subsequent research has shown that fluopyram has robust nematicidal activity on lesion nematodes and impact on Verticillium, the two main causal agents of potato early dying. This investigation demonstrates that low dose at-planting and post-emergence applications of Velum® Prime have efficacy on all major pathogenic contributors of the potato early dying complex.

Keywords: Fluopyram - Lesion Nematode - Early-die - Potato.

S5-P27

Premature and sudden death complexes: How and why both are important questions.

Ann MacGuidwin (aemacgui@wisc.edu)

University of Wisconsin-Madison, Madison, United States

Nematodes interact with fungi for two diseases, potato early dying (PED) and sudden death of soybean (SDS). In both cases, the nematode accelerates the severity and timing of symptoms typical for the fungal pathogens *Verticillium dahliae* (Vd) and *Fusarium virguliforme* (Fv), respectively. Compelling data demonstrate *Pratylenchus penetrans* (Pp) plays a role in PED. There is consensus that the interaction is synergistic at low pathogen population densities, among studies that vary for potato variety, soil type, and environment. The nematode-fungus synergism has been demonstrated for foliar symptoms and disease progression at multiple levels of resolution, as well as for potato yield metrics important to farmers. The synergistic model matches farmer's experience so well that the disease complex of PED is named differently than the fungus-only version of Verticillium wilt. Management decisions are made on the basis of the interaction, as recommendations reflect population densities of both the fungus and nematode. Studies on the interaction of *Heterodera glycines* (Hg) and Fv demonstrated that Hg influences SDS, but there is not compelling evidence of synergism at any levels of pathogen population densities. Consequently, there is a tendency for farmers to think of SDS as two versions, with and without Hg, rather than as the result of a biological interaction. Management recommendations of Fv inform farmers to include Hg in their consideration of environmental factors that affect SDS. Is there a different public interpretation of how these two diseases occur because of the way the research was executed or messaged? Or can it be attributed to differences in management options for the two diseases? The "why is there an interaction" question has not been answered for either disease complex. That detail is not very important to farmers because academic and industry partners have demonstrated how to manage the disease profitably – for now. Studies have demonstrated, for both systems, that the role of the nematode partner goes beyond root wounding. Digging deeper to unravel the mechanisms of these interactions is critical if we are to finesse nematode management to meet higher production and environmental demands in the future.

Keywords: Potato early dying - Sudden death of soybean - *Heterodera glycines* - *Pratylenchus* penetrans - fungal pathogens.

S5-P28

Compatibility of Nemafric-BL phytonematicide and biocontrol agents for the management of *Meloidogyne* species.

Jacqueline Tinashe Madaure[1] (jacquelinemadaure@yahoo.com), Phatu William Mashela[1], Dirk De Waele[2]

[1] *Green Biotechnologies Research Centre, University of Limpopo, Sovenga, South Africa*; [2] *Laboratory of Tropical Crop Improvement, University of Leuven (KU Leuven), Leuven, Belgium*

A field study was conducted to investigate the compatibility of Nemafric-BL phytonematicide (N) with *Steinernema feltiae* (S) and *Trichoderma harzianum* (T) in suppression of root-knot (*Meloidogyne* species) nematodes and enhancing growth of tomato plants as environment-friendly alternatives to synthetic chemical nematicides [1]. The treatments in a 2 × 2 × 2 factorial study ($N_0S_0T_0$, $N_1S_0T_0$, $N_0S_1T_0$, $N_0S_0T_1$, $N_1S_1T_0$, $N_1S_0T_1$, $N_0S_1T_1$ and N1S1T1), were arranged in a randomised complete block design, with nine replicates. Each product was applied as recommended, whereas for double combination each was halved and for triple combination each was applied as one-third the lone quantity [2]. Since data did not meet the assumptions of analysis of variance, nematode and plant data were subjected to principal component analysis for multivariate analysis. The first two principal components accounted for 95.7% variability on nematode reproduction. Nematode reproductive potential, juveniles in roots and eggs in roots were the highest in the untreated control and lowest in $N_0S_1T_1$, $N_1S_1T_0$, $N_0S_0T_0$ and $N_0S_1T_0$ in PC1 (72.4%). Nemafric-BL phytonematicide alone and $N_1S_0T_1$ had the highest and lowest J2 in soil, respectively, in PC2 (23.3%). The first two principal components accounted for 84.6% variability in plant growth. Plant dry shoot mass, fresh fruit mass, plant height and stem diameter were positively correlated with PC1, where the variables were increased by $N_1S_0T_0$, $N_0S_1T_1$ and $N_1S_0T_1$ and reduced by $N_1S_1T_1$. Chlorophyll was positively correlated with PC2, where $N_1S_0T_0$ increased and $N_0S_0T_0$ reduced the growth variable. The first two principal components accounted for 97.9% variability in selected nutrient elements. Potassium, Mn, Na and P were increased by $N_1S_1T_1$ in PC1, and P by $N_1S_0T_1$ in PC2. Interactions of Nemafric-BL phytonematicide with biocontrol agents on *Meloidogyne* species, tomato plant growth and nutrient elements indicated that the products were compatible. Therefore, the products could be considered for integrated pest management programmes.

Keywords: Biological control - *Cucumis africanus* - Entomopathogenic nematodes - Integrated nematode management - Nematophagous fungi.

References:

- [1] Mashela et al., 2011. Sci Res Essay 6(33):6762-6768.
- [2] Madaure et al., 2019. ACTA AGR SCAND B-S P. 69(3): 235-240.

S5-P29

Influence of the length of sugar beet sludge storage on the amount and viability of beet cyst nematode embryos (*Heterodera schachtii* A.Schmidt, 1871)

Marie Manasova[1] (manasova@af.czu.cz), Miloslav Zouhar[1], Jana Wenzlova[1], Ondrej Douda[2], Pavel Rysanek[1]

[1] *Department of Plant Protection, Czech University of Life Sciences Prague, Prague, Czech Republic*; [2] *Division of Plant Health, Crop Research Institute, Prague, Czech Republic*

The sugar beet is usually cultivated in fields with extremely fertile soil. Along with the harvested sugar beet, humus rich soil is transported to the sugar factory. Tons of soil are stored in a sugar factory for 3-4 years after the processing of sugar beet bulbs. During this period, the soil is relocated several times. In stored soil, seeds of weeds, pathogens and pests, can survive for a long time. Sugar beet cyst nematodes aren´t an exception. Stored soil is usually used for the fertilization of free fields. This kind of soil transport can cause an infestation of a new locality. New methods of harmful organism eradication are sought. One of the promising techniques could be the extension of storage time. In 2019, a long-term storage experiment was established in three variants: stored for 1, 2 and 3 years. Each year, representative samples (5 kg) from all variants were used for analyses. Samples were homogenized and 100 g of soil was processed by Fenwick can in 10 repetitions. Cysts of nematodes were manually counted and the viability of embryos was determined via light microscopy. The results were evaluated by software Statistica 12.0 (StatSoft, Tulsa USA) using Analysis of Variance followed by the Tukey test. Results showed a statistically significant reduction of cysts and embryo number and decreasing embryo viability. The results also showed the risk of weed infestation of stored soil to increase nematode populations. Most of the weed species are listed in the host range of *Heterodera schachtii*. The absence of weeds on the storage site can reduce the possibility of the nematode population increasing. This work was supported by the project of the Technology Agency of the Czech Republic No. TH04030242

Keywords: Sugar beet - *Heterodera schachtii* - Sugar factory - Sugar beet sludge.

S5-P30

Depicting the drivers of the root-associated microbiome in fields infected by root-knot nematodes in Cambodia.

Anne-Sophie Masson[1] (annesophie.masson@ird.fr), Marie Simonin[2], Marie-Liesse Vermaire[1], Caroline Brunel[1], Pierre Czernic[1], Lionel Moulin[1], Stéphane Bellafiore[1]

[1] *IPME, IRD, Montpellier, France*; [2] *IRHS, INRA, Angers, France*

Root-knot nematodes (*Meloidogyne* spp.) are among the most damaging plant parasitic nematodes (PPNs) worldwide [1]. They are obligate and polyphageous soilborne parasites affecting the root morphology and physiology and making plants more susceptible to other diseases and abiotic stresses. *Meloidogyne graminicola* is particularly widespread in Asia where it compromises the production of rice, the main staple food. In the search for sustainable and self-sufficient solutions to protect plants from pests, interest for the plant-associated microbiome recently emerged due to the recognition of its central role in plant health [2]. In 2014 and 2015, the occurrence of PPNs have been evaluated on a lowland rice field in Cambodia managed under traditional (with tillage) or conservation agriculture (direct-seeding, crop rotation, no tillage) during four years. At that time, no diminution of the occurrence of PPNs was observed [3]. In 2018, seven years after the conversion to a conservation agricultural system, we observed that the soil contains less PPNs. We hypothesized that in this soil, modifications of the rice microbiome induced a diminution of the PPNs occurrence. To disentangle the drivers of the rice microbiome, we assessed the impact of the agricultural system (conservation system *versus* a traditional system) and of the rice genotype (two *indica* cultivars and two *japonica* cultivars including one resistant to *M. graminicola*) in structuring the rice root microbiome (communities of bacteria and fungi) and mesofaune (community of PPNs). Our first results showed that the agricultural system has a higher impact on the structure of the fungal community than on the structure of the bacterial community, and that the role of rice genotype is relatively low. During the survey, we also created a collection of bacteria that we screened to select candidates for antagonistic activity against *M. graminicola* that we are currently testing *in vitro*. These findings will contribute to identify indicator taxa for the diagnostic of PPNs occurrence, and bacteria for biocontrol solutions against the root-knot nematode *M. graminicola*. Moreover, assessing the impact of agricultural practices on the root microbiome features and biological activity in rice fields raises the possibility of finding solutions (via microbiome ingeneering for example) that could help to limit the diseases.

Keywords: Rice - Plant parasitic nematodes - Microbiome - Conservation agriculture - *Meloidogyne graminicola*.

References:

- [1] Jones J et al., 2013. Molecular Plant Pathology. 14(9), 946–961.
- [2] Pieterse C et al., 2016. Trends in Plant Science. 21(3), 171–173.
- [3] Suong M et al., 2019. Journal of Nematology. 51: 1-15.

S5-P31

Performance of a commercial heat-killed *Burkholderia rinojensis* bio-based product in agricultural crops.

Karla Medina (kmedinaortega@marronebio.com), Timothy Johnson

Marrone Bio Innovations, Davis, CA, United States

The use and availability of biopesticides in both conventional and organic agricultural systems is increasing around the world. A recently introduced product based on inactivated *Burkholderia rinojensis* cell broth and fermentation by-products, called Majestene in the US, is evaluated on several crop groups. Every crop system is different and the best fit and use of the product will vary based on the management practices of each crop. Data will be presented from conventional, organic, or double cropping systems on fruiting vegetables, berries, root vegetables, tropical fruits, and row crops. Application methods range from drench to seed treatments. Although the mode of action on nematodes is still been elucidated, reductions in nematode counts, root gall damage, egg laying and egg hatching, infective capacity after exposure, and repellency have been observed both in the field and lab assays. In addition to nematicidal activity, byproducts of *B. rinojensis* also have insecticidal activity on soil borne insects, like wireworms and white grubs. A nematode integrated management is key to soil health and to long term effective programs, and Majestene is a tool offering flexibility and ease of use to fit into these programs.

Keywords: biological - Majestene - *Radopholus* - *Meloidogyne* - *Pratylenchus.*

S5-P32

Meloidogyne-Fusarium interaction for the management of vascular wilt of *Physalis peruviana* plants in Colombia.

Gladis Emilia Munera Uribe (gladismunera@yahoo.com)

Corporacion Colombiana de Investigacion Agropecuaria - AGROSAVIA, Rionegro, Antioquia, Colombia

Physalis peruviana L. is a solanaceous plant grown in the Colombian highlands and one of the most important Colombian cash crops in export markets. The vascular wilt caused by *Fusarium oxysporum* f. sp. *physali* (Foph), is the main limiting factor in cape gooseberry production, with losses up to 100%, which forces the displacement of the crop towards new areas. Interactions between *Meloidogyne* spp. and *F.oxysporum* have been recorded on different crops. For this reason, four nematode studies were conducted: abundance and distribution, parasitism test, inoculum densities and *M. hapla* X *F. oxysporum* interaction. For the first study, a total of 400 samples from the rhizosphere of cape gooseberry plants were taken throughout the country. Two nematode parasitism tests were performed (reproduction factor calculated at 120 and 360 days). The effect of different densities of *M. hapla* (500 to 15.000 eggs per pot) on plant growth parameters were estimated. *M. hapla* (5.000 eggs) and *F. oxysporum* ($1x10^6$ conidia) were used for interaction experiment. Twelve taxa of plant parasitic nematodes were associated with the rhizosphere of. *P. peruviana;* however only six reproduced on cape goosberry plants: *Meloidogyne, Pratylenchus, Xiphinema, Trichodoridae, Aphelenchidae* and *Helicotylenchus*. The most abundant genus was *Meloidogyne with M. hapla* found in all of the sampled regions. *Pratylenchus bolivianus* Corbett was reported for the first time in Colombia. An inoculum density experiment indicated that plant length, fresh and dry weight, and leaf number decreased as inoculum densities of the *M. hapla* increased. Sequential and concomitant inoculation of the *M.hapla* and *F. oxysporum* f.sp. *physali* showed that vascular wilt was more severe than the fungus alone as indicated by the decrease of the plant growth parameters. Beacuse nematodes enhance *Fusarium* damage, integrated management programs against both nematodes and *Fusarium* in cape gooseberry crops should be established.

Keywords: *Physalis peruviana* - nematodes - *Fusarium* - Interaction - Colombia.

References:

- May and Abawi, 1987. Ann. Rev. Phytopathol. 25: 317-338.
- Powell, N.T., 1971. Ann. Rev. Phytopathol. 9: 253-274.

S5-P33

Potential of biofumigant cover crops (*Brassica* spp.) for suppression of stubby root nematodes (*Trichodorus* and *Paratrichodorus* spp.), associated with docking disorder in sugar beet (*Beta vulgaris*).

Nyambura Mwangi[1] (nmwangi@live.harper.ac.uk), Alistair Wright[2], Mark Stevens[2], Martin Hare[1], Simon Edwards[1], Matthew Back[1]

[1] *Agriculture and evironmental sciences, Harper Adams University, Newport, Shropshire, United Kingdom*; [2] *British beet research organisation, Norwich, Norfolk, United Kingdom*

Nematodes in the family Trichodoridae (*Trichodorus* and *Paratrichodorus* spp.), commonly known to as stubby root nematodes (SRN), are some of the most economically important free-living, plant parasitic nematodes in the UK. In alkaline sandy soils at Docking, Norfolk, and elsewhere in eastern England, sugar beet suffers a patchy stunting called 'Docking disorder', which is named after the village in Norfolk (UK), where it was first characterized. Seven species of SRN i.e., *T. primitivus* (de Man), *T. viruliferus* Hooper, *T. similis* Seinhorst, *T. cylindricus, T. teres* Hooper and *T. anemones* have been reported in fields where the docking disorder occurs. SRN feed on the root tips causing death of the taproot and thickening of the lateral roots, leading to a stubby /fangy root system. This leads to inability of the roots to uptake water and nutrients leading to poor root yields. The aim of this study was to assess the potential of biofumigants i.e., *Raphanus sativus* var. Longipinnatus-Daikon, *Brassica juncea* var. Brons and *Raphanus sativus*var. Terranova, for use in suppression of SRN. Biofumigants suppress pests through the release of isothiocyanates (ITC) following hydrolysis of their glucosinolates. This chemical process is exploited in biofumigation, which involves growing brassica cover crops until early flowering, and then chopping and incorporating the residues. A field experiment was conducted in SRN naturally infested site near Bury St. Edmunds, Suffolk. Treatments were randomly allocated into five blocks in a randomised complete block design (RCBD). A fallow control was also included in the study. Soil samples were collected before drilling of the cover crop (Pi), four weeks after cover crop drilling, shortly after incorporation of brassica residues and six weeks post incorporation (Pf). Soil sub-samples of 200 ml were processed using centrifugal floatation method with magnesium sulphate heptahydrate as the extraction fluid. Nematodes were then quantified under a compound microscope at 10X magnification to distinguish genera in the family Trichodoridae. Results showed that SRN densities were significantly lower ($P<0.001$) in plots with biofumigants compared to the fallow control, four weeks after planting. SRN reduction of 27% in plots with Daikon and 70% in plots with Brons and Terranova was observed at the end of the experiment (Pf). In conclusion, biofumigant cover crops were shown to have potential in the suppression of stubby root nematodes.

Keywords: Cover crops - Stubby root nematodes - Isothiocyantes - Biofumigation - Docking disorder.

References:

- Whitehead, A.G., and Hooper, D.J. (1970). Needle nematodes (Longidorus spp.) and stubby-root nematodes (Trichodorus spp.) harmful to sugar beet and other field crops in England. Ann. Appl. Biol 339–350.
- Gibbs, A. J. (1959). Docking disorder. Plant Pathology, 8(3), 93-94.
- Ngala, B.M., Haydock, P.P.J., Woods, S., and Back, M.A. (2014). Biofumigation with Brassica juncea , Raphanus sativus and Eruca sativa for the management of field populations of the potato cyst nematode Globodera pallida.
- Kruger, D. H. M., Fouri, J. C., & Malan, A. P. (2013). Cover crops with biofumigation properties for the suppression of plant-parasitic nematodes: A review. South African Journal of Enology and Viticulture, 34(2), 287–295.
- Matthiessen, J., & Kirkegaard, J. (2006). Biofumigation and enhanced biodegradation: Opportunity and challenge in soilborne pest and disease management. In Critical Reviews in Plant Sciences 25(3), 235–265.

S5-P35

Vertical management zones for enhancing yield and nematode control in Florida strawberry.

Joseph Noling (jnoling@ufl.edu)

Entomology and Nematology Department, University of Florida, Lake Alfred, Florida, USA

The presence of a subsurface traffic pan (a dense, highly compacted soil layer), was determined using soil penetrometers using sensors measuring differential frictional resistance with soil depth. This subsurface traffic pan was observed to restrict hydraulic conductivity and movement of fumigant gases. The objectives of these studies were to determine whether fumigant treatment of zones above and below the traffic pan, different vertical zones, is critical for enhancing nematode control and strawberry yield in Florida. Methods: Since 2015, multiple field studies were initiated on an annual basis to demonstrate the importance of deep fumigant placement and need for considering nematode control as a composite of vertical management zones. Soil samples utilizing two different tractor mounted hydraulic soil coring probes was used to characterize the spatial distribution of plant pathogenic nematodes in commercial strawberry fields to a depth of 122 cm. Sting and root-knot nematode were repeatedly shown to inhabit very deep soil profiles, below the traffic pan and well below the depths to which any of the current shank or drip applied fumigants diffuse. To target deep soil profiles, new fumigant application system was developed to make deep shank fumigant applications. Fumigants were generally applied in a 2 step, sequential process consisting of targeted delivery of fumigants to the two different soil depths. As a prebed treatment, the deep shank unit injected Telone II (15-18 l/ha) to a depth of 40 cm to the flat which was then immediately followed by a separately applied Telone C35 (46 L/ha), PicClor60 (38 L/ha), PicClor80 (35.2 L/ha), or Pic100 (33 L/ha) applied in-the-30 cm raised plant bed during the bedding operation. Soil population densities of sting and or root-knot nematode were generally significantly lower at seasons end with the deep shank 1,3-D applications compared to the bed applications alone. When a fumigant was applied to the raised plant bed, supplemental deep shank treatments of 1,3-D typically increased strawberry yields by 10 to 34%. Our results suggests that nematode damage potential to the crop occurs from migrating individuals from soil depths below which fumigants distribute. Adoption of these new technologies have largely resolved, non transplant nematode problems in Florida strawberry.

Keywords: Fumigant - Integrated Nematode Management - Chemical control.

S5-P36

Invasion and reproduction of *P. penetrans* on 'Maris Peer' potatoes.

Valeria Orlando[1] (vorlando@harper-adams.ac.uk), Simon Edwards[1], Thomas Prior[2], Roy Neilson[3], Matthew Back[1]

[1] *Harper Adams University, Newport, United Kingdom*; [2] *Fera Science Ltd., York, United Kingdom*; [3] *The James Hutton Institute, Dundee, United Kingdom*

The potato, *Solanum tuberosum*, represents one of the most important crops in Great Britain with 'Maris Peer' being the third most commonly grown cultivar. However, there is no information about damage of root-lesion nematodes on UK potato cultivars. Two controlled enviroment experiments investigated the impact of *P. penetrans* on the yield and quality loss of 'Maris Peer' potatoes. Experiment 1 examined the impact of mixed juveniles and adults of *P. penetrans*, ranging from 0.125 to 4 nematodes g-1 soil. Nematodes were detected within the roots of potatoes, revealing that *P. penetrans* was able to invade this cultivar but without affecting the yield at these population densities. Experiment 2 used a broader range of nematode densities, from 2 to 32 *P. penetrans* g-1 soil. As in the previous experiment, yield was not significantly affected by nematode densities, even though root lesions were present and nematodes were recovered from the roots of inoculated plants. In both experiments, all final population densities (Pf) were less than initial population densities (Pi), suggesting a limited reproduction on Maris Peer occurred.

Keywords: Controlled experiment - Cultivar - IPM - Pathogenicity - Root-lesion nematode.

S5-P37

Influence of Acibenzolar-S-methyl application on the penetration and development of *Pratylenchus brachyurus* in maize.

Heriksen Puerari[1] (heriksenhp@hotmail.com), Angélica Miamoto[2], Cláudia Regina Dias-Arieira[2]

[1] *Escola de Agronomia, Universidade Federal de Goiás, Goiânia, Goiás, Brazil*; [2] *Pós-graduação em Agronomia, Universidade Estadual de Maringá, Maringá, Paraná, Brazil*

Resistance induction is a promising method for the control of sedentary endoparasitic nematodes, but there is a lack of information about this method against migratory nematodes. The objective of this study was to evaluate the application of acibenzolar-S-methyl (ASM) on the penetration, development and multiplication of *P. brachyurus* in maize. For this, maize seedlings cv. Al. Bandeirantes with ten days of germination were inoculated with 800 nematodes. Two experiments were conducted in a completely randomized design in a 2 x 3 (experiment 1) and 2 x 6 (experiment 2) factorial scheme, with six replications. Treatments consisted of with or without ASM sprayed at the dose 0.5 g a.i. L^{-1}, combined with evaluation timings of 5, 10 and 15 days after treatment (DAT) (experiment 1) and 2, 4, 6, 8, 10 and 12 DAT (experiment 2). The ASM was sprayed on shoots the same day as the inoculation. At each evaluation timing the plants were removed, the roots washed and submitted to a staining process with acid fuchsin. The nematodes were counted and the length measured with an eyepiece reticule coupled to the optical microscope, in which the specimens were ranked in three groups: 0.10 to 0.29, 0.30 to 0.49 and 0.50 to 0.69 mm. At 30 DAT, the evaluation of the nematological variables was carried out. In relation to the nematode development, there was an increase in the proportion of specimens at an early stage (0.10 to 0.29 mm), and a lower proportion of specimens between 0.50 and 0.65 mm of length in plants treated with ASM compared to control. ASM reduced number of nematodes and eggs in the plant root system at 8 and 10 DAT and the final nematode population in maize roots at 30 DAT. Therefore, the product studied is capable of affecting the development and reproduction of the nematode.

Keywords: Alternative control - Resistance induction - Root lesion nematode - *Zea mays*.

S5-P38

Management of pepper varieties resistant to *Meloidogyne* spp. for nematode control in greenhouse pepper crops.

Caridad Ros Ibáñez (caridad.ros@carm.es), Carmen Mª Lacasa Martínez, Adriana Esteban López, Victoriano Martínez Alarcón, Mª Angeles Hernández Colucho, Mª Carmen Martínez Lluch, Alfredo Lacasa Plasencia

Crop Protection, Instituto Murciano de Investigación y Desarrollo Agrario y Medioambiental (IMIDA, La Alberca (Murcia), Spain

The pepper is a monoculture for more than 30 years in the greenhouses of the Region of Murcia. *Meloidogyne* spp. is one of the main soil pathogens. At present, most of the greenhouses are disinfected with 1.3 dichloropropene + chloropicrin which has an exceptional authorization of 120 days per year. Despite the disinfection of the soil, the problem caused by nematodes is difficult to control in some of them and it is necessary to use integrated control strategies. In a greenhouse contaminated by *M. arenaria*, management strategies of resistant commercial varieties (R) (Yazir 3 first years and Angus the 4th year, carrying the Me1 gene) in soil disinfected with 1.3 dichloropropene were evaluated to control the incidence of the nematode for 3 consecutive years. The experimental design was randomized blocks with three repetitions per treatment, taking as a reference the variety Tamarín (S) susceptible to the nematode, and in the third year Tamarin and plants grafted onto Robusto rootstock (Me1 carrier). The treatments tested were: in the first year i) repeated Tamarin crop (SS), ii) Resistant variety cultivated after Tamarín (SR) iii) Repeated resistant crop (RR); in the second year i) SSS; ii) SRS; iii) SRR, iv) SRR-soil without disinfection, v) RRS, vi) RRR. In the third year i) SSSS; ii) SRSR; iii) SRRR; iv) SRRR-no (where the last R is Robusto) on soil without disinfection iv) RRSR; v) RRRR. In the second year the cultivation of Yazir after Tamarín (SR) and its repetition, one (RR) or two years (RRR) reduced the incidence of the pathogen and the number of juveniles in the soil at the end of the crop (84.6 and 91.6%) with respect to the Tamarín. The cultivation of Tamarín after 1 or 2 years of Yazir (SRS and RRS) experienced similar damage to that of repeated Tamarín (SSS). On soil without disinfecting the Yazir variety (SRR-no) showed an incidence of the nematode higher than the crop in disinfected soil (SRR) however the population of nematode in the soil was reduced between 93.6 and 96.6% with respect to Tamarin (SSS). In the 3rd year, Angus plants (RRRR) reduced the number of juveniles in the soil at the end of the crop only 32% and showing high gall index, while the alternation of resistant (Angus) and susceptible variety (SRSR) or the resistant rootstock (SRRR-no) reduced them by 80 and 97% respectively, exhibiting low gall index.

Keywords: Greenhouse - Root-knot nematode - Capsicum annum - Me1 gene.

S5-P39

Biogas digestate as potential source for nematicides.

Andrea Ruthes[1] (andreacaroline.ruthes@agroscope.admin.ch), Erica Oldani[1], Alessandro Cabianca[2], Paul Dahlin[2]

[1] *Mycology/Plant Protection, Agroscope, Wädenswil, Switzerland*; [2] *Entomology and Nematology/Plant Protection, Agroscope, Wädenswil, Switzerland*

Tighter regulations on plant protection chemicals, growing popular demand for safer and greener alternatives to synthetic pesticides, and increasing pest resistance pressure, urge researchers to find innovative crop protection agents that can assist farmers and the agricultural industry in pursuing urgent sustainability challenges while meeting the growing global demand for food. Thus, we evaluated the potential of new plant protection products by screening active compounds generated during the biogas production process. Digestate, once considered as a low-value waste product, is commonly applied as organic fertilizer on arable land due to its high macro- and micronutrients content, and has been shown to play a role in inhibiting plant pests and pathogens, but its exact mechanism of action is not well understood. Assuming that the control effect is linked to bioactive compounds, liquid biogas digestates produced from two different substrates, a mixture of dairy cattle manure and agricultural wastes *versus* municipal bio waste, were evaluated. Both digestates showed great potential to control *Meloidogyne incognita*, the most widely distributed and economically important plant-parasitic nematode, *in vitro* and *in vivo*. A significant increase in J2 mortality (>50%) was observed after 24h exposure to diluted digestate (10% v/v), while >90% mortality was observed after 7 days exposure. Application of digestate at 10% and 5% dilution was also effective in preventing root galling in susceptible cultivars (tomato and cucumber). Liquid-liquid fractionation of potentially bioactive compounds was performed on the crude digestate, and the chemical nature of the isolated extracts was investigated by Gas Chromatography-Mass Spectrometry (GC-MS), giving insight into the chemical composition of the digestate itself. The present research is one of the first to introduce the prospect of digestate-based, eco-friendly industrial formulations for the control of plant parasitic nematodes, paving the way for a more detailed identification of nematotoxic molecules or metabolites in the digestate.

Keywords: *Meloidogyne incognita* - Nematicidal compounds - Biogas digestate - GC-MS.

S5-P40

Potential chemical control options for *Aphelenchoides besseyi* in ornamental plants.

Lindsay Wheeler (wheelm@ufl.edu), William Crow, Rachel Shepherd

Entomology and Nematology Dept., University of Florida, Gainesville, FL, United States

A well-documented pest among certain food crops such as rice and strawberry, foliar nematodes are now poised as an emerging problem in the ornamental plant industry. Many varieties of ornamental plant are documented hosts for these nematodes, and infestations can result in loss of sale value due to lesions on the leaves and reduced plant vigor. Few pesticides are currently labeled for the control of foliar nematodes on ornamental plants. In 2019, an experiment was conducted to assess the effectiveness of eight pesticides at suppressing nematode populations in chrysanthemums (*Chrysanthemum indicum* 'Olympia') inoculated with *Aphelenchoides besseyi*. Pesticides evaluated were commercial formulations of fluopyram, fluensulfone, abamectin, spirotetramat, chlorfenapyr, azadirachtin, acephate, and oxamyl. The experiment was conducted in a greenhouse using a randomized block design with 10 replications of 10 treatments. Treatments included the eight pesticide treatments, inoculated-untreated control, and non-inoculated-untreated control. At the conclusion of the trial, the plants were severed at the soil line and their top weight was recorded. Data was then collected on leaf mass alone, followed by hand-shredding of the leaves for nematode extraction in Baermann dishes. After extraction, the nematodes were collected and counted. Of the pesticide treatments, only oxamyl, chlorfenapyr, and spirotetramat yielded nematode counts per gram of leaf tissue that were significantly less than those of the inoculated-untreated plants ($P \leq 0.05$). As oxamyl can no longer be applied to ornamentals due to label restrictions, and chlorfenapyr already has a commercial formulation labeled for foliar nematodes, spirotetramat warrants further investigation as an option for foliar nematode management.

Keywords: Foliar nematode - Nematicide - Ornamental plant - ipm - *Aphelenchoides besseyi.*

S5-P41

Reaction of sorghum genotypes to *Pratylenchus brachyurus.*

Monique Thiara Rodrigues Silva[1] (moniquetrs@hotmail.com), Angelica Calandrelli[1], Angelica Miamoto[1], Claudia Regina Dias-Arieira[2]

[1] *Graduate Programme in Agronomy, State University of Maringá, Maringá, PR, Brazil*; [2] *Graduate Programme in Agricultural Sciences, State University of Maringá, Umuarama, PR, Brazil*

Sorghum is the fifth most important cereal crop in the world, and can be grown in multiple environments, being relatively more drought tolerant than other cereal grain crops [1]. However, this plant can be susceptible to *Pratylenchus brachyurus*, so it is necessary to know the reaction of cultivars in order to reduce the nematode reproduction. This study aimed to investigate the reaction of sorghum genotypes to *P. brachyurus*. The experiment was conducted in a greenhouse, in a completely randomized design, with eight replications per treatment. Each experimental unit consisted of one plant, sown in a pot containing 1 L of autoclaved substrate (soil: sand, 1:2). The treatments consisted of six sorghum genotypes (Nucover 100, Nugrain 320, Fox, Nugrain 430, Alvo and NTX S100), and soybean cv. M6410 IPRO as control. The plants were inoculated at seven days after germination with 2 mL of suspension containing 500 *P. brachyurus* and, 75 days after inoculation, the nematode reproduction was evaluated. Sorghum genotypes supported fewer *P. brachyurus* per gram of root than soybean, and the best results were observed with Nucover 100, Nugrain 320, Fox and Alvo genotypes, which differed from the Nugrain 430 and NTX S100. Reproduction factor (RF) in the treatments Nucover 100, Nugrain 320, Fox and Nugrain 430 ranged from 2.23 to 3.10, being lower than the control (4.58). The treatments Alvo (3.51) and NTX S100 (4.18) supported similar reproduction to the control. We concluded that all sorghum genotypes were susceptible to *P. brachyurus;* however, the RF was variable. It is important to choose genotypes with lower RF to *P. brachyurus* for planting in infested areas.

Keywords: Root lesion nematode - Susceptibility - *Sorghum bicolor*.

References:

1. Stamenković et al., 2020. Renew Sust Energ Rev. 124: 1-14.

S5-P42

Measurement of soil mobility of Tymirium[R] nematicide using three different types of diffusion assays.

Jeffrey Simmons (jeff.simmons@syngenta.com)

Crop Protection / Nematology, Syngenta, Vero Beach, FL, United States

Objective: Measurement Of Soil Mobility Of Tymirium® Nematicide Using Three Different Types Of Diffusion Assays. Methods: To measure two-dimensional movement of Tymirium®, 5cm x 30cm plastic tubes were filled with sterilized soil and watered to 100% water holding capacity. Product application was made at 24hrs in 50mls of water per tube. At 24 and 48 hours after treatment, 50mls of water was added to each tube. At 72 hours after treatment, soil was removed from the tubes. Five centimeter sections of soil were mixed and placed in a small cups. *Meloidogyne incognita* eggs and a cucumber seedling were placed in each cup and three weeks later nematode root galls were counted. For three-dimensional movement of Tymirium®, 15cm x 30cm plastic tubes were filled with sterilized soil and watered to 100% water holding capacity. At 24 hours, product application was in 100mls of water per tube. At 24 and 48 hours after treatment, 100mls of water was added to each tube. At 72 hours after treatment application, soil was removed from tubes. Thirteen copper tubes side by side were inserted into the plastic tubes to remove all soil. Soil was pushed out of each copper tube and cut in 5cm sections. Each section was placed into a plastic cup. *Meloidogyne incognita* eggs were added and a cucumber seed planted. Three weeks later, nematode root galls were counted. For the last set of experiments, Tymirium® was applied in the field under plastic. We took soil core samples at the point of application (emitter) and, 2.5cm, 5.0cm, 7.5cm, and 10cm from the point of application. Bio-assays were conducted as described above using the top and bottom of each soil-core. Results: Two-dimensional experiments, Tymirium® had no galls down to 15cm. Three-dimensional experiments, Tymirium® had no galls laterally 4cm from application point and horizontally 15cm down. Field-experiments, Tymiriam® had no galls from the application point out to 5cm and down to 15cm. Conclusion: Tymirium® nematicide showed excellent activity towards *Meloidogyne incognita* both vertically and horizontally from the point of application. Results of greenhouse experiments were verified in samples generated in the field.

S5-P43

Evaluate plant effects, efficacy and yield benefits of Tymiriam® against *Meloidogyne incognita* on potato.

Jeffrey Simmons (jeff.simmons@syngenta.com)

Crop Protection / Nematology, Syngenta, Vero Beach, Florida, United States

Objective:
Evaluate Plant Effects, Efficacy and Yield Benefits of Tymiriam® against *Meloidogyne incognita* on Potato.
Methods:
Field studies were conducted on potato in a *Meloidogyne incognita* infested field. Tymiriam® was applied as an in-furrow drench application and seed treatment at 200 and 300ga/ha[GNU1]. Plant stand, vigor and phytotoxicity were assessed 14 and 28 days after planting. Nematode root galling and yield were assessed at harvest.
Results:
For in-furrow applications, Tymirium®, no reduction in plant stand and no plant phytotoxicity was observed. At 14 days after planting (DAT), there were no differences in plant vigor. At 28 DAT, all Tymirium® treatments showed statistically greater vigor than the check. All Tymiriam® treatments statistically reduced nematode root galling compared to the untreated check. All Tymiriam® treatments statistically yielded more than the untreated check. For seed treatment applications of Tymiriam®, there was no effect on plant stand, phytotoxicity, or plant vigor. All treatments showed statistically lower nematode root galling than the untreated check. For yield, all treatments were statistically higher compared to the untreated check.
Conclusion:
Tymiriam® statistically reduces nematode root galling and increases yield in potato when applied as an in-furrow drench or seed treatment. Tymiriam® doesn't cause any negative plant growth effects.
[GNU1]

S5-P44

Synergistic interaction of plant biomass and rhizobacteria for the management of *Meloidogyne* sp. on *Solanum lycopersicum*.

Nokuthula Sithole (nooku.nt@gmail.com), Shubhpriya Gupta, Manoj Kulkarni, Jeff Finnie, Johannes Van Staden

RCPGD, University of KwaZulu-Natal, Pietermaritzburg, South Africa

The root-knot nematode is one of the main global problems in vegetable producing areas, with higher losses in yield quantity and quality, contributing to food insecurity. The use of biological controls (BC) and organic soil amendments derived from plant material is a potential alternative to chemical control of nematodes. This study was conducted to co-evaluate synergistic interaction between the ground biomass of eight medicinal plants (GMP), nine isolated rhizobacteria and two plant growth promoting rhizobacteria (PGPR) as a management strategy for *Meloidogyne incognita*. When screened using a micro-dilution assay, *Vernonia colorata* and *Searsia lancea* showed inhibitory activity for isolated rhizobacteria and the PGPR. Under *invitro* conditions, *Bacillus licheniformis* (BL) significantly (P≤*0.05*) showed the highest nematicidal activity. However, to confirm synergism, a combination of BL+GMP were assessed under greenhouse conditions on nematode infested *Solanum lycopersicum* cv. Roma vf. seedlings. The seedlings were transplanted in sterile soil in pots and inoculated with a mixture of 2000 eggs+juveniles of *M. incognita*/pot. Seven days after inoculation, GMP and BL were applied sequentially. At day 56, *Cucurbita maxima* and *Merwilla plumbea* ground material increased the abundance of rhizobacteria in the soil rhizosphere of *S. lycopersicum*. *Pelargonium sidoides* and *Croton sylvaticus* ground material showed moderate stimulation properties; however, *S. lancea* demonstrated inhibitory activity against rhizobacterial populations. The combination of *M. plumbea+*BL, *C. sylvaticus+*BL, and *C. maxima+*BL significantly increased plant growth and reduced root gall formation and the nember of nematode juveniles in soil compared to the controls. While *S. lancea*+BL significantly decreased plant growth and increased root gall formation. All plant extracts showed no mutagenic activity against *Salmonella typhimurium* strains (TA 102 and TA 1535). Results indicated that *M. plumbea+*BL, *C. sylvaticus+*BL and *C. maxima+*BL possess promising synergism for future management of *M. incognita* in the production of *S. lycopersicum.*

Keywords: Phytonematicide - Plant-parasitic nematodes - Tomato - Bio-agent - Biodegradable.

S5-P45

An integrated approach to manage soybean cyst nematode: rotation of the resistant sources, compost, and cover crops.

Sita Thapa[1] (thapasit@msu.edu), Emilie Cole[2], Brian Levene[3], Marisol Quintanilla[2]

[1] *Michigan State University, East Lansing, Michigan, United States*; [2] *Michigan State University, East Lansing, United States*; [3] *Bayer Science, Dewitt, United States*

Soybean cyst nematode (SCN) is the most economically devastating pathogen of soybeans in the United States and most soybean growing countries worldwide. The most effective SCN management practice is the use of resistant cultivars. SCN, however, still causes significant yield loss due to the continuous use of cultivars derived from a single line of resistance, resulting in selection for virulent populations. A four-year field study (2017-2020) was conducted to evaluate the impact of six rotation systems with varieties derived from Peking/PI 548402 (PDV) and PI 88788 (8DV) on the SCN population development, virulence, and yield. SCN population levels were significantly lower in the PDV/8DV cultivar rotation in 2018 compared to the PDV/S cultivar rotation. In 2020, SCN population levels were the lowest in the 8DV/PDV/8DV/PDV cultivar rotation system and highest in PDV/PDV/PDV/PDV system. In 2017, there were no substantial soybean yield differences among the system, but for the remainder of the trial, the PDV/8DV rotations had the highest yields. Evaluation of other SCN management practices like the use of compost/manure and cover crops are still ongoing and had found mixed results by several researchers. From 2021 we had field trials monitoring SCN development when exposed to various composts (swine manure, poultry manure, and layer ash blend) and cover crops (cereal rye, clover, oilseed radish, and white mustard) near Monroe, Michigan. In addition, laboratory assays on the effect of various composts on SCN egg hatching and juvenile mortality are ongoing. To support the field trial results, greenhouse trials are also established to evaluate the use of cover crops in SCN management. The expected output of these trials is to design integrated SCN management strategies and reduce soybean yield loss caused by SCN worldwide.

Keywords: Soyean cyst nematode - Rotation of resistance source - Compost - Cover crops - Management.

References:

- An integrated approach to manage soybean cyst nematode: rotation of the resistant sources, compost, and cover crops. Thapa, Sita1, E. Cole1, B. Levene2, and M, Quintanilla-Tornel1.1Michigan State University, Department of Entomology, East Lansing, MI 48824.2Bayer Crop Science, Dewitt, MI.

S5-P46

Compatibility of Salibro™ and Vydate® with *Pasteuria penetrans* spore attachment to *Meloidogyne javanica* and *M. incognita.*

Eleni Nasiou[1] (lenanasiou@gmail.com), Tim Thoden[2], Iro Pardavella[1], Emmanuel Tzortzakakis[1]

[1] *N.AG.RE.F., Heraklion, Greece*; [2] *Corteva Agriscience, Munich, Germany*

Salibro™ is a novel sulfonamide nematicide containing the active substance (a.s.) fluazaindolizine. Its compatibility with *Pasteuria penetrans*, a bacterial parasite of plant parasitic nematodes, was assessed in populations of *Meloidogyne javanica* and *M. incognita*. Spores of a single *P. penetrans* isolate and a blend of six isolates, were incubated in suspensions of Salibro™ and Vydate® (oxamyl) for 1, 7 and 21 days. Thereafter, the suspensions were washed through a cellulose filter so as to remove the nematicide suspension and the spores which were retained on the filter were suspended in water. Juveniles were exposed in spore suspensions in Petri dishes and the number of attached spores was recorded. Neither Salibro™ at the higher tested dosage of 250ppm (a.s.), nor Vydate® at 50ppm (a.s.), had any adverse effect in the rate of spore attachment.

S5-P47

Soil health and nematicides: Considerations for integrated nematode management.

Tim Thoden[1] (tim.thoden@corteva.com), Marta Garcia[2], John Wiles[3]

[1] *Corteva Agriscience, München, Germany*; [2] *Corteva Agriscience, Indianapolis, United States*; [3] *Corteva Agriscience, Cambridge, United Kingdom*

Due to challenging climatic conditions, the rising lack of fertilizer resources and the increasing demand for sustainable agriculture, the role of healthy soils is expected to become more important in the future. Healthy soils have the capacity to lower the impact of climatic stress, reduce the extent of soil-borne plant diseases and pests as well as directly support plant growth via efficient natural nutrient cycles and improved soil structure. These benefits have been linked to well-developed soil food webs, the presence of natural antagonists of pests and diseases, as well as plant growth promoting microbes. The sustainable management of plant-parasitic nematodes, that currently mainly relies on the use of synthetic nematicides, could highly benefit from these naturally occurring antagonists and beneficial organisms. Consequently, we need to better understand how soil applied nematicides interact with organisms that contribute to the overall soil health and incorporate these interactions into the design of integrated nematode management approaches. In this presentation we will share recent data on the compatibility of Salibro™, a novel nematicide developed by Corteva Agriscience, with various beneficial organisms including free-living nematodes, nematode and disease suppressive soil fungi as well as beneficial soil bacteria. We will also discuss integrated nematode management in a broader context of the soil rhizosphere.

S5-P48

Host status of lavender and lavandin cultivars to *Meloidogyne incognita.*

Gulsum Uysal[1, 2] (gulsumuysal.gu@gmail.com), Demet Bayındır[2], Adnan Tülek[3], Zübeyir Devran[2]

[1] *Batı Akdeniz Agricultural Research Institute, Antalya, Turkey*; [2] *Faculty of Agriculture Department of Plant Protection, Akdeniz University, Antalya, Turkey*; [3] *Trakya Agricultural Research Institute, Edirne, Turkey*

Lavender (*Lavandula angustifolia* Mill.) and lavandin (*Lavandula*x *intermedia* Emeric ex Lois.) are aromatic, medicinal herbs that were originally grown in Mediterranean region and mostly used in perfumery, pharmaceutical and cosmetic industries. They are among the most produced essential oil plants in the world. However, lavender and lavandin are attacked by soilborne pathogens including plant parasitic nematodes. Root knot nematodes (*Meloidogyne* spp.) are one of the major limiting and damaging factors in lavender and lavandin plantations. This research was carried out in the year 2021 with the aim to determine the reaction lavender and lavandin species to *Meloidogyne incognita*. In this study, the responses of two lavandin cultivars (Abrial and Eğirdir clone) and two lavander cultivars (Raya and Sevtopolis), were evaluated for the S6 isolate of *Meloidogyne incognita* under controlled conditions [1]. The experiment was conducted with a 1000 second stage juveniles (J2s) inoculation level of *M. incognita* and randomized block design with five replicates. Sixty days after inoculation, plants were uprooted and roots were evaluated according to the number of egg masses and galls. All cultivars tested were susceptible to the S6 isolate of *M. incognita*. These findings could be useful in lavender and lavandin breeding and for integrated nematode management.

Keywords: Aromatic plant - *Meloidogyne incognita* - Response.

References:

- [1] Devran and Söğüt, 2009. Journal of Nematology. 41(2): 128-133.

S5-P49

RNAi-mediated *Minc03328* gene silencing for the management of *Meloidogyne incognita* in transgenic *Arabidopsis thaliana.*

Valdeir Junio Vaz Moreira[1, 2, 3] (valdeirjmoreira22@gmail.com), Isabela Tristan Lourenço-Tessutti[1], Marcos Fernando Basso[1], Maria Eugênia Lisei-de-Sa[1], Carolina Vianna Morgante[1], Bruno Paes de Melo[1], Fabrício Barbosa Monteiro Arraes[1], Maria Cristina Mattar da Silva[1], Janice de Almeida Engler[4], Maria Fatima Grossi-de-Sa[1]

[1] *Laboratorio de Interaçao Molecular Planta Praga (LIMPP), Embrapa Genetic Resources and Biotechnology, Brasília, DF, Brazil*; [2] *Laboratorio de Interaçao Molecular Planta Praga (LIMPP), Federal University of Brasilia, UNB, Brasília, DF, Brazil*; [3] *Laboratorio de Interaçao Molecular Planta Praga (LIMPP), Biotechnology Center, PPGBCM, UFRGS, Porto Alegre, RS, Brazil*; [4] *Nematology laboratory, INRAe, Université Côte d'Azur, CNRS, ISA, Sophia Antipolis, France*

Meloidogyne incognita is the most economically important species of root-knot nematode (RKN) and causes severe damage to crops worldwide. *M. incognita* secretes several effector proteins to suppress the host plant defense response, manipulate the plant cell cycle and other plant processes facilitating its parasitism. Different secreted effector proteins have already been identified in *M. incognita*, but not all have been characterized or have had the confirmation of their involvement in nematode parasitism in their host plants. Herein, we characterized the *Minc03328*(*Minc3s00020g01299*) effector gene, confirmed its higher expression in the early stages of *M. incognita* parasitism in plants, as well as accumulation of the Minc03328 effector protein in subventral glands and its secretion. We also discuss the potential simultaneous silencing of its paralogous *Minc3s00083g03984* gene. Using the *in planta* RNA interference strategy, *Arabidopsis thaliana* plants expressing small interfering RNAs (siRNAs) were generated specifically targeting and downregulating the *Minc03328* during nematode parasitism. Transgenic Minc03328-siRNA lines that significantly downregulated *Minc03328* gene expression during *M. incognita* parasitism were significantly less susceptible. The number of galls and egg masses, as well as the [galls/egg masses] ratio, were reduced in these transgenic lines by up to 85, 90, and 64 to 87%, respectively. Transgenic Minc03328-siRNA lines showed the presence of fewer and smaller galls indicating that parasitism was hindered. Overall, data herein strongly suggest that Minc03328 effector protein is important for *M. incognita* parasitism establishment. As well, the *in planta* Minc03328-siRNA strategy demonstrated the high biotechnological potential for developing crop species that might efficiently control the field.

Keywords: Crop-protection - Effector protein - In planta RNAi - New biotechnological tools - Plant-nematode interaction.

References:

- Rutter WB, Hewezi T, Abubucker S, Maier TR, Huang G, Mitreva M, Hussey RS, Baum TJ (2014) Mining novel effector proteins from the esophageal gland cells of Meloidogyne incognita. Mol Plant-Microbe Interact 27 (9):965-974.
- Lisei-de-Sá ME, Rodrigues-Silva PL, Morgante CV, de Melo BP, Lourenço-Tessutti IT, Arraes FBM, Sousa JPA, Galbieri R, Amorim RMS, de Lins CBJ, Macedo LLP, Moreira VJ, Ferreira GF, Ribeiro TP, Fragoso RR, Silva MCM, de Almeida-Engler J, Grossi-de-Sa MF (2021) Pyramiding dsRNAs increases phytonematode tolerance in cotton plants. Planta 254 (6):121. doi:10.1007/s00425-021-03776-0.
- Holbein J, Grundler FMW, Siddique S (2016) Plant basal resistance to nematodes: an update. J. Exp Bot 67 (7):2049-2061. https://doi.org/10.1093/jxb/erw005
- Vieira P, Gleason C (2019) Plant-parasitic nematode effectors - insights into their diversity and new tools for their identification. Curr Opin Plant Biol 50:37-43. https://doi.org/10.1016/j.pbi.2019.02.007
- Mukhtar MS, Carvunis AR, Dreze M, Epple P, Steinbrenner J, Moore J, Tasan M, Galli M, Hao T, Nishimura MT, Pevzner SJ, Donovan SE, Ghamsari L, Santhanam B, Romero V, Poulin MM, Gebreab F, Gutierrez BJ, Tam S, Monachello D, Boxem M, Harbort CJ, McDonald N, Gai L, Chen H, He Y, Vandenhaute J, Roth FP, Hill DE, Ecker JR, Vidal M, Beynon J, Braun P, Dangl JL (2011) Effectors Converge onto Hubs in a Plant Immune System Network.

S5-P50

ELISOL environnement: a private French structure specialized in nematology R&D: soil bio-indication and crop protection.

Cécile Villenave (cecile.villenave@elisol.fr), Anne Jimenez, Camille Chauvin, Manon Trambolho, Hélène Cérémonie

ELISOL environnement, Congénies, France

ELISOL environnement is a French company (South of France) created in 2011 which develops and markets innovative tools for in situ assessment of soil quality for natural or anthropized ecosystems, including agrosystems. Our studies are based on the use of nematodes as bioindicators to characterize the soil health. Nematodes can be used as tools for monitoring soil quality and helping decision-making for the management of polluted sites, for preservation or restoration, diagnosis and decision support tools for agricultural soil management (evaluation of the effect of agricultural practices, inputs) and urban green spaces. In fact, ELISOL environnement is developing internal and external research programs in nematology with public and private partners, on several axes.
Axis 1: Methodological developments: 1) benchmarks for interpretation of nematofauna analyses in bio-indication (DANE - funding PIA Biodiversity PME), 2) Methods in nematology using molecular biology and image analysis (Meninnov – funding Occitanie region and FEDER; partner, IRD Nefonev), 3) Meta-analysis of the effect of agricultural practices on soil nématofaune (IPANEMA – funding AFB; partner, IRD Eco&Sols).
Axis 2: Use of bio-indicators in different research contexts (funding ADEME): 1) Effects of pollutant cocktails on soil toxicity using bio-indication and ecotoxicological tests on *C. elegans* (APPOLINE and BIOTERA project: Leb Aquitaine transfer partner, BRGM), 2) Refunctionalization of urban wasteland by the creation of anthroposols (Bio-Tubes project; partners, Valorhiz, BRGM (poster), 3) Use of various organic residual products (PRO) in agriculture (Proterr project; partners, INRA, CIRAD).
Axis 3: Private partners (funding PIA): 1) BIODERA led by Centre Mondial d'Innovation (Groupe ROULLIER): development of biosourced nematostatic/nematicide products, 2) AgroEcoSol led by Auréa: development of the industrial use of several soil bio-indicators, 3) ELISOL environnement is also a laboratory for phytonematological analyses, a training organization in soil biology and a research and development center.

Keywords: Nematofauna - Ecotoxicological test - Biosourced nematicide products - Phytonematological analyses - Bio-indicators.

S5-P51

Inundation: an effective method to control the root knot nematode *Meloidogyne chitwoodi*.

Johnny Visser (Johnny.Visser@wur.nl), Leendert Molendijk

Field crops, Wageningen University and Research, Lelystad, Netherlands

The root-knot nematode *Meloidogyne chitwoodi* is an important endoparasite with worldwide distribution. *M. chitwoodi* has a wide host range including economically important crops such as potato, grain (wheat, barley), maize and carrots and therefore cannot be easily controlled by crop rotation. *M. chitwoodi* has been listed as a quarantine organism in the EU since 1998. Because of its quarantine status, propagation material like seed potatoes must be free of this nematode species. Very low densities (even below detection level) may cause infestation of seed potatoes. Therefore, seed potatoes and other propagation material can only be grown on *M. chitwoodi* free soil. Inundation is successfully used by bulb growers in the Netherlands to eliminate infestations of stem and root lesion nematodes. In 2015 a field experiment was started to measure the effect of inundation on *M. chitwoodi*. A field of thirteen ha that was naturally infested with *M. chitwoodi*, was flooded for fourteen weeks. In addition, retrievable bags with heavily infested soil were buried in the field soil. After the inundation period the field was sampled again. No *M. chitwoodi* individuals could be found; neither in the natural infested field soil nor in the bags with artificially infested soil. After the inundation treatment, in consecutive years sugar beet (*Beta vulgaris*, poor host), onion (*Allium cepa*, poor host) and potato (*Solanum tuberosum*, good host) were grown on this field. In October 2018, after harvest of the potatoes, the field was sampled again. Even after growing three host crops, including the very good host potato, no *M. chitwoodi* was found, indicating that the inundation fully eliminated the infestation of *M. chitwoodi*. These promising results were confirmed in 2018 when the effect of inundation on natural and artificial M. chitwoodi infestations was measured on an additional three arable fields. Inundation has proven to be most effective when performed in (late) summer (temperature >16 oC) and the field is flooded for at least 14 weeks. A mesocosm experiment was performed under controlled conditions to investigate the effect of temperature and of amendment of organic material as a possible means to accelerate the disinfestation process of inundation. Results of this experiment and opportunities for farmers to integrate inundation in their nematode control strategy for control of this quarantine nematode species will be discussed.

Keywords: Inundation - Root-knot nematode - *Meloidogyne chitwoodi* - Disinfestation.

S5-P52

Damage threshold and host-plant status of spinach (*Spinacia oleracea*) for *Meloidogyne chitwoodi* and *Pratylenchus penetrans.*

Lirette Taning[1] (lirette.taning@ilvo.vlaanderen.be), Leakena Chann[2], Nathalie Cap[3], Sander Fleerakkers[4], Ellen Formesyn[5], Wim Wesemael[1]

[1] *ILVO, Merelbeke, Belgium*; [2] *Ghent University, Gent, Belgium*; [3] *PCG, Kruisem, Belgium*; [4] *PSKW, Sint-Katelijne-Waver, Belgium*; [5] *Inagro, Rumbeke, Belgium*

Problems with plant-parasitic nematodes remain a major challenge in most field vegetable crops worldwide. In agricultural fields in Belgium nematode problems increased in recent years. In open field vegetable crops the root-knot nematode *Meloidogyne chitwoodi* and the root-lesion nematode *Pratylenchus penetrans* pose most threats. Knowledge on host plant status and damage threshold can help farmers in their decision on rotation schemes. Therefore, both host plant status and damage threshold of spinach cultivars for *M. chitwoodi* and *P. penetrans* was determined. The three most commonly grown Spinach cultivars (Gnu, Whale and Meerkat) in Flanders were tested in pot test and field experiments. For the pot tests a range of 11 nematode densities was used. The yield data and the final population densities of *M. chitwoodi* fitted to Seinhorst's yield and population dynamic models, respectively. The damage threshold value (*T*) for the relative fresh weight yield was 0.14, 1.08 and 11.50 second-stage juveniles (J2) per 100 cm³ soil for Gnu, Meerkat and Whale respectively. The minimum yield (*m*) of the three cultivars was 0.84, 0.64, 0.52 g and maximum yield (Ymax) was 12.51, 12.44 and 9.60 g, respectively. The maximum multiplication rates (*a*) were 2.40, 2.75 and 3.85, while the maximum population densities (*M*) were 1550, 1508, and 857 J2 per 100 cm³ soil for Gnu, Meerkat and Whale respectively. For *P. penetrans* we could not determine a damage threshold. The *a* values for *P. penetrans* were 1.06, 1.19, 1.08 and the M values were 967, 880, 1757 nematodes per 100 cm³ soil for Gnu, Meerkat and Whale respectively. For both, *M. chitwoodi* and *P. penetrans*, field experiments were done on two locations with a natural infestation. On the four fields we established a nematode gradient covering a low, moderate and high infestation. Both nematode species had a significant impact on the spinach yield. The higher the nematode density the lower the yield and *vice versa*. Based on the pot experiments, the tested spinach cultivars were good hosts for both nematode species. However, this was not shown in the field. Under field conditions the growing period of spinach is 40 to 60 days which might be too short for the nematodes to complete a generation. Crops with a short growing period limit population build ups and allow subsequent management options.

Keywords: *Meloidogyne chitwoodi* - *Pratylenchus penetrans* - Spinach - Host plant status - Damage threshold.

S5-P53

Field evaluation of sugarbeet varieties resistant to sugarbeet cyst nematode.

Becky Westerdahl[1] (bbwesterdahl@ucdavis.edu), Kelly Richardson[2], Oli Bachie[3]

[1] *Entomology and Nematology, University of California, Davis, Davis, CA, USA*; [2] *USDA-ARS, Salinas, CA, USA*; [3] *University of California, Cooperative Extension, Holtville, CA, USA*

Sugarbeet cyst nematode (SBCN, *Heterodera schachtii*) is found in more than 40 countries. Eleven percent of the sugarbeet growing area in the Imperial Valley of California has been shown to be infested. Breeding programs to develop sugarbeets resistant to SBCN have been in progress for many years. Several new varieties were tested by a partnership composed of UC Davis CE Specialists, USDA-ARS, Imperial County UC CE, UC Agriculture and Natural Resources (ANR) Sugarbeet Workgroup, UC ANR Desert Research and Extension Center, Growers, California Beet Growers Association (CBGA), Holly Sugar, seed companies, Pest Control Advisors (PCAs), and industry. A trial was established in the Fall of 2016 at the University of California Desert Research and Extension Center in Holtville, CA (DREC). The trial had 18 treatments with 6 replicates per treatment. A standard treatment of 1,3-Dichloropropene (1,3-D) at 74 liters/ha was applied on 27 September 2016. The trial was planted on 12 October followed by irrigation on the same day. The 1,3-D treatment and untreated control were planted to the susceptible variety SV401. The 16 varieties tested were: SV2012, SV2013, SV2014, SV2015, SV2016, BTS 5499, BTS 5460, BTS 541 N, BTS 566 N, BTS 52RR45, 14927-4-309E, P1518-411, P1407-311, P1528, N1512-446, and 14929-227(F). The trial was harvested on 12 April 2017. Beets were hand dug from each replicate and analyzed at the tare and sugar labs at Spreckels Sugar factory in Brawley, CA. Nematode samples were taken from each replicate and analyzed for juveniles of SBCN. At P=0.05, the following treatments had a greater yield than untreated: SV2014, SV2015, SV2016, P1407-311, BTS 5460, BTS 541 N, and BTS 566 N. At P=0.05, SV2014, P1518-411, 14929-227(F), BTS 566 N, and BTS 52RR45 had fewer SBCN juveniles in soil than untreated. Results demonstrated that these new varieties are resistant to cyst nematode and produce yields similar to those achieved following traditional fumigation.

Keywords: *Heterodera schachtii* - Resistant variety - Sugarbeet - Sugarbeet cyst nematode.

S5-P54

Sugar beet field storage, a possible source of sugar beet cyst nematode (*Heterodera schachtii* A.Schmidt, 1871).

Miloslav Zouhar[1] (zouhar@af.czu.cz), Marie Manasova[1], Jana Wenzlova[1], Ondrej Douda[2], Pavel Rysanek[1]

[1] *Department of Plant Protection, Czech University of Life Sciences Prague, Prague, Czech Republic*; [2] *Division of Plant Health, Crop Research Institute, Prague, Czech Republic*

Sugar beet cyst nematode *Heterodera schachtii* is one of the major pests of the sugar beet. The whole life cycle of the nematode takes place in the soil, nematodes infest roots and mature cysts often remain on the roots until harvesting of sugar beet. During harvesting machinery, infected bulbs and roots are transported et the field storage and there they are left in piles for up to several months. Before transport to the sugar factory, the bulbs are mechanically cleaned and fragments of infected bulbs, roots and soil remain in the field. This material is the source of nematode inoculum, which is normally spread on the free field without any treatment. The nematodes could be spread also to the new locality in this way. In the years 2019 - 2021, the occurrence of sugar beet cyst nematodes in sugar beet field storage was monitored in the area of the Czech Republic. Representative samples (10 kg) of plant material together with soil were taken from 24 sites. Samples were dried and homogenized by mechanical mixing. The sample of 100 g was processed by Fenwick can extraction method in 10 repetitions per site. Cysts of nematodes were manually counted and the viability of embryons was determined via light microscopy. The results were evaluated by software Statistica 12.0 (StatSoft, Tulsa USA) using of analysis of variance followed by the Tuckey test. The results show a statistically significant difference in the number of cysts and viable embryons at the different localities. No correlation between the locality and the number or presence of nematode cysts was found. In some cases, the number of nematode cysts significantly exceeds the economic threshold. The field storage sites pose a threat to the spread of nematodes. It should be noted that this storage can be used for monitoring nematode occurrence and also for their eradication. This work was supported by the project of the Technology Agency of the Czech Republic No. TH04030242.

Keywords: Sugar beet cyst nematode - Field storage - Sugar beet - Soil - *Heterodera schachtii*.

S5-P55

Evaluating new commercial cotton (*Gossypium hirsutum*) cultivars for resistance to *Rotylenchulus reniformis* and *Meloidogyne incognita*

Zane Grabau[1] (zgrabau@ufl.edu), Chang Liu[2]

[1] *University of Florida, Gainesville, FL, US*; [2] *University of Florida, Gainesville, FL, United States*

In the United States, *Meloidogyne incognita* and *Rotylenchulus reniformis* are major pests of cotton (*Gossypium hirsutum*). Recently, new tools for managing these nematodes have become available as commercial cultivars with dual resistance to both of these nematodes have been released. Resistance to *M. incognita* has been available, but commercially viable cultivars with resistance to *R. reniformis* had not previously been available. The objectives of this research were to evaluate these new cultivars for resistance to *R. reniformis* or *M. incognita* and agronomic performance, particularly in comparison to or combination with nematicide treatments. To complete these objectives, three small plot field trials were completed in northern Florida in 2021. In the first trial, resistant cultivar Deltapine 2141NR (DP 2141NR) and susceptible standard Deltapine 1646 (DP 1646) were tested in combination with commercial seed coating and in-furrow nematicides for *R. reniformis* management. In that trial, DP 2141NR was the most effective treatment for reducing *R. reniformis* infection. Among nematicides tested, only aldicarb showed efficacy at reducing *R. reniformis* infection. The susceptible cultivar DP 1646 had significantly better yield than the resistant cultivar DP 2141NR, with none of the nematicides significantly influencing yield. In the second trial, susceptible DP 1646, with or without in-furrow fluopyram, was compared with DP 2141NR for management of *M. incognita*. The resistant cultivar DP 2141NR was effective against *M. incognita*, significantly reducing infection and final root galling compared with DP 1646 without nematicide, while DP 1646 with fluopyram was effective at reducing infection and intermediate at reducing galling. Yield was not significantly affected by treatments. In the final trial, Phytogen 443 (PHY 443) resistant cultivar as well as Phytogen 444 (PHY 444) susceptible cultivar with or without fluopyram nematicide were tested against *R. reniformis*. The resistant cultivar PHY 443 was the most effective treatment for both reducing *R. reniformis* infection or populations and increasing cotton yield. In summary, new cultivars had effective nematode resistance, but varied yield performance in initial tests.

Keywords: Resistance - *Gossypium hirsutum* - *Rotylenchulus reniformis* - *Meloidogyne incognita* - Nematicide.

S5-P56

Potato as a catch crop in late summer to control Potato Cyst Nematodes.

Leendert Molendijk (leendert.molendijk@wur.nl), Johnny Visser

Field Crops, Wageningen University and Research, Lelystad, the Netherlands

The use of potato as a catch crop to control potato cyst nematodes (PCN) has been known for a long time (Carroll J. & McMahon, 1937). At the end of the last century, Wageningen University and Research investigated application of potato as a catch crop in the Netherlands and concluded this could be an effective and reliable measure (Molendijk & van Beers, 2005). In the Netherlands, the use of potatoes as a catch crop is officially recognized measure, by the Dutch Plant Protection Service, to control PCN. At the moment this method is only permitted in spring, based on the assumption that in the second half of summer dormancy could prevent hatching, causing ineffectiveness. The efficacy of potato as a catch crop in late summer was investigated in a greenhouse experiment in 2017 and a mesocosm experiment in 2018. In the greenhouse the susceptible cultivar Bintje was grown on three soils naturally infested with *Globodera pallida* for 40 or 90 days in July, August and September. Full hatching and reproduction occurred under greenhouse conditions and hatching was not hampered in late summer. In the outdoor mesocosm experiment the resistant potato cultivar Arsenal (Relative Susceptibility Pa3 = 2.53%) was used as a catch crop to investigate the effect of late planting (July 10th, July 27th and August 10th) and growth conditions (two test locations; Lelystad and Valthermond) on the population density of three *G. pallida* infested soils (three populations). Forty days after planting the potato plants were killed with the herbicide glyphosate. Just before planting and four weeks after killing the plants soil samples were taken to determine the effect on the population densities of *G. pallida*. The late summer applications were effective, diminishing the population levels beween 50% up to 96%. Planting date had a significant effect on hatching. Potatoes planted on 27th of July reduced PCN populations most (84%). The late planting date of August 10th was less effective, decreasing the PCN population by 64%. No effect of the test location was observed. One (virulent) population multiplied on the resistant cultivar Arsenal, indicating that in late summer, a growth period of forty days of potato as a catch crop is too long. Applied field research and a search for an alternative for glyphosate is needed to develop potato as a catch crop in late summer into an effective and reliable method to control PCN.

Keywords: Catch crop - Potato - *Globodera pallida.*

References:

1. [1] Carroll J. & E. McMahon, 1937. Journal of Helminthology, Vol XV, No 1 pp21 – 34.
2. [2] Molendijk, L. P. G. , T. G. v. B., 2005. Projectreport https://edepot.wur.nl/120333.

S5-P57

Root preservation in epoxy resin to highlight in-season treatment response in potato

Justin Luangkhot (Justin.luangkhot@bayer.com), Kyle Strachila

Bayer Crop Science Division, Kennewick, Washington, United States

Meloidogyne chitwoodi infested potato tubers cause significant yield losses in the Columbia Basin. Unmanaged *M. chitwoodi* populations will continue to grow and increase economic losses. The use of nematicides to manage nematode populations is an accepted practice to mitigate these losses. Infested roots were harvested to highlight in-season treatment response to Velum Prime (fluopyram) in potatoes. *M. chitwoodi* infested potato roots were harvested 83 days after planting from research plots in Connell, Washington, USA. Once rinsed, the roots were flattened between two sheets of expanded metal to keep roots in place and left to dry via continuous forced air for 2 weeks. Once drying of the root systems was complete each root system was coated in 3 layers of clear coat spray, which was to prevent air bubbles in the finished product. Epoxy castings were done in custom built molds that were made of melamine board and large enough to accommodate the root system to allow for even sizing. Silicone sealant was used at each point of contact for the boards to keep liquid epoxy from leaking, and all-purpose acrylic adhesive seaming tape was used to aid in releasing the finished epoxy block from the mold once cured. Once custom molds were complete, epoxy resin was mixed to begin the casting process. A single first layer was poured and allowed to cure before placing the roots on the layer. More epoxy was poured over the root system to encase the roots. Once this layer was cured a final layer of epoxy was poured to finish the block. A plumbers' torch was used after each layer of epoxy was poured in order to release any air bubbles on the surface of the epoxy and to draw other bubbles from inside the epoxy to the surface. After the epoxy blocks were cured, they were removed from the custom boxes. All sides of the blocks were wet sanded to enhance the appearance from all sides. The results of the epoxy blocks highlight the importance of the in-season nematicides to reduce economic losses.

S5-P58

Nematode population dynamics in tomato nethouses over a three year period

Francinah L. Matlala[1] (u10238663@tuks.co.za), MS Daneel[2], H. Fourie[3]

[1] *Natuurboedery Research Centre, ZZ2 Bertie van Zyl, Mooketsi, South Africa;* [2] Agricultural Research Council – Institute for Tropical and Subtropical Crops, Nelspruit, South Africa; [3] *Unit for Environmental Sciences and Management, North-West University, Potchefstroom, South Africa*

Continued monocropping by commercial farmers under protected structures such as nethouses provide ideal environmental conditions for the crop, resulting in prolonged harvesting period and good quality fruits. However, under these structures, pests and diseases including plant-parasitic nematodes, become difficult to manage. The microclimate supported by the nets and mulch make the tomato crop vunerable to attack by nematodes. Tomatoes grown under nethouses at ZZ2 farms were suveryed for three consecutive years, investigating nematode population dynamics, from six nethouses located at three different climatic regions. Collected data was subjected to ANOVA using Statistica 10 and Principal Component Analysis (PCA) using ADE-4.
Infestation levels fluctuated between the years, however, those of *Meloidogyne* significantly increased irrespective of location or climate from the first year to the third year for all nethouses included. Nethouses differed also significantly between each other concerning the levels of *Meloidogyne* infestations. Chemical soil analysis indicated that nethouses with high K, Ca, pH and acid saturation had lower *Meloidogyne* numbers while those with higher Mg:K and Na:K were related with higher *Meloidogyne* densities. *Meloidogyne* densities were higher in sandy soils, where higher rainfall occurred and where higher humidity levels prevailed while planting time seemed to have less influence on *Meloidogyne* densities. Rootstocks also seemed to play a role in nematode infections with rootstocks RS2, RS3 and RS13 having lower *Meloidogyne* densities compared to RS11 and scion ZZX132. Another observation was that according to PCA, *Meloidogyne* and *Helicotylencus* were found opposite one another, indicating that when Helicotylenchus was present, *Meloidogyne* numbers were lower.

Keywords: Spiral nematodes - Root-knot nematodes - Monocropping - Climatic variation.

POSTERS

S6. Legal and regulatory aspects of nematode management

S6-P01

Method validations for reliable plant-parasitic nematode diagnosis: the example of *Heterodera glycines* identification.

Aude Letort (aude.letort@anses.fr), Sylvie Gamel

Nematology unit, Anses - Plant Health Laboratory, Le Rheu, France

The plant health regulations in force in the different countries worldwide require reliable analytical methods to identify regulated pests. Although several diagnostic methods for the specific identification of plant-parasitic nematodes (PPNs) have been published, an evaluation of the different existing methods is necessary to determine which ones are the most specific and sensitive in order to adopt their implementation for the purpose of the analysis (e.g. surveillance survey, outbreak management and import control). As *Heterodera glycines* is a regulated nematode species in several countries and, therefore prohibited for trade, an assessment of 4 different identification tests published [1, 2, 3, 4] was carried out to evaluate their performance criteria. The published tests were performed by PCR and real-time PCR techniques. Specificity was first evaluated to distinguish tests that detect target species only. Sensitivity, repeatability and reproducibility have been determined only for the most specific ones according to EPPO recommendations (PM7/98). Up to 15 *Heterodera* species and 36 populations were tested in this study. Some of the PCR tests confirmed their specificity only for the *H. schachtii* group and could be used as a screening method for cyst detection instead of performed by morphology. The published real-time PCR assays are the best performing for the *H. glycines* specific identification. These results also demonstrated that assessment is useful if a large number of nematode populations of target species and genetically related non-target species are available. This study also showed the necessity of using different nematode populations for the development and assessment of diagnostic methods to improve the reliability of the diagnostic results.

Keywords: Specificity - Sensitivity - Assessement - Diagnostic method - Performance criteria.

References:

- [1] Ko et al., 2019. Plant Pathology Journal. 35(6): 654-661.
- [2] Ye, 2012. Journal of Nematology. 44(3): 284-290.
- [3] Ou et al., 2008. Nematology. 10(3): 397-403.
- [4] Subbotin et al., 2001. Nematology. 3(4): 365-371.

S6-P02

Study of Cyclobutrifluram mode of action using on *Caenorhabditis elegans*.

Fariba Heydari[1, 2] (fariba_heidari70@yahoo.com), Majid Pedram[2], Chantal Wicky[1]

[1] *Department of Biology, University of Fribourg, Fribourg CH-1700, Switzerland;* [2] *Department of Plant Pathology, Faculty of Agriculture, Tarbiat Modares University, Tehran, Iran*

Today, human and veterinary medicine, as well as agriculture are challenged with parasitic nematode infections. Resistance is developed against currently available drugs. Most nematicides affect nervous system functions and the ability to reproduce. However, their mode of action is still unknown. There is an urgent need to study the effects of these chemicals on both target and non-target organisms. *Caenorhabditis elegans* has been successfully used as model organism in toxicological and pharmaceutical studies, in particular to test anthelmintic drugs and nematicides. So, we are using *C. elegans* wild type worm and other strains as well as stress resistant mutants. New nematicide that formulated by Syngenta company named Tymirium by active compound of cyclobutrifluram was used. In this study, we observed a significant increase in the number of apoptotic germ cells in exposed nematicide compared to controls in worms. Also, in enzyme expression SDH (sdha-1:: GFP) strain and sdhb-1 (that approximately L2 arrest), there are not significant difference between exposed worms on nematicide and control. In high concentrations, reproductive rate (Brood size) and survival rate are decrease. To get access to the affected genes and their regulatory pathways, next generation sequencing will be used to establish the transcriptional repertoire of the treated worms. Finally, data on nematicide mechanisms of action are mostly at physiological levels, and studies at genomic, transcriptomic, and/or epigenetic level remain to be done.

Keywords: Next generation nematicide - RNA-Seq - SDH subunits - Tymirium

S6-P03

Development of appropriated measures and methods to close pathways for the distribution of cyst nematodes.

Stephan König[1] (stephan.koenig@julius-kuehn.de), Matthias Daub[2], Beatrice Berger[3]

[1] *National and International Plant Health, Julius Kühn-Institute, Braunschweig, Germany*; [2] *Plant Protection in Field Crops, Julius Kühn-Institute, Elsdorf, Germany*; [3] *Plant Protection in Field Crops, Julius Kühn-Institute, Braunschweig, Germany*

The dissemination of soil tares from the potato processing industry is a main means for the spread of the potato cyst nematodes (PCN) to non-infected areas. According to the German regulation KartKrebs/ KartZystV annex 2 (to § 14), processed soils have to be treated to exclude contamination with viable PCN before soils are reused for any agricultural purpose. Currently, heat treatment, fumigation and soil inundation are proposed measures, but other potential treatments may be considered as efficient, too. Our project GlobRISK aims to validate and improve treatment procedures for disinfestation of tare soil from potato cyst nematodes. Here we present results on (a) microwave treatment and (b) soil inundation of PCN in soil tares. For all experiments, one sample consisted of 150 impeccable *Globodera pallida* cysts from our reference population «Kalle" enclosed in a mesh bag. The microwave treatment was conducted in a microwave tunel oven with a band-belt system. For this, the sample bags were inserted in potato soil tare and exposed to temperatures of 80 °C. The soil inundation experiments were conducted in ponds of a sugar factory. Cyst-containing bags were submerged within a solid construction in the ponds. Some bags were harvested every three weeks over a period of 90 days. In preliminary studies, a hatching assay with potato root diffusates (PRD), a trehalose assay and staining solutions (Meldola's blue and malachite green) for detecting nematode viability were compared and validated. Then, juvenile survival was determined based on the reproductive rates in a bioassay using the highly susceptible potato cultivar 'Desireé' that was inoculated with treated cysts. The hatching tests with PRD gave the most reliable results as testing method for viability. The microwave exposure of PCN within tare soil completely suppressed juvenile hatch at all tested exposure times. PCN from inundation ponds still showed some hatching activity after three weeks, but after six weeks hatching was no longer detectable**.** Both, short-term microwave treatment and 90 days soil inundation are potential measures to sanitise tare soil from PCNs. Outcomes of the experiments will be considered in the new EPPO Phytosanitary Standard on risks which result from transportation of soils associated with root crops and potatoes.

S6-P04

An experimental design optimized for rate-profiling of plant-parasitic nematodes in row crops.

Scott Moore[1] (scott.moore@syngenta.com), Amy Holm[2], Craig Eckel[3], Shannon Morsello[3], Jon Hamill[3], Matthias Gaberthueel[4]

[1] *Syngenta Crop Protection, LLC, Mer Rouge, LA, United States*; [2] *Syngenta Crop Protection, LLC, Stanton, MN, United States*; [3] *Syngenta Crop Protection, LLC, Greensboro, NC, United States*; [4] *Syngenta Crop Protection AG, Basel, Switzerland*

Field research on plant-parasitic nematodes in crops is well-known for being challenging, with a relatively low rate of trial success. The spatial distribution of nematodes within a field can be extremely variable, and traditional statistical designs often miss true population differences. Nearest neighbor trial design and analysis was evaluated for use in row crop nematicide testing. This strategy enhanced the success rate of nematicide trials. In a series of trials conducted from 2015 – 2019, the nearest neighbor trial design was compared to a traditional randomized complete block design for the ability to detect significant differences ($p<0.1$) between nematicide treatments and a non-nematicide control. In row crops, such as cotton and soybean, the nearest neighbor trial design resulted in a success rate of 89% compared to a 23% success rate in the randomized complete block design. This has led to an increase in efficiency and a greater return on investment of nematode trials in row crops. It has also catalyzed changes in trial design for other cropping systems, such as vegetables, and pests and diseases, such as Soybean Sudden Death Syndrome.

Keywords: Plant-parasitic nematodes - Nematicide - Trial design.

S6-P05

Detection and distribution of *Pratylenchus* spp. in UK potato fields.

Valeria Orlando[1] (vorlando@harper-adams.ac.uk), Simon Edwards[1], Thomas Prior[2], Roy Neilson[3], Matthew Back[1]

[1] *Harper Adams University, Newport, United Kingdom*; [2] *Fera Science Ltd, York, United Kingdom*; [3] *The James Hutton Institute, Invergowrie, Dundee, United Kingdom*

The potato (*Solanum tuberosum*) is an herbaceus plant that hosts various plant-parasitic nematodes including root-lesion nematodes (*Pratylenchus* spp.). In the UK, potato cyst nematodes are well documented to cause severe damage on different potato cultivars, but less is known about the significance of *Pratylenchus* ssp. The aim of this study was to assess the presence and distribution of root-lesion nematodes in UK potato growing land. The top fifteen counties with highest area of potato production were selected for sampling to produce 200 sites overall. The number of field sites per county was determined using a stratified method and soil samples were collected following potato harvest, between September and November, in 2017 and 2019. Nematodes were extracted from 200 g of soil from samples of the first year and then identified morphologically at species level. Root-lesion nematodes were detected in all counties and in 87% of soil samples, showing that they are widely distributed in UK potato fields. *Pratylenchus neglectus* and *P. crenatus* were the most prevalent species, often mixed together in the same sample, whereas *P. thornei* and *P. penetrans* were less frequently encountered. Additional nematode extractions, identification and quantification from samples collected in 2019 will provide the complete distribution of root-lesion nematodes in UK potato lands.

Keywords: Diagnostics - Plant-parasitic nematodes - Root lesion nematode - Sampling - *Solanum tuberosum*.

S6-P06

USDA regulations, decisions and operations for nematode management in the United States.

Tonia Quintero (tonia.g.quintero@usda.gov)

Animal and Plant Health Inspection Service, United States Department of Agriculture, Raleigh, NC, United States

Potato cyst nematodes (PCN) which includes *Globodera rostochiensis* (golden nematode) and *Globodera pallida* (pale cyst nematode), are major pests of potato crops. In 2006, *G. pallida* was detected in a routine exotic pest detection 'Cooperative Agreement Pest Survey' conducted jointly by Animal and Plant Health Inspection Service (APHIS) and the Idaho State Department of Agriculture (ISDA). In response to the detection, Canada, Mexico, and South Korea stopped the importation of Idaho potatoes, while Japan cut off the importation of chipping potatoes from the entire United States. *Globodera rostochiensis* was detected in 1941 in Long Island, NY. Furthermore, in 2006, *G. rostochiensis* was detected in Quebec, Canada. As a result of these actions, the Canada-United States Guidelines were developed to outline the phytosanitary measures for detection of PCN, provide guidance for the National Plant Protection Organizations on managment and release of fields in regulated areas and establish the requirements for the movement of regulated articles between the two countries.
Based on these guidelines, APHIS-Plant Protection and Quarantine (PPQ), and its various stakeholders established a cooperative domestic program with authorities derived from Federal regulations and State rules. With the formation of the cooperative PCN program, several goals were established. The goals were 1) to delimit the infestation, 2) prevent the spread, 3) restore lost foreign potato markets and preserve existing markets, and 4) eradicate PCN. By employing intensive soil surveys, the PCN infestation has been delimited to two counties in Idaho and eight counties in New York without evident spread beyond these areas. Currently, foreign markets have been restored. The USDA-APHIS continues efforts to eradicate PCN in the United States for safeguarding agriculture and trade.

Keywords: Eradication - Quarantine - Phytosanitary measures.

S6-P07

Molecular detection and distribution of root-knot nematode species in Florida

Gabrieli Riva (gabrieliriva@ufl.edu), Hung Bui, Mengyi Gu, Johan Desaeger

Entomology and Nematology, University of Florida, Wimauma, FL, United States

Root-knot nematodes (RKNs; *Meloidogyne* spp.) are widespread and known to cause considerable damage to several crops in the world. This is especially true in the subtropical sandy soils of Florida, USA. A survey to determine the RKN distribution in Central and South Florida was started in 2021. Among 95 samples collected, mostly from vegetable crops, 69 samples showed visible RKN infection. Species-specific primers were employed for the identification of *Meloidogyne* species and mitochondrial DNA was sequenced to confirm *Meloidogyne* species when needed. We identified five *Meloidogyne* species (*Meloidogyne arenaria, M. enterolobii, M. hapla, M. incognita,* and *M. javanica*) all of which can cause substantial economic damage to various crops in Florida. *M. incognita* and *M. enterolobii* were the most common, and were found in respectively 24 (35%) and 18 (26%) of the positive samples collected. *M. enterolobii* (*M. mayaguensis*) was first reported in Florida in 2004 and is considered a highly virulent RKN species capable of breaking RKN resistance in soybean, sweet potatoes and tomatoes. It is considered a major emerging threat to agriculture in the southeastern United States. Our survey is the first of its kind in Florida and clearly shows the widespread distribution of *M. enterolobii* in vegetable production in Florida. Accurate RKN species identification is critical for the development of integrated nematode management strategies for the management of RKN in Florida agriculture.

Keywords: Meloidogyne species - Species-specific primers - Molecular detection - Florida crops.

S6-P08

A cost-benefit and efficacy analysis of Meloidogyne management strategies in Mediterranean intensive horticulture.

Miguel Talavera (miguelf.talavera@juntadeandalucia.es)

Sustainable Plant Protection, IFAPA, Córdoba, Spain

Losses caused by phytoparasitic nematodes in crops depend directly on their soil densities at the start of the crop, so reducing their populations before planting is the main objective of nematological control. Efficacies in reducing *Meloidogyne* spp. soil populations of different RKN-IPM strategies, as agrochemicals, botanicals, soil solarization, biofumigation, soil steaming or soilless cultivation, were calculated based on multiple field trials carried out during 15 consecutive years. *Meloidogyne* soil densities were reduced by 78 to 87% after fumigation with 1.3-dichloropropene:chloropicrin or dimethyl-disulfide. Other chemical nematicides such as fluopyram, oxamyl, dazomet, fosthiazate, fenamiphos, azadirachtin, ethoprophos, abamectin and metam-sodium showed efficacies ranging from 51 to 64%, while garlic extract, ozone or hydrogen peroxide reduced *Meloidogyne* populations by 41 to 46% and chloropicrin alone and furfural showed less efficacy of 40%. The combination of solarization with organic manure (biosolarization) reduced soil nematode populations by 73%, an efficiency slightly lower than soil fumigation, but similar to that of other agrochemicals. An economical cost-benefit study, including social and environmental externalities, of several RKN-strategies (soil disinfestation methods, resistant cultivars and grafting, soilless cultivation) in Mediterranean intensive horticulture was performed and a comparison of them is presented.

Keywords: Meloidogyne - Horticulture - Soil disinfestation - Cost-Benefit.

S6-P09

Influence of relative humidity during drying on viability of *Globodera* cysts.

Evelyn van Heese[1] (e.y.j.vanheese@nvwa.nl), Anne Sophie van Bruggen[1], Jan Luimes[2], Toos Dekker[2], Loes den Nijs[3]

[1] *National Plant Protection Organization, National Reference Centre, Nematology, Wageningen, Netherlands*; [2] *NAK, Dutch General Inspection Service, Emmeloord, Netherlands*; [3] *Nassauweg 1, Wageningen, Netherlands*

Potatoes are one of the main crops in the Netherlands, the total area used for potato production (seed, starch and ware) covers around 175.000 ha. Monitoring the presence of potato cyst nematodes (PCN, *Globodera pallida* and *G. rostochiensis*) is essential in such an intensive growing system. Official control of PCN is warranted by legislation, EU Council Directive 2007/33/EC [1]. Due to this legislation ±140.000 soil samples of around 42.000 ha of seed potatoes have to be analysed for PCN every year. Furthermore, surveys are carried out on ware potato fields. This large number of samples cannot be extracted directly, storage before processing is needed. Before the cysts can be extracted it is necessary to dry the soil samples. Currently samples are dried at room temperature (RT) at relative humidity (RH) of 40% or higher, as lower than 40% might influence the viability [2]. In particular, wet soil samples do not dry quickly under these conditions, it takes about 7 days. When lowering the RH, these samples will dry faster. To determine the influence of drying at lower RH on the viability of *Globodera* cysts, open tubes containing 20 *G. rostochiensis* cysts each, were randomly distributed over six different treatments, n=10 (Not treated, RH 10%, RH 20%, RH ≥40%, RH ≥50% and cold storage). The viability of the non-treated cysts was assessed immediately. The other cysts were treated at the various RH's for 21 days at room temperature (except the cold storage). After the treatments the cysts were transferred into 1 ml water, crushed and stored at 2-8°C for two nights. Subsequently the viability was assessed visually by counting living eggs and second stage juveniles. The RH≥40% was considered as standard, as it is currently used. Variance analysis of the number of living second stage juveniles and eggs per treatment in percentage relative to the standard method showed no significant difference between the various treatments. Exposing *G. rostochiensis* cysts to a relative humidity of 10 to 50% at room temperature does not affect the viability. We assume this is also valid for *G. pallida*.

Keywords: Globodera - PCN - Viability - Relative humidity - Drying soil.

References:

- [1] Anonymous, 2007, Official Journal of the European Union, 156/12.
- [2] EPPO, 2013, Bulletin OEPP/EPPO Bulletin (2013) 43 (3), 471-495.

POSTERS

S7. Biological control of nematodes

S7-P01

Biological control of *Meloidogyne graminicola* in rice plant by cost-effective bionematicide formula.

Balawara Andika[1] (180210103042@mail.unej.ac.id), Iis Nur Asyiah[1], Sugeng Winarso[2], Lenny Widjayanthi[3], Dwi Nugroho[4], Kurniawan Firmansyah[5], Ankardiansyah Pandu Pradana[6]

[1] *Biology Education, University of Jember, Jember, East Java, Indonesia*; [2] *Soil Science, University of Jember, Jember, East Java, Indonesia*; [3] *Agricultural Extension, University of Jember, Jember, East Java, Indonesia*; [4] *Indonesian Coffee and Cocoa Research Institute, Jember, East Java, Indonesia*; [5] *Tiga Kreasi Bersama Limited Partnership, Bondowoso, East Java, Indonesia*; [6] *Plant Protection, University of Jember, Jember, East Java, Indonesia*

The root-knot nematode *Meloidogyne graminicola* has been reported to infect rice plants in Indonesia and cause high losses. This nematode infection is exacerbated by the increasingly difficult access to irrigation in some infected rice fields. The reduced water intake caused *M. graminicola* to grow more easily in the root area of rice [1]. In previous research, we have found a cost-effective bionematicide formula that can control *M. incognita* on tomato plants [2]. In this research, we evaluated the effectiveness of this formula for controlling *M. graminicola* in rice. This research was conducted in Bantul Regency, Special Region of Yogyakarta, Indonesia. Rice plants were planted on land infected by 135 *M. graminicola* per 100 g of soil, following a randomized block design pattern. We used 6 treatments and 5 replications, each replication consisted of 150 rice clumps. The treatments used were water control (T1), 5 g per plant of carbofuran active ingredient (T2), 1% bionematicide (T3), 2% bionematicide (T4), 3% bionematicide (T5), and 4% bionematicide (T6). The variables observed included plant growth, yields, and pathological variables. Data were analyzed using Analysis of Variance, and Tukey's Test with 5% α. The application of cost-effective bionematicide has a significant effect on plant growth and yield variables, and can reduce infection rates and nematode populations in the rhizosphere and roots. Plants that were given bionematicide treatment showed 3.7-8.4% higher plant height. When compared with the control, the bionematicide treatment gave 13.4-27.7% more panicles. The average seed weight per panicle after bionematicide application increased by 16.4-33.14%. Plant root length increased by 26.4-34.6%. In pathological variables, the application of bionematicides can reduce the scale of root damage (38.4-71.4%), the amount of *M. graminicola* in the soil (14.9-40.5%), and the number of *M. graminicola* J2 in the roots (19.5-42.6%). The treatment that showed the highest effectiveness was T6 (4% bionematicide), but the most efficient and effective treatment at the same time was T5 (3% bionematicide). The results of this research can be used as a reference for decision making to control *M. graminicola* using cost-effective bionematicide in Indonesia.

Keywords: Root-knot - biocontrol - Pseudomonas - Bacillus - yield.

References:

- [1] Nurjayadi et al., 2015. Jurnal Fitopatologi Indonesia. 11 (4): 113-120.
- [2] Asyiah et al., 2021. Biodiversitas. 22 (6): 3256-3264.

S7-P02

Parasitic nematodes of the harlequin ladybird, *Harmonia axyridis* in Hungary.

Luca Eszter Balog[1] (baloglucaeszter@gmail.com), Oleksandr Holovachov[2]

[1] *Dep. of Systematic Zoology and Ecology, Eötvös Loránd University, Budapest, Hungary*; [2] *Department of Zoology, Swedish Museum of Natural History, Stockholm, Sweden*

Harlequin ladybird, *Harmonia axyridis* (Coleoptera: Coccinellidae) is the most common invasive ladybird species in the world and most countries of Europe. It is native to East Asia and was introduced in Western Europe in the late 1990s for the biological control of agricultural pests. *Ha. axyridis* is a highly effective predator for aphids and can appear very quickly anywhere. It poses a threat to native ladybirds and causes economic loss and harm to humans. The European population of harlequin ladybirds was mainly affected by different natural enemies, but the co-infection of the parasites and parasitoids is understudied. This investigation aims to determine the natural enemies species are present in the Hungarian populations and whether potential biological control properties against the harlequin ladybird can be found in this insect host. In this study adult *H. axyridis* specimens were collected from 21 different places in Hungary. A total of 581 insect specimens were dissected under a stereomicroscope and screened for ecto- and endoparasites and parasitoids. Natural enemies were identified using light microscopy. The interaction between different parasites was analyzed using statistical methods. Investigations revealed that in Hungary, the collected harlequin ladybirds were infected by the parasitic nematode *Parasitylenchus bifurcatus* (Nematoda: Allantonematidae), the ectoparasitic fungus *Hesperomyces virescens* (Ascomycota: Laboulbeniales), and the parasitoid *Dinocampus coccinellae* (Hymenoptera: Braconidae). This is the first evidence of the presence of *P. bifurcatus* in Hungary. The nematode parasite was present in all localities where fungi-infected ladybirds appeared. The prevalence of recorded natural enemies varied by the locality: for *P. bifurcatus* it ranged from 1% to 8%; for *He. virescens* from 2% to 36%; and for the parasitoid it was 4%. Co-infection of fungi and nematode was found in samples from mountainous sites with a statistically significant positive correlation using Spearman's correlation. New parasite species have not yet been identified. A study on the multiparasitism of harlequin ladybirds has not yet been performed in Hungary. Our study area is located between two distant areas in Europe where co-infections had already been observed, indicating that this is probably not a sporadic phenomenon, but that parasitism of the harlequin ladybird by multiple natural enemies might be prevalent throughout the area.

Keywords: Harlequin ladybird - Potential biocontrol - Entomoparasitic nematode.

References:

- Balog et al., 2021, Redia, 104, 125-137.

S7-P03

The effects of root lesion nematodes (*Pratylenchus thornei*) on chickpea plant and rhizobium bacteria.

Tohid Behmand[1] (tohid.behmand63@gmail.com), Jens Berger[2], İbrahim Halil Elekcioğlu[3], Abdullah Kahraman[4]

[1] *Cukurova University, Plant protection department, Adana, Turkey*; [2] *Centre for Environment and Life Sciences, CSIRO Agriculture, Perth, Avustralia*; [3] *plant protection department, Cukurova university, Adana, Turkey*; [4] *field crops, Hrran university, Sanliurfa, Turkey*

Legumes like chickpea depend on nitrogen provided by the activity of rhizobium bacteria to grow, but they are also infected by parasitic worms called nematodes, which steal plant nutrients. The root lesion nematodes (*Pratylenchus thornei*) are common and economically important pests described as one of the major limiting factors in agriculture and the growing chickpea field in Turkey. The effects of this nematode on rhizobium bacteria activity and abundance were investigated in different wild chickpea genotypes under laboratory conditions. Here, we tried to test whether susceptibility to nematodes is one such tradeoff between legumes and nitrogen-fixing bacteria (rhizobia), and how do parasitic nematodes impact the rhizobium bacteria activity. In this study, we inoculated all *cicer* genotypes with *Pratylenchus thornei*. It was observed that chickpea genotypes differed in susceptibility to nematode infection, and between the number of rhizobia (nodules) and nematode number differed in infectivity. Finally, we observed that nematodes affect the rhizobium bacteria activity in the chickpea and nematode infection decreased nodule number and total nodule biomasses between 12-26% in the chickpea roots depending to the cultured and wild varieties.

Keywords: Chickpea - Rhizobium bacteria - Pratylenchus thornei - Genetic correlation.

S7-P04

Arbuscular mycorrhizal fungus mobilization against root-knot nematodes of tomato and pepper roots.

Renaud Canaguier[1] (canaguier@nixe.fr), Melvin Rodriguez[2], Laurent Lapeyre[2], Caroline Caporalino[3], Michel Ponchet[3], Marie Offroy-Chave[4], Sonia Braquet[2], Renaud Canaguier[2]

[1] Direction, NIXE, Sophia-Antipolis, France; [2] Nixe, Sophia-Antipolis, France; [3] INRAe-ISA, Sophia-Antipolis, France; [4] INRAe, Centre de Recherche Antilles Guyanne, Petit-Bourg, France

Developing biocontrol products and methods is one of the preferred ways to reduce the use of pesticides and to contribute to the protection of crops in a more eco-friendly way. Biocontrol is based on highly complex natural regulations such as interactions between plants and soil microorganisms. Designing new biocontrol practices and better understanding of regulatory processes are two major issues. Among the microorganisms of the soil, Arbuscular mycorrhizal fungi (AMF) establish symbiosis with the roots of 80% of the vascular plants and form networks that connect plants with one another (Common Mycorrhizal Networks or CMN). Mycorrhizae contribute to plant growth and bioprotection against various soil-borne pathogens such as root-knot nematodes that cause major damage globally on several crops. The role of mycorrhizal networks for bioprotection of plants is still not fully understood. Practices of direct inoculation and mobilization of the mycorrhizal network were evaluated for colonizing tomato roots and the bioprotective effects. Effects of AMF (*Rhizoglomus irregulare* BEG72 and *Funneliformis mosseae* BEG 234, (AEGIS - Italpollina France)) against root knot nematodes on tomato and pepper were analyzed. Under controlled conditions, the mobilization of a mycorrhizal network from a donor plant (e.g. sorghum) was faster than direct inoculation for tomato mycorrhization, and AMF, once established, promoted the bioprotection of pepper against root-knot nematodes (30% reduction of egg masses) and reduced the wilting symptoms on tomato. Analysis of potential processes showed that mycorrhization induced a modification of the root exudate content. Mobilization of the mycorrhizal networks has potential as an efficient way in integrated protection strategies, for the biocontrol of root-knot nematodes in vegetable crops.

Keywords: Common Mycorrhizal Network - Bioprotection - Tomato - Pepper - Sorghum - Root-knot nematodes.

References:

- Djian-Caporalino C. (2010). Nématodes à galles, des ravageurs de plus en plus préoccupants. Résultats de 3 ans d'enquête dans quinze régions françaises. Phytoma : La défense des végétaux, 638, 43-49.
- Djian-Caporalino C. (2012). Rootknot nematodes (Meloidogyne spp.), a growing problem in French vegetable crops. EPPO Bulletin, 42, 127–137.
- Djian-Caporalino C., Védie H., Arrufat A. (2009). Gestion des nématodes à galles : lutte conventionnelle et luttes alternatives. L'atout des plantes pièges. Phytoma : La défense des végétaux, 624, 21-25.
- Offroy-Chave M. (2014). Étude du potentiel de plantes mycorhizotrophes pour la gestion de la bioprotection de la tomate contre le flétrissement bactérien. DOCTORAT, SPÉCIALITÉ EN SCIENCES AGRONOMIQUES ET BIOTECHNOLOGIES ALIMENTAIRES. Université des Antilles et de la Guyane.

S7-P05

Symbiotic bacteria of entomopathogenic nematodes for the biocontrol of dagger nematode *Xiphinema index.*

Carlos Castaneda-Alvarez[1] (carlos.castaneda@uchile.cl), Simona Prodan[1], Ernesto San-Blas[2], Erwin Aballay[1]

[1] *Departamento de Sanidad Vegetal, Universidad de Chile, Santiago, RM, Chile*; [2] *Instituto de Ciencias Agroalimentarias, Animales y Ambientales (ICA3), Universidad de O'Higgins, San Fernando, O'Higgins, Chile*

Xenorhabdus and *Photorhabdus* are symbiotic bacteria genera of entomopathogenic nematodes (EPN) that have been evaluated for controlling several pests; however, their effect on plant-parasitic nematodes is not widely known. This study aimed to evaluate the effect of these bacteria on *Xiphinema index* under *in vitro* and semi-field conditions. A series of three bacteria isolated from *Heterorhabditis bacteriophora*, *Heterorhabditis atacamensis*, and *Steinernema unicornium*, identified in this study as *Photorhabdus* thracensis 31926 and two novel species *Photorhabdus* sp. nov (*under review*) UCH-936 and *Xenorhabdus lircayensis* VLS, respectively, were considered for nematicidal assessments. The bacteria isolates were recovered from EPN, cultivated in agar plates, and then grown in liquid media of Luria broth. Supernatants of bacteria were eliminated by centrifugation, then bacteria were resuspended in phosphate-buffered saline (PBS) and adjusted to 1 x 10^6 and 1 x 10^7 CFU mL^{-1} for laboratory and semi-field assays, respectively. Direct effect of the bacteria was evaluated on a population of *X. index* (25 individuals in 0.5 mL of sterilized water; juveniles 30 % and adults 70 %) in Petri dishes. A bacteria aliquot of 3 mL was added to each Petri dish, assessing at 24 h and 72 h the mortality of *X. index*. Cell bacteria (1 x 10^7 CFU mL^{-1}) were applied in the semi-field assay by 30 min-dipping grapevine roots in the bacterial suspension. Afterward, these plants were established in 5 L pots filled with naturally infested soil and immediately inoculated with 350 mL of the same bacterial suspension. A completely randomized design was used with five treatments, including two controls (PBS medium and water), five replicates (*in vitro*), and six (in semi-field). Bacteria were identified using the 16S rRNA gene and the *gyrB* housekeeping gene or whole genome in case of new species. The effects of the symbiotic bacteria on *X. index*, under *in vitro* conditions, were mainly nematicidal at 24 h. All bacteria results were significantly higher than control, reaching 100 % of mortality on *X. index*, after 72 h exposition ($p < 0.001$). Similarly, assays in semi-field conditions showed a significatively reduction of *X. index* population ($p \leq 0.05$). These symbiotic bacteria are good candidates for further assesses in field conditions. Likewise, additional analyses must be carried out to determine the mode of action, enzymes, and metabolites associated with their nematicidal aptitude.

Keywords: Nematode vector - Biological control - Symbiotic bacteria - Entomopathogenic nematodes - Grapevine fanleaf virus (GFLV).

S7-P06

New member of the Cytolysin A family : Cry6Aa controls nematocidal activity through glycosphingolipids.

Hanqiao Chen (chenhanqiao@webmail.hzau.edu.cn), Jinshui Zheng, Donghai Peng, Ming Sun

Huazhong Agricultural University, Wuhan, China

Pore-forming toxins (PFTs) are virulence factors produced by various pathogenic bacteria. PFTs promote pathogen infection, growth and dissemination, and play an important role in human health. Cry6Aa is a nematocidal crystal protein produced by *Bacillus thuringiensis* during sporulation. In structural analysis, Cry6Aa was identified as a new member of the classic α-PFTs Cytolysin A (ClyA) family. Cry6Aa has a similar structure as ClyA family toxins but has a different host specificity, especially similar to non-haemolytic tripartite enterotoxin (Nhe) and the B component of haemolysin BL enterotoxin (Hbl)[1]. The breadth of toxicity and identity of specific receptors of this family is generally unknown[2]. Recent studies have shown that cholesterol stabilizes the pore-forming intermediate state of ClyA toxin but it still does not explain this puzzling problem [3]. In our research with Cry6Aa-*C. elegans* as model, the absence of cholesterol and sphingosine lipid components resulted in significant resistance of *C. elegans* to Cry6Aa. At the same time, Cry6Aa can interact with the total lipid components of *C. elegans* origin, liposomes rich in glycosphingolipids and form a 12-mer complex. There is a key “β-tongue” segment on Cry6Aa that interacts with the cell membrane. Through the modification of this segment, the interaction of toxin proteins with specific membrane components and the formation of a multimeric state are different from ClyA were determined. This segment caused the disorder of the multimeric state of the toxin protein, resulting in loss of pore-forming ability. More details in the interaction of Cry6Aa and glycosphingolipids are being investigated. Our work can not only explain the confusion of receptor specificity in the ClyA family, but also provide guidance to excavate biological resources for nematodes control.

Keywords: Pore-forming toxins - Cry6Aa - Cytolysin A - Receptor - Glycosphingolipids.

References:

- [1] Jinbo Huang et al., 2016, Biochemical and Biophysical Research Communications, 478(1): 307-313.
- [2] Dal Peraro et al., 2016, Nature reviews microbiology, 14(2): 77.
- [3] Sathyanarayana P et al., 2018, Proceedings of the National Academy of Sciences, 115(31): E7323-E7330.

S7-P07

Interkingdom cooperation between rhizosphere bacteria and nematodes modulates the infectivity of plant-nematodes.

Feng Chen (fengmy@webmail.hzau.edu.cn), Ling Chen, Jinshui Zheng, Jianwei Shi, Donghai Peng, Quanrong Liu, Ming Sun

Huazhong Agricultural University, Wuhan, China

There are thousands of bacteria that colonized the plant rhizosphere and contribute to plant resistance to pathogens. It is reported that infection of plant-parasitic nematodes (PPNs) can change the structure and diversity of the hosts' rhizosphere bacterial community, but the relationship between nematodes and rhizosphere bacteria is still not clear. The aim of our study was to determine the influence of bacteria in the same niche on the infectivity of *Ditylenchus destructor*, an important PPN for global production of potato and sweet potato. Firstly, it was found that the infectivity of nematodes sterilized with antibiotics decreased 78.3% compared with unsterilized nematodes. With a filter the bacterial community in the same niche was obtained from the lixivium of infected tissue of sweet potato. In infection assays, co-inoculation of the bacterial community with the nematode complemented the reduced infection numbers of sterilized nematodes in sweet potato from 19.9% to 87.1%. Amplicon sequencing of 16S rRNA demonstrated that the presence of *D. destructor* altered the structure and diversity of bacterial community in the nematode-infected site. To determine the exact bacterial species that was responsible for enhancing nematode infectivity, 182 bacterial strains involving 20 genera were isolated from the bacterial community. In co-inoculation assays with sterilized nematodes, one strain (0130LB-2) increased the colonization number of sterilized nematode 4.73-fold. In additional infection assays of co-inoculation between 0130LB-2 and *D. destructor*, the isolated 0130LB-2 did not only recover the colonization ability of sterilized nematodes, but also additionally enhanced the infectivity of unsterilized nematodes by ~2-times. Furthermore, the enhancing effect on nematode colonization of 0130LB-2 depended on the bacterial abundance. Concomitantly, the relative abundance of 0130LB-2 in the infection site could be maintained by the presence of *D. destructor*. Thus, it is revealed that there is a mutualistic interaction between *D. destructor* and 0130LB-2 in the niche of sweet potato. Genomic analysis suggested that the mutualistic interaction might be achieved by metabolic complementary mechanism. Our work gives new insights into the relationship between PPNs and rhizosphere bacteria and will be helpful for the development of new strategies against PPNs.

Keywords: Plant-parasitic nematodes - Rhizosphere bacteria - Interkingdom cooperation - Mutualistic interaction - Nematode infectivity.

S7-P08

Effects of *Trichoderma* secondary metabolites on the fitness of Root-Knot Nematodes.

Diana Carvalho[1] (dpatriciafc@gmail.com), Sheridan L. Woo[2], Giada d'Errico[3], Martina Sinno[3], Isabel Abrantes[1], Isabel Conceição[1]

[1] *Centre for Functional Ecology - Science for People & the Planet (CFE), Department of Life Sciences, University of Coimbra, Coimbra, Portugal*; [2] *Department of Pharmacy, University of Naples Federico II, Naples, Italy*; [3] *Department of Agricultural Sciences, University of Naples Federico II, Naples, Italy*

Root knot nematodes (RKN), *Meloidogyne* spp., are considered among the economically most important plant pests affecting the production and quality of diverse crops. *Meloidogyne arenaria*, *M. hapla*, *M. incognita* and *M. javanica* have a wide geographic distribution, large host ranges and high destructive potential. Although chemical nematicides are widely used for their control, their impact on the environment and human health has resulted in enforced regulations or the total ban of various active substances. Thus, the efficacy of sustainable alternatives for nematode suppression must be further evaluated. Soilborne filamentous fungi such as *Trichoderma* spp. Are already used for biocontrol of fungal pathogens and could have potential for nematode suppression. The organisms are knonw to enhance plant growth and development that can also have possible applications in containing plant-parasitic nematodes. It has been demonstrated that metabolites produced by isolates of *Trichoderma* spp. have some effects against RKN (1). In this study, two secondary metabolites produced by *Trichoderma* spp., an hydrophobin (HYTLO1) and a 6 pentyl-alpha-pyrone (6PP), were evaluated for their impacts on *M. incognita* and *M. hapla.* Fitness parameters such as hatching and second-stage juveniles (J2) mortality when used in several concentrations (1 x 10-6 M ; 3 x 10-6 M ; 5 x 10-6 M ; 7.5 x 10-6 M) were measured. Infectivity was assessed, but only at the highest concentrations of both fungal compounds. For all the assays, 5 replicates were carried out and the experiments were repeated twice at different times. Results were analyzed using ANOVA from MiniTab 18. Hatching of *M. incognita* juveniles was more affected by HYTLO1, whereas that of *M. hapla* by 6PP. However, it was noticed that HYTLO1 had greater potential nematicidal activity towards the J2 of both species. In fact, at the highest concentration, 100% J2 mortality occurred after 24 h exposure to HYTLO1. Both molecules affected infectivity of *M. hapla* and *M. incognita*. Nevertheless, when exposed to HYTLO1, infectivity of *M. hapla* was reduced by 50% and that of *M. incognita* by 10% compared to the control.

Keywords: Biological control - Hydrophobin - Meloidogyne - Mortality - Infectivity.

References:

- [1] Al-Hazmi and TariqJaveed, (2016). Saudi J. Biol. Sci., 23, pp. 288-292.

S7-P09

Compatibility of selected pesticides with *Pochonia chlamydosporia*.

Rui P. Carvalho (pg35021@alunos.uminho.pt), M. Clara Vieira dos Santos, M. Teresa M. Almeida, Sofia R. Costa

CBMA - Centre of Molecular and Environmental Biology, Department of Biology, University of Minho, Campus de Gualtar, Braga, Portugal

Fresh market tomato, mainly produced in protected cultivation in Portugal, is susceptible to several pests and diseases. Root-knot nematodes (RKN), *Meloidogyne* spp., are widely distributed pathogens of tomato, but other organisms, such as late blight *Phytophthora infestans* and the tomato leaf miner *Tuta absoluta* are major concerns in these systems. The biology and ecology of *Pochonia chlamydosporia* (Pc) is well known, but its success as biological control agent is often compromised in the field by confounding factors not anticipated in laboratory conditions. In protected cultivation, conventional farmers undertake a thorough synthetic pesticide application plan to protect their crop not only from RKN but also from the attack of other pests and pathogens. The toxicity of these pesticides towards Pc is mostly unknown. We tested the *in vitro* effects of six fungicides, five insecticides and one nematicide, all applied along the same growing season in a conventional farm growing fresh market tomato in protected cultivation, on radial growth and chlamydospore viability of a promising Portuguese Pc isolate PE1 [1]. All fungicides, tested at range of 1 to 100 ppm active substance (AS) significantly inhibited radial growth of Pc in corn meal agar-amended medium in a dose-responsive manner, with the exception of a metalaxyl formulation. Radial growth was inhibited by two of the 5 insectides tested at the same range of concentrations, with results comparable to untreated control for insecticides containing AS methiocarb and spiromesifen. The nematicide formulation of oxamyl did not significantly affect radial growth of Pc. At 100 ppm AS, all fungicides and the nematicide inhibited chlamydospore viability when compared to control ($p<0.01$). Among the insecticides, formulations containing AS indoxacarb, methiocarb and spinosade did not significantly affect chlamydospore germination in sorbose agar medium at the tested concentration. Results suggest Pc success as a biological control agent may be compromised by pesticides that target pests and pathogens other than RKN in tomato crops under conventional management. Toxicity testing of pesticides is therefore of utmost importance in the selection and design of Pc-compatible crop protection management strategies.

Keywords: Fungal growth - Chlamydospore viability - Nematophagous fungus - Fungicide - Insecticide.

References:

- [1] Vieira dos Santos et al., 2019. Phytopathol Mediterr 58(1): 187-99.

S7-P10

Trichoderma spp. isolates as potential resistance inducers and biocontrol agents of *Meloidogyne javanica* on banana.

Nayane Almeida[1] (nayaneagro@yahoo.com.br), Camilla Martins[2], Cirano Ulhoa[2], Murillo Lobo Junior[3], Mara Rubia Da Rocha[1]

[1] *Escola de Agronomia, Universidade Federal de Goias, Goiania, Goias, Brazil*; [2] *Instituto de Ciências Biológicas, Universidade Federal de Goias, Goiania, Goias, Brazil*; [3] *Embrapa - Arroz e Feijao, Santo Antonio de Goias, Goias, Brazil*

Plant parasitic nematode biological control has been widely studied especially in annual crops such as soybean, corn and cotton, but scarcely studied on fruit crops. Banana plantations are often attacked by nematodes, with the genus *Meloidogyne* one of the most frequent. Depending on the nematode population, it can lead to yield losses or even to plant death. There are several commercial products based on *Trichoderma* species registered in Brazil either as fungicides or nematicides but only a few are recommended for the control of *Meloidogyne* spp. The objective of this study was to evaluate the effect of the isolates *Trichoderma harzianum* (ALL42 and IBLF006) and *T. asperellum* (T00) on the control of *Meloidogyne javanica* and as inducers of systemic resistance on banana. Enzymatic activity of the three *Trichoderma* sp. isolates were evaluated in two *in vitro* experiments using parabolized rice as substrate. Higher activity of chitinase (CHI) and b 1,3 glucanase (GLU) was found on the two isolates of *T. harzianum* (ALL42 and IBFL006). Experiments in a greenhouse were conducted in order to evaluate the capacity of the *Trichoderma* sp. isolates to promote plant growth and to control *M. javanica* populations using the banana cultivar Grand Naine. Enzymatic activity of CHI and GLU were also evaluated on banana leaves at 7, 14 and 21 days after inoculation (DAI). The isolates did not increase the plant growth. *T. harzianum* ALL42 and *T. harzianum* IBFL006 reduced the *M. javanica* population on banana roots by 43.56% and 60.34%, respectively. The isolate of *T. asperellum* (T00) showed no efficacy in reducing the nematode population. Enzymatic activity of CHI and GLU in non-treated and non inculated plants was higher at 7 DAI, reducing afterwards. At 7 DAI CHI activity was higher on plants treated with the isolate ALL42 and inoculated with *M. javanica*. CHI and GLU activity were higher at 21 DAI when treated with isolate IBFL006 and inoculated with the nematode. The effect of *T. harzianum* isolates (ALL42 and IFBL006) was also tested on *M. javanica* J2 mortality *in vitro*. The treatments were: suspension of conidia, autoclaved filtrate, non-autoclaved filtrate and control. All treatments resulted in J2 mortality higher than the control. The non-autoclaved filtrate of both isolates ALL42 and IFBL006 resulted in 38.83% and 44.88% higher J2 mortality than the control, respectively.

Keywords: Root-knot nematode - Musa sp. - Enzyme activity - J2 mortality.

S7-P11

Alternative management of Sugarbeet nematode (*Heterodera schachtii*).

Ondřej Douda[1] (douda@vurv.cz), Marie Maňasová[2], Miloslav Zouhar[2]

[1] *Crop Research Institute, Praha 6 - Ruzyně, Czech Republic*; [2] *Czech University of Life Sciences, Praha 6 - Suchdol, Czech Republic*

Currently the management of sugarbeet cyst nematode (*Heterodera schachtii*) heaqvily relies on growing of tolerant cultivars. Hypothetically, dependence on this single method of *Heterodera schachtii* management could bear the risk of breaking of the tolerance. Therefore alternative methods of *Heterodera schachtii* management should be investigated. The main objective of our research was to evaluate effects of field application of *Pleurotus ostreatus* on population densities of *Heterodera schachtii*. The experiment was established in a field scheduled for sugar beet production, soil samples were taken before treatment. Cysts of *Heterodera schachtii* were extracted from soil using the Fenwick can. The number of living juveniles was estimated for each sample. These data were used for staking of experimental plots in areas of highest nematode presence. The inoculum of *Pleurotus ostreatus* was obtained from a commercial producer in the form of straw packs containing the mycelium. Straw packs were spreader with a manure spreader and that way the mycelium inoculum was applied to experimental plots; the field was ploughed subsequently. The applications were made in November and after ploughing, the field was left undisturbed until the subsequent spring. In March, sugar beet of nematode susceptible cultivar Alpaca was sown into experimental plots, an untreated control variant was also established. Sugar beet was cultivated under standard conditions until harvest which in October. Soil samples from both experimental variants were taken, and the presence of nematodes was investigated. Our results showed a statistically significant decrease of *Heterodera schachtii* juveniles in plots treated with *Pleurotus ostreatus* mycelium. According to our findings, application of *Pleurotus ostreatus* is an effective alternative to growing of tolerant sugar beet cultivars as measure of *Heterodera schachtii* management.
Research was financed by project of Czech Technology Agency, project nr. TH04030242.Research was financed by project of Czech Technology Agency, project nr. TH04030242.

Keywords: Heterodera schachtii - Management - Sugar beet - Pleurotus ostreatus.

S7-P12

Application of starch citrate biopolymer for controlled release of carbofuran for *Meloidogyne incognita* management.

Oluwatoyin Fabiyi[1] (fabiyitoyinike@hotmail.com), Oluwaseyi Saliu[2]

[1] *Department of Crop Protection, University of Ilorin, Ilorin, Kwara, Nigeria*; [2] *Industrial Chemistry Department, University of ilorin, Ilorin, Kwara, Nigeria*

The undesirable environmental impacts of inappropriate application of pesticides have brought about research into new matrices for controlled release of pesticides. Application of controlled release systems in pest management reduces environmental pollution, while achieving the desired goal [1]. Starch is one of the most commonly used biopolymers in controlled release formulations [2]. Porous starch, and starch citrate biopolymer was prepared by freezing and thawing of starch gels. Carbofuran was incorporated into the starch microspheres at three dosages (10g, 20g, and 30g) for comparative evaluation of the effectiveness of controlled release methods on juvenile mortality and egg hatch of *M. incognita* in the laboratory. The biopolymers were characterized using Fourier Transform Infrared Spectroscopy (FTIR), Scanning Electron Microscopy (SEM) and Thermo-Gravimetric Analysis (TGA) for functional group, surface morphology and thermal stability properties respectively. The SEM revealed highly stabilized porous starch citrate biopolymers with non-porous structures and gradients suitable for controlled release studies. The transmittance bands at 3347, 1714 and 1073 cm^{-1} for O-H, C=O and C-O-C stretching vibrations further confirms the successful synthesis of the biopolymer. TGA showed an increase in the thermal stability after citric acid modification with one-step decomposition from 290 to 500°C. From Korsemeyer-Peppas model, the carbofuran-porous starch citrate (CBFN/PRS/STH/CTRT) followed a lower diffusion release model with gradual increment in all the quantity of carbofuran loaded. An accelerated rate of diffusion percentage was seen in direct application of carbofuran. Egg hatch and mortality of juveniles were recorded on a daily basis for seven days. Direct application of carbofuran (CBFN/DRT) and carbofuran-porous starch citrate biopolymer gave the best results with significant ($p<0.05$) reduction in egg hatch and higher percentage mortality. However, significantly ($p<0.05$) lower juvenile mortality and a hundred percent (100%) egg hatch was recorded in the control experiment. The rate of release of carbofuran from the starch citrate bio polymer matrix was significantly lower than the direct application, and in spite of the slow rate of release, higher juvenile mortality and reduction in egg hatch was achieved. Controlled release formulations could be employed in *M. incognita* population control while minimising environmental pollution.

Keywords: Carbofuran - Porous starch - Biopolymer - Starch citrate.

References:

- [1] Campos et al., 2015. Agron. Sustain. Dev. 35:47-66.
- [2] Jerobin et al., 2012. Carbohydrate Polymer. 90:1750-1756.

S7-P13

The biocontrol link between rhizosphere microorganism communities and *Meloidogyne* populations.

Gerhard Engelbrecht (gerhardengelbrecht38@gmail.com), Charlotte Mienie, Sarina Claassens, Hendrika Fourie

Unit for Environmental Sciences, North-West University, Potchefstroom, South Africa

Meloidogyne species or root-knot nematodes (RKN) are a major threat to global crop production. There has been increasingly more research done on biocontrol agents using secondary metabolites produced by bacterial species as the active substance. These biocontrol agents are expected to substitute the harmful chemicals used in the management of RKNs. Though little is known about rhizosphere microorganism communities with relation to different levels of infestation by *Meloidogyne* species, some rhizosphere microorganisms are capable of supressing root invasion and reproduction of plant parasitic nematodes. This study aimed to identify the correlation between nematode and bacterial communities. *Meloidogyne* population composition (assemblages) was determined using morphometrical and molecular techniques. The rhizosphere microorganism community composition was characterized using Next Generation Sequencing (NGS). Results indicate that there are differences in the rhizosphere microorganism community composition when compared to the different levels of infestation by *Meloidogyne* species, with several bacterial species being more abundant in sites with lower levels of *Meloidogyne*.

Keywords: Bacteria - Biocontrol - Meloidogyne - Microbial community.

S7-P14

Trichoderma spp., a growth promoter of tomato roots and *Meloidogyne enterolobii* populations.

Sheila Freitas de Almeida[1] (sheilafreitas92@hotmail.com), Ana Luiza Bezerra Cardoso[2], Vanessa da Silva Mattos[3], Rebeca Mesquita e Silva[4], Jansen Rodrigo Pereira Santos[5], Regina Maria Dechechi G. Carneiro[3], Sueli Correa Marques de Mello[6]

[1] *Phytopathology laboratory, University of Brasilia, Brasilia, DF, Brazil*; [2] *Phytopathology laboratory, Pioneer Union of Social Integration, Brasilia, DF, Brazil*; [3] *Laboratory of nematology, Embrapa Genetic Resouces and Biotechnolo, Brasilia, DF, Brazil*; [4] *Laboratory of nematology, University of Brasilia, Brasilia, DF, Brazil*; [5] *Department of Phytosanitary Defense, Center for Rural Sciences, Santa Maria, RS, Brazil*; [6] *Phytopathology laboratory, Embrapa Genetic Resouces and Biotechnolo, Brasilia, DF, Brazil*

Meloidogyne enterolobii is an emerging pathogen in Brazil, considered highly destructive to tomato (*Solanum lycopersicum* L.). There are few research results involving the fungi of the genus *Trichoderma* controlling this nematode. The objective of this work was to evaluate *Trichoderma* strains as growth promoters of tomato root and their effects on *M. enterolobii* populations. Thirteen-day-old tomato seedlings 'Santa Clara' were transplanted to pots containing soil treated with conidial suspension of different *Trichoderma* strains at the concentration of $1.43x10^6$ conidia / mL (10 mL of the suspension/ pot). Six days later the plants were inoculated with eggs of *M. enterolobii* (5,000 eggs / plant). Eight strains of *Trichoderma* spp. (CENARGEN collection) and three other commercial products (Trichoplus®, Trichodermil® and Quality®), and their monosporic isolates were used in the bioassay. The treatments were arranged in a completely randomized design with eight replications. After three months, the plants were evaluated for fresh root weight, galls index, egg mass index, total eggs, number of eggs/gram of root, and reproduction factor (RF=final population/inicial population). The data were submitted to the analysis of variance and the means compared by the Scott knott's test ($p < 0.05$). The correlation between the parameters was analyzed by Pearson's correlation ($p < 0.05$). The application of CEN288, CEN289, CEN290, CEN316, CEN1153, CEN1399, just like Quality® and Trichodermil®, both monosporic and commercial products significantly increased fresh root weight when compared to the control. All of these treatments, except CEN316, also promoted a significant increase in the RF nematode compared to the nematode-inoculated control. The variable RF was positively and significantly correlated with fresh root weight, galls index, egg mass index, eggs / grams of root and total eggs. Most *Trichoderma* strains, including commercial products at the recommended doses, promoted root growth and increased nematode populations. None of them controlled significantly *M. enterolobii.* These results are of concern because commercial *Trichoderma*-based products are marketed in Brazil for the control of nematodes, without any evidence of their effectiveness under field conditions.

Keywords: Disease management - Solanum lycopersicum L - Biological control.

S7-P15

Two nematicidal *Bacillus* strains revealed a wide range of possible virulence factors.

Nik Susič[1] (nik.susic@kis.si), Sandra Janežič[2], Maja Rupnik[2], Barbara Gerič Stare[1]

[1] *Agricultural Institute of Slovenia, Ljubljana, Slovenia*; [2] *Centre for Medical Microbiology, NLZOH, Maribor, Slovenia*

Bacillus firmus nematicidal bacterial strains are used to control plant parasitic nematode infestation of crops in agricultural production. Proteases are presumed to be the primary nematode virulence factors in nematicidal *B. firmus* degrading the nematode cuticle and other organs. We determined and compared the whole genome sequences of two nematicidal strains: a commercial bionematicidal isolate *Bacillus firmus* I-1582 and a wild type *Bacillus* sp. ZZV12-4809. Resulting *B. firmus* I-1582 and *Bacillus* sp. ZZV12-4809 genome assemblies have a length of 4,597,711 bp and 5,245,841 bp; an average G+C content of 41.70 % and 41.17 %; and harbour 5,048 and 5,671 predicted genes, respectively. Among those, 71 and 81 annotated genes included homologs to known transporters, virulence factors, drug targets and antibiotic resistance genes; as well as 18 and 19 homologs to nematode-virulent proteases were found in *B. firmus* I-1582 and *Bacillus* sp. ZZV12-4809 genome assemblies, respectively. Secondary metabolite gene clusters were also found in both genomes, suggesting the genetic capacity for nematode virulence. The results of this study point to the genetic capability of *B. firmus* and related species for nematode virulence through a range of direct and indirect mechanisms.

Keywords: Bacillus firmus - Complete genomes - Bioinformatics - Nematicidal activity - Virulence factors.

S7-P16

Effects of application timing on the efficacy of *Xenorhabdus* and *Photorhabdus* metabolites for control of *Meloidogyne incognita*.

Abolfazl Hajihassani[1] (ahajihassani@ufl.edu), Denis Gitonga[1], Patricia Timper[2], David Shapiro-Ilan[3]

[1] *Entomology and Nematology Dep., Fort Lauderdale Research and Education Center, University of Florida, Davie, FL, United States*; [2] *Genetics and Breeding Research Unit, USDA-ARS, Tifton, GA, United States*; [3] *Southeastern Fruit and Tree Nut Research Laboratory, USDA-ARS, Byron, GA, United States*

Bacteria in the genera *Photorhabdus* and *Xenorhabdus* are symbionts of entomopathogenic nematodes (EPNs) in the genera *Heterorhabditis* and *Steinernema*, respectively. Studies have shown that metabolites of EPN symbiotic bacteria are toxic to a wide range of plant-parasitic nematodes [1-3]. We evaluated the efficacy of application timing [5 days before planting (DBP) and at plant (AP)] of two bacterial metabolites, *X. szentirmaii* and *X. bovienii*, against the root-knot nematode (*Meloidogyne incognita)* on cabbage in two environmental conditions. At-plant applications of *Paecilomyces lilacinus* strain 251 (MeloCon WG) and secondary metabolites of *Burkholderia rinojensis* strain A396 (Majestene) and oxamyl (Vydate) were also included in the trials for comparison. Plants were infected with 2,500 *M. incognita* second-stage juveniles (J2), soil-drenched with the metabolites or nematicides, and grown for 450 degree days using a base temperature of 10 °C. Plants treated with water were considered the untreated control, and each treatment had six replications arranged in a completely randomized design. In the greenhouse, Vydate and *X. szentirmaii* at 5 DBP had a lower ($P < 0.05$) root gall rating than the control, while all metabolite and Vydate treatments had significantly lower root galling compared to Majestene, MeloCon, and the control. The metabolites and Vydate reduced ($P < 0.05$) *M. incognita* egg counts per gram of root compared to other treatments in the greenhouse; however, there were no differences in the egg count between the metabolites and Vydate. In the screenhouse, AP application of *X. szentirmaii and X. bovienii* at 5 DBP reduced the egg count compared to Majestene and the control; however, no differences in the egg counts between the metabolites and Vydate were observed. This study shows that natural metabolites produced by the EPN symbiotic bacteria can control root-knot nematodes regardless of application timing and can be considered a potential alternative to nematicides in organic production systems where chemical use is not allowed.

Keywords: Application timing - Metabolite - Control - Entomopathogenic nematodes - Root-knot nematode.

References:

- [1] Shapiro-Ilan et al., J. Nematol. 38:449–454.
- [2] Hazir et al., Eur. J. Plant Pathol. 146:369–381.
- [3] Kepenekci et al., Crop Protec. 108:31–38.

S7-P18

Virulence and microbial activity of different nematophagous fungi and chemicals against root-knot nematodes, *Meloidogyne incognita,* on tomato.

Manzoor Hussain[1] (manzoor.sahi@gmail.com), Miloslav Zouhar[2], Pavel Rysanek[2]

[1] *Czech University of Life Sciences,Prague, Prague, Czechia*; [2] *Plant Protection, Czech University of Life Sciences,Prague, Prague, Czechia*

Root-knot nematodes (Meloidogyne spp.) are some of the most destructive polyphagous pests with a wide host range that includes field crops, vegetables, and ornamental plants. These nematodes contribute to more than 10% of crop production losses worldwide. The problem is increased due to lack of our farmer's knowledge, repeated cultivation of the same crop in the fields, and unawareness of pesticide applications. During the selection of suitable pesticides, several critical factors including low toxicity to human and wildlife populations, limited environmental impact, and limited chemical residues in the food supply are considered. Ten isolates of different fungi, *Arthrobotrys oligospora*, *Arthrobotrys robusta, Dactylella oviparasitica*, *Clonostachys rosea*, *Stropharia rugosoannulata*, *Lecanicillium muscarium*, *Trichoderma harzianum Pleurotus ostreatus*, *Drechslerella aphrobrochum*, *Esteya vermicola*, and two bacterial strains, *Bacillus thuringiensis, Bacillus velezensis* and three commercially available chemicals, Vydate and Basamid (G), and Velum were evaluated against root-knot nematodes, *Meloidogyne incognita,* on tomato in a growth chamber. All fungi and chemicals proved to be efficient in reducing the infestation level of *Meloidogyne incognita* and providing better growth of tomato except Velum as compared to their controls. Maximum reductions in the nematode population were observed in the plants treated with *Lecanicillium muscarium* and chemicals. *Lecanicillium muscarium* treatments alone or with nematodes had significant ($P = 0.01$) positive effects on plant root shoot growth compared to the growth of control plants and all other treatments in the experiment. The overall microbial activity was drastically reduced in the plants treated with all commercial chemicals whereas it was significantly enhanced in the plants treated with *L. muscarium, Stropharia rugosoannulata and Bacillus thuringiensis.*

Keywords: Nematodes - Meloidogyne incognita - Nematophagous fungi - Lecanicillium muscarium - Tomato.

S7-P19

Potential of plant growth promoting bacteria as biocontrol agents against the root knot nematode *Meloidogyne javanica*.

Thomais Kakouli-Duarte (thomae.kakouli@itcarlow.ie), Aoife Egan

Department of Science and Health, Institute of Technology Carlow, Carlow, Ireland

Plant growth promoting (PGP) bacteria are plant colonisers in a mutualistic relationship with their host plant. Such a relationship entails several benefits for the plant, including plant growth promotion and biological control against plant pathogens. The current and main applied practice for the management of plant parasitic nematodes (PPN) is mostly the usage of conventional synthetic chemical nematicides, although various alternatives are also used to some extent. As conventional synthetic chemical nematicides are hazardous for the environment, non-target biota, and consumers' health, these compounds are heavily regulated. In addition, lack of efficacy has been observed in their application as well as PPN resistance. These factors make the need for the identification and development of viable alternatives even more pressing. A recent approach towards this aim is the exploitation of the potential biocontrol properties of PGP bacteria in plants and the work presented here has this focus. Specifically, this presentation will highlight (1) the capacity of PGP bacteria and their antibiotic component 2,4 – DAPG to attract (or repel) the PPN *Meloidogyne javanica in vitro*, and (2) the interactions among beneficial bacterial strains, *M. javanica* and tomato plants, in a tritrophic model, and in the context of plant growth promotion and nematode biocontrol. This study revealed that the antibiotic repelled *M. javanica*. Tomato plant growth was positively influenced by various PGP bacterial treatments and by that of 2,4-DAPG, even during PPN infection. At the same time, low levels of nematode infection were initially observed in plants (at 20 days post infection) but these increased with time (at 50 and 70 days post infection). An induced systemic resistance response was noted in plants treated with the bacterial strain *Pseudomonas* L321. The potential of these PGP bacterial agents as alternatives to *M. javanica* management will be discussed.

Keywords: Root knot nematodes - Plant growth promoting bacteria - 2,4-DAPG, antibiotic - Biological control.

S7-P20

Effects of an Alltech® soil health product on entomopathogenic and plant parasitic nematodes *in invitro* bioassays.

Anusha Pulavarty[1*], Karina Horgan[2], Thomais Kakouli-Duarte[1]

[1]Molecular Ecology and Nematode Research Group, enviroCORE, Department of Applied Science, Institute of Technology Carlow; [2]Alltech Bioscience Centre, Dunboyne, County Meath, Ireland

Biological control is a highly preferred, effective, non-polluting and environmentally safe approach that should be considered while adopting any sustainable pest management approach. Alltech® Crop Science (ACS) provides nature-based solutions to agronomic and horticultural challenges faced by producers worldwide. A formulation of Alltech®, which is a proprietary blend of fermentation products and plant extracts with micronutrients (ACS5075), was evaluated against the root-knot nematode (RKN) *Meloidogyne javanica* and the potato cyst nematode (PCN) *Globodera rostochiensis.* In the context of product sustainability, preliminary bioassays were conducted to determine LC_{50} of this product against three strains of entomopathogenic nematodes (EPN). Subsequently, experiments were conducted at dilutions below EPN LC_{50} concentration to determine RKN and PCN sensitivity. The effectiveness of the product against RKN and PCN was recorded in terms of percentage juvenile hatching and mortality. Attraction assays were performed by adding product to 20% (w/v) Pluronic F-127 gel to study behaviour of RKN and PCN second-stage juveniles (J2) towards host roots. The product was found safe for EPN at concentrations upto 4%. However, the product reduced *M. javanica* juvenile hatching from 100% of J2 in untreated, to only 10% when 0.5% of the product was used and to no hatching with higher concentrations. A 10-fold reduction in percentage survival was observed at 0.5% treatment compared to untreated control, however survival dropped to zero with product concentrations of 1% and above. Similarly, at 0.5% and 1%, a 13 and 43 fold reduction, respectively, in PCN percentage survival was recorded compared to untreated. ACS5075 completely inhibited attraction of RKN and PCN J2 towards roots when compared to the untreated control. Based on these *in vitro* bioassays, we conclude that, under the conditions of this study, this product was efficient in controlling PPN while being safe for the beneficial EPN.

Keywords: Sustainability - Root-knot nematodes - Potato cyst nematodes - Entomopathogenic nematodes.

S7-P21

Tomato rhizosphere under RKN attack - Deciphering the *Meloidogyne incognita* pathobiome.

Enoch Narh Kudjordjie (enochnarh@agro.au.dk), Susana Santos, Mette Vestergård

Department of Agroecology, Aarhus University, Slagelse, Denmark

Background:
Plant parasitic nematodes including root-knot nematodes (RKN - *Meloidogyne* sp.) are major threats to crop production worldwide [1]. Plants harbour a diverse assembly of microorganisms in the root endo- and rhizosphere that they can select, promote and (de)activate to enlarge the pool of bioavailable nutrients and maximize local disease suppressiveness [2]. Rhizosphere interactions can be between microorganisms from the same kingdom or different kingdoms, and can involve plant-beneficial microorganisms as well as pathogens [3]. The **RKNpathobiome**, i.e. the microbial community associated with a pathogen varies over time and between host tissues [4]. Our aim was to decipher the microbiome potentially interacting with *Meloidogyne incognita* during root invasion. We characterized the multi-trophic inter-kingdom microbiome associated with *M. incognita* infection in tomato plants. We hypothesize that the RKN infection is accompanied by microbiome shifts in the root and rhizosphere, causing changes in a variety of functional traits associated with plant's defence system.
Methods:
We used a metabarcoding approach to characterize the root and rhizosphere microbiomes associated with RKN-inoculated tomato. Interactions were deciphered by combining linear discriminant analyses and ecological network analyses.
Results:
Our results indicate that infection with *M. incognita* is accompanied by significant changes in the composition of root and rhizosphere microbiome communities. Community analysis of root-associated microbiomes in healthy and nematode-infected tomatoes indicated that nematode infections were associated with variation and differentiation of the root endophyte and rhizosphere bacterial populations. Specific bacterial, fungal and protists ASVs appeared to interact directly with *M. incognita* with some of them showing potential antagonistic activity according to the inferred microbial network.
Conclusion:
With crop yields approaching their maximum capacity, increasing pests and extreme climatic events adding additional stress to productivity in many regions, the world needs innovative solutions to avoid crop losses by RKN. By revealing putative antagonistic interactions of the RKN pathobiome, this study contributes to improving the biological control of RKN.

Keywords: Root-knot nematodes - Ecological network analysis - Metabarcoding - Microbiome - Plant-pathogen interaction - Pathobiome.

References:

- [1] Zhou D, et al., 2019. Microbial Ecology, 78:470–81.
- [2] Toju H, et al., 2018. Nat Plants, 4:247–57.
- [3] Jakuschkin B, et al., 2016. Microbial Ecology, 72:870–80.
- [4] Bass D, et al., 2019. Trends Ecol Evol, 34:996–1008.

S7-P22

Effects of a chicken manure fertilizer on beneficial nematode communities in a vineyard

Laurent Lapeyre[1] (lapeyre@nixe.fr), Gaetan Marcq[2], Renaud Canaguier[1]

[1] *Nixe Laboratoire, Sophia-Antipolis, France*; [2] *Italpollina, Sophia-Antipolis, France*

The company Italpollina (www.italpollina.fr) addresses the problems of fertilization and crop biostimulation by combining traditional, sustainable agriculture, and agroecological innovative processes. Italpollina produces liquid and solid fertilizers by using dehydrated livestock manure and by-products from the agro-food industries. Biological activity of soil improves the use of nutrients, and the growth, the health and the quality of crops. Traditionally, farmers increase that biological activity by (a) providing organic and mineral plant nutrients to stimulate the development of plants and soil microorganisms, and (b) bringing to the soil, useful microorganisms, like the spore forming bacteria occurring in manure, *Bacillus thuringiensis, B. cereus, B. subtilis, B. pumilus, B. firmus, B. toyonensis, Lysinibacillus sphaericus*, which have potential nematocidal effects (Zheng et al., 2016) and plant biostimulant activities. The Italpollina pelletized chicken manure, thermally sanitized according to Regulation (CE) 1069/2009, meets these two requirements. The analysis of nematode communities from vineyard soils (AOC Baux-de-Provence, Mouriès, France) confirm the interest of dried chicken manure. In Mouriès vineyard assay, Italpollina chicken manure (800 kg/ha, NPK 4-4-4, 71 % organic matter (OM) NF U 42-001) was compared to 2 organic soil amendments (800 kg/ha, > 65 % OM, NF U 44-051), one yeast fractions “soil activator” (200 kg/ha) and one fish residues organic fertilizer (800 kg/ha, NPK 2.5-3.5-2, NF U 42-001). Analysis of nematode communities, 1 month after application, fulfilled by Elisol-Environnement, Congénies, France, according to Villenave et al. (2010), showed an effective action of Italpollina organic fertilizer on nematode populations, with an increase in beneficial nematode communities (saprophytic and phytoparasites, Djian-Caporalino et al., 2018), and absence of *Meloidogyne* or *Xiphinema*. The main observed phytoparasite genera do not present any infection risk for the grapevine, but reflect the biological activity of the green cover and soil between 0 and 15 cm. Italpollina organic fertilizer promoted the dynamic of organic matter and nutrient recycling in green cover more than in the crop. The other organic products had lesser impacts than Italpollina, perhpas because they do not contain simultaneously NPK and the required bacteria.

Keywords: Italpollina - Chicken manure - Nematodes communities - Vineyard.

References:

- Djian-Caporalino C., Mateille T., 2018, Impact de la diversité des communautés de nématodes phytoparasites sur la durabilité de systèmes de culture maraîchers visant spécifiquement le contrôle des nématodes à galles : de la pertinence d’une approche « diversité », Innovations Agronomiques 69, 83-89.
- Renco M, 2013, Organic amendments of soil as useful tools of plant parasitic nematodes, Helminthologia, 50, 1:3-14.
- Villenave, C., Saj, S., Pablo, A.L., Sall, S., Djigal, D., Chotte, J.L., Bonzi, M. 2010, Influence of long-term organic and mineral fertilization on soil nematofauna when growing Sorghum bicolor in Burkina Faso, Biology and Fertility of Soils 46, 659-670.
- Zheng Z., Zheng J., Zhang Z., Peng D., Sun M., 2016, Nematicidal spore-forming Bacilli share similar virulence factors and mechanisms, Sci Rep, 6, 31341.

S7-P23

Hatching of cyst nematodes in soil drenched with root exudates under controlled environmental conditions.

Nicolas Mariette[1] (nicolas.mariette@anses.fr), Bruno Ngala[2], Mélina Ianszen[2], Pauline Dewaegeneire[2], Marie-Christine Denis[3], Catherine Porte[3], Christophe Piriou[3], Emilie Robilliard[4], Antoine Couetil[4], Eric Nguema[5], Virginie Gobert[2], Amélie Beury[2], Anne-Claire Le Roux[6], Josselin Montarry[3], Sylvain Fournet[3]

[1] *ANSES - INRAe, Le Rheu, France*; [2] *RD3PT, Achicourt, France*; [3] *IGEPP, INRAe, Le Rheu, France*; [4] *SILEBAN, Gatteville le Phare, France*; [5] *CMI-Roullier, Saint-Malo, France*; [6] *RD3PT, Le Rheu, France*

Cyst nematodes account for substantial annual yield lost in vegetable production worldwide. Concerns over environmental and health issues resulting from the use of chemical nematicides mean alternative sustainable and integrated solutions are urgently required. Hatch induction of encysted eggs in the absence of host plants, i.e. 'suicide-hatching', could be a sustainable alternative in reducing population densities in infested soils. Here we examined *in-situ* hatching of encysted eggs of *Globodera pallida, G. rostochiensis, Heterodera carotae* and *H. schachtii* at varying soil depths, following exogenous applications of host root exudates in repeated glasshouse experiments. Cysts were retrieved 30 or 43 days post-incubation, depending on the nematode species, and assessed for hatching rates relative to the initial number of viable larvae per cyst. Hatching of potato cyst nematodes depended on both soil moisture and effective contact with root exudates, and to a lesser extent, on exudate concentration. *Globodera rostochiensis* hatched better under *in-situ* conditions as compared with *in-vitro* hatch. *Heterodera carotae* had over 75% hatch induced by root exudate irrespective of the concentration, with better hatch induction at 20 cm as compared with 10 cm soil depth. Hatching of *H. schachtii* largely depended on the soil moisture level at constant temperatures, rather than the type or concentration of exudates applied. We therefore concluded that exogenously applied host root exudates could play a major role in inducing *in-situ* hatch of encysted eggs of potato and carrot cyst nematodes in the absence of host plant under favourable soil temperature/moisture conditions.

Keywords: Globodera - Heterodera - Plant root exudates - Hatching - In situ.

S7-P24

An evaluation of small grain cover crops to reduce *Meloidogyne incognita* population density in cotton fields.

Sloane McPeak[1] (sem0070@auburn.edu), Kara Gordon[1], Kathy Lawrence[1], Dustin Herb[2]

[1] *Entomology and Plant Pathology, Auburn University, Auburn, Alabama, United States*; [2] *Research and Development, OreGro Seeds, Albany, Oregon, United States*

The objective of this study was to determine the host susceptibility of small grain cover crops to *Meloidogyne incognita*, the southern root-knot nematode (RKN). Greenhouse trials were initiated to determine the host suitability of RKN on eight triticale, five barley, four wheat, and three oat cultivars in comparison to corn (DEKALB DKC68-26), a known host for RKN. Seeds were planted with and without RKN to determine RKN reproductive factor (Rf) as measured by the number of RKN eggs per gram of fresh root evaluated 42 days after planting (DAP). Cultivars were replicated 5 times and arranged in a randomized complete block design (RCBD). All wheat and barley cultivars tested were confirmed to be hosts for RKN (Rf ranged from 1.11 to 3.93) and were more susceptible than the corn (Rf = 1.76) with the exception of one wheat and two barley cultivars. Two triticale cultivars, Forerunner and OG170039, and one oat, ORO4372, proved to be poor hosts for RKN with Rf of 0.38, 0.73, and 0.91 respectively. Three triticale, five barley, four wheat, and three oat cultivars were chosen to be tested in the field with the inclusion of three rye cultivars that were not tested in the greenhouse. Field evaluations were planted at E.V. Smith Research and Extension Center in Tallassee, AL, USA on 3 Oct. 2019, Plant Breeding Unit in Shorter, AL, USA on 20 Oct. 2019, and one on-farm location in Germanton, NC, USA 19 Oct. 2019. All tests were arranged in RCBD with 5 replications. Each location was sampled 30 to 45 DAP measuring for plant height and biomass. Forerunner and OG170039 triticale, poor hosts for *M. incognita* in the greenhouse, produced greater biomass (45% and 75% respectively) than the other triticale tested in the field. ORO4372 oat, also a poor host, had no significant increase in plant growth over other oat cultivars. Results suggest Forerunner and OG170039 triticale and ORO4372 oat are favorable cover crop selections for RKN infested cotton fields.

S7-P25

Soil actinobacteria with biocontrol potential against *Meloidogyne javanica.*

Ángela María Mogollón Ortiz[1] (ammogollon@gmail.com), Thalita Suelen Avelar Monteiro[2], Leandro Grassi de Freitas[3], Marisa Vieira de Queiroz[4]

[1] *Universidade Federal de Viçosa/ Universidad de los Llanos, Villavicencio, Meta, Colombia*; [2] *Department of Plant Pathology, Universidade Federal de Viçosa, Viçosa, Minas Gerais, Brazil*; [3] *Plant Pathology Department, Universidade Federal de Viçosa, Viçosa, Minas Gerais, Brazil*; [4] *Microbiology Department, Universidade Federal de Viçosa, Viçosa, Minas Gerais, Brazil*

The root-knot nematodes (RKN) *Meloidogyne* spp. are the most important nematode distributed around the world, causing considerable economic losses [1]. The genus *Streptomyces*is, is the most studied actinobacteria for the control of RKN [2,3]; however, other genera of bacteria remain to be explored. Nematode management strategies were focused towards sustainable crop production and being able to identify the control efficacy of several isolates of actinobacteria against *Meloidogyne* spp. In this study under *in vivo* and *in vitro* analysis, we selected actinobacteria isolates with biocontrol potential against the *M. javanica*. 10 actinobacteria were isolated from soil using the serial dilution method for selection. In experiments with *Solanum lycopersicum* plants of the susceptible variety Santa Cruz Kada, $2x10^4$ spores/g soil of each actinobacteria were applied to sterile soil at the time of transplanting and after five days *M. javanica* infection was performed with 800 eggs. The number of eggs and galls were evaluated 46 days after inoculation with nematodes, in addition to *in vitro* parasitism of *M. javanica* eggs. Plants treated with bacterial *isolates* 5N, 2AE, 6O and 4L, showed reduced percentages of *M. javanica* eggs of 63, 57, 51 and 50%, respectively, compared to the control treatment. The percentage reduction of nematode galls were also significant with values 28, 26, 25 and 16% in plants treated with 8S, 4L, 10U e 2AE, respectively, compared to the control treatment. Additionally, in *in vitro* tests conducted on water agar, microscopic observations showed that all the actinobacteria isolates had the ability to parasitize *M. javanica* eggs. Our results suggest that actinobacteria could be an excellent biocontrol agent against *M. javanica* by acting directly through egg parasitism.

Keywords: Root-knot nematode - Parasitism - Eggs - Bacteria.

References:

- [1] Moens et al., 2009. CABI. 1-17p
- [2] Kaur et al., 2016. Microbiological Research 192 (2016) 247–252.
- [3] Na et al., 2017. Journal of Integrative Agriculture 2017, 16(6): 1347–1357.

S7-P26

Evaluation of bacterial extracts of *Xenorhabdus, Photorhabdus,* and rhizobacteria to control *Meloidogyne ethiopica*.

Patricia Morales-Montero[1] (pzmorales@uc.cl), Simona Prodan[2], Carlos Castañeda-Álvarez[2], Marlene Rosales[1], Erwin Aballay[2]

[1] *Pontificia Universidad Católica de Chile, Santiago, Chile*; [2] *Universidad de Chile, Santiago, Chile*

Meloidogyne ethiopica is one of the most damaging plant-parasitic nematodes affecting vineyards in Chile. This research evaluated the nematicidal effect of the crude extracts of *Xenorhabdus bovienii, Photorhabdus thracensis,* and *Photorhabdus sp.* singly and their combination with the crude extracts of two rhizobacteria *Bacillus weihenstephanensis* and *Brevibacterium frigoritolerans* on *M. ethiopica in vitro* and greenhouse experiments. *In vitro* second-stage juveniles (J2) and eggs of *M. ethiopica* were exposed to the bacterial extract at two concentrations: 25 and 50% to study J2 mortality and egg hatching. In the greenhouse, grapevine roots were dipped in the crude bacterial extracts, 300 mL of the bacterial extract at 25% concentration was applied to the plant, and were infested with 2000 eggs of *M. ethiopica* afterward. To evaluate the effect, the number of eggs and galls per root system, number of larvae in 250 cm3 of soil, the reproductive index, and the weight of the roots system and stem of the plants were registered. The experiment was run for four months, and one extract application was made. *In vitro* was observed that *Photorhabdus* strains produced high mortality when used independently from rhizobacteria at the highest dose (50%), while *X. bovienii* at 25% showed higher mortality of *M. ethiopica* J2 when mixed with the rhizobacterial extract. The greenhouse experiments demonstrated that the extracts do not impact grapevine biomass or *M. ethiopica's* population under these conditions. These results might be due to the concentration of the extract used, extreme climatic conditions, and its effect on the stability of the natural products contained in the crude extracts of the bacteria. We are still missing greenhouse data such as the short-term effect of the extract on the plant-parasitic nematode population, the effect on other soil organisms, the use of higher concentration of the extract, and the effect of the extract on the plant at a metabolomic level. By conducting more experiments and using more combinations of symbiotic bacteria and rhizobacteria, we could find other options for chemical nematicides.

Keywords: Xenorhabdus - Photorhabdus - Plant parasitic nematodes - Biocontrol - Rhizobacteria.

S7-P27

Free-living nematodes and microorganisms in soil improvement materials for plant parasitic nematode control.

Kyoko Owada[1] (kyokoowa@chem.gunma-ct.ac.jp), Karen Kurihara[2], Aoi Toru[3]

[1] *Department of Chemistry and Materials Science, National Institute of Technology, Gunma College, Maebashi, Gunma, Japan*;
[2] *Advanced Environmental Engineering Course, National Institute of Technology, Gunma College, Maebashi, Gunma, Japan*; [3] *Department of Civil engineering, National Institute of Technology, Gunma College, Maebashi, Gunma, Japan*

We have developed a soil improvement material including free-living nematodes and microorganisms as biological indicators. It was made from biomass resources such as cedar bark, dried sludge and dehydrated soil. The suppressive effect of the soil improvement material for plant parasitic nematodes was expected when the soil improvement material included *Bacillus subtilis* ($1x10^7$/g dry weight) and free-living nematodes ($1x10^4$/10 g wet weight). It was known that *Bacillus subtilis* produces antibiotics and *Bacillus* strains have the biocontrol activity for plant pathogens [1]. The soil improvement material was introduced into the test fields of Konjac potato, *Amorphophallus konjac* and Chainese yam. The soil samples were collected once a week. For the extraction of nematodes from the soil samples the Baermann funnel method was used. Nematodes were counted alive and examined to confirm the morphology of mouth form. Genetic variations of free-living nematodes associated with the soil improvement material were also investigated. DNA was extracted from single nematode and the nucleic acid sequences of ITS1 and ITS2 regions were compared. Mycophagous nematodes, *Cephalobidae sp.* and *Phelenchoides sp*. were identified. Predatory nematodes were confirmed as *Rhyssocolpus* and *Paractinolaimus sp*. We observed that a predatory nematode from the soil sample was feeding on free-living nematodes. It was reported that predatory nematodes feed on soil microorganisms and plant parasitic nematodes [2]. They might control plant parasitic nematodes. We are investigating the resource of predatory nematodes. Both yields of Chinese yam and Konjac potato were the same as when chemical pesticides were used. It might be suggested that the soil improvement in this study had the effect of inhibiting cropping damage equivalent to chemical pesticides.

Keywords: Soil improvement material - Free-living nematodes - Parasitic nematodes.

References:

- [1] Kavitha et al., 2012. Nematol. Medit. 40:203-206.
- [2] Khan and Kim, 2007. Applied Soil Ecology. 35:370-379.

S7-P28

The combined use of *Metarhizium anisopliae* and *Trichoderma asperellum* bioeffectors in the control of *Meloidogyne incognita*.

Eszter Erdős[1] (erdos.esztike@gmail.com), Renáta Petrikovszki[1], Franciska Tóthné-Bogdányi[2], György Turóczi[1], Ferenc Tóth[1]

[1] *Plant Protection Institute, Szent István University, Gödöllő, Hungary*; [2] *Fővárosi Közterület-fenntartó Nonprofit Zrt., Budapest, Hungary*

There are many preventive and protective measures available against the soil borne *Meloidogyne incognita* that pose a serious risk to the production of vegetables in horticulture, especially in greenhouses by damaging the roots of crops. The search for cost-effective and environmentally friendly defense methods in still open ended. Many of the soil inoculants in use have beneficial organisms, namely fungi as active ingredients. The entomopathogenic *Metarhizium anisopliae* and the antagonist *Trichoderma asperellum* are microorganisms that have been shown to have potential as bionematicides by the results of previous research on their ability to suppress populations of harmful organisms, as well as their ability to parasitize nematodes. Yet, the combined use of these two beneficial organisms is an unexplored subject, so our study is the first to investigate the interaction and compatibility of these two plant protection bioagents *in vitro* and then in a pot experiment with artificial *M. incognita* infection. We found that the growth of *M. anisopliae* was inhibited *in vitro* when inoculated simultaneously with *T. asperellum*. However, when *T. asperellum* was inoculated subsequently, that is, after the inoculation of *M. anisopliae*, an increasing inhibitory effect by *M. anisopliae* was observed. Based on these preliminary results, we launched a pot experiment to investigate the damage caused by *Meloidogyne incognita* on "Dányi" landrace tomatoes using *M. anisopliae*, *T. asperellum* alone and their combination as treatments. Since no significant difference was found between any of the treatment combinations, we tested the effect of these two fungal strains on the activity of *M. incognita*. The larvae were observed to avoid colonies of *T. asperellum* the most. Interestingly, the same effect was observed with the control fungal colony of *Sclerotinia sclerotiorum*. Our results indicate that these two fungi can be combined with each other and they can develop together, and because of their properties, can control *M. incognita*, too. However, further studies are needed to determine the circumstances in which the use of these two fungal agents is even more successful.

Keywords: Metarhizium anisopliae - Trichoderma asperellum - Meloidogyne incognita - Biological pest control.

S7-P29

Efficacy of a cost-effective bionematicide to control *Pratylenchus coffeae* on robusta coffee.

Ankardiansyah Pandu Pradana[1] (pandu@unej.ac.id), Iis Nur Asyiah[2], Ninda Sintia Wulandari[2], Balawara Andika[2], Sugeng Winarso[3], Lenny Widjayanthi[4], Dwi Nugroho[5], Kurniawan Firmansyah[6]

[1] *Plant Protection, University of Jember, Jember, East Java, Indonesia*; [2] *Biology Education, University of Jember, Jember, East Java, Indonesia*; [3] *Soil Science, University of Jember, Jember, East Java, Indonesia*; [4] *Agricultural Extension, University of Jember, Jember, East Java, Indonesia*; [5] *Indonesian Coffee and Cocoa Research Institute, Jember, East Java, Indonesia*; [6] *Tiga Kreasi Bersama Limited Partnership, Bondowoso, East Java, Indonesia*

Coffee production in Indonesia is currently fluctuating and tends to decline. This phenomenon is caused by the attack of the root lesion nematode *Pratylenchus coffeae* in various coffee plantations in Indonesia [1]. Bionematicide application can be an effective and environmentally friendly method for controlling *Pratylenchus coffeae*. In previous research, we found a cost-effective bionematicide formula with the active ingredients of 3 rhizobacteria isolates and 1 endophytic bacterial isolate [2]. This research aims to test the effectiveness of bionematicides in controlling *P. coffeae* in the field. The field experiment used a randomized block design with 8 treatments, each treatment had 4 replications and 12 sub-replications. The test plant used in the field experiment was 5-year-old robusta coffee. The data obtained were analyzed for diversity using Analysis of Variance and continued with Duncan's Multiple Range Test with 5% α. The treatments used in the field experiment included control (T1), 5 L of organic matter (T2), 60 mL of bionematicide (T3), 30 mL of bionematicide + 5 L of organic matter (T4), 60 mL of bionematicide + 5 L of organic matter (T5), 90 mL of bionematicide + 5 L of organic matter (T6), and 120 mL of bionematicide + 5 L of organic matter (T7). The results showed that 1.5 months after bionematicide application, the population of *P. coffeae* in the soil was reduced up to 28.12%. At the same time, the nematode population in the roots was reduced up to 42.85%. We also observed the nematode population in the soil and roots 3 months after bionematicide application, which showed that the nematode population in the soil was reduced up to 81.1%, while the nematode population in the roots was suppressed up to 68.28%. The best bionematicide treatment in this research was T7.

Keywords: Root lesion nematode - Biocontrol - Bacillus - Pseudomonas - P. coffeae.

References:

- [1] Mutala'liah et al., 2018. Biodiversitas. 19 (1): 67-70.
- [2] Asyiah et al., 2020. Biodiversitas. 21 (10): 4702-4708.

S7-P30

Orchid mycorrhizal fungus *Waitea circinata* on the control of *Meloidogyne enterolobii* in tomato crop.

Denner Robert Faria[1] (denner.robertfaria@gmail.com), Igor Guimarães Barbosa[1], Felipe Gonçalves de Carvalho[2], José Feliciano Bernardes Neto[1], Gianlucca de Urzeda Alves[2], Amanda Abdallah Chaibub[2], Leila Garcês de Araújo[2], Mara Rúbia da Rocha[1]

[1] *Escola de Agronomia, Universidade Federal de Goiás, Goiânia, Goiás, Brasil*; [2] *Instituto de Ciências Biológicas, Universidade Federal de Goiás, Goiânia, Goiás, Brasil*

Tomato (*Solanum lycopersicum* L.) is one of vegetables produced and consumed worldwide. The attack of nematodes, especially the root-knot nematodes (*Meloidogyne* spp.), can encumber its yield. The species *Meloidigyne enterolobii* is of special concern for the crop due to the lack of plant resistance to this species. Plant-mycorrhizal fungi associations are promising on nematode control although most mycorrhizal fungi are plant dependent for production. *Waitea circinata* is an orchid mycorrhizal fungi (OMF) that grows in culture medium, such as PDA (popato dextrose agar), and therefore may be promising for commercial production. This study aimed to evaluate the effect of *W. circinata* on the control of *M. enterolobii* in tomato plants, under greenhouse conditions. The experiment was carried out in a completely randomized design, in a 6 x 2 factorial scheme, with six concentrations of the fungus mycelial suspension (0, 5, 10, 15, 20 and 25 $g.L^{-1}$), two evaluation timings (30 and 60 days after inoculation (DAI)) and seven replications. The OMF was cultured for 10 days at 28 °C in Petri dishes with PDA culture medium to obtain different concentrations of mycelial suspension. Tomato seedlings of 'Santa Clara-5300' were transplanted into 1 L-pots filled with soil and sand (1:1) previously autoclaved (102°C/20 min). At the time of transplanting, 50 mL of each mycelial suspension concentration were applied per plant via soil irrigation. After 24 h, 4000 eggs + J2 of *M. enterolobii* were inoculated. At 30 and 60 DAI the shoot and root length, shoot and root fresh mass were evaluated. Nematodes were extracted from the roots, and population density (nematodes/g of root) and reproduction factor (RF) were determined. Significant interactions were found between both factors (OMF concentrations and evaluation timings). Shoot length and fresh matter increased linearly as the OMF concentration increased at both evaluation timings. Root length and fresh weight increased up to the mycelial concentration of 15 g L^{-1}, followed by a reduction in subsequent concentrations, represented by polynomial regression. The nematode population density and RF reduced as the OMF concentrations increased. *W. circinata* was effective in controlling *M. enterolobii* and improving tomato plant development being an efficient biocontrol agent.

Keywords: Biological control - Mycorrhizae - Root-knot nematode - Solanum lycopersicum.

References:

- Carvalho et al., 2015. Tropical Plant Pathology. 40(3): 151-159.
- Carvalho et al., 2021. Pesquisa Agropecuária Tropical. 51: e66916.
- Peixoto et al., 2017. Genetics and Molecular Research. 16(2): 1-15.
- Silva et al., 2017. Journal of Nematology. 49: 77-85.

S7-P31

Microbial rhamnolipids as a powerful tool in modern agriculture nematode control.

Sandra Bredenbruch (bredenb@uni-bonn.de), Jennifer Müller, Florian M.W. Grundler, A. Sylvia S. Schleker

Molecular Phytomedicine, University of Bonn, Bonn, NRW, Germany

In today's agriculture, plant parasitic nematodes (PPNs) are a global threat to crop production which is still not easily addressed. Genetic control is restricted and adapted farming methods are often inefficient, not introduced or economically less attractive. The use of "old-generation" synthetic nematode control agents is frequently banned for reasons of environmental safety. Sustainability and the environmental footprint are a main focus in today's industry. In this respect, rhamnolipids, surface active agents of natural microbial origin, represent a substantial opportunity to match these criteria in modern nematode control. We demonstrate that these secondary metabolites interfere with the infection of two agriculturally relevant PPNs, *Heterodera schachtii* and *Meloidogyne incognita*, at concentrations far below the ecotoxicological threshold. In the presence of rhamnolipids, the infection by *H. schachtii* is reduced by up to 90% without indication of direct nematicidal activity. Furthermore, gall development of *M. incognita* is significantly impaired eventually resulting in a reproduction decline for both PPNs. The plant's reactive oxygen species profile induced by rhamnolipids further proves that the underlying control mechanism is of indirect nature. Our findings highlight the potential of rhamnolipids as biocontrol agent in a sustainable and ecologically friendly agriculture.

Keywords: Biocontrol - Biosurfactant - Root-knot nematode - Cyst nematode - Reactive oxygen species.

S7-P32

Endophytic fungi: a biological alternative for the management of root-knot nematodes.

Manuel Silva-Valenzuela[1] (manuel.silva@colpos.mx), Reyna I. Rojas-Martínez[1], Rosa H. Manzanilla-López[2], Martha L. Macías-Rubalcava[3], Sergio Aranda-Ocampo[1], Emma Zavaleta-Mejía[1]

[1] *Fitosanidad-Fitopatología Lab-209, Colegio de Postgraduados, Texcoco, Mexico*; [2] *CEPROBI, Instituto Politécnico Nacional, Yautepec, Morelos, Mexico*; [3] *Instituto de Química, Universidad Nacional Autónoma de México, Ciudad de México, Mexico*

Gall nematodes are a limiting factor for vegetable crop production in Mexico where control is mainly done with nematicides, which represent a health risk. As a result, current research focuses on developing biological alternatives, such as the use of endophytic mutualistic fungi [1, 2, 3]. The objective of our work was to isolate and select endophytic fungi with antagonistic activity against *Meloidogyne enterolobii*, *M. incognita* and *Nacobbus aberrans*, as well as to investigate the mechanisms involved in host protection by the fungi. Tomato plants with a low galling index were collected from nematode infested fields in Sinaloa State and Mexico City, Mexico, to isolate endophytic fungi. A total of 14 isolates were obtained from leaves and roots. The isolates were assessed for: i) their ability to promote chilli pepper and tomato plant growth; ii) their percentage parasitism on eggs and second-stage juveniles (J2); and iii) the nematostatic and nematicidal effect of culture filtrates against the J2. Three out of the 14 isolates tested stood out: *Echria macrotheca* (S3R3B), *Stagonospora trichophoricola* (XRC3) and *Chaetomium globosum* (S1JH2). Results showed that all three isolates promoted growth in one chili pepper cultivar. The S3R3B and XRC3 isolates in one tomato cultivar. The isolate S3R3B and XRC3 parasitized *N*. *aberrans* eggs and S1JH2 only parasitized *M*. *enterolobii* eggs while S3R3B parasitized *M*. *incognita* J2 and *N*. *aberrans* eggs. The S3R3B and XRC3 filtrates were nematostatic and nematicidal to *N*. *aberrans* J2 and the S1JH2 filtrate was only nematostatic to *M*. *incognita* and *N*. *aberrans* J2. Plant resistance induction and protection against nematode infection experiments were also performed with the three isolates. The treatment combinations that stood out were: S1JH2-*N*. *aberrans*-chilli pepper, S3R3B-*M*. *incognita*-tomato, XRC3-*N*. *aberrans*-tomato, and XRC3-*N*. *aberrans*-chilli pepper. All three isolates induced resistance, which was reflected in a reduction of the galling index, J2 numbers per gram of root and root necrosis. The dry weight of root and aerial parts was significantly higher in treated plants when compared to controls. Two ongoing assays will assess if endophytic fungi affect the process of J2 attraction to the roots and, using qPCR, the expression level of defence-related genes. The next step will be to determine whether these endophytic fungi can protect the plants against nematodes under greenhouse and field conditions.

Keywords: Biological control - Endophytic mutualistic fungi - Root-knot nematode.

References:

- [1] Schouten et al., 2016. Annu. Rev. Phytopathol. 54: 121-142.
- [2] Sikora, R. and Dababat, A.E.F. 2007. Nematology 9(6): 771-776.
- [3] Martínez-Medina et al., 2017. New Phytologist 213(3): 1363–1377.

S7-P33

Nematicidal and plant growth-promoting effects of *Bacillus* cf. *firmus* in white-fruited strawberries.

Nik Susič[1] (nik.susic@kis.si), Uroš Žibrat[1], Nika Cvelbar Weber[2], Saša Širca[1], Jaka Razinger[1], Andrej Vončina[1], Matej Knapič[1], Barbara Gerič Stare[1]

[1] *Plant Protection Department, Agricultural Institute of Slovenia, Ljubljana, Slovenia*; [2] *Department of Fruit Growing, Viticulture and Oenology, Agricultural Institute of Slovenia, Ljubljana, Slovenia*

Root-knot nematodes (RKN) are considered the most important group of plant-parasitic nematodes due to their wide range of plant hosts and their role in yield losses in agricultural production systems. A glasshouse and tunnel pot experiments were conducted to test the nematicidal as well as possible plant growth promoting effects of *Bacillus* cf. *firmus* on white-fruited strawberry plants (*Fragaria chiloensis* (L.) Mill.) infected with RKN *Meloidogyne hapla*. Effects of the nematode *M. hapla* and the bacteria *Bacillus* cf. *firmus* on strawberry plants were tested either in isolation or in combination. In the glasshouse pot experiment, the bacteria significantly reduced *M. hapla* population, which was comparable to the application of the chemical nematicide Velum (Bayer). In the tunnel pot experiment, *Bacillus* did not significantly reduce the nematode population compared to the untreated control. Inoculation of *Bacillus* cf. *firmus* spores on plant roots significantly increased the total microbial activity of the substrate and resulted in growth-promoting effects in *Bacillus*-treated plants in the tunnel experiment, in which significantly higher leaf area and crown fresh weight were measured. In the tunnel experiment, *Bacillus*-treated and nematode-infected plants also exhibited significantly higher performance in the physiological parameters photosynthetic rate and effective quantum yield of PSII, as well as higher relative chlorophyll content in leaves. Hyperspectral images of plants in the spectral range of 400–2500 nm followed by principal component analysis (PCA) and t-distributed stochastic neighbour embedding (t-SNE) were used for hyperspectral data exploration and visualization. Kernel PCA (kPCA) was used for dimensionality reduction and feature generation for classifications and regression analysis. Support vector machines approach aided both classifications and regressions. Hyperspectral analysis differentiated plants within the different treatments with overall classification success of over 90%. This demonstrates the usefulness of the remote sensing approach for potential high-throughput phenotyping applications in the field.

Funding: Horizon 2020, Grant agreement No. 817946 – EXCALIBUR.

Keywords: White strawberry - *Meloidogyne hapla* - Root-knot nematode - Biological control - *Bacillus* cf. *firmus*.

S7-P34

Evaluation of different isolates of *Trichoderma spp.* for antagonistic activity against *Meloidogyne incognita*.

Trifone D'Addabbo[1] (trifone.daddabbo@ipsp.cnr.it), Pasqua Veronico[1], Maria Teresa Melillo[1], Mariachiara Zonno[2], Claudio Altomare[2]

[1] *CNR Institute for Sustainable Plant Protection, Bari, Italy*; [2] *CNR Institute of Sciences of Food Production, Bari, Italy*

Although better known as mycoparasites, fungi of the genus *Trichoderma* have been reported to control phytoparasitic nematodes *in vitro* and in the field. In this study, a series of experiments were carried out to assess the activity of different isolates of *Trichoderma* spp. against the root-knot nematode *Meloidogyne incognita*. An *in vitro* screening of culture extracts of *Trichoderma* strains was carried out to assess the production of anti-nematode compounds. Fungal extracts were tested *in vitro* for their activity on egg hatching and nematode juveniles (J2) mobility. Mortality rates of *M. incognita* J2 treated with the fungal extracts varied largely among the strains tested, ranging from no activity to almost complete kill of J2 within 24-hours. Based on these results, greenhouse trials were conducted to assess the anti-nematode capability of *Trichoderma* isolates *in planta* (semi-field conditions) on the tomato susceptible cv. Regina. Fungal suspensions (10 mL of a conidial suspension 10^7 Cfu/mL) were applied either by seedling tray drench 2 weeks before transplanting or at the same time of transplanting in pre infested soil (5 eggs and J2 per g/soil). The control of *M. incognita* infestation was assessed by nematode multiplication rate and gall formation on tomato roots 60 days post infestation (dpi). Soil treatments with fungal spore preparations were found to suppress both nematode multiplication and gall formation on tomato roots, but the suppressive potential was largely variable among the different fungal isolates. Nematode response to soil treatment with the same *Trichoderma* isolates was also tested in growth chamber experiments, on tomato plants grown in 50 ml clay pots, each treated with 5 mL of a 10^7 cfu/mL conidial suspension. Nematode penetration and development in tomato roots were observed at 2, 7, 15 and 35 dpi under a stereomicroscope. Reduction in gall number and female fecundity were observed, particularly when the fungal spores were applied at the same time of J2 inoculation. These results indicate that an appropriate selection of *Trichoderma* isolates can allow the development of effective biocontrol formulations for safe management of root-knot nematodes.

Keywords: Biological control - *Trichoderma* - Root-knot nematode - Tomato.

S7-P35

Cold water extract of *Cucurbitaceae* as basis for future nematode control agents

Eva Degroote[1, 2, 3] (eva.degroote@ugent.be), Jonas De Kesel[1], Stefaan Pockelee[2], Sven Mangelinckx[3], Tina Kyndt[1]

[1] *Epigenetics and Defence Group/ Biotechnology, University of Ghent, Ghent, Belgium (België)*; [2] *Lima Europe, Rumst, Belgium (België)*; [3] *Synthesis, Bioresources and Bioorganic Chemistry Research Group/ Green Chemistry, University of Ghent, Ghent, Belgium (België)*

Plant pathogenic nematodes are responsible for annual agricultural losses worth an estimated 100 billion dollar. Worldwide, nematicides are used to control nematodes, but the regulation concerning these agrochemicals is becoming more strict every year. That is why the need for new and more sustainable products is increasing constantly. In this research, cold water extracts of *Cucurbitaceae*peelings were evaluated as a control strategy against root-knot nematodes. Since rice, a staple crop, and tomato, a cash crop, both suffer a great deal from damage due to root-knot nematodes, *Meloidogyne graminicola* and *Meloidogyne incognita* respectively, the focus of this research was directed to these two pathosystems. Cold peelings extracts (COPE) from different members of the *Cucurbitaceae* family, *e.g.* melon, pumpkin and zucchini, were tested for their resistance inducing capacities. Results showed that the number of galls in rice as well as tomato formed by root-knot nematodes was significantly reduced upon foliar application of melon, pumpkin and zucchini COPE (mCOPE, pCOPE and zCOPE respectively).

Further research was focused on mCOPE. It was revealed that mCOPE not only induces plant resistance against nematodes, but also has a direct nematicidal effect on *M. graminicola* as well as on *M. incognita*. Via RNA-sequencing in rice, our data indicate that the induced resistance might be regulated by ethylene, a hormone known to play a crucial role in the immune system of plants. Gene expression analyses using qRT-PCR showed that in tomato, mCOPE stimulates genes that are important in the immune system upon nematode infection, such as a strong induction of *NIMIN2*, which is described to be regulated by salicylic acid, an important hormone in plant defence against nematodes.

In conclusion, our results show that cold water extracts of Cucurbitaceae peelings can protect rice and tomato via inducedresistance against root-knot nematode infection. Next to that, we revealed that mCOPE has a direct nematicidal effect on these root-knot nematodes. Lastly, in rice as well as in tomato, we have demonstrated that plant hormones play an important role in establishing induced resistance.

Keywords: Root-knot nematodes - Plant extracts - Induced resistance.

References:

- J. De Kesel et al., 2022, Frontiers in Plant Science, (12) 15.

S7-P37

Towards high throughput phenotyping of banana for nematode resistance

Joseph Kisitu[1] (J.Kisitu@cgiar.org), Kanan Saikai[2], James Kisaakye[2], Daniel L. Coyne[2]

[1] *International Institute of Tropical Agriculture (IITA) Uganda office, Kampala, Uganda*; [2] *International Institute of Tropical Agriculture (IITA), Nairobi, Kenya*

Banana is an important staple among small holder farming communities in East Africa. Owing to several biotic constraints to production, banana yield has followed a declining trend with a 4.6% reduction between 2012 and 2019. Plant parasitic nematodes including the root burrowing nematode (*Radopholus* spp), the spiral nematode (*Helicotylenchus* spp), the lesion nematode (*Pratylenchus* spp) amongst others have been sighted as some of the key pests causing banana yield reduction. Breeding efforts to address this challenge produce hundreds of banana lines. Existing banana nematode phenotyping bioassays can hardly accommodate this volume, considering a short time frame. The current study explored the possibility of developing a high throughput bioassay using in-vitro phenotyping approach by modifying the conventional Murashige and Skoog (MS) media to optimize plant growth and provide near natural conditions for contamination free nematode survival under invitro conditions. A nematode resistant diploid banana genotype (SH3142), and a susceptible triploid banana genotype (Valery) were used for this experiment. An initial population of 50 sterile infective root burrowing nematode *(Radopholus similis)* juveniles were used to inoculate individual plants 40 days post introduction of plants on media. After sixty (60) days post nematode inoculation, assessment of nematode reproduction revealed successful nematode colonization in roots in which significant differences were observed between the susceptible (Valery) and resistant (SH3142) genotypes.

Keywords: High throughput - Phenotyping - Banana - *Radopholus similis*.

POSTERS

S8. Nematode omics, metabolism and physiology

S8-P01

Manipulating lipid metabolism in plants as a novel plant parasitic nematode control measure.

Scott Anderson (scott.d.anderson@wsu.edu)

Plant Pathology, Washington State University, Pullman, Washington(WA), United States

Plant parasitic nematodes (PPNs) cause upwards of $180 billion in crop loss worldwide annually. Of these agricultural pests, the root-knot nematodes (*Meloidogyne* spp.) are the most economically destructive. Unfortunately, the removal of chemical nematicides due to health and environmental concerns, along with the dearth of resistant cultivars, has left growers at a disadvantage when it comes to combatting these destructive animals. This research explored an alternative form of PPN control based on manipulating plant lipid metabolism to deliver toxic chemicals to PPNs. This research was based on data from *Caenorhabditis elegans,* a free-living bacteriophagous nematode. When *C. elegans was* fed a specific dietary fatty acid, dihomo-γ-linolenic acid (DGLA), the nematodes became sterile. The sterility was due to a form of programmed cell death called ferroptosis in developing germ line cells. A bioinformatics analysis of the genome of *Meloidogyne incognita* showed that the nematode contained genes homologous to *C. elegans* that were involved in the modulation of ferroptosis as well as de novo lipid biosynthesis and desaturation**.** Using gas chromatography/mass spectrometry (GC/MS) analysis of fatty acid methyl esters, we found the most abundant 20-carbon polyunsaturated fatty acid in *C. elegans* was also present in *M. incognita.* Based on this data, and the highly conserved nature of ferroptosis pathway in animals, we hypothesize that root-knot nematodes would be susceptible to germ-line cell death when exposed to DGLA. Root-knot nematodes only feed on plants, but plants do not make DGLA. Therefore, our goal was to generate plants that produce DGLA by introducing into them two genes, a desaturase gene from a picoalga and an elongase gene from moss. We generated stably transformed Arabidopsis and tomato plants to express these genes, and GC/MS analysis of the roots of the transformed plants indicated that they could make DGLA. Initial root-knot nematode infection data suggested that Arabidopsis roots that produce DGLA have less galling compared to the control plants. Overall, our data on nematode fatty acids and ferroptosis will provide a better understanding of the core lipid biosynthesis pathways in parasitic nematodes and provide a novel method for nematode control.

S8-P02

The search for "parasitism-genes" readers in the world's most damaging plant-parasitic nematode, *Meloidogyne incognita.*

Caroline Bournaud[1] (bournaudcaroline8@gmail.com), Priscila Grynberga[2], Martine Da Rocha[3], Etienne G.J. Danchin[3], Peter Thorpe[4], Sebastian Eves-van den Akker[5]

[1] *University of Cambridge, Cambridge, Cambridgeshire, United Kingdom*; [2] *EMBRAPA, Brasília, Brazil*; [3] *INRA Sophia Antipolis, Antibes, France*; [4] *University of St Andrews, St Andrews, United Kingdom*; [5] *University of Cambridge, Cambridge, United Kingdom*

Cyst- and root-knot nematodes are the two most devastating groups of plant-parasitic nematodes for worldwide agriculture. Like other pathogens and parasites, plant parasitic nematodes have evolved sophisticated strategies to manipulate plant development, physiology and immunity. They deliver a number of 'effector proteins', produced primarily in dorsal and sub-ventral glands, into the host plant to hijack and perturb cellular and other processes. Elucidating the molecular mechanisms involved in the regulation of effector gene expression may provide novel attractive targets for nematode disease control. The discoveries of the DOG box (a 6 bp motif highly enriched in promoters of cyst nematode dorsal gland effectors), and more recently the STATAWAARS box (a 10 bp motif highly enriched in promoters of pine wilt nematode pharyngeal gland effectors) provide tangible insights into the transcriptional regulation of effectors and therefore the molecular mechanisms upstream of effector biogenesis. Ultimately, these putative cis-regulatory elements may lead to the discovery of unifying regulatory proteins/factors (aka readers) that coordinate effector gene expression. In the present project, we propose to extend this knowledge to the most damaging and widespread plant-parasitic worm: the root-knot nematode *Meloidogyne incognita*. A preliminary *in silico* search has revealed a DNA motif enriched in the upstream regions of *M. incognita* effector genes specifically expressed in dorsal gland cells. This sequence is unrelated but conceptually similar to the DOG box in other plant-parasitic nematodes, and we tentatively termed it 'Mi-DOG' box. Future goals of this project will be to identify candidate 'readers' of Mi-DOG, and to understand how these candidate readers actually work *in vivo*. Ultimately, the goal is to highlight parasitism-readers as promising novel targets for nematode control.

Keywords: Plant-parasitic nematode - Root-knot nematode - Promoter motif - Effectors.

S8-P03

A bioinformatics pipeline for the characterization of small RNAs involved in the plant-root knot nematode interaction.

Martine Da Rocha[1] (martine.da-rocha@inrae.fr), Clemence Medina[2], Laetitia Perfus-Barbeoch[3], Bruno Favery[1], Stéphanie Jaubert-Possamai[1]

[1] *Institut Sophia Agrobiotech, INRAe, Université Cote d'azur, CNRS, Sophia-Antipolis, France*; [2] *micropep, Toulouse, France*; [3] *Institut Sophia Agrobiotech, Université Nice Cote D'Azur, INRAe, CNRS, Sophia-Antipolis, France*

Root-knot nematodes (RKN), genus *Meloidogyne*, are plant parasitic worms that have the striking ability to transform plant cell fate from root vascular cylinder into hypertrophied, multinucleate and metabolically highly active feeding cells. Since RKN are able to induce similar feeding cells in roots of thousands of plant species, these worms are thought to manipulate essential and conserved plant molecular pathways. Redifferentiation into feeding cells is the result of a massive transcriptional reprogramming of root cells targeted by RKN with a large repression of plant gene expression. Small noncoding RNAs include microRNAs and small interfering RNAs (siRNAs) are 20-24 nucleotides RNAs that play a key role in the regulation of gene expression in eukaryotes at the transcriptional and the post-transcriptional level. Our work aims to investigate the role of plant and nematode microRNAs and siRNAs, in the control of the massive transcriptional reprogramming observed during redifferentiation of root cells into feeding cells. Understanding the role of small-RNAs in this biological process was initiated through sequencing of small RNA extracted from tomato galls induced by *Meloidogyne incognita,* from uninfected roots and from preparasitic *M. incognita* second stage juveniles. Analysis of these datasets, in the context of plant-RKN interaction, is challenging due to the complexity of i) the biological samples composed of both plant and nematode tissue and ii) of the difference of smallRNAs processing pathways between plant and animals. Here, we present a pipeline we developed for smallRNAs analysis in this biological context. This pipeline includes bioinformatic prediction of new small-RNAs, annotation of microRNAs with comparison with microRNAs listed in miRBase, and comparison of expression level in the various samples using DESeq2. This pipeline identified the tomato smallRNAs differentially expressed in galls compared to uninfected plants and identified the smallRNAs loci in the genome of *M. incognita*.

Keywords: Root-knot - siRNAs - microRNAs - Bioinformatic prediction - Small noncoding RNAs.

S8-P04

Early life stress promotes aggression and inhibits learning in male nematodes.

Johnathan Dalzell (j.dalzell@qub.ac.uk), Neil Warnock, Lloyd Steele-Nicholson, Daniel Bunney

Queen's University Belfast, Belfast, Northern Ireland, UK

Developmental experience is known to alter aggression levels across various organisms, including humans. However, our understanding of this phenomenon is obscured by a lack of tractable model organisms. Nematodes within the *Steinernema* genus are obligate insect pathogens. Uniquely among nematodes, *Steinernema* spp. males will engage in lethal fighting with other males to secure access to resources. This lethal fighting phenotype is altered as a factor of developmental experience. Males that develop directly from the Infective Juvenile (IJ) stage are significantly more aggressive than males that develop through the entire life-cycle *in vivo*, avoiding the IJ stage. This phenotype switch allows us to study the impact of early life stress on transcription and behaviour through comparing males derived immediately from IJs versus those that are not immediately derived from IJs. We aimed to use *Caenorhabditis elegans* as a 'behavioural outgroup' as it can transition through the dauer stage, which is triggered by early life stress representing an analogous stress control in a nematode species that has not been reported to engage in lethal fighting. However, we found that dauer experience leads to significantly decreased associative learning capacity in *C. elegans* males. Using transcriptomic and small RNA sequencing, we have begun to characterise the transcriptional landscape of early life stress in these nematode species revealing intriguing similarities and differences underpinning behavioural changes. These nematodes represent interesting model systems to study the mechanistic impact of early life stress.

Keywords: *Steinernema carpocapsae* - *Caenorhabditis elegans* - Transcritpome - Non-coding RNA - Behaviour.

S8-P05

Temperature modulates tomato gene expression networks, root exudate composition and parasite interactions.

Johnathan Dalzell (j.dalzell@qub.ac.uk), Steven Dyer, Brian Reilly, Deborah Cox, Neil Warnock

Queen's University Belfast, Belfast, Northern Ireland, UK

Climate change is predicted to alter crop – parasite interactions, however we have little insight to the molecular and physiological basis of these changes. We have found that *Globodera pallida* and *Meloidogyne incognita* second-stage juveniles respond differently to root exudates collected from developmentally matched tomato seedlings that were grown at 18, 23, or 28°C. *G. pallida* is significantly more attracted to root exudates collected from seedlings grown at 18°C, whereas *M. incongnita* is significantly more attracted to exudates collected from seedlings grown at 28°C. In both cases, the temperature for PPN behavioural assays was controlled, indicating that the behavioural change arises from modulation of root exudate composition. Metabolomic analysis of the exudates by LC-Qtof- and GC-MS revealed substantial changes in exudate composition. Transcriptome and small RNA sequencing of tomato seedlings grown at each of the experimental temperatures revealed substantial changes in the expression of genes, long non-coding RNAs, microRNAs and phased siRNAs, contributing to a complex temperature-modulated regulatory landscape. These data provide insight to the impact of temperature on tomato biology, and parasite behaviour ex planta, underpinning efforts to develop new sources of climate-smart resistance.

Keywords: Plant Parasitic Nematode - *Meloidogyne incognita* - *Globodera pallida* - Plant root exudate - Temperature.

S8-P06

Parasitic success without sexual reproduction: what more than 10 years of root-knot nematode genomics revealed?

Etienne Danchin (etienne.danchin@inrae.fr)

Institut Sophia Agrobiotech, Sophia Antipolis, France

Plant-parasitic nematodes are responsible for the destruction of ca. 11% of the worldwide life-sustaining crop production every year. Feeding an ever-growing human population without affecting more negatively our planet, will require important efforts aiming at reducing the agricultural losses caused by plant pests. In this context, understanding how these parasites have evolved and how adaptive they are is highly important. Plant parasitism has evolved at least four times independently in the phylum Nematoda and the root-knot nematodes (RKN) are the most devastating of them. Curiously, the RKN that display the wider range of compatible host plants and the broader geographic distribution reproduce without sex and meiosis. This parasitic success without sex has long been considered an evolutionary mystery.

In 2008, we coordinated the genome sequencing and analysis of the RKN *Meloidogyne incognita*, using 1st generation sequencing technology (Sanger). This was the first genome for a plant-parasitic animal and the first for a metazoan species reproducing without sex and meiosis. In 2017, we published a more complete genome assembly for *M. incognita* and produced genome sequences for two other devastating asexually reproducing RKN, *M. javanica* and *M. arenaria,* using 2nd generation sequencing technology (illumina + 454). This provided the most comprehensive set of protein-coding genes for a plant-parasitic nematode and enabled comparative genomics analyses. Phylogenomics analyses based on these new genomes and gene sets revealed the polyploid hybrid origin of these species. This also allowed demonstrating that the resulting gene copies differ in their expression patterns and some have underwent diversifying selection. Recently, using population genomics on the genome of *M. incognita* allowed confirming the absence of sexual recombination in this species, which reinforced the paradoxical nature of the parasitic success.

In this presentation, I will summarize what the 1st and 2nd generation genomics analyses have allowed to learn and understand about the genome structure of parthenogenetic RKN in relation to their surprising parasitic success despite their lack of sexual reproduction. I will also give an overview of the latest progresses obtained thanks to 3rd generation long read sequencing and the new opportunities opened.

Keywords: Evolutionary genomics - Parthenogenesis - Polyploidy - Hybridization.

S8-P07

The transcriptional regulation of plant-nematode parasitism.

Sebastian Eves-van den Akker[1] (se389@cam.ac.uk), Helen Beasley[2], Clement Pellegrin[2], Caroline Bournaud[2], John Jones[3], Peter Urwin[4], Margarida Espada[5], Manuel Mota[5], Etienne Danchin[6]

[1] *University of Cambridge, Cambridge, Cambridgeshire, United Kingdom*; [2] *University of Cambridge, Cambridge, United Kingdom*; [3] *University of St Andrews, St Andrews, United Kingdom*; [4] *University of Leeds, Leeds, United Kingdom*; [5] *Universidade de Évora, Evora, Portugal*; [6] *INRA Sophia Antipolis, Antibes, France*

Plant-parasitic nematodes are estimated to cost world agriculture over $100 billion per year. Like many pathogens, parasites, and symbionts, plant-parasitic nematodes deliver 'effectors' into their host during parasitism to manipulate plant delvelopment and immunity. What makes plant-parasitic cyst nematodes different from many other plant pathogens is that they have three major specialised gland cells in which effectors are produced. Concerted expression of effectors necessitates concerted regulation of effectors. We have taken advantage of the genomics revolution in plant-parasitic nematology to identify a number of putative cis-regulatory elements associated with subsets of nematode effector repertoires. In the potato cyst nematodes, the dorsal gland Box (DOG box) is a 6 bp motif of the consensus sequence ATGCCA that is enriched approximatley 150 bp upstream of the coding start in dorsal gland effectors. We exploited this discovery for utility by predicting a putative effector superset associated with this motif. We validate gland cell expression in two novel genes by *in situ* hybridisation and catalogued a dorsal gland promoter element-containing effector repertiore from available cyst nematode genomes. The DOG box appears to be conserved in all cyst nematode genomes we have studied to date from the Heterodera and Globodera genera. More recently we have expanded these analyses to identify conceptually similar, but sequence unrelated, motifs in other economically or ecologically important plant-parasitic nematode genomes (e.g. the root-knot nematode MiDOG box, and the pine wilt nematode STATAWAARS). Current efforts are focused on identifying the putative regulators that may recognise these motifs.

Keywords: Plant-parasitic nematodes - Promoter motif - Regulation - Effectors.

S8-P08

Chromatin Landscape Dynamics in the Early Development of the Plant Parasitic Nematode *Meloidogyne incognita.*

Rahim Hassanaly-Goulamhoussen[1] (rahim.hassanaly-goulamhoussen@inrae.fr), Augusto R. De Carvalho[2, 3], Nathalie Marteu-Garello[4], A Péré[4], Bruno Favery[4], Martine Da Rocha[4], E. G. J. Danchin[4], Pierre Abad[4], C. Grunau[2], Laetitia Perfus-Barbeoch[4]

[1] *ISA, Université Côte d'Azur, INRAe, CNRS, Sophia Antipolis, France*; [2] *IHPE, Univ Perpignan, CNRS, IFREMER, Univ Montpellier, Perpignan, France*; [3] *ENS, Lyon, France*; [4] *Université Côte d'Azur, INRAe, CNRS, Sophia Antipolis, France*

In model organisms, epigenome dynamics underlies a plethora of biological processes. The role of epigenetic modifications in development and parasitism in nematode pests remains unknown. The root-knot nematode *Meloidogyne incognita* adapts rapidly to unfavorable conditions, despite its asexual reproduction. However, the mechanisms underlying this remarkable plasticity and their potential impact on gene expression remain unknown. This study provides the first insight into contribution of epigenetic mechanisms to this plasticity, by studying histone modifications in *M. incognita*. The distribution of five histone modifications revealed the existence of strong epigenetic signatures, similar to those found in the model nematode *Caenorhabditis elegans*. We investigated their impact on chromatin structure and their distribution relative to transposable elements (TE) loci. We assessed the influence of the chromatin landscape on gene expression at two developmental stages: eggs, and pre-parasitic juveniles. H3K4me3 histone modification was strongly correlated with high levels of expression for protein-coding genes implicated in stage-specific processes during *M. incognita* development. We provided new insights in the dynamic regulation of parasitism genes kept under histone modifications silencing. In this pioneering study, we establish a comprehensive framework for the importance of epigenetic mechanisms in the regulation of the genome expression and its stability in plant-parasitic nematodes.

Keywords: *Meloidogyne incognita* - Epigenetics - Histone modification.

S8-P09

The roles of neuropeptide genes *flp16* and *flp18* in *Pratylenchus vulnus*.

Wan-Chun Lee (r06633006@ntu.edu.tw), Jiue-In Yang

Plant Pathology and Microbiology, National Taiwan University, Taipei, Taiwan

Pratylenchus spp. are considered amongst the most damaging plant parasitic nematodes globally, and growers rely heavily on broad spectrum nematicides for management, a practice that often leads to human health concerns. FMRFamide-like peptides (FLPs) are neuropeptides widely expressed in the nematode nervous system and regulate their behavior, thus could therefore serve as specific nematicide targets. In this study, we aimed to identify the functions of *flp* neuropeptide genes in *P. vulnus*. 3' Rapid Amplification of cDNA Ends (RACE) PCR followed by cloned sequencing were performed to acquire sequences of *flp* genes. Behavior assays were designed to examine the roles of the neuropeptide genes in migration and invasion ability using the *flp* mutants obtained by dsRNA soaking method. *In situ* hybridization was performed to localize *flp* expression sites [1] and neuron reference analysis was performed. As results, a novel *pv-flp18* gene was identified and the existence of *pv-flp16* was confirmed. The migration rate of *pv-flp16* and *pv-flp18* mutants were significantly decreased by 24.2% and 24.6%, respectively. The invasion rate of *pv-flp18* mutants was significantly increased by 73.6%, while *pv-flp16* mutants showed no difference to controls. Microscopic imaging of DIG-labelled *pv-flp* transcript patterns from *in situ* hybridization were consistent with behavioral assay results. Both *pv-flp16* and *pv-flp18* transcripts revealed similar patterns in female, with expressions in VC4, VC5, HSN motor neurons, and uv1 cells. In juveniles, *pv-flp16* transcripts were found in AVE and BDU interneurons while *pv-flp18* transcripts are in M2 pharyngeal motor neuron and ventral nerve cord motor neurons. This is the first FLP research on migratory plant parasitic nematodes. We revealed the roles of *flp16* and *flp18* genes in *P. vulnus*, and shed a light on using FLPs as a potential target for new *Pratylenchus* spp. control strategy development.

Keywords: Root lesion nematode - FMRFamide-like peptides (FLPs) - RNA interference (RNAi) - Nematode behavior assays.

References:

- Thisse, C., and Thisse, B., 2008. Nature protocols. 3(1), 59.

S8-P11

Molecular characterization and functional importance of b-1,4-endoglucanases from the root-lesion nematode *Pratylenchus loosi.*

Seyedeh Negin Mirghasemi[1] (neginmirghasemi@yahoo.com), Franscesca Deluca[2], Elena fanelli[2], Salar jamali[3], Mohamad mehdi sohani[3]

[1] *Plant Pathology Departman, University of Guilan, Iran, Rasht, Guilan, Iran*; [2] *Consiglio Nazionale delle Ricerche (CNR), Istituto per la Protezione Sostenibile delle Piante (IPSP),, Bari, Italy*; [3] *University of Guilan, Iran, University of Guilan, Iran, Rasht, Iran*

The tea root lesion nematode, *Pratylenchus loosi* Loof 1960, is a serious nematode pest causing yield losses in tea plantations all over the world. Plant parasitic nematodes secrete different enzymes, among these several β-1,4 endoglucanases (*ENG*), essential for development and parasitism inside the roots of the host plants.. The current study reports on the isolation and functional characterization of an endoglucanase gene, *Pl-eng*-2, from *P. loosi*.

Using degenerate primers, two amplified fragments were obtained on cDNA template, both coding for *eng* genes. One of the two cDNA fragments was fully sequenced by using RACE experiments and the corresponding gene was isolated and characterized. Real-time PCR shows that Pl-eng-2 has the highest transcriptional level in adult males and females thus confirming that adults are also parasitic in *P. loosi*. Tissue localization of *Pl-eng-2* mRNA in *P. loosi* was at level of subventral pharyngeal glands suggesting an important role during the parasitism. The effect of silencing was followed analyzing nematodes mobility and reproduction rate on carrot disks and the results will be discussed. The comparison of *Pratylenchus* endoglucanase gene structures and their evolutionary significance will be described.

Keywords: β -1,4 endoglucanas - , in Situ Hybridization - Plant Parasitism - RNA Interference - Real-time RT-PCR..

References:

- Castillo, P., & Vovlas, N. (2007), Pratylenchus (Nematoda: Pratylenchidae): Diagnosis, biology, pathogenicity and management. In Nematology monographs and perspectives (Vol. 6, p. 529). Leiden: Brill. https://doi.org/10.1163/ej.9789004155640.i-523.
- Fanelli, E., Troccoli, A., Picardi, E., Pousis, C. and De Luca, F. (2014), "Molecular characterization and functional analysis of four b-1,4-endoglucanases from the root-lesion nematode Pratylenchus vulnus", Plant Pathology, 63, 1436–1445.
- Haegeman, A., Jacob, J., Vanhome, B., Kyndt, T. & Gheysen, G. (2008), "A family of GHF5 endo-1,4-beta-glucanases in the migratory plant-parasitic nematode Radopholus similis", Plant Pathology, 57:581-590.
- Peng, H., Peng, D., Long, H., HE, W., Qiaq, F., Wang, G. & Huang, W., (2014), Characterisation and functional importance of b-1,4 endoglucanases from the potato rot nematode, Ditylenchus destructor, Nematology, 16: 505-517.
- Uehara, T., Kushida, A., & Momota, Y. (2001), "PCR-based cloning of two beta-1,4-endoglucanases from the root-lesion nematode Pratylenchus penetrans", Nematology, 3:335-341.

S8-P12

Comparative genomics of parasitic nematodes.

Makedonka Mitreva (mmitreva@wustl.edu)

Department of Internal Medicine, Washington University School of Medicine, St. Louis, MO, United States

Parasitic nematode infections in humans cause substantial mortality and morbidity, especially in tropical regions of Africa, Asia, and the Americas, and in agricultural animals and plants. They disproportionately reduce nutritional resources of the global poor. Progress in the development of novel therapeutics and diagnostics has been impeded by the complex biology and lifecycles of the parasitic nematodes. Our objective was to produce high quality assembly and genome annotations from species spanning the phylum Nematoda and facilitate broad comparative studies. Comparative studies of nearly 60 nematode genomes (generated by us and others), included major parasitic species. Gene family births and gene family expansions at key nodes in the phylogeny that are relevant to parasitism, and reconstruction and comparison of metabolic pathways revealed extensive lineage-specific differences in core metabolism. We complemented the genetic and metabolic potential of these parasites with a wealth of expression data at gene and protein level identifying stage- and tissues-specific genes and gene products for a subset of parasitic species. In advanced bioinformatic analysis of these multi-omics datasets, molecular processes of importance to organism's development and of importance to specific tissue types were determined. These nematode omics data provide an invaluable resource to enable the development of postgenomic tools that provide a basis for a multi-omics approach to obtain omics-based discovery of information essential for developing of novel diagnostics, vaccines and anthelmintics. It is essential to follow-up to the genome project by depicting the complexity of the nematode proteome and metabolome (structure and function), evolutionary insights gained from comparative analysis, and subsequent laboratory studies to confirm the multi-omics findings and predictions.

Keywords: Parasitic nematodes - Genomes - Transcriptomes - Proteomes - Multi-omics.

S8-P13

Elucidating the mode of action of a novel nematicide, fluensulfone, using plant parasitic nematode, *Globodera pallida*.

Emily Feist (ef1g12@soton.ac.uk), Matthew Terry, Vincent O'Connor, Lindy Holden-Dye

Biological Sciences, University of Southampton, Southampton, United Kingdom

Fluensulfone (FLS) is a novel nematicide with a distinct profile of effects on nematode behaviour in comparison to other nematicides, suggesting a unique mode of action (Kearn et al. 2014). The effect of FLS on PPN *Globodera pallida* is underpinned by a progressive metabolic insult, observed as impaired motility, reduced metabolic activity and sustained levels of stored lipid (Kearn et al. 2017). Here, we use techniques including histological staining, spectroscopy, mass spectrometry and biochemical analysis to delineate this metabolic insult. Exposure to FLS was investigated in *G. pallida* J2s and compared to known metabolic inhibitors. Fatty acid oxidation inhibitor, perhexiline (PER), elicited a similar profile of effects to FLS. In healthy J2s, MTT staining, which is an indicator of metabolic activity, is most intense in the head, consistent with a high concentration of mitochondrial activity in this region. In J2s exposed to FLS and PER, we observed an accumulating paralysis that progressed to death, accompanied by a loss of MTT staining. Prior to a complete loss of MTT staining, there was a redistribution of staining that manifest as a loss of intense anterior staining and a shift to predominant posterior staining in both treatments. This phenomenon was not common to other metabolic inhibitors tested, suggesting it is not as a result of a generalised metabolic insult. It has been shown that MTT formazan localises in lipid droplets. As lipid stores in J2s are selectively associated with their posterior region, the results are consistent with the known mode of action of PER in preventing fatty acid oxidation and suggest that FLS may be acting via a common mechanism. Coherent anti-Stokes Raman scattering (CARS) spectroscopy was used to visualise lipids following FLS treatment and suggested that it resulted in an impairment of lipid utilisation. To further understanding of FLS's mechanism of action and its effect on lipid utilisation, we have used mass spectrometry to determine the lipid species and fatty acid profiles of treated *G. pallida* J2s. These results were compared to PER treatment and performed in parallel with behavioural analysis. Furthermore, biochemical assays to measure the concentration of triglycerides, which is the predominant lipid species in PPNs, was performed. The range of techniques used have allowed us to further the understanding of the mechanism of action of FLS on PPNs and, specifically, its effect on lipid metabolism.

Keywords: Lipids - Metabolism - Coherent anti-Stokes Raman scattering spectroscopy - Mass spectrometry - Perhexiline.

References:

1. Kearn et al., 2014. Pesticide Biochemistry and Physiology. 109: 44-57.
2. Kearn et al., 2017. Pesticide Biochemistry and Physiology. 142: 83-90.

S8-P14

Unravelling the demographic history of a Pleistocene nematode.

Laura Pettrich (l.pettrich@uni-koeln.de), Laura Villegas, Philipp Schiffer, Ann-Marie Waldvogel

Departement for Biology, Institute of Zoology, University of Cologne, Cologne, NRW, Germany

Various models allow inferences of population history by reconstructing the genealogy of sequences from genomic data. However, a general problem with any model is that they are simplifications of processes in nature, which is the reason for constant optimization of the developed models or the development of entirely new models. This is also true for coalescence models inferring past population demography since they disregard life-traits like dormancy or asexual reproduction.
The recently released eSMC (ecological Sequentially Markovian Coalescent, [1]) approach accounts for precisely these life-cycle traits and thus promises to overcome known inaccuracies when inferring population histories. Here, we tested eSMC on a particularly challenging case that is reconstructing the demography of an ancient species along evolutionary large (i.e. geological) time-scales. The recently revived nematode species *Panagrolaimus kolymaensis,* which likely persisted in the cryobiotic state in the Russian permafrost since the late Pleistocene, is an ideal test-case for the eSMC approach, as it also exhibits other difficult traits such as asexuality or tetraploidy.
Our goals were 1) to establish a pipeline and 2) to identify potential pitfalls arising while working with genomic data of an ancient species, as well as 3) to reconstruct the demographic history of this ancient species. We additionally benchmarked results against the likewise inferred population history estimates of an arthropod species.

Keywords: Population history - Population genomics - eSMC - Ancient species.

References:

- [1] Sellinger et al., 2020. PLOS Genetics. 16(4): e1008698.

S8-P15

Temperature Response of Metabolic Activity of an Antarctic Nematode.

Colin Robinson[1] (cr464@byu.edu), Lee Hansen[2], Xia (Summer) Xue[3], Byron Adams[4]

[1] *Department of Microbiology and Molecular Biology, Brigham Young University, Provo, Utah, United States*; [2] *Department of Chemistry and Biochemistry, Brigham Young University, Provo, Utah, United States*; [3] *Precision Medicine Center, Zhengzhou University, Zhengzhou, 450000, China*; [4] *Department of Biology, Brigham Young University, Provo, Utah, United States*

Because of climate change, the McMurdo Dry Valleys of Antarctica (MCM) have experienced increased summer temperatures and surface ice and snow melting events (Nielsen et al., 2011). In response to these environmental changes, some nematode species in the MCM have experienced steady population declines over the last three decades, but *Plectus murrayi,* a mesophilic nematode species, has responded with a steady increase in range and abundance (Andriuzzi et al., 2018). To determine how *P. murrayi* responds to increasing temperatures we measured metabolic heat and CO2 rates and calculated O2 consumption rate as a function of temperature at 5°C intervals from 5 to 50°C (Criddle et al., 1990). Heat, CO2 production, and O2 consumption rates increase approximately exponentially up to 40°C, a temperature never experienced in the polar habitat. Metabolic rates decline rapidly above 40°C and are irreversibly lost at 50°C due to thermal stress and mortality. *Caenorhabditis elegans,* a much more widespread nematode that is found in more temperate environments reaches peak metabolic heat rate at just 27°C, above which it experiences high mortality due to thermal stress. At temperatures from 10 to 40°C, *P. murrayi* produces about 6 times more CO2 than the O2 it consumes, a respiratory quotient indicative of either acetogenesis or *de novo* lipogenesis. No potential acetogenic microbes were identified in the *P. murrayi* microbiome. We, therefore, conclude that *P. murrayi* is producing increased CO2 as a byproduct of *de novo* lipogenesis. This phenomenon, in conjunction with increased summer temperatures in their polar habitat, will likely lead to increased CO2 production, population abundance, and range expansion.

Keywords: Antarctica - Carbon cycling - Climate change - Soil temperature - Respiration rates.

References:

- Andriuzzi et al., 2018, Ecology 99, 312–321.
- Criddle et al., 1990, Thermochimica Acta 172, 213–221.
- Nielsen et al., 2011, Polar Biology 34, 1701–1711.

S8-P16

A newly identified volatile sex pheromone of *Caenorhabditis elegans*.

Marika Sagawa[1] (jasmine.0915.snow@gmail.com), Matthew Gronquist[2], Frank Schroeder[3], Paul Sternberg[4], Ryoji Shinya[5]

[1] *School of Agriculture, Meiji University, kawasaki, Kanagawa, Japan*; [2] *Department of Chemistry, State University of New York at Fredonia, Fredonia, New York, United States*; [3] *Boyce Thompson Institute, Cornell University, Ithaca, New York, United States*; [4] *Division of Biology and Biological Engineering, California Institute of Technology, Pasadena, California, United States*; [5] *School of Agriculture, Meiji University and JST PRESTO, Kawasaki, Kanagawa, Japan*

Caenorhabditis elegans utilizes volatile pheromones, which are secreted conditionally: *C. elegans* hermaphrodites that do not harbor sperm produce volatile sex pheromones to attract adult males [1]. Previous research has shown that the male chemotactic behavior to volatile sex pheromones required amphid sensory neurons (AWA neurons) and the male-specific cephalic neurons (CEM neurons) [2, 3]. However, it remains unknown what compounds act as the volatile sex pheromone. Recently, we successfully identified one of the active candidate compounds in the volatile sex pheromone (hereinafter, referred to as "X"). We performed the chemotaxis assay to test whether the response to X is sex-specific and whether CEM neurons and AWA neurons were involved in responding to X. First, we tested *C. elegans* wild-type males and hermaphrodites for their attraction to X. Males were attracted to X over a wide concentration range while hermaphrodites were not attracted at low concentrations. Second, we tested CEM and AWA neuron defective mutants. CEM-defective mutant strain completely lost their chemoattraction response toward X while AWA-defective mutant strain responded well to X. These results show that X was a sex pheromone compound that specifically attracts males through the male-specific CEM neurons.

Keywords: Sex attractant - Pheromone - *Caenorhabditis elegans*.

References:

- [1] Leighton et al., 2014. PNAS 111(50): 17905-10.
- [2] Chasnov et al., 2007. PNAS 104(16): 6730-35.
- [3] Wan et al., 2019. EMBO reports 20(3):e46288.

S8-P17

Beyond omics: establishing new nematode model systems to study the evolution of parthenogenesis (and development).

Philipp Schiffer (p.schiffer@uni-koeln.de)

Worm-Lab, University of Cologne, Köln, Germany

The transition from costly sexual to asexual (parthenogenetic) reproduction is rare in animals. It has many implications on the biology of animals, including on their development and genome structure, where systems have to drastically and quickly change to allow for reproduction without male input. Nevertheless, in nematodes we find several genera with many parthenogenetic species each, such as in *Plectus*, *Meloidogyne*, *Acrobeloides*, or *Panagrolaimus*. Oddly, the model genus *Caenorhabditis* appears to be void of any parthenogens, despite the presence of several hermaphrodites among the ~50 or so identified species. Thus, to understand the genomic and developmental consequences of life without sex, we cannot rely on the model species *C. elegans*, nor any of its congeners. In this talk, I will firstly show results from our recent study on meiotic parthenogenetic and sexual *Panagrolaimus* nematodes. I will illustrate how we combined classical cytological and omics analyses to study polyploid genome structures, and identify traits connected to the transition to parthenogenesis in these nematodes. Secondly, I will then discuss an interesting comparison to the close hermaphroditic outgroup *Propanagrolaimus*, which in comparison to *Panagrolaimus* is not cryptobiotic. We identified several groups of genes potentially connected to the evolution of cryptobiosis, including some gained through horizontal gene transfer.
Finally, I will explain how we are now starting to use *Panagrolaimus* as a model to study the evolution to parthenogenesis, using omics data to identify key genes for detailed molecular analyses. In particular, I will also show why we need to study developmental systems in a wide array of nematodes to understand if mechanisms allowing the repeated establishment of parthenogenesis are convergent or rooted in parallel evolution.

Keywords: Sex - Parthenogenesis - EvoDevo - *Panagrolaimus* - Model system.

S8-P18

Highly Polymorphic Regions in the Genome of *Meloidogyne chitwoodi* Reveal Potential Effectors

Joris van Steenbrugge (joris.vansteenbrugge@wur.nl), Sven van den Elsen, Hein Overmars, Muhammad Maulana, Geert Smant, Mark Sterken, Johannes Helder

Laboratory of Nematology, Wageningen University and Research, Wageningen, Netherlands

Plant-parasitic nematodes deploy a concoction of effectors into the host to enable parasitism. The exact composition of this mixture ultimately defines virulence. Consequently, genetic variation, even between populations of the same species may cause differences in virulence. In *Meloidogyne chitwoodi* populations, pathogenicity has been characterised using differentials, however the genetic basis is unknown. We studied this genetic basis by first assembling a near chromosome scale genome assembly of the Dutch Mc31 population, using PacBio sequencing. This reference genome was used to compare the genetic variation between the Dutch population and three populations from the United States that each represents a different pathotype. Our analyses show that there is little variation present over-all, except for four regions that contain a high degree of polymorphisms. We identified four genes in these highly diverse regions that appear to be under diversifying selection and potentially act as effectors. We conclude that while there are many more effector genes present in the genome that are and important for parasitism, these four are the most likely candidates to explain the differences in virulence of these populations.

Keywords: *Meloidogyne chitwoodi* - Genetic variation - Population Genomics - Effectors.

S8-P19

Functional characterization of a highly expanded superfamily of dorsal gland effector proteins in cyst nematodes.

Anju Verma[1] (anju.verma@uga.edu), Marriam Lin[1], Dante Smith[1], Demos Chronis[1], Michael Gardner[1], Ying Lin[1], Tarek Hewezi[2], Thomas Baum[3], Melissa Mitchum[1]

[1] *Dept. of Plant Pathology and Institute of Plant Breeding, Genetics, and Genomics, University of Georgia, Athens, GA, United States*; [2] *Department of Plant Sciences, University of Tennessee, Knoxvlle, TN, United States*; [3] *Dept. of Plant Pathology and Microbiology, Iowa State University, Ames, IA, United States*

Cyst nematodes are one of the most damaging groups of phytonematodes to world agricultural production. These obligate parasites deliver a suite of effector proteins via a stylet to root cells to establish and maintain a metabolically hyperactive feeding site. Stylet-secreted effectors hijack host cellular machinery to promote feeding site formation by modulating the balance between plant development and defense. Despite great progress, the functions of a large number of identified cyst nematode effectors remain unknown. Two novel stylet-secreted effectors of the soybean cyst nematode (SCN) *Heterodera glycines* parasitome, 16B09 and 2D01 [1], belonging to the same superfamily of effectors, are highly expanded in the genome, share the same gene structure, harbor conserved protein domains, and exhibit the same spatial and temporal expression in the dorsal gland cell during parasitism, but exhibit differential toxicity to yeast. Host-induced gene silencing demonstrated a requirement of this effector protein family for successful cyst nematode parasitism. Yeast two-hybrid analysis identified a specific interaction of 2D01 with a plant protein kinase. We further demonstrated that this protein kinase was expressed in feeding sites; plants unable to produce this kinase were less susceptible to infection. A comparative genomics analysis of this effector family across populations of SCN differing in virulence on resistant soybean is currently under investigation.

Keywords: Soybean cyst nematode - Effectors - Superfamily - Yeast two-hybrid - Comparative genomics.

References:

- Guo et al., 2003. Mol Plant Microbe Interact. 16 (8): 720-726.

S8-P20

POSTERS S8

Growing the tree: an update of the Onchocercidae evolutionary history with a multi-locus phylogeny.

Jules Rodrigues[1] (jules.rodrigues@edu.mnhn.fr), Emilie Lefoulon[2], Shigehiko Uni[3], Ricardo Guerrero[4], Nathaly Vallarino-Lhermitte[5], Coralie Martin[1]

[1] *UMR7245 Molécules de Communication et Adaptation des Microorganismes, Natural History Museum, Paris, France*; [2] *School of Animal and Comparative Biomedical Sciences, University of Arizona, Tucson, USA*; [3] *Institute of Biological Science, University of Malaya, Kuala Lumpur, Malaisie*; [4] *Laboratorio de Biologia de Vectores y Parásitos, Universidad Central de Venezuela, Caracas, Venezuela*; [5] *Natural History Museum, Paris, France*

Objective:
Filariae are parasitic roundworms belonging to the Onchocercidae, a family of Nematodes. They are vectorized by haematophagous arthropods and they infect terrestrial vertebrates. Based on their morphology, around 600 species have been described distributed in 90 genera and 8 subfamilies. Previous phylogenetic studies have shown that this family is monophyletic. However, the number of available taxa was limited and few genes were accessible. These factors restricted the global evolutionary history understanding of this family. By sampling a larger number of filariae isolated from the wild fauna and by using a multi-locus sequence typing, combining mitochondrial (*cox1*, 12S rDNA) and nuclear genes (18S rDNA, 28S rDNA, *hsp70*, *MyoHC*, *rbp1*), a more comprehensive phylogeny was inferred in 2015. The three most derivated subfamilies (Dirofilariinae, Onchocerciinae and Splendidofilariinae) topology, by being paraphyletic, are not congruent with the morphological delineations and a new way to classify phylogenetically the Onchocercidae was then proposed (ONC1 to ONC5). Since then, new species has been molecularly identified. In widening and reviewing a filariae phylogeny on all the available Onchocercidae data, the objective is to better understand the higher taxonomic level of the Onchocercidae.
Methods:
Sequences were produced at the MNHN or came from GenBank. A sequence of reference from Genbank was selected based on the consensus score of the sequence, the number of degenerated nucleotides and the length of the sequence. For each genes the sequences were aligned, then the datasets were concatenated to perform a phylogeny using a Bayesian inference.
Results & Conclusion:
This phylogeny confirms the discrepancies of the subfamily taxonomy. A new version of the ONC classification is proposed, with a sixth clade added. The new ONC clade is linked to the genus *Dipetalonema*, due to being too distinct from the others ONC4 taxa. Another subfamily incongruence is the taxa of the Dirofilariinae which are distributed in two distinct clades.
One sixth of the filariae species has been molecularly characterized, although many lack nuclear genes which limits the global evolutionary history understanding of the Onchocercidae. But for the first time, albeit for one gene, all the subfamilies are present in a comprehensive phylogeny.

Keywords: Nematode - Filariae - Phylogeny - Evolution.

POSTERS

S9. Entomopathogenic nematodes

S9-P01

Diversity of entomoparasitic nematodes in the rose chafer, *Cetonia aurata* grub.

Luca Eszter Balog[1] (baloglucaeszter@gmail.com), Oleksandr Holovachov[2]

[1] *Dep. of Systematic Zoology and Ecology, Eötvös Loránd University, Budapest, Hungary*; [2] *Department of Zoology, Swedish Museum of Natural History, Stockholm, Sweden*

Intensive nematological research is essential for effective biological control of insect pests. The majority of studies focus on only two entomopathogenic nematode families: Steinernematidae and Heterorhabditidae. However, there are other entomoparasitic nematode species that have recently been found in scarab beetles from Hungary and may have biocontrol potential. These species differ in their evolutionary strategies. Obligate parasitic nematode species spend their whole life inside the host's body, as opposed to the facultative entomopathogenic nematodes, which have both free-living and insect-parasitic stages. Current knowledge about host specificity among obligate parasitic and necromenic species and their impact on insects is still very incomplete. In this study, rose chafer, *Cetonia aurata* (Coleoptera: Scarabaeoidea) grubs were collected from soil and compost from nine different places in Hungary. More than 100 insect specimens were dissected under stereomicroscope. Some of the isolated nematodes were cultured using a nematode growth medium. Nematodes were identified using light and scanning electron microscopy. The intestine of many grub specimens was infected with Thelastomatidae nematode species, while the insect's coelom was mainly infected with nematode species from the Rhabditidae and Panagrolaimidae families. In addition, several free-living nematode species were found and identified from the insect grubs. This study discovered many species of nematodes that had not previously been registered in Hungary and that were new to the insect host species. Most studies in the literature focused on only a few genera of entomopathogenic nematodes, and less attention has been paid to other entomoparasitic, necromenic, phoretic, entomopathogenic and obligate parasitic groups. Our results show that there is a high diversity of insect-associated nematodes other than Steinernematidae and Heterorhabditidae, that has not been recognized so far. For instance, nematodes from the families Rhabditidae and Panagrolaimidae found in the grubs could in future be investigated as alternative bio-control agents.

Keywords: Rose chafer beetle grubs - Entomopathogenic nematodes - Necromenic nematodes - Obligate parasitic nematodes.

S9-P02

Host-seeking behavior of the native entomopathogenic nematode *Steinernema unicornum* and its phylogenetic relationship with the exotic strains marketed in Chile.

Nicolás Betanzo[1] (n.betanzo01@ufromail.cl), Graciela Berríos[2], Ivonne Alvarez[2], Patricia Navarro[2]

[1] *Laboratorio de Biocontrol, Universidad de La Frontera, Temuco, Chile*; [2] *Laboratorio de Entomología, INIA Carillanca, Vilcún, Chile*

Steinernema unicornum is a native entomopathogenic nematode (EPN) isolated in 2007 from Tierra del Fuego archipelago at the southern extremity of South America in Chile. Scarce information is available about its biology, ecology and phylogenetic relationship with other native and exotic EPNs. So far, this nematode has been evaluated against different insect species; however, its foraging strategy is unknown. The objective of this study was to evaluate the host-seeking behavior of *S. unicornum*, *Steinernema feltiae* strain Koppert and *Steinernema feltiae* strain Biobee and their phylogenetic relationship. To achieve this objective, both horizontal and vertical movement were evaluated. Additionally, the phylogeny of these species was determined and their distances compared. Molecular analyses were conducted amplifying the sequences of the 18S (Primers 18S and 26S), LSU (391 and 501), 28S (TW81 and AB28) and D2D3 regions (D2F and 536r). Data obtained from the mobility tests were analyzed with ANOVA and differences among means were determined using Tukey´s test. Sequencing data were compared using BLAST, the alignment was performed using the bioinformatic software Geneious v.4.8 and the phylogenetic analyzes using the neighbor-joining and maximum likelihood approaches (MEGA v.7.0 software). Mobility results showed an ambusher foraging behavior for all species evaluated. The vertical movement of *S. unicornum* ranged from 5 to 10 cm, and *S. feltiae* strain Koppert and *S. feltiae* strain Biobee moved between 5 and 20 cm. For the horizontal dispersion test all EPNs showed averages lower than 10 millimeters of displacement after 30 minutes. Phylogenetic analysis showed that *S. unicornum* shares a common ancestor with *Steinernema oregonense* (from Arizona) and *Steinernema kraussei* (from Poland), both species belonging to the *feltiae* group. The species *S. feltiae* strain Koppert and *S. feltiae* strain Biobee share a common ancestor with the species *S. weiseri* (from Eastern Europe) and *S. puntauvense* (from Costa Rica). The results obtained not only showed the reduced range of movement of all evaluated EPN species, which was phylogenetically confirmed, but also it is possible to conclude that these nematodes are not a good candidate for insect pests located deep in the soil profile.

Keywords: *Steinernema unicornum* - *Steinernema feltiae* - Foraging strategy - Phylogeny.

S9-P03

Steinernema siamkayai (Rhabditida: Steinernematidae): notes on its morphology, bionomy and distribution from the Indian subcontinent

Aashaq Hussain Bhat[1] (aashiqhussainbhat10@gmail.com), Ashok Kumar Chaubey[1], Jiří Nermuť[2], Vladimir Půža[2]

[1] *Nematology Laboratory, Chaudhary Charan Singh University Meerut, Meerut, Uttar Pradesh, India*; [2] *Nematology, Institute of Entomology, Biology Center, Branišovská, České Budějovice, Czechia*

The entomopathogenic nematode *Steinernema siamkayai* has been a subject of several detailed studies but its morphological, molecular and ecological characterization is still incomplete. In the present work, we studied morphology of several strains of *S. siamkayai* using morphometry and SEM and we aimed to complete its molecular characterization by obtaining the sequence of its D2-D3 region of the 28S rDNA. Furthermore, we studied the life cycle of *S. siamkayai* and we attempted to assess its distribution using ITS sequence based meta-analysis of the GenBank records because the natural occurrence of EPN species facilitates its use in particular areas [1]. Finally, we performed a molecular and biochemical characterization of the bacterial symbiont of *S. siamkayai* and we also tested them in a laboratory assay for pathogenicity against two major pests, *Helicoverpa armigera* and *Spodoptera litura.* The SEM study revealed the presence of the ad-anal pair of genital papillae (papillae at the edge of the cloacal aperture) in males, not observed in previous descriptions [2,3]. Duration of the life cycle of the Indian strains did not differ for the previous records [4]; however, we have observed production of the infective juveniles by the first generation adults not reported in the original description. For the first time in EPNs, we have recorded intraindividual variability in the flanking D2-D3 region of the 28S rDNA and we discuss potential implications of this phenomenon for the use of this marker in EPN systematics. Interestingly, we found that the D2D3 sequence of all the tested strains contained variation in the 3-base segment. Our analysis of distribution has shown that *S. siamkayai* is ubiquitous throughout the Indian subcontinent, but it is rarely found in South East Asia. The sequences of bacterial recombinase A and gyrase B genes and biochemical tests have shown that the symbiont of *S. siamkayai* is closely related to *Xenorhabdus stockiae*, which suggest that this bacterium is widespread among South Asian nematodes from the ''*carpocapsae*'' group [5]. In the virulence assay, strains caused a 100% mortality of both tested insects after 48 h, even at the lowest doses of 25 infective juveniles per insect. These findings and high prevalence of this species throughout the tropical Asia suggest that a biocontrol product based on this species could be used for biocontrol purposes throughout this region.

Keywords: Meta-analysis - Genital papillae - D2D3 region - Intraindividual - *S. siamkayai.*

References:

- [1] Bhat et al., 2019. Acta Parasitol. https://doi.org/10.2478/s11686-019-00061-9
- [2] Stock et al., 1998. Syst Parasitol. 41, 105-113.
- [3] Tabassum et al., 2010. Int J Nematol. 17, 217-224.
- [4] Chongchitmate et al., 2005. Kasetsart J (Nat Sci). 39, 431-439.
- [5] Bhat et al., 2017. J Nematol. 49, 92–102.

S9-P04

Impact of differentiated farming practices on the native entomopathogenic nematodes in DOCa Rioja vineyards (Northern Spain).

Ruben Blanco-Pérez[1] (ruben.blanco@icvv.es), Ignacio Vicente-Díez[1], José Luis Ramos-Sáenz de Ojer[2], Vicente Santiago Marco-Mancebón[3], Ignacio Pérez-Moreno[3], Raquel Campos-Herrera[1]

[1] *Dpto. Viticultura, ICVV-CSIC, Logroño, La Rioja, Spain*; [2] *Investigación Agraria y Sanidad Vegetal, Gobierno de La Rioja, Logroño, La Rioja, Spain*; [3] *Dpto. Agricultura y Alimentacion, Universidad de La Rioja, Logroño, La Rioja, Spain*

Current viticulture is still widely based on intensive tillage and agrochemical applications to control pests, diseases, and weeds. These practices compromise beneficial soil organisms such as the entomopathogenic nematodes (EPNs). We hypothesized that organic farming and the implementation of alternative strategies to tillage might enhance the EPN occurrence and activity. In autumn 2019, we collected two composite soil samples from 80 vineyards distributed all across the Designation of Origin (DOCa) Rioja region, selected to belong to two categories of two factors: soil management (regular tillage and cover cropping, 48 and 32 plots, respectively) and pest management system (integrated "IPM" and organic "OPM", 40 plots each). Using the traditional insect bait method, we assessed three soil activity rates associated with the percentage of *Galleria mellonella* larvae that (i) die (total activity), (ii) exhibit nematode emergence (nematode activity), and (iii) confirmed Koch's postulates (EPN activity). Moreover, the nematofauna was isolated through sucrose-gradient centrifugation, and nematode species were identified by qPCR using species-specific primers-probe sets. We recorded significantly ($P < 0.05$) higher soil activity rates for OPM than IPM (11.3% *vs*. 4.2% for total activity, 3.2% *vs*. 0.4% for nematode activity, and 2.1% *vs*. 0.3% for EPN activity), while not between soil management practices. On the other hand, we found no differences in total EPN abundance for any evaluated treatment. However, *Steinernema feltiae*, the predominant EPN species in this study, recorded significantly ($P < 0.01$) higher numbers for OPM than IPM. Thus, we conclude that organic viticulture could support the native EPN community better than IPM, possibly translating into increased resilience against potential arthropod pests in vineyards.

S9-P05

Steinernema abbasi (isolate CS_2)-*Xenorhabdus indica* complex and *Helicoverpa armigera* immune response.

Ashok Chaubey[1] (akc.nema@gmail.com), Istkhar Rao[2]

[1] *Nematology Laboratory/Zoology, Chaudhary Charan Singh University, MEERUT, UTTAR PRADESH, India*; [2] *Bioscience & Biotechnology, Banasthali Vidyapith, Tonk, Rajsthan, India*

Insects like other organisms are bestowed with immune systems which are comprised of both humoral plasma-borne factors and cellular or hemocytes-associated molecules that are mobilized in response to parasitic and pathogenic infection. Many of these elements act cooperatively during an immune response, revealing a complex level of interaction between cell-based and humoral factors. This study dealt with the influence of *Steinernema abbasi* (CS2)-*Xenorhabdus indica* complex on total hemocyte counts (Miranpuri et al. (1992), differential hemocytes counts (Mowlds et al. 2010) and phenol oxidase activity (Kalia et al. 2001) in *Helicoverpa armigera*. Based on the results of bioassay experiments, *H. armigera* along with controls were exposed to 20 IJ/larva each in six well plates. Hemolymph was collected after each 6 h post infection (PIP) period for 24 h. An average of 10.2×10^5 hemocytes/mm^3 (9.7×10^5-10.9×10^5) was recorded during the experimentation time set for 0 to 24 h duration. However, for total hemocytes count this average was significantly higher (One-way ANOVA, p=0.05) with 11.75×10^5 cells (8.0×10^5-15.5×10^5) in hemolymph of larvae infected with IJ of *S. abbasi* (CS_2). The efficiency of nematode-bacterium infection was evidenced significantly from 3-9 h duration. An increment of 23.53% was observed in average total hemocytes from 10.2×10^5 hemocytes/mm3 at 0 h to 12.6×10^5 hemocytes/mm^3 at 3 h. Six types of hemocytes (Kalia et al. 2001) were observed in the larval hemolymph of *H. armigera* viz., prohemocytes, granulocytes, plasmatocytes, spherulocytes, oenocytoids and adepohemocytes. Granulocytes (24.75-25.73%) and plasmatocytes (40.22-41.81%) contributed more than 60% of the total population of hemocytes whereas the spherulocytes (11.72-12.25%) were the third most abundant cells followed by adepohemocytes (8.26-9.29%) and oenocytoids (7.91-8.93%). Prohemocytes contributed a small fraction of the cell types. Phenoloxidase (PO) activity was highest at 9 h PIP and lowest at 24 h in all three sample types. PO activity in total and cellular component of hemolymph was positively correlated with prohemocytes, granulocytes and oenocytoids.
The study showed that the hemocytes and PO accounted as active immune responses against nematode infection. The results provide the first insight to understand the hemolytic activity, quick immunosuppression responses of *Steinernema abbasi* (CS2)-*Xenorhabdus indica* complex on *Helioverpa armigera*.

Keywords: *Steinernema abbasi* - *Xenorhabdus indica* - *Helicoverpa armigera* - Phenoloxidase activity - Hemolymph.

References:

- Kalia et al., 2001. Indian J Exp Biol. 39:1123-1129.
- Miranpuri et al., 1992. J Invertebr Pathol. 60:274-282.
- Mowlds et al., 2010. Microb Infect. 12:146-153.

S9-P06

Natural populations of entomopathogenic nematodes on sweetpotato farms in southern Ghana.

Yaw Danso (abiacea@yahoo.co.uk)

Nematology Lab, CSIR-Crops Research Institute, Kumasi, Ghana

Cylas species cause sweetpotato root losses ranging from 5 to 97%. The weevils infest the fleshy tuber roots and basal stems, tainting them with bad oduor and bitter taste. This greatly reduces marketability of the storage roots. Applying farm chemical insecticides and cultural practices (crop rotation and earthing-up) are conventional management tactics against *Cylas* species. These management practices are however not sustainable due to high cost as well as human and environmental health concerns, respectively. Entomopathogenic nematodes (EPNs) have proven to be effective biological insecticides against *Cylas* species. One hundred and thirty-five sweetpotato farms were prospected for EPNs in 9 districts of southern Ghana between February and September 2017. The 3rd and 4th instar *Galleria mellonella* (Lepidoptera: Pyralidae) larvae were used to recover EPNs from rhizosphere soil samples employing the Insect Baiting Technique (IBT). The *G. mellonella* cadavers were cultured in an incubation chamber (25 ± 2 ºC and 85% RH) using modified White traps and infective juveniles were collected after 8 days. Of the 675 *G. mellonella* cadaver populations across the study area, 504 (74.7%) tested positive for heterorhabditid EPN parasitism and 171 (25.3%) contained steinernematids. The highest population of 11,959 IJs/5 *G. mellonella* cadavers were recovered from Fanteakwa district, whilst the lowest of 33 IJs/5 *G. mellonella* cadavers were recovered from Akatsi South district. Presence of entomopathogenic nematodes in sweetpotato rhizosphere soils renders the prospect of using these beneficial nematodes for *Cylas* species and other insect pests' management in Ghana promising.

S9-P07

Arthropod community responses reveal potential predators and prey of entomopathogenic nematodes in a citrus orchard.

Alexandros Dritsoulas[1] (alexdritsi@gmail.com), Sheng-Yen Wu[2], Homan Regmi[3], Larry Duncan[3]

[1] *Entomology, Athens Agricultural University, Athens, Greece*; [2] *College of Plant Protection, Fujian Agriculture and Forestry University, Fujian, China*; [3] *Citrus Research and Education Center, University of Florida, Lake Alfred, FL, United States*

The contributions of soil arthropods to entomopathogenic nematode (EPN) food webs are mainly studied in highly artificial conditions. We investigated changes in arthropod and nematode communities in a citrus orchard following the inundation of soil with *Steinernema feltiae* or *Heterorhabditis bacteriophora*. We hypothesized that arthropod taxa which decline or increase in EPN-treated compared to untreated plots represent potential key prey or predators of EPN, respectively. Soil samples were taken 3, 7, 14, and 28 days after nematodes were applied to eight, three-tree plots per treatment. DNA was extracted from the organisms that were separated from soil by sucrose centrifugation, libraries prepared, and the ITS2 and CO1 gene regions were sequenced according to Illumina protocol. Arthropod species from 107 microarthropod (mites and collembola) families and 121 insect families were identified. EPN applications reduced the insect species richness from 32.6 molecular operational taxonomic units (MOTU) in untreated plots to 26.8 in plots with *S. feltiae* (P=0.02) and 30.4 in those with *H. bacteriophora* (NS). Most insect taxa were rare, but several were encountered frequently and their responses to EPN treatments as well as correlations with endemic *H. indica* suggests an important role for an ant (Formicidae) and a non-biting midge (Chironomidae) in maintaining EPN in this orchard. Between days 3 and 28, the numbers of *H. bacteriophora* declined by 80%, more than twice the rate for *S. feltiae* (46%). Consistent with the different EPN disappearance rates, there were 44% fewer microarthropod reads in plots treated with *S. feltiae* than with *H. bacteriophora* (P=0.01). Microarthropod species richness also tended to be higher (P=0.051) in plots treated with *H. bacteriophora* than with *S. feltiae*. The responses over time of the individual microarthropod species suggest that regulation of EPN resulted from a cumulative response by many species, rather than by a few key species. Ascomycete and Basidiomycete sequence reads were reduced (P=0.001) by applications of both EPN species. The effect of EPN on *Fusarium oxysporum* epidemiology as measured by qPCR was congruent with the HTS measurements.

Keywords: Food web - Metagenomics - Conservation biocontrol.

S9-P08

Protein source impact on the recovery and yield of entomopathogenic nematodes, using *in vitro* liquid culture.

Murray David Dunn (mdunn94@sun.ac.za), Antoinette Paula Malan

Department of Conservation Ecology and Entomology, Stellenbosch University, Stellenbosch, Western Cape Province, South Africa

The effective mass production of local entomopathogenic nematodes (EPNs) of the family Steinernematidae, as biological control agents for commercial use against pest insects, is an important step in their future use in an integrated pest management system for many countries [1,2]. EPNs have been successfully produced using *in vitro* liquid culture technology in such areas as Europe and the USA. An important ingredient of the artificial liquid media diet is a suitable protein source that encourages the rapid recovery of infected juveniles to enable the start of their feeding, development and reproduction. Protein has been shown to be essential to the nematode, both as an energy source and for muscle development. Moreover, a good protein source has a significant impact on the growth of the all-important symbiotic bacteria, which, in turn, initiates rapid recovery and high yield. In this investigation, a standard protocol for nematode production was followed, using 30 ml of a complex medium in 250-ml Erlenmeyer flasks on an orbital shaker at 140 rpm at 25°C [3]. Three powdered protein sources, consisting of egg yolk, soy protein and black soldier fly, separately and in combination, were used to determine the optimal protein source for the *in vitro* liquid mass culture, in terms of recovery and yield. The nematodes used were the South African EPN species, *Steinernema jeffreyense* and *S. yirgalemense* [4,5]. Results showed that egg yolk on its own is, by far, the superior source of protein for the recovery and yield of both *Steinernema* species. Egg yolk was found to be rich in essential amino acids and high in cholesterol, which is crucial to nematode development and production, although further research is necessary to confirm the assumption.

Keywords: *in vitro* - Protein - *Steinernema jeffreyense* - *Steinernema yirgalamense*.

References:

1. Malan et al. 2018. Afr. Entomol. 26 (1): 14-29.
2. Malan & Moore, 2016. Afr. Entomol. 24 (2): 489-501
3. Dunn et al., 2019. BioControl https://doi.org/10.1007/s10526-019-09977-7
4. Steyn et al., 2019. Biocontr. Sci. Technol. 27 (11): 1265-1278.
5. Steyn et al., 2019. Biol. Control https://doi.org/10.1016/j.biocontrol.2019.104052

S9-P09

In vitro liquid mass production of *Steinernema jeffreyense,* using a designer desktop bioreactor.

Murray David Dunn[1] (mdunn94@sun.ac.za), Antoinette Paula Malan[1], Prasanna Belur[2]

[1] *Department of Conservation Ecology and Entomology, Stellenbosch University, Stellenbosch, Western Cape Province, South Africa*;
[2] *Department of Chemical Engineering, National Institute of Technology, Karnataka, Surathkal, Mangalore, India*

Entomopathogenic nematodes (EPNs) of the family Steinernematidae are effective biological control agents that can be mass-produced in bioreactor vessels, for use as biocontrol agents against agriculturally important insect pests. The bioreactor mass production of EPNs uses technologically advanced, low-cost *in vitro* liquid culture technology to produce nematodes of a high quantity and quality. The first step in the *in vitro* mass production of nematodes begins with shake flasks, with the process being upscaled to larger bioreactors to meet the scale of demand. The South African EPN species, *Steinernema jeffreyense*, has shown potential to be a good candidate as a biological control agent against various agricultural insect pests of grapevine in the Western Cape Province. To implement the species as a biological control agent on a commercial scale, mass production in bioreactors is required. The study was undertaken to optimise the in vitro liquid culture mass production of *S. jeffreyense* in a 10-L glass designer desktop bioreactor vessel, and to optimise the design of the bioreactor to suit the biology of the nematode concerned. Various challenges were overcome in terms of the design and method, such as the sterility of the environment, the provision of oxygen using an air defuser, the speed of rotation, and the shape and number, of stirred impellers, the addition of a stainless-steel baffle, and the consideration of shear force damage on the growth and development of the nematodes. Successful reproduction, without contamination, was achieved in the bioreactor,with yields of up to 81 000 IJs/ml, using different bacteria and IJ inoculum densities. This is the first time that *S. jeffreyense* has been cultured in a bioreactor vessel. Future prospects include assessing the growth dynamics of the nematode, as well as that of the symbiotic bacteria in the bioreactor vessel, the addition of new measuring devices for dissolved oxygen, and the upscaling to larger vessels of 100 L and 1000 L that requires optimisation through future research

Keywords: Entomopathogenic nematodes - bioreactor - *in vitro* - *Steinernema jeffreyense* - *Steinernema yirgalemense.*

References:

1. Malan et al. 2018. Afr. Entomol. 26 (1): 14-29.
2. Malan & Moore, 2016. Afr. Entomol. 24 (2): 489-501.
3. Dunn et al., 2019. BioControl https://doi.org/10.1007/s10526-019-09977-7
4. Steyn et al., 2019. Biocontr. Sci. Technol. 27 (11): 1265-1278
5. Steyn et al., 2019. Biol. Control https://doi.org/10.1016/j.biocontrol.2019.104052

S9-P10

Heterorhabditis bacteriophora: An excellent model for genetic improvement of biocontrol traits.

Ralf-Udo Ehlers (ehlers@e-nema.de)

e-nema GmbH, Schwentinental, Germany

More than 150 different invertebrates are currently used in biological control of insect pests. For breeding insects and mites, producers of invertebrate biocontrol agents largely depend on sampling and characterising natural populations. Entomopathogenic nematodes (EPN), especially *Heterorhabditis bacteriophora* are different. This contribution will describe the relevant biological peculiarities and briefly introduce relevant techniques to show why this nematode is an excellent model for genetic improvement. EPN biology permits production of inbred lines through self-fertilisation by the hermaphrodite and production of hybrids through crosses of second generation amphimictic adults. The genetic pool can be preserved by storage in liquid nitrogen. Mass production is done in industrial scale bioreactors in liquid culture. EPN have a short life cycle allowing rapid progress by genetic selection. Several traits have been improved, e.g. reproduction, longevity, field persistence and stress resistance to heat, desiccation, reactive oxygen species and nematicides. EPN can be subjected to EMS mutagenesis. Progress of genetic selection is easily lost through outcrossing during mass production. As *Heterorhabditis* spp. are unable to mate in liquid media, reproduction is only through self-fertilising hermaphrodites. The use of well characterised inbred lines can overcome problems of trait deterioration when production is done in liquid media. A large pool of molecular genetic information and tools are available to support breeding of heterorhabditid biocontrol agents.

Keywords: Genetic improvement - Heterorhabditis - Breeding - Inbred lines - Beneficial traits.

S9-P11

Bioactive molecules produced by *Heterorhabditis bacteriophora* affects the phenoloxidase system of *Galleria mellonella*

Sara Eliáš[1] (saraelias43@gmail.com), Hurychová Jana[1], Pavel Dobeš[1], Duarte Toubarro[2], Nelson Simões[2], Hyršl Pavel[1]

[1] *Department of Experimental Biology, Masaryk University, Brno, Czech Republic*; [2] *CIRN and Department of Biology, University of Azores, Ponta Delgada, Portugal*

Entomopathogenic nematodes are able to produce bioactive molecules referred to as excreted/secreted products (ESPs). The ESPs are small molecules, proteins and nucleic acids that can interfere with the host immune system [1,2,3,4] in order to increase the chance of entomopathogenic nematodes reproduction. We tested the effect of ESPs obtained from *Heterorhabditis bacteriophora* on immune system of *Galleria mellonella* larvae. The results indicated suppression of phenoloxidase activity after administration of isolated ESPs. Subsequently, the isolated ESPs were purified in two steps to obtain and identify the active fraction. Firstly, the ESPs were purified based on their charge resulting in five distinct fractions. Two of them inhibited phenoloxidase activity of *G. mellonella* larvae. These active fractions were subjected to a second purification step in which ESPs were separated based on their molecular weight. We obtained seven fractions and we tested their effect on phenoloxidase activity. The purified molecules in the fraction with molecular mass of 38 kDa significantly inhibited phenoloxidase activity of *G. mellonella* larvae. Oncoming analysis of purified active molecules are focused on their identification by mass spectroscopy. The immunosuppressive nature of ESPs is possible to utilize as the biological control of insect pests in agriculture. It is of note that some ESPs contain also compounds with high bioactivity that are able to affect the mammalian immunity or act as the antibiotic agent. Because of these properties, the ESPs can be used as an interesting source of bioactive molecules with potential use in pharmacology. This study was supported by grant from The Ministry of Agriculture of the Czech Republic QK1910286.

Keywords: Entomopathogenic nematodes - Excreted/secreted products - *Heterorhabditis bacteriophora* - *Galleria mellonella* - phenoloxidase activity.

References:

- [1] Balasubramanian et al., 2009. International Journal for Parasitology. 39(9): 975-84.
- [2] Balasubramanian et al., 2010. Parasite Immunology. 32(3): 165–75.
- [3] Toubarro et al., 2009. International Journal for Parasitology. 39(12): 1319-30
- [4] Toubarro et. al., 2013. PLoS ONE. 8(7): e69161

S9-P12

The successful story of Entomopathogenic Nematodes against foliar pests: our silver bullet.

Magda Galeano[1] (mgaleano@koppert.es), Kay Moisan[2], Mireya Baños[1], Kelly Arkoumanea[2], Olga Kostenko[2], Marlies Dissevelt[2], Roxina Soler[2], Jose E. Belda[1]

[1] *R&D department, Koppert Spain, Vicar, Almería, Spain*; [2] *AgDev department, Koppert B.V, Berkel en Rodenrijs, The Netherlands*

Entomopathogenic Nematodes (EPNs) are excellent biocontrol agents against many arthropod species (1, 2) that may also be applied onto the canopy of crops to control agricultural foliar pests (3, 4, 5). Yet, results so far are contrasting, indicating that foliar application of EPNs does not systematically lead to the control of the pests. Fundamental understanding of the EPN biology and engineering technology are key to develop biological control agents and to ensure their successful application. We present here the work carried out by a dedicated R&D team of Koppert which has developed over the past years successful biological products based on the EPNs *Steinernema feltiae* and *S. carpocapsae*. We conducted experiments to break down and characterize the effective steps of the EPNs against foliar pests using a model system. We explored the survival time of nematodes on leaves and the time needed to enter the insects as these two steps are among the most critical for the efficacy of the EPNs against foliar pests. Additionally, we tested the infectivity of the two species of *Steinernema* through efficacy bioassays against a range of Lepidoptera pests in greenhouse settings as well as under field conditions. Application of the EPNs led to more than 70% of mortality of all tested pests. Together, our data show that EPNs are successful biological control agents that can successful target foliar pests.

Keywords: *Steinernema* spp. - Lepidoptera - Temperature - Survival - Efficacy.

References:

- [1] Campos-Herrera, R. 2015. 2015. Springer International Publishing, p. 531.
- [2] Batalla-Carrera et al., 2010. BioControl,55:523–30.
- [3] Lacey et al., 2015. Journal of Invertebrate Pathology, 132:1–41.
- [4] van Damme et al., 2013. Pest Management Science 72:1702–9.
- [5] Mutegi et al., 2017. World Journal of Agricultural Research 5:233–9.

S9-P13

How to Successfully Apply Entomopathogenic Nematodes in an IPM System?

Magda Galeano[1] (mgaleano@koppert.es), Kay Moisan[2], Mireya Baños[1], Kelly Arkoumanea[2], Olga Kostenko[2], Roxina Soler[2], Jose E. Belda[1]

[1] *R&D department, Koppert Spain, VICAR, Almería, Spain*; [2] *AgDev department, Koppert B.V, Berkel en Rodenrijs, The Netherlands*

Several studies have demonstrated the potential of entomopathogenic nematodes (EPNs) to control soil and foliar insect pests (1, 2, 3, 4) under laboratory and semi field conditions. However, scaling-up and implementing EPNs in field conditions within the grower's crop management practices can be challenging. These practical aspects have often been overlooked though they can drastically shorten the EPN survival and limit their effectiveness to control pests. In this study, we investigated the effects of application techniques on 3 Koppert products based on the commercial species *Steinernema feltiae*, *S. carpocapsae* and *Heterorhabditis bacteriophora*, as well as the compatibility of these EPNs with the most common practices of growers. We explored the tolerance of EPNs to high spraying pressure by monitoring their survival and efficacy after application. Additionally, we screened for the compatibility of EPNs with the most common adjuvants used in foliar applications in the Mediterranean areas. Last but not least, we tested the compatibility of the EPNs with the most common beneficial insects used in biological control in the view of integrated pest management. Our knowledge is gathered on our side effects App (compatibility with chemicals and effect against beneficial insects and mites) for easy access to growers and partners. To sum up, our results show that EPNs are resilient and can be fully integrated in crop and pest management strategies with certain considerations. In this poster, we describe the key factors to consider for the successful application of EPNs in a crop management strategy or IPM system.

Keywords: *Steinernema* spp. - Natural enemies - Adjuvants - Spraying pressure.

References:

- [1] Lacey et al., 2015. Journal of Invertebrate Pathology, 132:1–41.
- [2] van Damme et al., 2013. Pest Management Science 72:1702–9.
- [3] Mutegi et al., 2017. World Journal of Agricultural Research 5:233–9.
- [4] Batalla-Carrera et al., 2010. BioControl 55(4):523-530.

S9-P14

Metformin as enhancer of entomopathogenic nematode performance.

Lenin González-Paz[1, 2] (lgonzalezpaz@gmail.com), Ernesto San-Blas[2, 3], Carla Lossada[4], Edgar Portillo[2], Ysaias J. Alvarado[4]

[1] *Molecular Genetics Laboratory, University of Zulia, Maracaibo, Zulia, Venezuela*; [2] *Plant Protection Laboratory, Venezuelan Institute for Research, Maracaibo, Zulia, Venezuela*; [3] *Laboratorio de Nematología - ICA3, Universidad de O`Higgins, O'Higgins, Chile*; [4] *Molecular Characterization Laboratory, Venezuelan Institute for Research, Maracaibo, Zulia, Venezuela*

Entomopathogenic nematodes (EPNs) of the genera *Heterorhabditis* and *Steinernema* they are excellent biological control alternatives of insect pests, but they can present failures in the field. Lipoperoxidation (LPO) is a lipid oxidation process and can cause loss of infectivity and survival in EPN due to thermal stress. However, the effect of LPO on EPNs has not been studied[1-4]. Therefore, we studied the effect of LPO on the infectivity and survival of *Steinernema feltiae* and *Heterorhabditis indica* exposing 100IJ at intervals between 10º-40ºC, evaluating every 24h for 5days. Additionally, the 0.025µM antioxidant effect of Metformin was tested. Infectivity was determined by multiplication in *G. mellonella* and survival by absence of larvae movement. The LPO was determined by quantification of conjugated dienes and trienes (DC and TC) and total proteins (TP) using visible absorption spectrophotometry. The viable nematodes for the group without Metformin of *S. feltiae* reached 63% and 100% infectivity in ≈4days. Metformin promoted infectivity at ≈3days and viability of 79% under all conditions. At 40°C *S. feltiae* with metformin increased their viability more than 20% after 120h. The viability of the *H. indica* was 68% with infectivity of ≈3days, while with metformin, viability rose 83%, no changes in infectivity, allowing viability >25% after 120h at 40°C. We found significant differences in viability with and without metformin ($p<0.01$) with a high correlation with the loss of infectivity ($R2\approx70$). The concentration of DC and TC in *H. indica* was 2.434 and 0.457µM/mgTP, and in *S. feltiae* 2.571 and 0.854µM/mgTP, with metformin, respectively. The LPO increased with temperature and time, with maximum values at 35°C. High oxidative damage at these temperatures is related to the decrease of viability and infectivity. However, with metformin the concentration of DC and TC averaged 1.065 and 0.453µM/mgTP in *H. indica*, and 2.102 and 0.853µM/mgTP in *S. feltiae*, respectively. There was a significant decrease of ≈56% of DC in *H. indica*, while for *S. feltiae* it was ≈18%. No significant differences were observed in the TC concentration. We found that metformin exerts a reduction and modulation effect on LPO, even at >25°C, allowing greater viability and infectivity of EPN under all storage conditions tested. Our subsequent stress studies at pH, Hypoxia and osmotic pressure, measurement of other LPO markers in conjunction with computational analyzes support these findings.

Keywords: Oxidative stress - Aging - Infectivity - Survival - Lipoperoxidation.

References:

- Long et al., 2000. Nematology. 2(3): 309-317.
- Hass et al., 2002. J. Nematol. 34(2): 151–158.
- Hirao & Ehlers. 2009. Appl Microbiol Biotechnol. 84(1): 1061–1067.
- Joyner et al., 2011. Genetics. 189(1): 1439–1447.

S9-P15

Isolation of entomopathogenic nematodes on agricultural land in Croatia.

Dinka Grubišić (djelinic@agr.hr), Zrinka Drmić, Martina Kadoić Balaško, Darija Lemić, Renata Bažok, Maja Čačija

Department of Agricultural Zoology, University of Zagreb Faculty of Agriculture, Zagreb, Croatia

Entomopathogenic nematodes (EPNs) are lethal obligatory parasites of insects. They are ubiquitously distributed and have been recovered from variety of soil textures and habitats, including pastures, forests and crop fields. EPNs are a promising alternative to chemical insecticides. Various species and strains of EPNs exhibit differences in survival, infectivity and reproduction that make them more or less efficacious in pest control. The use of endemic nematodes may present less risk to non-target organisms than introduced species. The aim of this investigation was to verify the occurrence of EPNs in agricultural land in Croatia. In three years of investigation (2017-2019) a total of 135 soil samples were collected randomly [1] from 27 localities in 7 Croatian counties (Bjelovarsko - bilogorska, Grad Zagreb, Koprivničko - križevačka, Međimurska, Virovitičko - podravska, Vukovarsko - srijemska and Zagrebačka) from corn, soya, potato and alfalfa field, hazel, apple and cherry orchard and pasture. The insect baiting technique [2] was used to recover the nematodes from the soil. Using modified White trap [3] and molecular biology analysis, EPNs belonging to genus *Steinernema* were recovered from two soil samples, taken in a corn crop in Koprivničko - križevačka county and in apple orchard in Vukovarsko - srijemska county. Isolates caused 100% mortality of wax caterpillars in the process of isolation from soil samples. In order to determine its effectiveness in the control of other pest species, the investigation needs to be continued.

Keywords: Biological control - EPNs - Croatia - Steinernematidae.

References:

- [1] Orozco, R.A., Lee, M.M., Stock, S.P. , 2014. Soil sampling and isolation of entomopathogenic nematodes (Steinernematidae, Heterorhabditidae). Journal of visualized experiment (89), e52083, doi: 10.3791/52083.
- [2] Bedinng, R.A., Akhurst, R.J., 1975. A simple technique for detection of insect parasitic rhabditid nematodes in soil. Nematologica 21: 109-110.
- [3] White, G.F., 1927. A method for obtaining infective nematode larvae from culture. Parasitology 4:147-154.

S9-P16

Morphological responses of *Steinernema feltiae* exposed to purified active ingredient of Nemarioc-AL phytonematicide.

Jacqueline Tinashe Madaure[1] (jacquelinemadaure@yahoo.com), Phatu William Mashela[1], Dirk De Waele[2]

[1] *Green Biotechnologies Research Centre, University of Limpopo, Sovenga, South Africa*; [2] *Laboratory of Tropical Crop Improvement, University of Leuven (KU Leuven), Leuven, Belgium*

Steinernema feltiae is highly tolerant to Nemarioc-AL phytonematicide, with the characteristic feature of body length versus increasing concentrations of the product exhibiting positive quadratic relations [1]. However, it is not known whether the observed tolerance could hold at a similar concentration of the purified active ingredient of Nemarioc-AL phytonematicide, namely, cucurbitacin A. Infective juveniles (IJ) of *S. feltiae* in aqueous solutions were exposed to cucurbitacin A at 0, 2, 4, 8, 16, 32 and 64 µg/mL (ppm) in 96-well plates and incubated 18°C for 7 days. Samples were fixed on slides and morphometric measurements taken using OMAX microscope equipped with Toup view software [2]. Concentration data were expressed as an exponential series and transformed using log22x prior to subjecting the data to lines of the best fit. Body length (R2 = 0.73), rectum length (0.78), anus diameter (0.56), excretory pore (0.44) and cuticle thickness (0.97) versus log-transformed concentration exhibited negative quadratic equations. In conclusion, *S. feltiae* IJ failed to be tolerant to concentration of cucurbitacin A at concentrations similar to those of Nemarioc-AL phytonematicide.

Keywords: Cryptobiosis - Cucumis myriocarpus - Dauer activation - Dauer recovery - Entomopathogenic nematodes.

References:

- [1] Mashela et al., 2019. PLos One (In Press).
- [2] Shokoohi et al., 2015. J. Nematol. 47(4):370.

S9-P17

In vitro-cultured entomopathogenic nematodes to control the false codling moth, *Thaumatotibia leucotreta.*

Vernon Murray Steyn (vmsteyn1@gmail.com), Antoinette Paula Malan, Pia Addison

Department of Conservation Ecology and Entomology, Stellenbosch University, Stellenbosch, Western Cape, South Africa

False codling moth (FCM), *Thaumatotibia leucotreta* (Lepidoptera: Tortricidae), is an important pest of citrus, stone fruit and grapes in South Africa. Biological control using entomopathogenic nematodes (EPNs) has not been explored for use against FCM in grapevine. However, EPNs have been shown in previous studies to provide exceptionally good control against the larvae of FCM in laboratory bioassays in citrus, avocado, macadamia and litchi field trials. EPNs are particularly attractive for the control of FCM, as the pre-pupae, larvae and emerging moths occur in the soil. No soil treatment is currently targeted to control the soil stages of FCM. In this study, the pathogenicity, quality and age of *in vitro* liquid-cultured *Steinernema jeffreyense* was screened in the laboratory and tested in a vineyard field trial. Additionally, pre- and post-application pathogenicity tests with FCM larvae were conducted to assess the virulence of *in vitro*-cultured infective juveniles (IJs) and in the field by means of applying IJs to the vineyard soil. *Steinernema jeffreyense* at concentrations of 0, 10, 20, and 30 IJs/cm^2 were applied to 40 × 1 m^2 experimental plots that were artificially infested with FCM larvae. The insects were retrieved from the soil 48 h after application, to allow for the assessment of the immediate effect on FCM infection after application. The FCM-loaded cages were replaced over a period of four weeks, to determine the persistence of the original application. In a second trial, following the same procedure, *in vitro*-cultured *S. jeffreyense* and *Steinernema yirgalemense* were compared with regard to virulence and persistence over a four-week period. In the laboratory, the *in vitro*-cultured EPNs proved to be of similar quality to the *in vivo*-cultured *S. jeffreyense*, with a percentage FCM larval infection of >80%, while *in vitro*-cultured *S. jeffreyense* stored for 136 days at 14°C caused 87% infection. The semi-field study showed promising results, with the immediate effect yielding up to 77% mortality of FCM larvae, which remained >35% over the four-week period after application. The results compare favourably with those of previous field studies, using *in vivo* EPN, proving that EPNs would be a valuable addition to the current integrated pest management programme aimed at the control of FCM.

Keywords: *in vitro*-cultured - Semi-field trials - S*teinernema jeffreyense* - *Steinernema yirgalemense* - Bioassays.

References:

- [1] Dunn et al., 2019. BioControl https://doi.org/10.1007/s10526-019-09977-7
- [2] Malan et al. 2018. Afr. Entomol. 26 (1): 14-29
- [3] Malan & Moore, 2016. Afr. Entomol. 24 (2): 489-501.
- [4] Steyn et al., 2019. Biocontr. Sci. Technol. 27 (11): 1265-1278.
- [5] Steyn et al., 2019. Biol. Control doi.org/10.1016/j.biocontrol.2019.104052

S9-P18

Isolation, characterization, and virulence of entomopathogenic nematodes in Davao del Sur, philippines against superworm *Zophobas morio.*

Mara Louisa Navarez (maralouisa.navarez@g.msuiit.edu.ph), Nanette Hope Sumaya, PhD.

Department of Biological Sciences, PRISM-Flora Biodiversity Laboratory, Mindanao State University-Iligan Institute of Technology, Iligan, Philippines

Heterorhabditidae and Steinernematidae families consist of entomopathogenic nematodes (EPNs) that occur in soils of most part of the globe, are parasites of a wide range of insects, and have symbiotic relationships with the bacteria *Photorhabdus* and *Xenorhabdus*, respectively. This study aims to evaluate the distribution, characterize and assess the efficacy of the entomopathogenic nematodes isolated from the superworm, *Zophobas morio*. Through baiting techniques, 15 soil samples (75%) retrieved entomopathogenic nematodes out of 20 randomly collected soil samples from four agronomic farms (Banana, Mango, Coconut and Corn) in Davao del Sur. Using morphological keys, four nematode isolates were identified which predominantly belonged to the genus *Heterorhabditis.* For molecular analyses the 28S rRNA region and the D2D3 expansion segment will be used. Laboratory assays against *Z. morio* are currently being conducted to further investigate the efficacy of the locally isolated entomopathogenic nematodes. All tested isolates will be subjected to 100 IJs per insect larva. Mortality rate of the isolates will be recorded and compared to the four commercially available EPNs: *Heterorhabditis bacteriophora, Steinernema carpocapsae, Steinernema feltiae and Steinernema saimkayai*. The results from this study may contribute to the biological control programs in the country.

Keywords: Entomopathogenic nematodes - Biological control - Zophobas morio - Virulence testing - Heterorhabditis.

References:

- Shapiro-Ilan et al., 2002. Factors affecting commercial success. 265-287.
- Nguyen, K. B., & Smart, G. J. 2004. Taxonomy of insect parasitic nematodes. 795-878.
- Orozco, R. A. et. al., 2014. Journal of Visualized Experiments (89).
- Navarez, M.L. et. al., 2021. Egyptian Journal of Biological Pest Control. 31(1).

S9-P19

Steinernema australe display chemotaxis towards volatiles identified from blueberry roots

Rubén Palma[1] (rubenandrespalma@gmail.com), Ricardo Ceballos[2], Patricia Navarro[1], Julio Urzúa[2], Juan Alveal[2], Almendra Monje[1]

[1] *Insect Science, Instituto de Investigaciones Agropecuarias (INIA), Vilcún, Región de La Araucanía, Chile*; [2] *Chemical Ecology, Instituto de Investigaciones Agropecuarias (INIA), Chillán, Región de Ñuble, Chile*

Steinernema australe is an entomopathogenic nematode isolated in Chile, which has shown promising characteristics for controlling larvae of the weevil *Aegorhinus superciliosus*, one of the most damaging blueberry pests in this country. Recently, chemical volatile compounds were isolated and characterized from *Vaccinium corymbosum* damaged and non-damaged roots by this curculionidae (unpublished data). The capability of these compounds to elicit chemotaxis upon *S. australe* was determined establishing their potential role as attractants to find their host (*A. superciliosus* larvae). To reach this aim, we used a two-sided olfactometer to perform bioassays that measured the preference of infective juveniles (IJs), towards eight compounds, in five concentrations each: 1,000, 100, 10, 1, and 0.1 µg mL-1. Double-distilled hexane was used as control. The olfactometer consisted of two plastic Petri dishes into an external 90 mm plate. The internal components were divided into halves representing treatment and control, separated by the decision area. Three hundred (±50) infective juveniles (IJs) of *S. australe* were suspended in one milliliter of distilled water and inoculated over the decision area. Olfactometers were randomly oriented and kept in a dark room at 14 (± 2 °C) for 15 hours. Results showed that in dose-response tests, *S. australe* was attracted to all-five tested concentrations of methyl salicylate, 1-nonine, α-terpineol and 2-carene; and by 100 µg mL-1 of 10-undecyn-ol; 0.1 and 100 µg mL-1 of linalool; 100 µg mL-1 of limonene; whereas eucalyptol elicited no attraction or repellency. These results suggest that some volatiles released from *V. corymbosum´*s damaged roots may attract *S. australe*, which may have implications for the biocontrol of subterranean pests.

Keywords: VOCs - tri-trophic - Entomopathogenic nematodes - Chemical compounds - Blueberry.

S9-P20

The effect of aqueous extracts of mulch materials on entomopathogenic, slug-parasitic and root-knot nematodes.

Renáta Petrikovszki[1] (petrencsi@gmail.com), Robin Jakusovszky[1], Franciska Tóthné Bogdányi[2], Ferenc Tóth[3], Péter Nagy[1]

[1] *Department of Zoology and Ecology, Hungarian University of Agricultural and Life Sciences, Gödöllő, Hungary*;
[2] *ImMuniPot®Independent Research Group, Gödöllő, Hungary*; [3] *Hungarian Research Institute of Organic Agriculture, Budapest, Hungary*

While organic mulch decreased the damage done by *Meloidogyne incognita* and increased tomato yield in our previous long-term field experiment [1], new pest problems emerged: there was a general increase in the damage to fruits done by wireworms, and by snails and slugs in mulched plots. As we observed, wireworms were sensitive to soil moisture content, therefore they did not distribute evenly in the soil, but remain close to the surface, where mulching preserves water. Looking for a new technique in plant protection, we envisioned a combination of entomopathogenic fungi and nematodes applied to the soil surface, protected by mulching. In order to confirm the viability of this technique, preliminary studies were performed to test the mortality caused by the aqueous extracts of different mulch materials on entomopathogenic (EPN), slug-parasitic (SPN) and root-knot nematodes. Mortality caused by leaf litters (walnut and Norway maple), and green yard waste compost were tested in four concentrations (0.1, 0.5, 1 and 5%) on four EPN species (Heterorhabditis bacteriophora, *Steinernema carpocapsae*, *S. feltiae, S. kraussei*) and on the SPN (*Phasmarhabditis hermaphrodita*). In addition, walnut leaf litter extract was tested in six concentrations (0.78, 1.56, 3.125, 6.25, 12.5 and 25%) on *Ph. hermaphrodita* and on *M. incognita*. The 5% treatment of leaf litter extracts caused 100% mortality after 24 hours, except for *S. carpocapsae* (35% mortality) in the case of Norway maple. Compost treatments resulted in low overall mortality (under 20%). There was no lethal effect on *Ph. hermaphrodita* by the 0.78% walnut leaf litter extract, while all the J2 individuals of *M. incognita* died at this concentration. We found that *S. carpocapsae* was less sensitive to leaf litter extracts than the other examined species. Compost materials may act as a carrier medium for EPN and SPN species, and also are suitable for mulching. Since certain concentrations caused 100% mortality on the larvae of *M. incognita* and cause no harm to the SPN species, this finding may lead to an inventive crop protection protocol.
Our work and presentation were supported by New National Excellence Program (ÚNKP-18-3) and by the Higher Education Institutional Excellence Program (1783-3/2018/FEKUTSTRAT) of the Ministry of Human Capacities. The work was supported by the EFOP-3.6.3-VEKOP-16-2017-00008 project. The project is co-financed by the European Union and the European Social Fund.

Keywords: Common walnut - Green yard waste compost - leaf litter - Norway maple - Toxicity.

References:

- [1] Petrikovszki et al., 2016. Columella 3(2): 35-46.

S9-P22

Co-cultivation of entomopathogenic fungi and entomopathogenic nematodes in search of improved biocontrol against *Spodoptera litura*.

Rea Sebumpan (rea.sebumpan@g.msuiit.edu.ph)

MSU-IIT, Iligan City, Philippines

Utilization of entomopathogens against agricultural pests avoids the excessive use of harmful chemical insecticides. A new technology of co-cultivation of entomopathogens has been rising recently for more highly effective and sustainable agricultural practices. This study aims to assess the individual and combined effects of locally-occurring entomopathogenic fungi (EPFs) and entomopathogenic nematodes (EPNs) for potential biocontrol of *Spodoptera litura.* EPFs were isolated from different areas of Bukidnon and identified using universal primers, ITS 1/ITS 4 and TEF region. For the preliminary lab assay, 5 species of EPFs, *Trichoderma virens, Penicillium polonicum, Penicillium expansum, Acremonium cellulolyticus, & Diaporthales* sp. were most virulent against *Zophobas morio.* The efficacy of these species including *Metarhizium anisopliae & Beauveria bassiana* were further studied in a bioassay with different developmental stages ,larvae 1-2, 4-5; 2 and 5 day old pupae, of *S. litura*. The most virulent against L1-2 was *B. bassiana* with 100% mortality (day 3); L4-5 was *M. anisopliae* with 100% mortality (day 6); on 2 day old pupae was *T. virens* with mortality rate of 72.50%, 80%, and 76.66% (day 9); on 5 day old pupae was *Diaporthales* sp. with mortality rate of 92.50%, 100%, and 96.66% (day 7), post treatment of $1.6x10^7$, $4.3×10^8$, $3x10^9$ conidia/ml, respectively. Efficacy of *M. anisopliae* was further tested against L3 of *S. litura* in a bioassay with 3 EPNs namely, *Oschieus* sp. *Oschieus columbiana, & Oschieus carolinensis*. All EPFs and EPNs were applied either alone or in combination (EPF+EPN). Results shows that co-cultivation of *M. anisopliae & O. carolinensis* rprovided the highest larval mortality rate with 97.5% on day 2 when treated with *M. anisopliae* suspension containing 3 x 10^9 conidia/ml & 500 IJs/ml of *O. carolinensis* compared to the larval %mortality of *Oschieus* sp.,75%, *O. columbiana,* 85%, *O. carolinensis* 90%, *M. anisopliae,* 87.5%, *M. anisopliae & Oschieus* sp., 92.5%, *M. anisopliae & O. columbiana,* 85%. Results showed the potential of co-cultivation technology using EPF & EPN as an effective option to kill L3 of *S. litura* populations. Further studies are recommended for co-cultivation of EPF & EPN against all developmental stages of *S. litura* in order to determine whether the combination of *M. anisopliae* and *O. carolinensis* is consistent with its effects, and to further assess its efficiency in semifield/field conditions to validate the lab results.

Keywords: Entomopathogenic nematodes - Biological control - Soil microorganisms - *Spodoptera litura*.

References:

- Karuppiah et al., 2019. Front Microbiol. 10: 1068.

S9-P26

Potential of local entomopathogenic nematodes for control of the vine mealybug, *Planococcus ficus.*

Nomakholwa Stokwe (nfs@sun.ac.za), Antoinette Malan, Thomas Platt

Conservation Ecology & Entomology, Stellenbosch University, Stellenbosch, Western Cape, South Africa

Planococcus ficus, the vine mealybug, is the dominant mealybug pest of grapes in South Africa [1]. To provide an alternative for chemical control, entomopathogenic nematodes (EPNs) were investigated as a biological control agent to be used in an integrated pest management system. Four local EPN species were screened for efficacy against female *P. ficus*, the most potent of which were *Heterorhabditis noenieputensis*, with 90% mortality, and *Steinernema yirgalemense*,with 63%. Since *S. yirgalemense* was previously shown to be highly effective against a range of pests, the effects of temperature and humidity on the infectivity of *S. yirgalemense* to female *P. ficus* were also assessed. The application of *S. yirgalemense* at 25°C yielded the highest mortality, of 72%, followed by 45% mortality at 30°C, and only 9% mortality when applied at 15°C. *Steinernema yirgalemense* performed best at 100% relative humidity (RH), resulting in 70% mortality. Decreasing RH levels resulted in decreased mortality (61% mortality at 80% RH, 40% mortality at 60% RH). As a soil-based organism, *S. yirgalemense* is most effective as a biocontrol agent of *P. ficus* under conditions of moderate temperature and high humidity. Its lethality to *P. ficus*, and its status as an indigenous species, indicate its promise as a potential biocontrol agent of the vine mealybug.

Keywords: Grapevine - Heterorhabditidae - Integrated pest management - *Steinernematidae* - *Planococcus ficus*.

References:

- [1] Walton, V.M., 2003. ficus (Signoret), in vineyards in the Western Cape province, South Africa. Thesis, Stellenbosch University,South Africa.

S9-P27

Distribution, characterization and virulence of the isolated entomopathogenic and entomophilic nematodes in selected vegetable and root crop farms in Bukidnon province, Philippines against cotton cutworm *(Spodoptera litura).*

Mayvel Suan[1, 2] (mayvel.suan@g.msuiit.edu.ph), Nanette Hope Sumaya[1, 2], Prakaijan Nimkingrat[3]

[1] *Department of Biological Sciences, College of Science and Mathematics, Mindanao State University- Iligan Institute of Technology, Iligan City, Philippines*; [2] *FBL-Nematology Research Group, Premier Research Institute of Science and Mathematics (PRISM), Iligan City, Philippines*; [3] *Department of Entomology and Plant Pathology, Faculty of Agriculture, Khon Kaen University, Khon Kaen, Thailand*

In recent years of searching for a non-hazardous and environmental friendly pest control agent, entomopathogenic including entomophilic nematodes emerged as an alternative biocontrol agent to reduce and manage the pest population. This study sought to understand the distribution, characterize and evaluate virulence of the locally isolated nematodes. From randomly selected 24 sites of vegetable and root crop farms in Bukidnon, Philippines; 33 soil samples (27.97%) harbored nematodes out of 118 collected soil samples. Through morphological key characters and analyses of the D2-D3 expansion segment of the 28S rRNA, 18S rRNA, ITS and mitochondrial COI regions, we have identified a total of 6 EPN species from 2 genera, *Oscheius*: *O. carolinensis* (20 isolates), *O. colombiana* (2 isolates), *O. myriophila* (2 isolates), *Oscheius* spp. (2 isolates) and *Metarhabditis*: *M. blumi* (3 isolates) and *M. rainai* (4 isolates). Notably, we have not recovered nematodes from the genera *Heterorhabditis* and *Steinernema*. The biocontrol potential of the six nematodes species was further investigated against the cotton cutworm (*Spodoptera litura*) under laboratory conditions. Four developmental stages of *Spodoptera litura* L1/L2, L4/L5, 2 and 5 days old pupae were exposed to 100, 300 and 500 IJs/ml. All tested isolates were pathogenic against *S. litura* in all developmental stages, particularly, *O. colombiana* inducing a 100% mortality rate in both L1/L2 and L4/L5 stages in the 4th day post nematode inoculation. For comparison, we used the commercially available *Steinernema saimkayai* from Thailand which recorded a 100% mortality after 4 and 5 days post nematode inoculation for both L1-L2 and L4-L5, respectively. After 8 days, *O. colombiana* recorded more than 70% and 96% mortality rate in both 2 and 5 days old pupae whereas *S. saimkayai* had more than 72% and 97% mortality rate, respectively. Thus, the entomopathogenicity bioassay of the locally isolated nematodes strains revealed an insecticide potential against cotton cutworm, *S. litura*. Screenhouse and field trials are currently being conducted to further investigate the efficacy of the nematode isolates.

Keywords: Entomopathogenic nematodes - Entomophilic nematodes - Biological control - Spodoptera litura.

S9-P28

Isolation and biocontrol potential of entomopathogenic and entomophilic nematodes from Talakag, Bukidnon and Claveria, Misamis Oriental, Philippines.

Nanette Hope Sumaya (nanettehope.sumaya@g.msuiit.edu.ph), Cynthia Ann Dichusa, Mary Flor Maureal, Jill Kate Lluisma, Janna Andalan

Department of Biological Sciences, Mindanao State University-Iligan Institute of Technology (MSU-IIT), Iligan, Lanao del Norte, Philippines

Amidst rising concerns over rampant synthetic chemical applications and their deleterious effects, the identification of alternative biopesticides has become paramount. Entomopathogenic and entomophilic nematodes (EPNs) of the families Steinernematidae, Heterorhabditidae and Rhabditidae are used as biocontrol agents (BCAs) against several insect pests. In this study, local nematodes as BCAs were explored in different agricultural areas of Talakag, Bukidnon and Claveria, Misamis Oriental. Insect-baiting using the larvae of the superworm, *Zophobas morio* and greater wax moth, *Galleria mellonella* was carried out. Out of 70 soil samples collected, only 10 samples harbored nematodes from the different sampling areas in Talakag (carrot, cabbage, broccoli, bell pepper, maize, mungbean and potato) and in Claveria (cabbage, eggplant, chayote, cassava, dragon fruit, maize and pineapple). Through morphology and analyses of ITS and SSU rDNA regions, we identified and confirmed nematode isolates belonging to family Rhabditidae*: Oscheius* and *Metarhabditis* spp. from Talakag and Claveria. Unexpectedly, the widely recognized EPNs *Heterorhabditis* and *Steinernema* were not recovered in this study. Furthermore, the efficacy of the local nematode isolates from Talakag was assessed against the last instar larvae of pineapple white grub, *Anomala orientalis* under laboratory conditions. At 48 hours post inoculation, the mean mortality induced by *O. carolinensis*, *O. colombiana* and *M. amsactae* were 100%, 100% and 55.6%, respectively. For nematode isolates from Claveria, *Oscheius* sp. and *Metarhabditis* sp., the efficacy will be tested against the cotton cutworm, *Spodoptera litura* and then compared with the commercially available *Steinernema siamkayai*. Our findings highlight the potential of locally-occurring nematodes to be utilized as BCAs of agricultural target pests after further comprehensive trials.

Keywords: Biocontrol - Morphology - Molecular characterization - Anomala - Efficacy.

References:

- Dichusa, C. A., Ramos, R., Aryal, S., Sumaya, N. P. D., & Sumaya, N. H. (2021). Survey and identification of entomopathogenic nematodes in the province of Cotabato, Philippines, for biocontrol potential against the tobacco cutworm, Spodoptera litura (Fabricius)(Lepidoptera: Noctuidae). Egyptian Journal of Biological Pest Control, 31(1), 1-10.
- Leonar, A. L. C., & Sumaya, N. H. N. (2019, November). Isolation and identification of entomopathogenic nematodes from selected areas in Lanao del Norte, Philippines and their infectivity against Zophobas morio (Coleoptera: Tenebrionidae). In Entomology 2019. ESA.
- Navarez, M. L., Sangcopan, R., Aryal, S., Sumaya, N. P. D., Bhat, A. H., & Sumaya, N. H. (2021). Native Philippine Heterorhabditis indica isolates from banana and rice fields and preliminary results of their virulence against the larvae of super worm (Zophobas morio Fabricius Coleoptera: Tenebrionidae). Egyptian Journal of Biological Pest Control, 31(1), 1-10.

S9-P30

Breeding for improved virulence and post-application longevity of *Heterorhabditis bacteriophora* dauer juveniles.

Bart Vandenbossche (b.vandenbossche@e-nema.de), Carlos Molina, Mike Barg, Verena Dörfler, Erika Consoli, Alejandra Centurion, Sitiram Ayral, Chris Ogaya, Nanette Hope Sumaya, Olaf Strauch, Ralf-Udo Ehlers

e-nema GmbH, Schwentinental, Germany

Entomopathogenic nematodes to control the Western Corn Rootworm are applied into the soil together with the maize seeds at sowing time, when *Diabrotica* eggs are still in diapause. Therefore, genetic improvement of persistence is crucial as nematodes have to survive and remain infective for 4 to 6 weeks until insect larvae appear. Wild-type strains were phenotypically characterized for virulence towards WCR larvae and larvae of *Tenebrio molitor*. Nematode concentration at which 50% (LD50) of *Tenebrio molitor* larvae were infected ranged between 1.4 to 30.5 DJs per insect. Nematode persistence was analysed by subsequently baiting the soil using T. molitor as trap insect. The tested strains survived and were able to infect the target pest after six weeks of incubation at 17°C in the absence of hosts. A wild type strain with best performance in virulence and persistence was selected over several cycles in sand bio-assays with WCR larvae and resulted in an increased virulence in four consecutive selection cycles. Results of persistence from sand assays correlate with longevity and oxidative stress resistance assays. Nematode persistence at two application dosages correspondent to the current commercial application (2 billion DJs ha-1) and the target dose (1 billion DJs ha-1) did not significantly differ as analysed by non-destructive baiting. These results support the hypothesis that more persistent and virulent strains enable the reduction of the application dosage and consequently reduce nematode application costs. Phenotypic data were combined with genotypic data and molecular SNP markers were identified that will assist in further nematode breeding programmes.

Keywords: Entomopathogenic nematodes - Persistence - Selection.

S9-P31

Plasticity in the use of *Xenorhabdus nematophila* and *Photorhabdus laumondii* against *Botrytis cinerea.*

Ignacio Vicente-Díez[1] (ignacio.vicente@icvv.es), Rubén Blanco-Pérez[1], Maryam Chelkha[2], Alicia Pou[1], Raquel Campos-Herrera[1]

[1] *Institute of Grapevine and Wine Sciences (ICVV), Logroño, La Rioja, Spain*; [2] *Centre Eau, Ressources Naturelles, Environnement et Développement Durable, Mohammed V University, Rabat, Morocco*

The fungus *Botrytis cinerea* (Pers.: Fr) is one of the most challenging problems for fruit and vegetable crops worldwide and especially relevant in postharvest fruit decay. Industrialized agriculture has predominantly used synthetic chemical pesticides to protect crops from this phytopathogen to maintain the highest standards of food production rates. However, concern for environmental damage due to excessive reliance on chemical pesticides and increased antibiotic resistance has expanded the interest in new biological control products. *Xenorhabdus* spp. and *Photorhabdus*spp. are Gram-negative γ-Proteobacteria that occur in symbiosis with nematodes and produce potent antifungal compounds. The objectives of this study were to (i) assess the antibiotic effect of unfiltered ferments (UFs) and cell-free supernatants (CFSs) of *X. nematophila* and *P. laumondii* on *Botrytis cinerea* mycelium growth and (ii) compare the activity of bacteria isolated from a bio-fermenter with the commercial *Bacillus amyloliquefaciens* (Serenade®ASO, Bayern CropScience). The UFs and CFSs were mixed with sterilized Potato Dextrose Agar (PDA) medium (1:11) on Petri dishes. For the second study, the isolated bacteria were seeded on one border of the PDA plate. Twenty microliters of *Gamborg*suspension with a concentration of 107 conidia/mL of *B. cinerea* were applied in the middle of all the plates (ten per treatment). The experiments were conducted three times with their corresponding controls. We calculated the fungal pathogen growth area and inhibitory growth rate (IR). In agreement with previous studies, the secondary metabolites of *X. nematophila* showed the highest fungicide effect on *B. cinerea* mycelium growth. The UF and CFS of *X. nematophila* inhibited ~95% and ~80% of fungus growth, respectively, while both UF and CFS of *P. laumondii* inhibited ~40%. The isolated bacteria of *Photorhabdus* controlled the mycelial growth as commercial *B. amyloliquefaciens* during three days (inhibition ~80%), although isolated *X. nematophila* did not suppress fungal mycelial growth. Our findings indicate the potential of UF and CFS of *X. nematophila* for control *B. cinerea*. The application of *P. laumondii* isolates should also be further investigated for controlling *B. cinerea* on a larger scale.

Keywords: *Botrytis cinerea* - Control - *Photorhabdus laumondii* - *Xenorhabdus nematophila*.

S9-P32

Concomitant species of entomopathogenic nematodes alter dispersal behavior and increase insecticidal efficiency

Sheng-Yen Wu (sywu@ufl.edu), Larry Duncan

Entomology and Nematology, University of Florida, Citrus Research and Education Center, Lake Alfred, Florida, United States

Combinations of entomopathogenic nematode (EPN) species are sometimes either more or less efficacious than individual species for management of insect pests1,2. The mechanism of these synergistic or antagonistic outcomes are unknown. The dispersal rates of three Heterorhabditis species and six Steinernema species were assessed using image analysis of videos of the nematodes on water agar. Dispersal rates between the genera differed significantly (P=0.002), but were unrelated to putative foraging strategy and/or body volume. Heterorhabditis indica dispersed more rapidly when combined with Steinernema diaprepesi, but not with S. glaseri. In a soil column assay, more Diaprepes root weevil larvae were killed by concomitant H. indica and S. diaprepesi than by equivalent numbers of individual species (P=0.022). Communication between disparate EPN species upon dispersal appears to be an initial step in a suite of behaviors that can increase insecticidal efficacy.

Keywords: Entomopathogenic nematode - Dispersal behavior - Interaction - Biological control.

References:

- [1] Jabbour et al., 2011. Biol. Control. 59(2): 277-283.
- [2] Heve et al., 2018. J. Pest Sci. 91(2): 799-813.

S9-P33

Broad phenotyping of DJ-recovery in *Heterorhabditis bacteriophora* using highly homozygous mutants and WT-inbred lines

Zhen Wang[1, 2] (stu225777@mail.uni-kiel.de), Christopher Ogaya[1, 2], Bart Vandenbossche[2], Verena Dörfler[2], Mike Barg[2], Ralf-Udo Ehlers[2], Carlos Molina[2]

[1] *The Faculty of Agricultural and Nutritional Sciences, Christian-Albrechts-Universität zu Kiel, Kiel, Germany*; [2] *e–nema GmbH, Schwentinental, Germany*

The entomopathogenic nematode (EPN) *Heterorhabditis bacteriophora* is an effective biological control agent against insect pests. The dauer juvenile (DJ) is the free-living developmentally-arrested stage in charge of seeking for new hosts and carrying inside the symbiotic bacteria *Photorhabdus luminescens*. Once the DJ invades a target insect, bacterial cells are released into the insect's haemocoel. Then these bacteria override the insect's immune system, grow exponentially, and provide nutritions for nematodes. Upon infection, the DJ perceive signals from the insect that trigger the development until reproductive maturity. This event called DJ recovery, is crucial for the reproductive success in EPN. For commercial EPN-production, DJ recovery also determines in great part the final yield of monoxenic liquid cultures [1]. In this scenario, DJs recover upon contact with pre-cultured *Photorhabdus* bacteria. The identity of the so called "bacterial food signal" [2], triggering DJ recovery is unknown and major regulators from the nematode side are still not clear. To enhance the understanding of the possible genetic components regulating DJ recovery in *H. bacteriophora* we have combined large-scale phenotyping and genotyping approaches. We have produced a collection of more than 100 highly homozygous EMS-mutant lines and 48 wild type (WT) inbred lines [3, 4]. All lines were evaluated by systematically testing the recovery induced by bacterial cultures as well cell-free bacterial supernatants, and as complementary traits, we evaluated the DJ longevity and virulence of line-subsets. Parallel to phenotyping, we carried out genotyping by sequencing (GBS) in all EMS-mutants and WT-inbred lines. From more than 500 identified single nucleotide polymorphisms (SNPs), at least 13 SNPs showed potential association to the recovery trait. We have as well confirmed a strong correlation between recovery in bacterial supernatant and the performance of WT-inbred lines in monoxenic liquid cultures. Further, we have identified robust subsets of four EMS-mutant lines with highly contrasting recovery (3.9 vs 76.6 %), DJ-virulence against mealworms (*Tenebrio molitor;* 22 vs 78 %), and mean time survived under oxidative stress (MTS50; 9.3 vs to 150.8 h). The present phenotypic and genotypic results are the basis for future physiologic and molecular approaches directed to the identification of major regulatory elements in complex quantitative traits in *H. bacteriophora*.

Keywords: *Heterorhabditis bacteriophora* - Dauer juvenile - EMS-mutant - Wild type - DJ-recovery.

References:

- [1] Ehlers, R.-U. (2001). Mass production of entomopathogenic nematodes for plant protection. Applied Microbiology and Biotechnology 56: 623–633.
- [2] Strauch, O. & Ehlers, R.-U. (1998). Food signal production of Photorhabdus luminescens inducing the recovery of entomopathogenic nematodes Heterorhabditis spp. in liquid culture. Applied Microbiology and Biotechnology 50: 369-374.
- [3] Brenner, S. (1974). The genetics of Caenorhabditis elegans. Genetics 1: 71-94.
- [4] Sumaya, N. H., Gohil, R., Okolo, C., Addis, T., Doerfler, V., Ehlers, R.-U. & Molina, C. (2018). Applying inbreeding, hybridization and mutagenesis to improve oxidative stress tolerance and longevity of the entomopathogenic nematode Heterorhabditis bacteriophora. Journal of Invertebrate Pathology 151: 50-58.

POSTERS

S10. Future of nematology, education and training

S10-P01

Nematology lab of INIAV: a 10-year overview of research, training and services.

Maria Inacio (lurdes.inacio@iniav.pt)

Plant Health/ Nematology, INIAV, Lisbon, Oeiras, Portugal

NemaINIAV, the nematology lab of INIAV (National Institute for Agrarian and Veterinary Research) located in Oeiras, near Lisbon, integrates the National Reference Laboratory (NRL) for Plant Health in Portugal. The mission of this laboratory is to conduct research activities and provide community services for the detection of plant-parasitic nematodes, aiming to prevent the spread and introduction of nematodes from plant material and plant products, and also to ensure compliance with local and EU legislation. As a NRL, NemaINIAV is responsible for the testing of samples coming from national surveys. In the last decade, several thousands of wood and insect samples were processed each year, for detection of the pinewood nematode (*Bursaphelenchus xylophilus*). Similarly, hundreds of soil cores were screened for the presence of potato cyst nematodes (*Globodera pallida* and *G. rostochiensis*) and species of root-knot nematodes (*Meloidogyne* spp). Diagnostics are based on nematode morphology and validated by molecular and/or biochemical studies whenever needed. Additionally, the lab performs the detection of quarantine nematodes such as the rice leaf nematode (*Aphelenchoides besseyi*) from seeds, and for the certification of orchards, vineyards and ornamentals.
Technical support is continuously given to private farmers, farmer associations and forest owners through phytosanitary diagnostics and consultancy. Besides these activities, the NemaINIAV team leads and participates in several ongoing research projects, both at national and international levels. Teaching and training the next generation of nematologists is also a paramount engagement and core value of the staff. Here, we present the distribution of different activities and the evolution of projects, supervisions, training and publications and the numbers of tested samples over the last ten years at NemaINIAV.

Keywords: NemaINIAV - National reference laboratory - Diagnostics - Training.

S10-P02

Nematology and the environment: a worm's tail.

Thomais Kakouli-Duarte (thomae.kakouli@itcarlow.ie), Anna Karpinska, Stephen Boyle, Aoife Egan

Department of Science and Health, Institute of Technology Carlow, Carlow, Ireland

Nematodes are microscopic (mostly), non-segmented round worms, with great environmental importance, inhabiting nearly every habitat on the planet. They may be parasitic to animals and plants or they may be free-living in their environment, feeding on bacteria, fungi, or other nematodes. As they occupy intermediate positions in food webs and their assemblages are sensitive to environmental parameters, disturbance in these assemblages reflects disturbance in other ecosystem elements. These facts, together with the facts that nematodes are representative of their habitats and are easily extractable from soils and sediments, enable them to be ideal as indicators of environmental change. In addition, certain species can be easily cultured and thus easily amenable to laboratory based ecotoxicity experiments. Interest in nematode research, however, is also relevant in the context of food security, particularly in sustainable crop protection practices. Entomopathogenic nematodes (EPN), with part of their life cycle being parasitic to insects, provide effective alternatives for integrated pest management systems against economically important soil and cryptic insect pests. Plant parasitic nematodes (PPN) are a global challenge for crop production due to vast crop loses they cause. At the same time, PPN have developed resistance in some cases to conventional chemical nematicides, which are heavily regulated because of environmental and consumer health concerns. This poster presentation will provide an overview on the environmental nematode research carried out by the Molecular Ecology and Nematode Research Group of enviroCORE, in the Institute of Technology Carlow, in recent years. Highlights will include the utilisation of nematodes as environmental bioindicators and, specifically, the development of the EPN *Steinernema feltiae* as a sentinel of heavy metal toxicity, in the context of environmental biomonitoring, and in the ReNu2Farm project (www.nweurope.eu/renu2farm). The ReNu2Farm projects seeks to develop recycling derived fertilisers (RDF) with an aim to replace the environmentally detrimental conventional fertilisers, and turning waste into a valuable resource, for soil nutrient sustainability. Nematodes are used for the environmental risk assessment of RDF. In the context of sustainable food security, the presentation will focus on studies on the interactions of PPN with plant growth promoting bacteria towards sustainable PPN management approaches.

Keywords: Environmental nematology - ReNu2Farm - Sustainability - Plant parasitic nematodes - Biomonitoring.

S10-P03

Thirty years of plant-parasitic nematode research in America.

Lizzete Dayana Romero Moya (ldromerom@unal.edu.co)

Institute of Plant Protection, Chinese Academy of Agricultural Sciense, Beijing, R. P. China

Phytoparasitic nematodes are associated with nearly every important agricultural crop and represent a significant constraint on global food security. The crop yield loss due to nematodes is estimated at US$157 billion each year. The challenge of feeding the current and future world population is widely recognized and the knowledge of plant diseases has an important role in overcoming this. Science is fundamental to protect plants, and plant pathologists work around the world in the identification and understanding of diseases caused by plant-parasitic nematodes. This analysis aims to trace the state of research in America on nematology, as well as knowing the main lines of work, their priorities, and their evolution over the 2012-2022 period. A synthetic bibliometric analysis of the studies on plant-parasitic nematodes published in America over the 2012-2022 period was performed using 9697 documents retrieved from the Scopus, Web of Science, and Science Direct bibliographic databases. The progression of scientific outputs by countries, years, and authors was characterized. The results showed a significant increase in the number of publications per year until 2021. United States and Brazil led the countries with most publications on nematodes. United States was the country with the most prolific contribution to agricultural nematology research in the Americas or China. Within the keywords analyzed, the lines of research and genders that received the most attention were biological control, resistence, *Meloidogyne* sp. and *Heterodera* sp., respectively. The study of phytoparasitic nematodes in America is limited to a few research groups that have generated important advances in plant pathology. Although American countries have increased the number of their scientists and research institutions in recent years, the gap between countries is substantial.

Keywords: Nematology - Phytoparasitic - Bibliometry - Bibliographic data bases - Scientific Research.

References:

- Aballay, E. (2018). Nematology In South America. [Abstract]. 50th Reunión Anual 50th Annual Meeting. Organization of Nematologists of Tropical America. NEMATROPICA Vol. 48, No. 2.
- Ciocca, D.R. and Delgado, G. (2017). The reality of scientific research in Latin America; an insider's perspective. . Cell Stress and Chaperones 22, 847–852 . https://doi.org/10.1007/s12192-017-0815-8
- Food and Agriculture Organization (FAO). (2020). FAOSTAT Statistical database of the United Nation Food and Agriculture Organization (FAO) statistical division. Retrieved from http://faostat.fao.org/
- Mesa, C. M., Garrido, J. A., Cebrian, J., Talavera, M. and Manzano, F. Global Research on Plant Nematodes (2020). Agronomy; 10(8):1148. https://doi.org/10.3390/agronomy10081148

S10-P04

The Journal of Nematology.

David Shapiro-Ilan (David.Shapiro@ars.usda.gov)

USDA-ARS, Byron, Georgia, United States

The *Journal of Nematology* is the official technical and scientific communication publication of the Society of Nematologists since 1969. The journal publishes original papers on all aspects of basic, applied, descriptive, theoretical or experimental nematology and adheres to strict peer-review policy. Other categories of papers include invited reviews, research notes, abstracts of papers presented at annual meetings, and special publications as appropriate. The journal is open access and publishes articles in a continuous fashion. The journal is indexed widely. The journal's impact factor has continued to rise (the impact factor was 1.386 in 2018) and number of views and downloads per article has also continued to rise. Please see more information about the journal and how to submit articles at https://www.exeley.com/journal/journal_of_nematology

Keywords: Journal - Nematology.

S10-P05

The nematode collections at the NPPO, Wageningen, The Netherlands.

Evelyn van Heese (e.y.j.vanheese@nvwa.nl), Anne-Marie de Heij, Patricia Bor, Gerrit Karssen

National Plant Protection Organization, National Reference Centre, Nematology, Wageningen, Netherlands

Invertebrate collections are essential as reference material for the identification of species and for research purposes [1]. An overview is presented of all the nematode collections present at the National Plant Protection Organization (NPPO) in Wageningen, The Netherlands. These collections include living nematode populations, fixed nematodes and preserved plant symptoms. All are used for identification, teaching and scientific studies. The oldest collection, with nearly 200 sealed jars, are formalin fixed plant symptoms induced by plant-parasitic nematodes. For fixed nematodes on slides two collections are available: a type slide and a regular slide collection. The regular slide collection, includes about 50.000 slides, with a focus on terrestrial nematodes from Europe. The type collection (Wageningen Nematode Collection) maintains nearly 4.000 accessions of types from terrestrial, freshwater and marine habitats worldwide. This collection, together with a nomenclatorial database, can be searched at www.nce.nu. Living nematode collections include in total around 225 populations and are maintained in a greenhouse, in vitro on carrot disks and agar plates with fungi or in micro plots outside. The focus of these collections is quarantine and related or economic important plant-parasitic nematodes. Some collection items are available upon request at the NPPO, Wageningen, The Netherlands.

Keywords: Collections - Taxonomy - Identification - Nematodes.

References:

- Hockland, 2005, EPPO Bulletin: 35, 165-169.

S10-P06

Preliminary development of an automated system for identification and quantification of nematodes.

Carina Lopes[1] (carina@nemafito.com), Cristine Carneiro[2], Ramon Franco[2], Juan Jaramillo[3], Beatriz Seidel[2]

[1] *Nemafito Laboratory, Luis Eduardo Magalhães, Bahia, Brazil*; [2] *Universidade Federal do Oeste da Bahia, Barreiras, Bahia, Brazil*; [3] *Vita Tecnologia, Limeira, São Paulo, Brazil*

The diagnosis of nematodes is currently based on classical taxonomy by optical microscopy and molecular tools. These analyses require advanced training of technicians, thus being time consuming and expensive for specialized laboratories. In Brazil, there are few nematology laboratories with the appropriate level of specialization, burdening producers with the costs of transporting soil samples over long distances. In this context, the development of fast, reliable, and easily accessible diagnostic technologies would increase the analytical capacity of laboratories and democratize the access of farmers to very important information about the populations of nematodes in agricultural cultivation areas. The objective of the present work was to develop an automated nematode quantification and identification process. For this purpose, an optical microscopy image scanning system was built. This system takes digital images as input. The first step, image preprocessing, is performed by filtering the images to mathematically improve their characteristics for the following steps. In the second step, the preprocessed images are treated by applying spatial region growth mathematical algorithms. In the third stage, the morphological regions of interest are marked, and transitions between these regions are identified using different mathematical descriptors of contour and optical density. Finally, in the fourth step, the processed images are grouped for transmission. When any region with biological material is detected, the region is designated by the system as a region of interest for analysis. This exploited image processing system enables the identification of nematodes and soil debris using the Watershed algorithm and non-linear regression techniques. This model has already been successfully tested in medical diagnosis. Next steps for this research are related to the improvement of nematode identification by the development of a recognition process through supervised and unsupervised learning, supported by the image bank that is being created.

Keywords: Nematodes diagnosis - Automated systems - Microscopy.

E-POSTERS ONLINE VIEWING

PineWALL Project: Linking pine cell wall composition and structure to pinewood nematode resistance under climate change.

Ricardo da Costa[1, 2] (rmfdcosta@uc.pt), Luís Fonseca[2], Joana Cardoso[2], Isabel Abrantes[2], Joana Vieira[3], Filipe Campelo[2], Jorge Canhoto[2], Marisa Gaspar[4], Silvia Coimbra[5], Inês Santos[1], Maria Paula M. Marques[1, 6], Luís Batista de Carvalho[1]

[1] *Molecular Physical-Chemistry R&D Unit, Department of Chemistry, University of Coimbra, Coimbra, Portugal*; [2] *Centre for Functional Ecology-Science for People & the Planet, Department of Life Sciences, University of Coimbra, Coimbra, Portugal*; [3] *Collaborative Laboratory for Integrated Forest & Fire Management, ForestWISE, Vila Real, Portugal*; [4] *Chemical Process Engineering and Forest Products Research Centre, Department of Chemical Engineering, University of Coimbra, Coimbra, Portugal*; [5] *Associated Laboratory for Green Chemistry, REQUIMTE, University of Porto, Porto, Portugal*; [6] *Department of Life Sciences, University of Coimbra, Coimbra, Portugal*

The pinewood nematode (PWN), *Bursaphelenchus xylophilus*, infects *Pinus* spp., namely *P. pinaster*, one of the most susceptible species, widely distributed in Portugal, whereas other species, such as *P. pinea* and *P. halepensis* are less susceptible. The PWN uses its stylet to perforate cell walls (CW), while releasing hydrolytic enzymes against all groups of CW glycans. Thus, understanding how the CW chemistry and structure can influence host susceptibility to PWN infection is essential. Moreover, it is crucial to better understand how varying environmental conditions affect host susceptibility. To achieve this goal, the innovative transdisciplinary project PineWALL (FCT-PTDC/ASP-SIL/3142/2020) was established. *Pinus* spp. (*P. pinaster*, *P. pinea*, *P. halepensis*) will be inoculated with PWN and disease progression will be monitored under various controlled temperatures and soil water contents to simulate climate change scenarios. *Pinus* spp. morphology/anatomy will be compared, to understand whether anatomical features have implications on disease progression. Furthermore, high-throughput vibrational spectroscopy techniques, Raman and Fourier transform infrared (FTIR), will be applied for a global characterisation of pine CW, as well as chemical analytical methods to characterize the CW fractions. Immunological approaches will also be employed to unveil the CW glycome profile and reveal *in muro* glycan distributions by *in situ* immunolabelling. At the end of the project, all collected data will be integrated to identify structural CW biomarkers that may be used to predict pine susceptibility/resistance to PWN, ultimately contributing to the modelling of a *P. pinaster* ideotype, better adapted to climate change.

Keywords: Cell wall - Climate change - Glycome profile - *Pinus* spp. - Vibrational spectroscopy.

Bursaphelenchus spp. inhabitants of a centennial *Pinus pinea* tree of public interest.

Ricardo da Costa (rmfdcosta@uc.pt), Luís Fonseca, Joana Cardoso, Joana Vieira, Filipe Campelo, Isabel Abrantes

CFE, Department of Life Sciences, University of Coimbra, Coimbra, Portugal

With the introduction of the 'Trees of Public Interest Law' in 1938, Portugal created one of the oldest laws for tree protection in Europe. This law evolved throughout the 20th century, which led to the implementation of a National Registry, where all trees of public interest are catalogued and classified as living monuments. As a consequence of their age and size, these tree monuments represent biodiversity hotspots, being crucial habitats for a large diversity of organisms. Recently, a monumental centennial wilted stone pine, *Pinus pinea*, was felled at the University of Coimbra, Portugal. Thus, the diversity of *Bursaphelenchus* species was assessed, including a survey for the presence of *B. xylophilus*, the pinewood nematode. This study combined dendrochronological and nematological studies in order to investigate the age of the tree and *Bursaphelenchus* species diversity. The differences in tracheid features between earlywood (thin-walled cells) and latewood (thick-walled cells) were used to identify tree rings, which revealed an age of approximately 160 years. Nematodes, extracted from wood samples from the trunk and branches, were isolated and propagated in *Botrytis cinerea* cultures. Afterwards, the species were characterized morphologically (species-specific characters) and molecularly (ITS-RFLP). Although *Bursaphelenchus xylophilus* was not found, *B. fungivorus* and *B. sexdentati* were identified, in addition to four *Bursaphelenchus* isolates which were detected as co-occurring in the *P. pinea* centennial tree and are currently being studied. These findings highlight that long-established wilted pine trees can host several *Bursaphelenchus* species acting as nematode diversity hotspots.

Keywords: *Bursaphelenchus* - Centennial tree - Dendrochronology - Diversity - *Pinus pinea*.

Assessing the influence of yam genotype on the performance of abamectin-treated banana paper to manage nematodes.

Antoine Affokpon[1] (affokpona@gmail.com), Tahira Pirzada[2], D. N. Elodie Coffi[1], H. Charles Opperman[2], H. Richard Guenther[3], Steve Koenning[3], Reny Mathew[3], L. Tim Sit[3], V. Medwick Byrd[3], Lokendra Pal[3], L. Eric Davis[3], A. Saad Khan[3], L. Danny Coyne[4]

[1] *Faculty of Agricultural Sciences, University of Abomey-Calavi, Abomey-Calavi, Benin*; [2] *North Carolina State University, Raleigh, North Carolina, United States*; [3] *North Carolina State University, Raleigh, United States*; [4] *IITA, Nairobi, Kenya*

Yam (*Dioscorea* spp.) is among the most important staple crops in West Africa. To protect yam against the threat of nematode pests, a banana-fibre paper, impregnated with micro-doses of abamectin, has been developed. Initial participatory field trials using the paper to wrap seed material at planting has demonstrated its protective effect in Central Benin. The objective of the current study was to assess the influence of yam cultivar on the efficacy of the treated banana paper against *Scutellonema bradys*. Using four yam cultivars, Klatchi, Môrôkô, Aïmon and Baniouré, popular in different yam production zones of Benin, the paper was assessed in a split-plot design in field trials. Compared with untreated controls, results showed that at tuber maturity *S. bradys* tuber peel densities were significantly lower by between 50% to 86%, depending on cultivar. By wrapping yam seed with abamectin-treated banana paper at planting, tuber yields were also increased, from 7 to 20% when compared with control plots. No interaction however was observed between control method and yam cultivar, indicating that yam cultivar did not influence the performance of abamectin-treated banana paper. This study demonstrates the protective effect of banana-fibre paper-based matrices impregnated with abamectin against plant-parasitic nematodes, irrespective of yam cultivar. Considering the high yam diversity in West Africa, this technology presents a worthy option for nematode management in yam.

Keywords: Benin - *Dioscorea* spp. - Food security - Plant-parasitic nematodes - *Scutellonema bradys*.

Addressing the challenges of High-throughput nematode identification using metabarcoding.

Mohammed Ahmed (mohammed.ahmed@nrm.se)

Nematology, Zoology, Swedish Museum of Natural History, Stockholm, Sweden

Over the years, metabarcoding helped unravel great insights into nematode communities in both aquatic and terrestrial environments. Despite the promise that this approach holds, there are still a number of factors that limit its widespread use within nematology. Among these are (i) the problem of identifying a suitable marker of the required taxonomic coverage and species resolution for discriminating between nematodes (ii) the inability to accurately predict relative abundance of taxa in mixed samples of any group of organisms based on their read frequencies from metabarcoding data (iii) the need for a huge repository of sequences of characterized species to serve as references for identification of unknown sequences. This study presents key findings in metabarcoding studies of nematodes, how these findings have helped address some of the above-mentioned challenges, and how they have opened future research opportunities. The identification of nematodes in bulk communities through the use of metabarcoding was tested with success. However, it is the inability of this approach to satisfactorily quantify the composition of bulk community samples that has been very difficult to realize. And efforts to resolve the the relationship between sequence read abundance and biomass of taxa in samples continue. Approaches such as improved sample preparation, alternative bioinformatic analysis and the use of correction factors based on prior knowledge of primer mismatch and whole genome sequences have been tested with some promising results, although not satisfactory for applion to real data. The issue of available reference sequences in public databases is one that needs time and increased efforts in ensuring that only sequences of accurately identified organisms are allowed into the database.

Plant-parasitic nematodes in potato crops in Portugal: patterns and influencing factors.

Ana Martins (anamartins@bio.uminho.pt), M. Teresa M. Almeida, Sofia R. Costa

CBMA, University of Minho, Campus de Gualtar, Braga, Portugal

Potato crops are susceptible to several pests and pathogens, including plant-parasitic nematodes (PPN), that reduce yield of this important food crop. Numerous studies on key genera such as *Globodera* spp. are available, but the communities and associations between different PPN genera of potato crops have seldom been considered. As part of the TREND project (POCI-01-0145-FEDER-029289; PTDC ASP-PLA/29289/2017) we characterised the PPN communities in 40 potato fields in the major potato cropping regions of Portugal. Data on PPN assemblages was then related to crop and abiotic factors to infer about their ecology and driving factors. Nematodes were extracted from soil samples collected at full plant growth, identified to family (for free-living nematodes) or to genus level (for PPN) and quantified. Enrichment, Structure and MI-family indices [1] were calculated using NINJA [2] to infer on soil ecological parameters. Climatic, edaphic, short-term cropping history and crop performance values were collected from available databases (Portuguese Institute for Sea and Atmosphere, United States Geological Survey – Sentinel 2, iGEO – Geographical information) and from farmer interviews for each sampling site. Data on abundance and distribution of PPN clustered the 14 genera detected into four main clusters reflecting their associations: *Pratylenchus, Tylenchorhynchus* and *Helicotylenchus*, the most abundant nematodes (prevalence above 65%) clustered together, along with tricodorid nematodes and *Meloidogyne*. The A2-listed *Globodera* was frequently associated with *Rotylenchus*, *Paratylenchus* and *Heterodera*. Correspondence analysis related the PPN assemblages with several biotic and abiotic parameters: climate (temperature and precipitation), latitude, soil properties (organic matter and P content) as well as plant performance (Normalized Difference Vegetation Index). Overall, nematode community analyses indicated simplified and N-enriched soil foodwebs, conducive to pests and diseases, but the ecological indices could not be related to PPN assemblages. Our results identify association patterns between different PPN genera that can be related to environmental or cultural conditions. This can be further explored to support the design of resilient potato cropping systems and prevent yield losses due to PPN.

Keywords: Climatic variables - NDVI - Soil properties - Bioindicators - Multivariate.

References:

- [1] Ferris et al., 2001. App Soil Ecol 18: 13–29.
- [2] Sieriebrienikov, 2014. Eur. J. Soil Biol. 61: 90:93.

Diversity and geographical distribution of plant parasitic nematodes of the federal capital territory Abuja, Nigeria.

Agatha Anih (esath_agatha@yahoo.com)

Biological Science Department, Nigeria Police Academy, Wudil, Kano, Nigeria

Plant-parasitic nematodes are known as one of the most important pests attacking various plants in the world, and investigating the nematode component is very essential for management of this pest and prevent damage to plants in general. This research was conducted to isolate and identify the different plant parasitic nematodes that exist in farming sites of Federal Capital territory, Abuja. Community structure and diversity of the nematodes were assessed from the 250g from soil rhizosphere of different Monocotyledonous and dicotyledonous crops from different area councils in Federal Capital Territory and were analysed by carrying out extraction and identification of the isolated of plant-parasitic nematodes. Quantification was also determined using Cobb sieving andmodified Baermann funnel method of extraction. Identified nematode groups were comprised of both endo and ecto-parasitic nematodes from ten selected crops collected from the six Area councils. Six major plant parasitic nematodes with *Helicotylenchus* sp. (20,560), *Scutellonema* sp. (8700), *Ditylenchus* sp. (7430), *Circonemoides* sp. (4590), *Meloidogyne* sp. (5100) and *Tylenchulus* sp. (3700) were detected. Kuje Area council had the highest rate of nematode infestation with a total population of 13,410. This is followed by Kwali (10,500), Bwari (8900), Gwagwalada (7090), Abaji (6050) and AMAC (5230) in the order of decreasing population. These data were analysed statistically to determine the rate and the prevalence of the nematode infestation in respect to each locality of the Federal Capital Territory.

Keywords: Community Analysis - Diversity - Federal Capital Territory - Geographical Distribution - Plant parasitic nematode.

In silico analyses of genes responsible for somatic sex determination in plant-parasitic nematode, *Heterodera glycines.*

Muhammad Shahzad Anjam[1] (shahzadanjam@bzu.edu.pk), Azra Yasmeen[1], Muhammad Ibrahim[2], Nageena Zahid[1]

[1] *Institute of Molecular Biology and Biotechnology (IMBB), Bahauddin Zakariya University, Multan, Pakistan*; [2] *Department of Bio sciences, COMSATS University, Sahiwal Campus, Pakistan*

Plant-parasitic cyst nematodes are sedentary endoparasites, develop hypermetabolic feeding sites inside the roots of the host plant. They are sexually dimorphic and reproduce strictly amphimictically. However, sexual differentiation of the nematodes occurs only after feeding from the host plant. No sex chromosomes are present, and the environment plays a role in the development of sexual phenotype. Under favorable conditions, when plenty of nutrients are available with vigorous growth of the host plant, the majority of the nematodes develop into females. Whereas, high number of males develop under adverse conditions like resistant genotype and reduced availability of nutrients. However, molecular cascades leading to somatic sex determination in cyst nematodes are still unexplored. In current studies, we used well described global pathway of somatic sex determination in *Caenorhabditis elegans* as reference for genes involved in sexual differentiation. We accessed the transcriptome databases of *Heterodera glycines*, a soybean cyst nematode from Sequence Read Archive (SRA) available at NCBI. There are more than 15 genes in the pathway of somatic sex determination in *C. elegans*. In ongoing analyses, we found 4 genes of *Heterodera glycines* showing significant amino acid homology with *sex-1*, *fem-2*, *tra-1* and *tra-3* genes in *C. elegans*. TRA-1 is a transcription factor and terminal gene of the pathway which is under the direct regulation of FEM-2, a PP2C phosphatase protein. We predicted the tertiary structure of putative Hg-FEM-2 protein by ab-initio method. The structural analyses were performed using I-TASSER, ERRAT and CHIMERA. Moreover, functional, comparative and interactional features were studied using PFAM, MEME, COACH, TM-align and protein string. Our *in-silico* results revealed significant structural, functional and interactional homology of putative Hg-FEM-2 with Ce-FEM-2. We propose involvement of putative Hg-FEM-2 in the sex determination of *H. glycines* that may mimic the function of Ce-FEM-2 in *C. elegans*. The studies are important to devise nematode-control strategies by manipulating their sexual differentiation in the field.

Keywords: Sex determination - FEM-2 - Sexual differentiation.

Understanding damage signals propagation required to build resilient cell walls in plant roots.

Muhammad Shahzad Anjam[1, 2] (muhammad.anjam@slu.se), Luciano Martìn Di Fino[1], Xuemin Ma[1], Julei Guerreiro[1], Shahid Siddique[3], Florian M.W. Grundler[4], Peter Marhavy[1]

[1] *Umeå Plant Science Centre, Department of Forest Genetics and Plant Physiology, Swedish University of Agricultural Sciences, UMEÅ, Sweden*; [2] *Institute of Molecular Biology and Biotechnology, Bahauddin Zakariya University, Multan, Pakistan*; [3] *Department of Entomology and Nematology, University of California, Davis, United States*; [4] *Molecular Phytomedicine, Rheinische Friedrich-Wilhelms-University of Bonn, Bonn, Germany*

Cyst nematodes are biotrophic endoparasites that have developed a very complex infection strategy that involves manipulating plant developmental systems in order to generate neoplastic feeding cells within the host root. The second stage juveniles (J2s) of beet cyst nematode, *Heterodera schachtii* invade generally near the elongation zone, move across numerous tissue layers to reach the nutrient-rich vascular cylinder to develop permanent feeding sites. During the migratory phase, nematodes release cell wall degrading enzymes, which compromise the cellular integrity of distinct cell files. As a result, when plants detect the collapse of their own cell wall components and the release of cytoplasmic fluid, a number of defensive reactions are triggered. Here, we identified that for restricted, single-cell wounding in different root tissues caused by cyst nematodes or laser ablation, plant roots rapidly localize lignin accumulation leading to delay in nematode development. Surprisingly, nematode and laser ablation does not induce a robust lignin accumulation response in root, but regionally activates genes involved in the phenylpropanoid pathway, which is implicated in the biosynthesis of lignin. This lignin activation depends on ethylene biosynthesis activities, as well as of NADPH oxidases. Overall, the regional signals caused by single-cell wounding stimulation of lignin deposition is not a default effect for general healing, but a specific mechanism to strengthen defense responses around the vascular cylinder, thus appears to constitute a relevant root immune response against small invaders.

Keywords: Root wounding - Lignin accumulation - Single cell damage - Laser ablation, - Cyst nematodes.

Diversity of the entomopathogenic nematodes in Australia and their potential for the control of Queensland fruit fly.

Sitaram Aryal[1] (s.aryal@westernsydney.edu.au), Uffe N Nielsen[1], Craig Wilson[2], Ian Broughton[2], Nanette Hope Sumaya[3], Markus Riegler[1]

[1] *Western Sydney University, Richmond, NSW, Australia*; [2] *Ecogrow Environment Pty Ltd, Westgate, NSW, Australia*; [3] *Department of Biological Sciences, College of Science and Mathematics, Mindanao State University-Iligan Institute of Technology, Iligan City, The Philippines*

Entomopathogenic nematodes (EPNs) are important biocontrol agents of insect pests with soil-inhabiting stages. They may be useful for the control of Australia's most significant horticultural pest, Queensland fruit fly, *Bactrocera tryoni*. The exploration of Australian EPNs may lead to isolates with higher efficacy against *B. tryoni* due to their adaptation to *B. tryoni*, local climate and soil conditions. A total of 116 soil samples were collected from three different habitats (citrus orchard, forest and grassland) across ten sites in New South Wales (NSW). Two EPN isolation techniques were compared; insect baiting and Baermann funnel extraction. For each soil sample, twenty bait individuals of *Tenebrio molitor*, *Galleria mellonella* and *B. tryoni* were used. Soil moisture, pH, texture and SOM were measured. EPN recovery frequencies were 52%, 22% and 9%, respectively. With the Baermann funnel extraction, 47% of soil samples were EPN-positive. Four different species of EPN were found: *Steinernema feltiae*, *Heterorhabditis zealandica*, *H. marelatus* and *H. indica*. *Heterorhabditis marelatus* is newly recorded for Australia while *H. indica* and *H. zealandica* are newly recorded for NSW. The infectivity of the EPNs were assessed against *T. molitor* larvae and *B. tryoni* larvae and pupae. The lethal dosages (LD50) were 4.83± 0.83 to 23.16± 6.62 for *T. molitor*; 23.97± 5.20 to 176.50± 20.61 for *B. tryoni* larvae and 88.77± 8.16 to 412.33± 104.67 for *B. tryoni* pupae. *Heterorhabditis zealandica* had higher infection rates against *T. molitor* and *B. tryoni* larvae. Among the 16 tested strains of the four different EPN species *S. feltiae* was found most effective against *B. tryoni* pupae. Detailed morphological and molecular characterization of all EPNs and their bacterial symbionts is ongoing. Amplicon sequencing will be performed to further characterise and quantify EPN communities directly from the soil.

Keywords: Biocontrol - Nematode distribution - DNA barcoding - Insect pests - Lethal dosage.

Identification of soil factors associated with root-knot nematode density in green manure-applied fields.

Erika Asamizu[1] (easamizu@agr.ryukoku.ac.jp), Kazuma Tanioka[1], Haruki Sawada[1], Yuzuka Senoo[1], Yusaku Nezu[1], Daisuke Yoshimura[2]

[1] *Department of Plant Life Science, Ryukoku University, Otsu, Shiga, Japan*; [2] *Department of Bioresource Science, Ryukoku University, Otsu, Shiga, Japan*

Green manure is widely used in fields to suppress plant-parasitic nematode density in the soil. Typical examples are crotalaria (Fabaceae), which traps nematodes in the roots but prevents their growth; marigold (Asteraceae), which secretes nematicidal substances from the roots; and leaf mustard (Brassicaceae), which contains glucosinolates in leaves that act as fumigant by plowing them into the soil. These nematode suppressive effects are direct effects of green manure plants. Application of green manure changes various soil environmental factors that would indirectly affect plant-parasitic nematode density in the soil. The microbiome and autotrophic nematode flora are such variable factors. When the soil is rich in microbiome, the autotrophic nematodes feeding on bacteria and fungi will be increased and conversely, the plant-parasitic nematode density will antagonistically be suppressed. In this study, we are focusing on the indirect effects of green manure in terms of inorganic soil elements (pH, EC, nitrate nitrogen, available phosphate, exchangeable K, Mg and Ca), microbiome and autotrophic nematodes. The interaction of these factors with the plant-parasitic nematode density observed in a field test will be discussed.

Keywords: Green manure - Plant-parasitic nematode - Autotrophic nematode - Microbiome.

Novel endophytic nematode antagonistic fungi- potential for nematode biocontrol.

Samad Ashrafi[1] (samad.ashrafi@julius-kuehn.de), Abdefattah A. Dababat[2], Wolfgang Maier[1]

[1] *Institute for Epidemiology and Pathogen Diagnostics, Julius Kuehn Institute (JKI)—Federal Research Centre for Cultivated Plants, Braunschweig, Germany*; [2] *International Maize and Wheat Improvement Center (CIMMYT), Ankara, Turkey*

Parasitism caused by cereal cyst nematodes is a major limiting biotic factor in cereal cropping systems. A sharp reduction in population of *Heterodera filipjevi* was observed in experimental wheat fields of the International Maize and Wheat Improvement Centre (CIMMYT) in Turkey. Microscopic observations revealed the presence of many fungal infected nematode cysts colonising their egg contents. Accordingly, a study was conducted to isolate and identify potentially new nematophagous fungi associated with nematode eggs by establishing a specific isolation technique, study the antagonistic interaction of these fungi with *H. filipjevi*, and to identify the natural compounds with nematicidal effect produced by these fungi. A focused screening approach was applied, in which only those nematode eggs exhibiting symptoms of fungal infection were individually cultured. This approach has so far resulted in finding several new fungal species including the recently described species *Ijuhya vitellina*, *Monocillium gamsii*, and *Polyphilus sieberi*. The fungal species *P. sieberi* is the first report of a dark septate endophyte (DSE) parasitising nematode eggs. In addition, three hitherto undescribed pleosporalean species were isolated, two of which were also DSEs reported during several independent ecological studies. Screening of *the newly obtained fungi for secondary metabolites with* has so far resulted in identification of several known and new natural compounds such as chaetoglobosin A, new peptides including an arthrichitin-like lipodepsipeptide, several new peptaibols, the new cyclic lipodepsipeptide ophiotine, artrichitin B and C, and a new xanthocillin-like alkaloid, xanthomide Z. Several of these compounds showed moderate nematicidal activity. Light microscopy of the antagonistic interaction between these fungi and *H. filipjevi* showed that fungal hyphae enter the body cavity of developing juveniles inside the eggs and develop into microsclerotia. These microsclerotia were rich in oil-like droplets and germinated on artificial media, suggesting that they might be appropriate candidates for mass production of this group of fungi. Parasitism of nematode eggs caused by the DSEs *P. sieberi*, and the pleosporean strains JKI72996, and JKI73017 suggests a multifunctional lifestyle that could potentially benefit the plant host by antagonising the nematodes as well as by translocating nutrients from colonised nematodes to the host plant.

Keywords: Antagonist - Biocontrol - Endophyte - New species.

Root lesion nematode biocontrol in potato.

Pedro Barbosa[1] (pbarbosa@uevora.pt), Jordana Branco[2], Cláudia Vicente[1, 3], Ana Cristina Figueiredo[4], Manuel Mota[5]

[1] *NemaLab/ Mediterranean Institute for Agriculture, Environment and Development, Institute for Advanced Studies and Research, Évora University, Évora, Portugal*; [2] *InnovPlant Protect CoLab, Elvas, Portugal*; [3] *INIAV, I.P., National Institute for Agrarian and Veterinarian Research, Oeiras, Portugal*; [4] *Centro de Estudos do Ambiente e do Mar (CESAM Lisboa), Centro de Biotecnologia Vegetal, Faculdade de Ciências da Universidade de Lisboa, Lisboa, Portugal*; [5] *NemaLab/ Mediterranean Institute for Agriculture, Environment and Development, Department of Biology, School of Science and Technology, Évora University, Évora, Portugal*

Potatoes (*Solanum tuberosum*) are one of the world's most common vegetables, as asource of many essential nutrients, like vitamin C and potassium. The diversity of pests and diseases affecting potato is high, with special emphasis towards fungi, insects and nematodes. The root lesion nematode, *Pratylenchus penetrans*, ranking third in terms of agronomic economic losses among plant parasitic nematodes, has a wide host range and despite being mobile during the majority of his life cycle, they are root dependent as feeding site. *P. penetrans* is the most common *Pratylenchus* species in Portuguese soils, although species distribution and abundance are largely unknown. Current control methods are costly and hazardous to the environment and to humans, and in today´s context of sustainable development, the selection of environmentally safe and effective ways to control plant parasitic nematodes, become imperative. Thus, there is a need to search for sustainable alternatives, and plant natural products may play an important role in nematode control. Nematicidal activity of 30 molecules (18 oxygen-containing- and 12 hydrocarbon compounds) was assessed following standard methodology. At 2 mg/mL after 24h-exposure, two oxygen-containing terpenes (carvacrol and thymol) and a benzoic acid derivative (benzaldehyde) achieved 100% mortality, followed by the fatty alcohol 3-octanol (99%) and benzoic acid derivative, methyl salicylate, with around 76%. The mortality from the monoterpene hydrocarbons was <10%. On-going research is evaluating the minimum inhibitory concentration from the compounds able to achieve full mortality. Future steps include evaluating the phytotoxic effects of the selected compounds in healthy potato plants, and the nematode–potato response to the addition of such compounds in terms of plant physiology, biochemistry, and defensive pathways.

This work was funded by National Funds through FCT under the PhD grant SFRH/BD/134201/2017, and projects PratyOmics (PTDC/ASP-PLA/0197/2020), MED UIDB/05183/2020, CESAM UIDP/50017/2020+UIDB/50017/2020+LA/P/0094/2020.

Keywords: Biocontrol - Plant-response - *Pratylenchus penetrans* - *Solanum tuberosum*.

Strategies to enchance efficacy of entomopathogenic nematodes for management of diamondback moth and imported cabbageworm.

Sabina Budhathoki (sbudhathoki@ufl.edu), Koon-Hui Wang

Plant and Environmental Protection Sciences, University of Hawaii at Manoa, Honolulu, Hawaii, United States

The foliar application of entomopathogenic nematodes (EPNs) to manage diamondback moth (DBM) and imported cabbageworm (ICW) in the field is not promising due to its susceptibility to environmental stress. Greenhouse trials were done to test the efficacy of adjuvants in protecting *Steinernema feltiae*. Adjuvants 1) Oroboost® 2) Kinetic® and 3) Exit® mixed at 3.9 ml/L, and 4) no adjuvant in 9-cm diameter plates containing 500 IJs freshly harvested infective juveniles (IJs) were exposed for 0, 30, 60, 120 and 180 min. Oroboost® enhanced the survival rates of *S. feltiae* for 2 hours compared to no adjuvants, whereas Kinetic® and Exit® showed signs of EPN toxicity with lower survival rates. In another greenhouse trial where 10 DBM 4th instar were introduced on cabbage seedlings in pots, Oroboost® outperformed the other adjuvants in reducing DBM number 2 days after EPN spray. A laboratory and greenhouse experiment were done to test the dosages of *S. feltiae* at 0, 0.625, 1.25, and 2.5 IJ/cm2 mixed with Oroboost®. Both experiments showed that 0.625 IJ/cm2 was as effective as the commercial (1.25 IJ/cm2) and higher dosage (2.5 IJ/cm2) in reducing DBM larvae. Two field trials were done by integrating trap cropping (kai choi) with EPN sprays (1.25 IJs/cm2 of *S. feltiae* + Oroboost®) to manage DBM and ICW, on head cabbage and kale. Both experiments were arranged in 2 × 2 (trap crop × EPN) factorial design with 4 replicated plots. It was hypothesized that trap crop would lure DBM away from cash crops and reduce the pest pressure for EPN to be effective. Planting trap crop suppressed ICW (73%), DBM (47%), and their damage on head cabbage ($P \leq 0.05$). On the other hand, EPN suppressed the abundance of ICW larvae by 43.4% compared to no EPN but had no effect on DBM. On kale, trap cropping did not affect DBM abundance and damage but reduced ICW damage by 12.6%. However, EPN significantly suppressed DBM numbers on kale by 87.5% ($P \leq 0.05$) probably due to low numbers of DBM in the kale (< 0.2 DBM/plant) than in the cabbage (> 0.3 DBM/plant). No interaction between trap crop and EPN was observed indicating that trap cropping did not improve efficacy of EPN against DBM and ICW. This study concluded that while Oroboost® at 3.9 ml/L could extend persistency and enhance the efficacy of *S. feltiae* foliar spray against DBM, its effect is short-lived. Field application of EPN is only effective when the population densities of DBM are lower than the economic threshold (0.5 DBM/plant).

Histopatholoy of cotton resistant line CNPA 17-26 B2RF to *Meloidogyne incognita* obtained by marker-assisted selection.

Juvenil E. Cares[1] (cares@unb.br), Carina M. L. Lopes[1], Ana Cristina M. M. Gomes[2], Guilherme F. Nascimento[2], Regina M. D. G. Carneiro[2]

[1] *Dep. Fitopatologia, Universidade de Brasília, Inst Ciências Biológicas, Brasília, DF, Brazil*; [2] *Lab. Nematologia, Embrapa Recursos Genéticos e Biotecnologia, Brasília, DF, Brazil*

The root-knot nematode (RKN), *Meloidogyne incognita,* is a major pathogen of cotton crops. Breeding plants for genetic resistance is the most desirable strategy to control this pathogen. This study objective was to clarify the resistance mechanisms, in the germplasm derived from the M-315 resistance source. The resistant line CNPA 17-26 B2RF (triple cross [BRS 368RF x M-315] x [BRS 430B2RF]) was selected for histopathological characterization of plant-nematode interaction, and compared with the susceptible FiberMax 966 (FM 966). Plants were inoculated with 10,000 J2s, and some roots were stained with acid fuchsin and other processed according Pegard et al. (2005) [1] at 2, 4, 6, 9, 12, 15, 19, 23, 26, 30, 34 and 40 days after inoculation (DAI). The nematode penetrated equally in both genotypes. In root thin sections using microscopy UV excitation or stained with toluidine blue, a strong blue fluorescence or dark blue color was visualized in the tissues around the nematode (hypersensitivity reaction, HR), in the cortex and central cylinder of the resistant plant, indicating accumulation of phenolic compounds in the roots, mainly at the beginning (from 2 to 6 DAI). At 9 DAI, giant cells in the early stage of subdivision next to nematodes were observed in the central cylinder of the resistant plant, and phenolic compounds were also shown around the nematode. At 12–40 DAI these initial cells were completely degraded with phenolics affecting the nematodes and initial giant cells. No fully developed giant cells or mature females were observed, only fourth-stage juveniles and males were visualized at 34 DAI. This resistance mechanism characterizes near-immunity, and so no enlarged females, nor egg production were observed. In susceptible control, well developed feeding sites were observed from 6 to 30 DAI. Females reached maturity at 26 DAI, and eggs were observed at 30 DAI. Our results suggest that the resistance (near-immunity) of the line CNPA 17-26 B2RF was related to early (2–12 DAI) defense responses that totally prevented nematode reproduction.

Keywords: *Gossypium* - Host reaction - Near-immunity - Root-knot nematode.

References:

- [1] Pegard et al., 2005. Phytopathology 95: 158-165.

Comparative and evolutionary analysis of RNAi pathways in plant parasitic nematodes.

Xinyue Cheng[1] (chengxy@bnu.edu.cn), Bingyan Xie[2]

[1] *College of Life Sciences, Beijing Normal University, Beijing, China*; [2] *Department of Plant Protection, Institute of Vegetables and Flowers, Chinese Academy of Agricultural Sciences, Beijing, China*

Objective: Plant-parasitic nematodes (PPNs) are a group of organisms that cause tremendous economic losses in agriculture and are intractable to control. RNA interference (RNAi) provides a promising technology for developing a novel control strategy to these PPNs. So far, the repertoires of related proteins in RNAi pathways are well known in *C. elegnas* and its related nematodes, but few are known in plant-parasitic nematodes, although RNAi is an established experimental technique in plant-parasitic nematodes, especially in root-knot nematodes and cyst nematodes. Available genome sequences provide opportunity to investigate the repertoire of core proteins associated with small RNA biogenesis and RNAi effectors, and further explore the origin and evolution of RNAi machinery in plant-parasitic nematodes. Methods: Utilizing available genome sequences deposited in NCBI, in addition to three new genomes we sequenced, totally 26 PPN genomes were obtained, which comprised 7 genera 17 species. Then, using known protein sequences involving in RNAi pathways of *C. elegans* and other nematodes as a query, by BLASTp against the PPN genome dataset, orthologous sequences were identified. The maximum likelihood phylogenetic tree of orthologues was constructed. Results: We found that RNAi effector repertoires in PPN genomes are richer than those reported before. Most of *C. elegans* proteins involving in RNAi pathways have orthologues in PPN genomes. Notably, there are more components in RNAi machinery in migratory PPNs than those in sedentary PPNs. Phylogenetic trees of these proteins were constructed, and their origin and evolution were discussed. Conclusion: By comparative analysis, we determined the conservation and divergence of proteins in RNAi machinery in migratory and sedentary PPNs, and explored the origin and evolution of RNAi pathways in nematodes.

Keywords: Plant-parasitic nematode - RNA interference - RNAi effors - Small RNA biosynthesisy - RNAi pathways.

References:

- Dalzell JJ, McVeigh P, Warnock ND, Mitreva M, Bird DM, et al. 2011 . PLoS Negl Trop Dis 5(6): e1176
- Ghildiyal M, Zamore PD. 2009. Nat Rev Genet.; 10(2): 94–108.
- Buck AH, Blaxter M. 2013. Biochem. Soc. Trans.41: 881–886

Occurrence and abundance of parasitic nematodes of papaya (*Carica papaya* Linnaeus) in western region of Burkina Faso.

Laëtitia Coulibaly (laetitiacoulibali@yahoo.fr)

Doctoral School of Science and Technology/Nematology Laboratory, Joseph KI ZERBO University/INERA, Bobo-Dioulasso, Burkina Faso

The production of papayas is expanding rapidly in Burkina Faso but remains under the pressure of many pests such as plant-parasitic nematodes (PPNs) which were poorly characterized until now. Our work aimed to perform a comprehensive survey of PPNs associated with papaya and to assess their structure and distribution in regions growing papaya in Burkina Faso. In this study, we collected 69 soil and root samples in Western region of Burkina Faso (2 regions, 7 localities). About 5 sub-samples were collected per farm to form a composite sample and nematodes were extracted from soil and roots [2, 3]. Among 69 isolates, nine genera from eight families were identified [1], such as *Meloidogyne*, *Rotylenchulus*, *Helicotylenchus*, *Pratylenchus*, *Scutellonema*, *Tylenchorhynchus*, *Xiphinema, Criconemella*, and *Paratrichodorus*. The amount of PPNs varied from 0 to 18,927 nematodes per dm^3 soil, and 0 to 62 nematodes per gram of roots. The highest PPN prevalences were recorded for *Rotylenchulus* (97.1 %), *Helicotylenchus* (95.6 %) and *Meloidogyne* (81.1 %). The highest nematode densities (mean; min-max per dm^3 soil) were observed for *Rotylenchulus* (3,989; 0 to 28,240) in the Cascades region, followed by *Helicotylenchus*(1,349; 0 to 7,940) and *Meloidogyne* (354; 0 to 3,840) in the Hauts-Bassins region. Based on these preliminary results, PPNs surveys were now extended to new papaya-producing regions and molecular characterization (rDNA, SCARs) is underway to identify PPN species associated with papaya, focusing on *Meloidogyne*, *Pratylenchus* and *Rotylenchulus*. This study provides an update for the PPNs associated with papaya in Burkina Faso and will allow implementing sustainable management strategies in orchards.

Keywords: Plant-parasitic - Nematodes - Papaya - Identification - Burkina Faso.

References:

- [1] Mai et Lyon, 1975. United Cornell University Press, 219P.
- [2] Seinhorst, 1962. Nematologica, 8 : 117-128.
- [3] Seinhorst, 1950. Tijdschrift Plantenziekten, 56 : 291-349.

Pathotype and resistance classification of *Heterodera avenae* and *H. filipjevi* in Huanghuaihai Valley of China.

Jiangkuan Cui[1] (jk_cui@163.com), Bo Zhou[1], Haohao Ren[1], Shijun Jiang[1], Shiming Liu[2], Deliang Peng[2], Abdelfattah Dababat[3]

[1] *College of Plant Protection, Henan Agricultural University, Zhengzhou, Henan, China*; [2] *Institute of Plant Protection, Chinese Academy of Agricultural Sciences, Beijing, China*; [3] *Soil Borne Pathogen (SBP) Program, CIMMYT, Ankara, Turkey*

To better understand cereal cyst nematode (CCN) pathotypes, resistance and chemical efficacy in the Huanghuaihai Valley, the major wheat-production area in China, four typical CCN populations, Xuchang, Tangyin, Qihe, and Juye, were tested. Four barley cultivars Ortolan (Rha1+), Morocco (Rha3+), Bajo Aragon1-1, and Martin 403-2 were all resistant, while both KVL 191 (Rha2+) and Dalmatische were susceptible, to these four populations. Wheat cultivars, except Iskamish-K-2-light, were moderately resistant to the Qihe and Juye populations, while the other cultivars were highly susceptible to all four populations. *H. avenae* from Tangyin, Qihe and Juye and *H. filipjevi* from Xuchang were classified as pathotypes Ha91 and "West", respectively. In this study, the 56 most promising varieties of winter wheat, collected from the International Wheat and Maize Improvement Center (CIMMYT) and the Huanghuaihai Valley, China, were screened against CCNs. Among them, twenty-two cultivars were highly susceptible (HS), 5 susceptible (S), 3 moderately susceptible (MS), 6 moderately resistant (MR), and 6 resistant (R) to pathotype Ha91; 19 cultivars were HS, 7 S, 3 MS, 9 MR, and 4 R to pathotype "West". Aizao8-15, MV17/3/Azd/Vee»s»//Seri82/Rsh/4/Fln/Acc//Ana/3/Pew»s»/5/Rsk/CA8055//Cham6 and MIRZABEY2000 were all R to both *H. avenae*, and *H. filipjevi*. GA951079-3-5/Neuse, Wo0608 and Xi'nong509 were all R to *H. avenae* but MR to *H. filipjevi*. Under natural field conditions, coating wheat seed with abamectin (Aba) + isopycnic imidacloprid (Imi) and methylene (bis) thiocyanate + 10% thiamethoxam (MTT) decreased the numbers of *H. avenae* cysts by 56.41% and 53.84%, but increased the wheat yield by 26.61% and 20.18% (net income valued at 175.9 and 136 CNY per 667 m^2), respectively. Meanwhile, these treatments decreased the numbers of *H. filipjevi* cysts by 45.69% and 47.02%, but increased the wheat yield by 19.23% and 9.43% (net income valued at 100.4 and 43.9 CNY per 667 m2), respectively. These results indicate that the *H. avenae* population from the Huanghuaihai Valley were inferred to be pathotype Ha 91 and the *H. filipjevi* population was pathotype "West". The main wheat cultivars in China were all S or HS varieties, however, the R resources are very scarce. Seed coating was still the most suitable means against CCN under the natural filed conditions, in China.

Keywords: Cereal cyst nematode - Thiamethoxam - Pathotype Ha91 - Wheat resistance - Seed coating compounds.

Widespread DNA N6-methyladenine plays a crucial role in parasitic nematodes.

Dadong Dai (daidadong@webmail.hzau.edu.cn), Shurong Zhang, Chuanshuai Xie, Yayi Zhou, Shengqiang Mao, Jinshui Zheng, Donghai Peng, Ming Sun

Huazhong Agricultural University, Wuhan, China

DNA *N*6-methyladenine (6mA) is a noncanonical DNA modification that is present at low levels in different eukaryotes. Previous studies have verified the presence of 6mA methylation in *Caenorhabditis elegans* and the mutant demethylase NMAD-1 alone still has high viability in the 20th generation, but its prevalence and importance associate with genomic function in parasitic nematodes remains poorly understood[1,2].Here, using SMRT sequencing and confirmed by LC-MS/MS, immunoprecipitation of 11 plant/animal-parasitic nematode genomes, we found that 0.05%-0.27% of all adenines were methylated in nematodes. *Mi24650* potentially involved in 6mA demethylation in *Meloidogyne incognita* (Mi) and conserved in plant-parasitic nematode, verified by demethylating activity reduced the 6mA percentage in vitro, and raised the percentage and 6mAIPseq peaks by RNAi in vivo. The number of eggs to J2 was significantly decreased by *Mi24650*-RNAi, and transcription factor (TF) 17946 show the same phenotype after RNAi. To study the function of *Mi24650* and *TF17946* RNAi *in vivo*, we constructed transgenic tobacco expressing dsRNA of *Mi24650*, *TF17946* and *GFP*, respectively. Pot experiment show that the incidence of root-knots and egg number per gram root was dropped to near no in high expression strain, and the male proportion increased significantly. Verified by qRT-PCR, multiple essential parasitic associate effector genes Mi-XYl1, Mi-CRT, Mi8D05, MiPFN3, 16D10, MiSGCR1, MiIDL1 were sharply down-regulated 22, 126, 10, 28, 27, 5, 13-fold, respectively, suggests 6mA may regulate effector to help Mi complete lifecycle. Gene ontology (GO) analysis of *Mi24650*-RNAi and *TF17946*-RNAi revealed that the enrichment pathways are almost identical, associated with a similar phenotype after RNAi *in vitro* and *in vivo*, suggested that *TF17946* may be the main TF for demethylase to regulate downstream genes. Our data present the first study to confirm that 6mA is widespread in parasitic nematode, and provides new insights into the role of 6mA plays a crucially function in root-knot nematode (RKN) for the regulation of the most important weapon of effector. Transgenic plant target demethylase shows a high resistance to RKN imply a feasible strategy for biocontrol. Additionally, the *Mi24650* knock-down caused Mi hardly to complete lifecycle, compared with the *C. elegans* NMAD-1 mutants high viability, we speculate that epigenetics may play a more important role in parthenogenetic than sexual reproduction.

Keywords: 6mA - Methylation - Biocontrol - Effector.

References:

- [1] Greer EL, Blanco MA, Gu L, et al. DNA Methylation on N6-Adenine in C. elegans. Cell. 2015;161(4):868-78
- [2] Mondo SJ, Dannebaum RO, Kuo RC, et al. Widespread adenine N6-methylation of active genes in fungi. Nat Genet. 2017;49(6):964-968

Transcriptome analysis of *Meloidogyne graminicola* infected roots of resistant mutant rice.

Manoranjan Dash[1] (manoranjan1993@gmail.com), Vishal S. Somvanshi[1], Roli Budhwar[2], Rohini Sreevathsa[3], Uma Rao[1]

[1] *Division of Nematology, Indian Agricultural Research Institute, New Delhi, Delhi, India*; [2] *Bioinformatics, Bionivid Technology [P] Limited, Bangalore, Karnataka, India*; [3] *Plant Biotechnology, National Institute of Plant Biotechnolog, New Delhi, Delhi, India*

Plant parasitic nematodes are well established as devastating pests in agricultural systems. Rice (*Oryza sativa* L.) production is severely affected by rice root-knot nematode *Meloidogyne graminicola* which has emerged as a menace in upland and irrigated culture systems. In a previous study, T-DNA insertional mutants were developed in rice (acc. JBT 36/14) using an apical meristem-targeted *in planta* transformation strategy. A panel of mutants showing varying phenotypes was screened against *M. graminicola* that led to selection of a resistant event. Quantification of transcripts of 25 genes associated with host defence responses indicated corroboration with the phenotype exhibited by the mutant [1]. Here, we investigated the mechanism of resistance to this nematode by comparing the transcriptomes of roots from the resistant mutant and susceptible JBT 36/14 wild type (WT). Among the 22,241 transcripts found, 674 exhibited altered total expression levels between infected mutant and susceptible WT. Significant differences were observed in the expression levels of genes related to integral component of membrane, cell wall organization, oxidoreductase activity, protein kinase activity and protein phosphorylation. The up-regulated genes related to plant secondary metabolites, such as diterpenoids, phytoalexins, phenylpropanoid and cellulose may be responsible for resistance in the mutant to the *M. graminicola* infection compared with that of JBT 36/14 by affecting cell wall organization or biogenesis. Further, several defence related genes were significantly induced by *M. graminicola* in the infected resistant line. Similarly, expression of genes related to calcium binding activity were altered that might be responsible for elevated levels of hypersensitive reaction in the mutant compared to control. The site of integration of T-DNA is being investigated through Genome walking. This study may help in enhancing our understanding of the mechanisms underlying plant-nematode interactions.

Keywords: Transcriptome - *Meloidogyne graminicola* - Rice - Parasitism - Nematode.

References:

- [1] Hatzade, B.et al., (2019). Biologia, 74(9), 1197-1217.

Identification of new esophageal gland effector candidates from adult females of the root-knot nematode.

Raquel de Oliveira Rocha (r.rocha@uga.edu), Richard Hussey, Melissa Mitchum

Plant Pathology and Institute of Plant Breeding, Genetics, and Genomics, University of Geogia, Athens, Georgia, United States

The root-knot nematode *Meloidogyne incognita* secretes effector proteins to manipulate and transform selected host cells into a specialized feeding site. These effectors are secreted through a hollow, protrusible stylet, and their production originates from either dorsal or subventral esophageal glands, whose activities differ according to the life stage of the nematode. Considerable community-wide efforts have led to the identity of genes encoding nematode effectors, however, the majority of studies have focused on the early stages of parasitism. Here, we discuss a new approach to enrich for the highly active dorsal glands of root-knot adult females. Female heads were manually cut from the body and a combination of sonication and vortexing was used to dislodge the contents inside the heads. Dorsal gland-enriched fractions were collected by filtering using cell strainers. RNA extraction and sequencing of pre-parasitic second-stage juveniles (ppJ2), female heads (FH), and dorsal gland-enriched (DG) samples was conducted. Mapping to *M. incognita* v3 reference genome identified 37,160 unique genes across all samples. Pair-wise comparisons of DG with ppJ2 and DG with FH identified 17,089 and 2,286 differentially expressed genes (DEG), respectively. Of the 1,788 genes common in both comparisons, 458 were upregulated in the DG. Further data mining led to a final list of 83 genes containing a predicted signal peptide, no transmembrane domains, and no homology to proteins in the free-living nematode *Caenorhabditis elegans*. Of these, 19 were previously determined to be gland-expressed effector genes [1,2,3]. Thirteen previously unidentified genes were confirmed to be expressed within the DG of adult females by *in situ* hybridization. Additional analyses of the new dorsal gland effector candidates (13) showed that 10 had a DOG box promoter motif, 3 contained a nuclear localization signal (NLS), and 11 contained a predicted MERCI effector motif within their encoded protein sequences. Future work on the functional characterization of these candidate effector proteins will expand our understanding of the molecular and biochemical mechanisms underpinning root-knot nematode parasitism.

Keywords: Dorsal gland - Effector - Root-knot nematode.

References:

- [1] Huang et al., 2003. Mol Plant Microbe Interact. 16(5): 376-381
- [2] Rutter et al., 2014. Mol Plant Microbe Interact. 27(9): 965-74
- [3] Nguyen et al., 2018. New Phytol. 217(2): 687-699

Molecular and functional characterization of the *Ditylenchus destructor* voltage-gated calcium channel α1 subunits.

Zhong Ding (dingzh@hunau.net), Shan Ye, Rune Zeng

College of Plant Protection, Hunan Agricultural University, Changsha, Hunan, China

Voltage-gated calcium channels (VGCCs) mediate the entry of Ca^{2+} ions into cells in response to membrane depolarization and play fundamental roles in the nervous system. The α1 subunits are the main subunits of Ca^{2+} channels and are membrane-spanning proteins with a pore-forming structure in the centre. *Caenorhabditis elegans* possess genes encoding α1 subunits [1-3]; however, very few of these genes have been cloned in plant parasitic nematodes (PPNs). *Ditylenchus destructor* is a PPN and has been proposed as a new model to study the biology and control of PPNs [4]. To understand the structure and function of the VGCCs of this PPN, we first cloned and identified three full-length cDNAs of VGCC α1 subunit genes in *D. destructor* with the defining structural and conserved features of Cav_1 (L-type), Cav_2 (non-L-type) and Cav_3 (T-type). *In situ* hybridization assays demonstrated that the Cav_1 VGCC α1 subunit gene (*DdCα1D)* was present within body wall muscles. The Cav_2 VGCC α1 subunit (*DdCα1A)* was expressed in the oesophageal gland, vulva and vas deferens of the worm, and the Cav_3 VGCC α1 subunit (*DdCα1G)* was localized to the oesophagus and median bulb. In addition, using in vitro knockdown of L-, non-L- and T-type genes via RNAi, these genes were predicted to play a key role in modulating locomotion, foraging and reproduction. After silencing *DdCα1G*, the median bulb muscle of *D. destructor* was obviously contracted, and the foraging and reproduction abilities were significantly inhibited. This study provided insight into the structure and function of VGCC α1 subunits in *D. destructor.*

Keywords: *Ditylenchus destructor* - Voltage-gated calcium channels - α1 subunit - In situ Hybridization - RNAi.

References:

- [1] Lee R Y, et al. 1997, EMBO J. 16, 6066–6076.
- [2] Schafer W R, et al. 1995, Nature 375, 73–78.
- [3] Shtonda B, et al. 2005, J. Exp. Biol. 208, 2177–2190.
- [4] Zheng J, et al. 2016, Proc Biol Sci. 283(1835):20160942.

Comparative study of neem-derived pesticides on *Meloidogyne incognita* under *in vitro* and pot trials in a greenhouse.

Pratik Doshi[1] (pratik.doshi159@gmail.com), Ferenc Tóth[1], Péter Nagy[2], György Turóczi[1], Renáta Petrikovszki[1]

[1] *Plant Protection Department, Szent Istvan University, Godollo, Hungary*; [2] *Zoology, Szent Istvan University, Godollo, Hungary*

Two different neem-derived plant protection products, i.e neem leaf extract and a commercial product containing 1% azadirachtin were used to study their effects on *Meloidogyne incognita* under *in vitro* and pot trials under glasshouse conditions. *In vitro* experiment was performed in flat-bottom 96-well microplates in three replicates. *M. incognita* J2 mortality patterns were examined after 24 hours under different neem leaf extract and azadirachtin concentrations. In the pot experiment, two Hungarian tomato land races (Dányi and Ceglédi) were artificially infested with *M. incognita*, then treated with different neem leaf extract and azadirachtin concentrations. Experiments were terminated 9 weeks after the setup. Gall index was measured using three different scales at the end of the experiments. In the *in vitro* studies, the highest concentration (1%) of neem leaf extract resulted in more than 90% mortality in J2, whereas mortality caused by the commercial product did not differ significantly from the control. In the pot trials under glasshouse conditions, fresh shoot weights and number of fruits of both landraces were not statistically different among the treatments. The Zeck scale was found to be the most suitable for evaluation of gall formations compared to other scales (by Garabedian and Van Gundy and Mukhtar et al.,). Gall index was reduced in number but not significantly in all the treatments compared to positive control and 0.1% azadirachtin was significantly different from the control. This shows that neem-derived pesticides can reduce root galling and help control *M. incognita* infestation with proper planning and implementing of treatments

Keywords: Neem leaf extract - Azadirachtin - Root-knot nematodes - Biological control - Tomato.

Effect of *Meloidogyne incognita* and *Fusarium oxysporum* f. sp. *vasinfectum* resistance traits in cotton.

David Dyer (drd0013@auburn.edu), Kathy Lawrence

Auburn University, Auburn, AL, United States

The cotton fusarium wilt disease complex of *Meloidogyne incognita* (RKN) and *Fusarium oxysporum* f. sp. *vasinfectum* (FOV) occurs all around the world. Ten different races/genotypes of the FOV pathogen have been identified in U.S. cotton fields and have been found to coexist within a single field. The objective of this study was to evaluate the effects of RKN and FOV resistance on the incidence of the fusarium wilt. The study was established in a cotton field historically known to be infested with *M. incognita* race 3 and diverse FOV races/genotypes. Six cultivars of *Gossypium hirsutum* and two cultivars of *Gossypium barbadense* that ranged from highly susceptible to highly resistant to FOV and RKN were planted to assess the ability of these resistance traits to limit fusarium wilt. To identify the FOV races and genotypes present, symptomatic plants were collected from the field throughout the 2018 and 2019 seasons. FOV isolates collected were identified to race by sequencing portions of the Translation elongation factor, Beta-tubulin, and Phosphate permease genes. Nematode egg data were collected from cotton roots at the end of the growing seasons by means of gravity sieving and sucrose centrifugation. Eight different races/genotypes of FOV were identified in samples collected throughout the seasons with race 1, LA 108, and race 8 being the most common. The highest RKN population density was recorded on the two *G. barbadense* cotton cultivars included in the test, Phytogen 800 and Pima S7 (366 and 166 eggs/g of root respectively). However, low rates of FOV infection, 8% on and 7% respectively, were recorded on these cultivars which may be due to FOV resistance traits present in these cultivars. The highest infection rate of FOV (20%) was recorded on the Rowden cultivar which also supported the highest RKN population density (121 eggs/g of root) of any *G. hirsutum* cultivar. A similar rate of FOV infection (17%) was recorded on DP 1558NR B2XF. This is a RKN resistant cotton cultivar that supported a low population density of nematodes (11 eggs/g of root). This demonstrates that RKN resistance alone is not sufficient to protect against fusarium wilt if the cultivar is highly susceptible to FOV.

Ethylene Response Factor genes modulate plant root exudate composition and the attraction of plant parasitic nematodes.

Steven Dyer[1] (sdyer01@qub.ac.uk), Ryan Weir[1], Deborah Cox[1], Xavier Cheseto[2], Baldwin Torto[2], Johnathan Dalzell[1]

[1] *Queen's University Belfast, Belfast, United Kingdom*; [2] *ICIPE, Nairobi, Kenya*

Plant root exudates are compositionally diverse, plastic and adaptive. Ethylene signalling influences the attraction of plant parasitic nematodes, presumably through the modulation of root exudate composition. Understanding this pathway could lead to new sources of crop parasite resistance. Here we used Virus-Induced Gene Silencing to knock down the expression of two *ETHYLENE RESPONSE FACTOR* (ERF) genes, *ERF-E2* and *ERF-E3,* in tomato. Root exudates were significantly more attractive to the PPNs *Meloidogyne incognita* and *Globodera pallida* following knockdown of *ERF-E2*, which had no impact on the attraction of *Meloidogyne javanica*. Knockdown of *ERF-E3* had no impact on the attraction of *Meloidogyne* or *Globodera* spp. Gas Chromatography Mass Spectrometry analysis revealed major changes in root exudate composition relative to controls. However, these changes did not alter the attraction of rhizosphere microbes *Bacillus subtilis* or *Agrobacterium tumefaciens.* This study further supports the potential of engineering plant root exudate for parasite control, through the modulation of plant genes.

Variation of recognition mechanisms of predator species to other nematodes.

Taisuke Ekino[1] (t.ekino888@gmail.com), Natsumi Kanzaki[2], Ryoji Shinya[3]

[1] *Meiji University, Kawasaki-shi, Kanagawa, Japan*; [2] *Kansai Research Center FFPRI, Kyoto-shi, Kyoto, Japan*; [3] *Meiji University and JST PRESTO, Kawasaki-shi, Kanagawa, Japan*

Animals recognize the environmental condition in their own way based on their own nervous systems. The structure of nematode nervous systems exhibits a number of common features, although some variations exist between species. Thus, what is the "difference" in recognition mechanism among nematode species? In terms of *Pristionchus pacificus*, an omnivore species including bacteriophagous and predatory froms, the recognition mechanism has been studied in detail [1]. *P. pacificus* distinguishes some strain from closely-related strains by chemical recognition. To understand the variation of recognition mechanism of predator species, we studied the recognition mechanism of *Seinura caverna*, a stylet-borne predator species. First, we observed predacious behavior of *S. caverna* against conspecifics, closely-related predatory species, and prey species. *S. caverna* distinguishes conspecifics from prey species, but it could not distinguish conspecifics from closely-related predatory species. Second, we carried out a chemotaxis assay to measure the attraction of *S. caverna* to nematode extracts, and we observed the ultrastructure of the cuticle of *S. caverna* and related species. Our results suggest that *S. caverna* distinguish conspecifics and close relatives from prey species based on physical structure rather than chemical substance. Thus, our results suggest that *S. caverna* and *P. pacificus* have different recognition mechanism, *i.e.*, *S. caverna* mainly uses physical sensation.

Keywords: Recognition - Predator - Transmission electron microscope - Aphelenchoididae.

References:

- [1] Lightfoot et al., 2019. Science. 364: 86-89

Advancing knowledge of the root-knot nematode *Meloidogyne luci.*

Ivânia Esteves[1] (iesteves@uc.pt), Joana Cardoso[1], Duarte Santos[1], António Correia[1], Beatriz Cantarinho[1], Olímpia Sobral[1], Isabel Abrantes[1], Maria L. Inácio[2], Filomena Nóbrega[2], Leidy Rusinque[1, 2], Carla Maleita[1, 3]

[1] *Centre for Functional Ecology - Science for People & the Planet, Department of Life Sciences, University of Coimbra, Coimbra, Portugal*; [2] *Instituto Nacional de Investigação Agrária e Veterinária (INIAV, I.P.), Oeiras, Portugal*; [3] *Chemical Process Engineering and Forest Products Research Centre, Department of Chemical Engineering, University of Coimbra, Coimbra, Portugal*

The root-knot nematode (RKN) *Meloidogyne luci* is included in the Pest Alert List of the European Plant Protection Organization due to the potential damage it can cause to economically important crops. In order to assess the impacts of *M. luci* and to reach informed decisions on the development of effective programs to manage this RKN on agricultural crops, research is being carried out at UC and INIAV regarding: 1) the ecological requisites of *M. luci*; 2) the host status of cultivated plants locally cropped in subsistence farms, to a Portuguese *M. luci* isolate; and 3) the development of molecular methods for quick detection and reliable discrimination of this RKN species. Results revealed that the *M. luci* isolate is well adapted to the Portuguese climate conditions, being suited to soil temperatures ranging between 20-30°C, and reproduces on 24 out of 35 local cultivars (Rf=2.1-152.3; GI=4-5). Seven cultivars of different crops were classified as resistant-hypersensitive (Rf<1; GI>2) and four tomato cultivars (Actimino, Briomino, Vimeiro and Veinal), carrying the *Mi-1.2* gene, were resistant (0.0<Rf<0.1; GI<2), suggesting that crop rotation can be an effective control strategy if resistant cultivars are selected. Furthermore, a species-specific primer set was successfully designed based on a sequence characterized amplified region (SCAR) marker, providing the first diagnostic molecular method for the specific detection of *M. luci.* The PCR-SCAR methodology proved to be reliable in detecting *M. luci* DNA from second-stage juveniles and from tomato root galls. Additional studies are being conducted to increase knowledge on *M. luci* and to identify new effective strategies for its monitoring and sustainable management.

Funding: FCT/FEDER projects UIDB/00102/2021, PTDC/ASP-PLA/31946/2017, CENTRO-01-0145-FEDER-000007, CEECIND/02082/2017 and Instituto Ambiente Tecnologia e Vida.

Keywords: Crop susceptibility - Molecular diagnostics - Root-knot nematode - Thermal requisites.

Nematicidal activity of naphthoquinones – nematode generation of reactive oxygen species.

Carla Maleita[1, 2] (carla.maleita@uc.pt), Ivânia Esteves[2], Mara E. M. Braga[1], Isabel Abrantes[2], Hermínio C. Sousa[1]

[1] *Chemical Process Engineering and Forest Products Research Centre, Department of Chemical Engineering, University of Coimbra, Coimbra, Portugal*; [2] *Centre for Functional Ecology-Science for People & the Planet, Department of Life Sciences, University of Coimbra, Coimbra, Portugal*

The scarce availability of efficient nematicides to control root-knot nematodes (*Meloidogyne* spp.), together with environmental and health-related issues, has recently encouraged research towards the development of novel, safer and effective natural-origin nematicides. Naphthoquinones (NTQ) are naturally occurring compounds in several families of plants. Among NTQ, juglone (JUG) and 1,4-naphthoquinone (1,4-NTQ) were shown to have nematicidal activity, among other properties and, thus, are promising compounds to develop novel, natural and effective nematicides. However, knowledge on their potential mode(s) of action is still unknown. Therefore, this study aimed to infer JUG and 1,4-NTQ modes of action through the assessment of reactive oxygen species (ROS) generation by microscopic observation of *M. luci* second-stage juveniles (J2). *M. luci* J2 were incubated at 22°C for 3 days in JUG or 1,4-NTQ at 20, 50, 100, 150 and 250 ppm, with water and Tween® 80 included as controls. After exposure, nematodes were washed, incubated in 2',7'-dichlorofluorescein diacetate (ROS indicator), paralyzed by sodium azide and randomly observed and photographed. After nematode staining and fluorescence monitoring, no J2 treated with water or Tween® 80 generated ROS fluorescence or vacuolization of internal contents. *M. luci* J2 treated with JUG did not exhibit ROS fluorescence but the formation of small and multiple vacuoles associated to mortality was detected, while J2 treated with 1,4-NTQ (concentrations ≥100 ppm) showed evidence of ROS fluorescence and multiple giant vacuoles. The vacuolization of J2 after exposure to JUG and 1,4-NTQ, and the lack of fluorescence of nematodes exposed to JUG suggest that the mode of action of the two compounds are different or the pro-antioxidant activity of JUG inhibits the generation of ROS at low concentrations. Although these bioactive compounds have effects on nematode mortality, hatching, penetration and reproduction, thus being promising alternatives to the use of synthetic nematicides to control RKN, their mode of action should be further investigated. *M. luci* transcriptome analysis after exposure to these NTQ will help to better understand the mode of action of these compounds on RKN.

Keywords: Bionematicide - Juglone - Management - 1,4-naphthoquinone - Root-knot nematode.

Control of rice root knot nematode by flooding.

Cheng Feixue[1] (cfx937207@126.com), Wang Dongwei[1], Tang Bei[1], Wang Jian[2]

[1] *Institute of Plant Protection, Hunan Academy of Agricultural Science, ChangSha, China*; [2] *Agricultural Economy and Regional Planning Research Institute of Hunan, ChangSha, China*

Rice root-knot nematode disease is a major constraint in rice production. In recent years, with the area of direct-seeding rice increasing in China, the occurrence and spread of rice root knot nematode is rapid and has occurred in Guangdong, Guangxi, Fujian, Hunan Province, etc. At present, there is no low-cost, safe and effective control measure. In this study, effects of flooding methods to rice root-knot nematode disease were evaluated in field trials. The results showed that flooding could significantly inhibit the occurrence of rice root-knot nematode disease better than dry-wet alternate conditions (direct seeding rice). The inhibition rate of root-knot nematode disease was 94.26% and flooding reduced the second-stage juvenile(J2) density of *Meloidogyne graminicola* in soil by 77.63% at 45 d after sowing. At 75 days after sowing, the inhibition rate of root-knot nematode disease was 58.56% and the J2 density of *M. graminicola* in soil was reduced by 72.22%. Therefore, flooding methods can effectively inhibit the occurrence of root-knot nematode disease in the field caused by the rice root-knot nematode, which is a safe and effective control measure.

Keywords: Rice root-knot disease - *Meloidogyne graminicola* - Waterflooding method - Direct–seeding rice - Control effect.

References:

- Mantelin et al.,2017,Molecular Plant Pathology, 18(1): 3-15
- Song et al.,2017,Plant Disease, 101(12): 2153
- Upadhyay et al.,2014,Journal of Progressive Agriculture, 5(1): 5-8
- Zhou,et al.,2018,Journal of Plant Protection, 45(6): 1412-1418 (in Chinese)

Plant parasitic nematode biosecurity threats from a Northern Ireland and UK perspective.

Thomas Fleming (thomas.fleming@afbini.gov.uk), Margaret Hainon-McDowell, Deborah Cox, Trevor Martin, Colin Fleming

Agri-Food & Biosciences Institute, Belfast, United Kingdom

Plant biosecurity is a complex issue facing modern day authorities. The UK's live plant trade has increased 71% since 1999, and recent plant health emergency outbreaks such as sudden oak death (*Phytophthora ramorum*) and ash dieback (*Hymenoscyphus fraxineus*) have been devastating to both the plant trade industry and the wider environment. The plant health risk from nematodes does exist, albeit their threat is less likely to be known by the general public due to the more localised impacts of nematodes and the scale of any quarantine actions required. Nevertheless, in recent years we have identified several unique cases of plant-parasitic nematodes in the UK and Ireland. Most interesting was the detection of the Pacific shoot-gall nematode *Anguina pacificae* at an Irish golf course, which was the first report outside of its known distribution in California, USA. There have also been isolated cases of the EU A2 quarantine pest, *Meloidogyne fallax* in both crops and turfgrass. Importantly, we recognise and highlight the importance of exporting planting material with several cases of Root-knot, Sting and Lance nematodes affecting newly constructed turfgrass sports pitches in Morocco and United Arab Emirates. From our own surveillance and reports across the UK and Ireland we are observing increased incidences of plant parasitic nematodes year on year, highlighting the importance of maintaining and reviewing plant biosecurity measures.

Keywords: Plant parasitic nematode - Biosecurity - Plant Health - Turfgrass.

References:

- Fleming, Thomas R., et al. A first report of Anguina pacificae in Ireland. Journal of Nematology 47.2 (2015): 97.

Population dynamics of *Mesocriconema xenoplax* parasitizing sweet cherry trees in British Columbia, Canada.

Thomas Forge (tom.forge@agr.gc.ca), Paige Munro

Science and Technology Branch, Agriculture and Agri-Food Canada, Summerland, British Columbia, Canada

Sweet cherry (*Prunus avium*) production is increasing worldwide and particularly in the semi-arid interior of British Columbia (BC). The ring nematode, *Mesocriconema xenoplax*, has recently become recognized as a pest of sweet cherry trees in BC. Understanding the cumulative impacts of *M. xenoplax* on tree health, interpreting diagnostic sample data, and predicting the impacts of climate change on *M. xenoplax* population densities, all depend on knowledge of the temporal dynamics of *M. xenoplax* populations. However, the annual population dynamics of *M. xenoplax* under irrigated perennial crops are not well known. The objective of our study was to measure population densities of *M. xenoplax* on a monthly basis over four years, in relation to soil temperature and moisture regimes, in a mature irrigated sweet cherry orchard. In most years there were two peaks in population densities, in spring and fall. In three of four years the spring peak was observed when soil temperatures had warmed to approximately 15 C, usually in May. The exception was spring of 2020 following the only winter in which the soil did not freeze, when the peak population density was observed in early March and soil temperatures were still below 5 C. Population densities generally declined through summer when soil temperatures were between 15 and 25 C, and then increased again in the fall after soil temperatures declined to below 15 C. The exception was 2021, the hottest summer of the study, when population densities did not start to rebound until November after soil temperatures had declined below 10 C. There was no relationship between degree day accumulation within growing seasons and the levels of fall population growth. Because the orchard was irrigated to maintain uniformly optimum soil moisture for tree growth, there were no observable relationships between soil moisture fluctuations and *M. xenoplax* population densities. In conclusion, our data indicate that extreme events such as soil freezing or density-dependent processes are more important drivers of *M. xenoplax* population dynamics than growing season heat units, and for diagnostic purposes spring and fall sampling periods would be equally representative of *M. xenoplax* population densities in irrigated orchards in BC.

Keywords: Population ecology - Climate change - Nematode sampling - Threshold populations - Density dependent.

Survey and Development of Molecular Soil Test for Soybean Cyst Nematode in Manitoba.

Nazanin Ghavamishirehjin[1] (ghavamin@myumanitoba.ca), Mario Tenuta[1], Albert Tenuta[2], Dennis Lange[3]

[1] *Department of Soil Science, University of Manitoba, Winnipeg, Manitoba, Canada*; [2] *Ontario Ministry of Agriculture, Food and Rural Affairs, Ridgetown, Ontario, Canada*; [3] *Manitoba Agriculture, Food and Rural Development, Altona, Manitoba, Canada*

This study reports the first occurrence of the SCN in Manitoba and development of real-time PCR assays for quantification of densities in soil. Soybean Cyst Nematode (SCN), *Heterodera glycines*,is recognized as one of the major pest of soybean worldwide. In Canada, the SCN is present in Ontario and Quebec. The nematode is expected to soon be found in Manitoba, as it is spreading rapidly from the neighboring counties of North Dakota and Minnesota(U.S.). Precise identification is crucial to reducing the spread of the pathogen and limiting yield losses. The objectives of this study were to survey soybean fields in Manitoba for the presence of SCN, and develop molecular protocols for quantification of SCN calibrated to traditional microscopic counting of eggs. In October 2017, 30 commercial soybean fields in Manitoba near the U.S. border with history of soybean cultivation were sampled. A modified Fenwick elutriator based on the USDA cyst extractor was used to recover cysts. Cysts were identified based on morphological characters[1],PCR with species-specific primers for *H. glycines* (CoxIII and SCAR) [2,3], and DNA sequencing of several genes. Four fields were found positive for SCN and had 2,1,14,and 4 cysts /2.2 kg of soil. In July of 2021, an additional soybean field showed severe symptoms of SCN. A sampling grid was conducted in an infested patch and cysts were extracted by the wet-sieving method[4]. The SCN population density of samples collected from the patch ranged from 0 to 7797 eggs/100 cm^3 of soil. The five positive fields were from one of the Rural Municipalities of Thompson,Norfolk-Treherne,Rhineland,Emerson-Franklin and Montcalm.In July 2019, 20 fields with a range of SCN levels from Southern Ontario were sampled and used to optimize extractions as well as PCR reaction procedures. Cyst and egg extraction was performed in a 100cm^3 subsample. Total genomic DNA was extracted using the PowerSoil DNA Isolation Kit. A SYBR Green based real-time PCR assay was optimized using the CoxIII,as well as SCAR-based primer set which were validated in our laboratory for their ability to identify SCN populations. Calibration curves were obtained by adding a different number of SCN eggs (10,100,500,1000) to both suspension and soil debris. The melting profile supported the specific detection of the SCN. There were highly significant correlations between the number of eggs added and Cq values, indicating the sensitivity, specificity and quantification of the method.

Keywords: *Heterodera glycines* - Soybean cyst nematode (SCN) - Survey - Molecular technique.

References:

- [1] Subbotin and Baldwin, 2010. Brill.
- [2] Ou et al., 2008. Nematology 10(3):397-403.
- [3] Subbotin et al., 2001. Nematology 3(4): 365-371.
- [4] Niblack et al., 1993. Nematology 25(4): 880-886.

Ability of indigenous EPN isolates to control Bactrocera dorsalis and their formulations for mango fruit fly biocontrol in Benin.

Anique Godjo[1] (Godjoanique@gmail.com), Nadia Chabi[1], Perrin Dossou[2], Lionel Zadji[1], Olive Batcho[1], Hugues Baimey[1], Antonio A. Sinzogan[2], Aimé H. Bokonon-Ganta[2], Nicodeme Orou Wari[1], Anne Willems[3], Wilfrida Decraemer[3], Leonard Afouda[1]

[1] University of Parakou, Parakou, Benin; [2] University of Abomey Calavi, Abomey Calavi, Benin; [3] Ghent University, Gent, Belgium

Looking for a sustainable way to prevent damage caused by fruit flies mainly Bactrocera dorsalis to mango fruits in Benin as in the whole West Africa region, we investigated the use of entomopathogenic nematodes (EPNs) for the biological control of this insect pest in mango orchards. One isolate of Steinernema kandii (Thui) and two of Heterorhabditis taysearae (Hessa1 and Koroborорou F4) were investigated for their invasion time and virulence to third instar larvae of B. dorsalis in laboratory and semi field tests, respectively. In addition, the persistence of the same nematode isolates in soil under field conditions was tested. Furthermore, the effects of adjuvants such as kaolin, clay, starch and biochar on the survival, fitness and pathogenicity of these entomopathogenic nematode species have been assessed in liquid and solid formulation under laboratory conditions. Results showed that all three nematode isolates could penetrate insect larvae during 2 h of exposure time. Under semi field conditions, insect mortality was significantly different among EPN application times. The three nematode isolates were highly pathogenic to B. dorsalis with H. taysearae Hessa1 being the most virulent (70.84% ± 10.46 [SEM] mortality) when EPNs were applied three days before insect introduction in the experimental pots. Moreover, Steinernema kandii persisted in soil up to 32 weeks after nematode application whereas both H. taysearae isolates persisted 30 weeks post application in the mango orchard. In general, four weeks after nematode application, the density of infective juveniles decreased considerably and remained variable the following sampling dates. Preliminarily results show that all the three isolates survived well in the tested adjuvants with the clay-based formulation showing the better performance. Experiments are being repeated and continued in lab and glasshouse conditions to determine the suitable formulations of EPNs to be used in biocontrol of mango fruit flies.

Keywords: Biological control - Tephritid - Entomopathogenic nematodes - EPN formulations - Mangifera indica L.

References:

- [1] Clarke AR, Armstrong KF, Carmichael AE, Milne JR, Raghu S, Roderick GK, Yeates DK (2005) Invasive phytophagous pests arising through a recent tropical evolutionary radiation: the Bactrocera dorsalis complex of fruit flies. Annual Review of Entomology 50:293-319
- [2] Ferguson CS, Schroeder PC, Shields EJ (1995) Vertical distribution, persistence, and activity of entomopathogenic nematodes (Nematoda: Heterorhabditidae and Steinernematidae) in alfalfa snout beetle (Coleoptera: Curculionidae) infested fields. Environmental Entomology 24:149-158
- [3] Shapiro-Ilan, DI, Lewis, EE, Behle, RW, McGuire, MR (2001) Formulation of entomopathogenic nematode–infected–cadavers. Journal Invertebrate Pathology, 78:17–23.
- [4] Godjo A, Zadji L, Decraemer W, Willems A, Afouda L (2018) Pathogenicity of indigenous entomopathogenic nematodes from Benin against mango fruit fly (Bactrocera dorsalis) under laboratory conditions. Biological Control 117:68-77 doi:10.1016/j. biocontrol.2017.10.009
- [4] Hou B, Xie Q, Zhang R (2006) Depth of pupation and survival of the Oriental fruit fly, Bactrocera dorsalis (Diptera: Tephritidae) pupae at selected soil moistures. Applied Entomology and Zoology 41:515-520

Impact and management of *Meloidogyne enterolobii* in sweetpotato production in North Carolina, United States.

Adrienne Gorny[1] (agorny@ncsu.edu), Tanner Schwarz[1], Hunter Collins[1], Lina Quesada-Ocampo[1], Lindsey Thiessen[1], Eric Davis[1], Weimin Ye[2]

[1] *Entomology and Plant Pathology, North Carolina State University, Raleigh, North Carolina, United States*; [2] *Nematode Assay Section, North Carolina Department of Agriculture & Consumer Services, Raleigh, North Carolina, United States*

The Guava root-knot nematode (*Meloidogyne enterolobii* Yang and Eisenback) is an aggressive plant-parasitic nematode of sweetpotato (*Ipomoea batatas (*L.) Lam.) and numerous other agronomic and vegetable crops (Castagnone-Sereno, 2012). *M. enterolobii* is able to overcome known root-knot nematode resistance genes in many crop genotypes, including the *Mi-1*, *Mh*, *Mir1*, *N*, *Tabasco*, and *Rk* genes (Castagnone-Sereno, 2012; Ye et al., 2013). The state of North Carolina is the largest producer of sweetpotatoes within the United States, and thus the nematode poses a significant threat to sweetpotato production within the state. First confirmed in North Carolina in 2011, *M. enterolobii* has been observed infecting sweetpotatoes, cotton, and soybean in several agriculturally intensive regions (Ye et al., 2013). Producer response to *M. enterolobii* in North Carolina centers on identification of infested fields through soil sampling, avoiding introduction of the nematode into new locations, and reducing established nematode populations within infested fields. Research objectives were developed to support and enhance these management principles. A diverse array of sweetpotato germplasm was screened in the glasshouse to identify potential host resistance to *M. enterolobii*. This work differentiated several sweetptoato lines with reduced galling symptoms and low egg production, suggesting increased resistance, and which may be useful in future plant-breeding efforts. Field-based nematicide trials evaluated the efficacy of chemical programs in controlling *M. enterolobii* in sweetpotato and rotational crops. These trials identified best practices in the management of this species within the field. In a survey for *M. enterolobii* in North Carolina, the nematode was found in 13 separate counties (local regions) within the state. Future research opportunities and challenges to management are discussed.

Keywords: Root-knot nematode - Integrated pest management - Host resistance - Chemical control.

References:

- Castagnone-Sereno et al., 2012. Nematology. 14(2): 133-138.
- Ye et al., 2013. Plant Dis. 97(7): 1262.

Metabolic profile of tomato infected by *Meloidogyne javanica* in the presence of the fungus *Pochonia chlamydosporia.*

Angelica Gouveia[1] (angelica.s.gouveia@gmail.com), Túlio Morgan[1], Thalita Monteiro[2], Paulo Luiz[2], Leandro Freitas[2], Humberto Ramos[1]

[1] *Bioquímica e Biologia Molecular, Universidade Federal de Viçosa, Viçosa, Brazil*; [2] *Fitopatologia, Universidade Federal de Viçosa, Viçosa, Brazil*

This study aimed to evaluate the metabolic changes associated with fungus in the leaves, galls (structure composed of giant and female cells) and roots. To evaluate *M. javanica* + *Pochonia chlamydosporia* interaction, the plants were cultivated with *M. javanica* or *M. javanica* + *Pochonia chlamydosporia* and leaves, galls and roots were collected at 24 and 44 days after nematode addition. Leaf, gall and total root were evaluated to determine the composition of secondary metabolites. With this macerated material, metabolites were extracted with methanol, isopropanol and acetic acid solution (20: 79: 1 v / v / v) and the samples were analyzed by the Q-Tof spectrometer. The metabolite table containing the intensity of all ions detected in all runs was exported from the XCMS platform and used as input for statistical analysis by the MetaboAnalyst platform (http://www.metaboanalyst.ca/). The fragmentation pattern of the metabolites in the samples was compared with library standards to identify possible compounds present in the galls and roots. Preliminary results of this project indicated reduction in galls in the first evaluation of 28.65% and at the end there was a 43.18% reduction in gall number. Composition of secondary metabolites in the galls and leaves, in both evaluated times, showed a significant difference when *P. chlamydosporia* was present. In the galls, metabolic changes ranged from increasing some compounds to decreasing others. A possible compound detected in greater quantity in galls in the treatment containing the fungus (first time evaluated) was 5,8,11,14-Eicosatetraenoic (arachidonic acid). In the literature, there is only one study that relates this compound with plant-parasitic nematodes to which tomato seeds incubated with arachidonic acid and then cultivated showed a reduction of 33% in the number of galls formed by *M.incognita* [1]. Thus, the increase detected due to fungal action may be one of the contributions to the reduction in the number of galls observed in *P. chlamydosporia* treatment. The next step will be to understand the function of all metabolites identified in the plant + *M. javanica* + *Pochonia chlamydosporia* interaction and thus better understand the mechanisms used by the fungus to control these pathogens and the metabolic changes that this can cause in the plant.

Keywords: Secondary metabolites - Root-knot nematode - Pochonia chlamydosporia - Biological control - Metabolomic.

References:

- [1] Vasyukova N et al., 2009. Doklady Biological Sciences. 428: 448-450.

Efficacy of *Solanum sisymbriifolium* to control Globodera *pallida*.

Amanda Gray (agray@uidaho.edu), Louise-Marie Dandurand

Entomology, Plant Pathology, Nematology, University of Idaho, Moscow, Idaho, United States

The efficacy of *Solanum sisymbriifolium* as a trap crop over time for controlling the pale cyst nematode (PCN), *Globodera pallida* was evaluated. Under field conditions *S. sisymbriifolium* was shown to induce hatch of PCN eggs and confirmed to reduce the reproduction factor of PCN in a subsequent greenhouse bioassay with the susceptible variety Desiree. Samples taken at 6, 9 or 12 weeks, post establishment of fallow treatment indicated a reduction of encysted eggs by 20, 33 or 14 % respectively. Whereas, for the same time points, *S. sisymbriifolium* reduced the encysted eggs by 38, 41 or 47% respectively. This indicates that PCN hatched in the presence of *S. sisymbriifolium.* After the field trial, all samples were put into cold storage for 8 weeks. A bioassay was then conducted with the susceptible variety Desiree under greenhouse conditions for 12 weeks. The reproduction factor (Rf) from the bioassay after the fallow treatment was 13.2. Whereas from the *S. sisymbriifolium* treatment, the Rf was 0.3*. Solanum sisymbriifolium* reduced the Rf by 97% compared to the fallow treatment. In conclusion, this research demonstrates the applicability of *S. sisymbriifolium* in reducing reproduction of PCN when in rotation with potato.

Keywords: *Globodera pallida* - *Solanum sisymbriifolium* - Potato cyst nematode - Trap crop.

Integrative characterisation of eight plant-parasitic nematode species on olive trees in central Tunisia.

Ilhem Guesmi-Mzoughi[1] (guesmiilhem@yahoo.fr), Juan Emilio Palomares-Rius[2], Antonio Archidona-Yuste[2], Carolina Cantalapiedra-Navarrete[2], Hajer Regaieg[1], Fathia Hadj-Naser[1], Najet Horrigue-Raouani[1], Pablo Castillo[2]

[1] *Institut Supérieur Agronomique CM, Sousse, Tunisia*; [2] *Instituto de Agricultura Sostenible, Cordoba, Spain*

Olive trees host a large number of plant-parasitic nematodes (PPN) being estimated at about 250 species documented worldwide in the main olive producing countries. In this study, we aimed to unravel the diversity of PPN inhabiting the rhizosphere of cultivated olive trees (*Olea europaea* subsp. *europaea* var. *europaea*) in central Tunisia (Mahdia, Sousse, Kairouan and Kasserine). The olive growing area of this region of Tunisia is of high agriculture and socio-economic importance with a wide distribution of this crop. To this aim, we conducted a survey between 2013 and 2015, comprising 22 commercial olive orchards (cvs. Chemlali and Koroneiki). Integrative taxonomic approaches (morphological, morphometrical, molecular and phylogenetic analyses) were carried out for some species, among them, eight plant-parasitic nematode were identified to species level, with frequencies of prevalence as following: *Pratylenchus oleae* (4.5%), *Rotylenchus incultus* (18.2%), *R. eximius* (13.7%), *L. euonymus* (4.5 %), *L. glycines* (13.7 %), *Xiphinema conurum* (13.7 %), *X. meridianum* (13.7 %) and *X. robbinsi* (9.1 %). Molecular characterisation using D2-D3 expansion regions of 28S rRNA, partial 18S rRNA, ITS1-rRNA, and cytochrome c oxidase subunit 1 (*coxI*) was carried out and Bayesian inference analysis was used to reconstruct phylogenetic relationships among these species with other related plant-parasitic nematodes. These data suggest that plant-parasitic nematode species of the genera *Pratylenchus*, *Rotylenchus*, *Xiphinema* and *Longidorus* are predominant in olive as previously reported in other Mediterranean areas.

Keywords: Olive - *Pratylenchus* - *Rotylenchus* - *Xiphinema* - *Longidorus.*

Plant-parasitic nematode communities associated with olive trees in Central Tunisia.

Ilhem Guesmi-Mzoughi[1] (guesmiilhem@yahoo.fr), Hajer Regaieg[2], Fathia Hadj-Nasser[2], Sadreddine Kallel[3], Najet Horrigue-Raouani[2]

[1] *PROMAG Mabrouka, Ben arous, Naassen, Tunisia*; [2] *Institut Supérieur Agronomique chott-mariem, Sousse, Tunisia*; [3] *Institut National Agronomique de Tunisie, Tunis, Tunisia*

A high diversity of plant-parasitic nematodes (PPN) was reported on olive trees in many countries bordering the Mediterranean Sea [1]. However, in Tunisia the nematofauna are still not well known. So, this study aims to determine the structure and the diversity of PPN communities associated with cultivated olive trees (*Olea europaea* subsp. *europaea* var. *europaea*) in the main producing area in Central Tunisia (Sousse, Mahdia, Monastir, Zaghouan, Kairouan, Kasserine and Sfax). A total of 298 soil samples were collected, between 2013 and 2015, in 29 commercial olive orchards presenting different soil management systems (rainfed, irrigated and irrigated with cover crops). PPN were extracted, enumerated and identified to genus level with classical methods. The structure of PPN communities and taxonomical diversity were described and compared between different soil management systems. Then, the effect of soil physico-chemical properties on PPN community patterns was studied. Sixteen genera of PPN were detected; the most frequent were, in decreasing order, *Tylenchorhynchus* spp. (67%), *Pratylenchus* spp. (53%), *Helicotylenchus* spp. (51%), *Paratylenchus* spp. (46%) and *Meloidogyne* spp. (40%). The abundance of PPN, especially *Paratylenchus* spp. and *Meloidogyne* spp., and nematode diversity were higher in irrigated olive orchards in the presence of cover crops. *Pratylenchus* spp. predominated in irrigated soils; however, *Helicotylenchus* spp. was better adapted to rainfed conditions. The prevalence of the root lesion nematodes (*Pratylenchus* spp.) was highest in clay soils with high conductivity and exchangeable potassium, whereas sandy soils poor in organic matter were more favorable for root-knot nematodes (*Meloidogyne* spp.). According to these data, PPN communities in Tunisian olive orchards have low diversity with a predominance of *Helicotylenchus* spp., *Pratylenchus* spp. and *Meloidogyne* spp. The damage to olive trees by these nematode genera depends on the species. Therefore, additional investigations in other olive producing area in Tunisia, including species identification, are needed to better characterize these commmunities.

Keywords: Olive - PPN communities - Structure - Diversity - Soil management systems.

References:

- [1] Ali et al., 2014. C. R. Biol. 337: 423-442.

Plant-parasitic nematodes associated with medical hemp in Maryland.

Zafar Handoo (zafar.handoo@usda.gov), Mihail Kantor, Andrea Skantar, Lynn Carta

MNGDBL, USDA, Beltsville, MD, United States

Plant-parasitic nematodes are microscopic worms that cause an estimated ten billion dollars of crop losses each year in the United States and 100 billion dollars globally. One problem with nematodes is that growers have no idea of how many kinds of nematodes exist and the host ranges of nematodes on specific crop cultivars such as medical hemp (***Cannabis sativa indica***) which was recently deregulated for cultivation in the United States. A survey was conducted in October and November, 2019 from an organic medical hemp field in Baltimore County, Maryland, USA that was previously planted with strawberries. Seven samples were arbitrarily selected from different sections of the field. Nematodes were extracted from 100 cm3 soil through sieving and decanting, followed by sucrose centrifugal flotation. Mixed populations of 11 economically important plant-parasitic nematodes belonging to 11 genera, with 8 identified morphologically to species level using a compound light microscope at 10x to 100x magnifications. Anatomical features and measurements of females and males together with molecular analysis by sequencing were used for final nematode identification. *Boleodorus volutus*, *Criconema mutabile*, *Pratylenchus penetrans*, *Paratylenchus projectus*, *Helicotylenchus pseudorobustus*, *Tylenchus exiguus, Psilenchus hilarulus* and *Basiria siddiqii* were identified for the first time as associated with hemp in Maryland of which *Boleodorus volutus* and *Basiria siddiqii* represent new records for the United States. Other important plant-parasitic nematodes found in hemp rhizosphere were *Anguina sp*., *Ditylenchus sp*., and *Hemicriconemoides* sp. In addition, several free-living nematodes belonging to the order Rhabditida and Dorylaimida were recovered. Survey records showed new host plant records for most of the identified nematode species in Maryland. Further research is needed to assess the impact of these nematodes on growth, vigor and yield of hemp production.

Keywords: *Cannabis sativa indica* - Hemp - First report - Nematode - Survey.

References:

- Geraert, E. 2008.Gent, Academia Press. 540p
- Handoo, Z. A. and Golden, A. M. 1989. Journal of Nematology 21:202-218.

Establishment of laboratory growth system for potato cyst nematode.

Kotomi Hayashi (kotomi-hayashi@hokusan-kk.jp), Hiroko Miyashiro, Teiko Ono, Kyoko Wanibuchi, Akane Haseda, Yukiko Matsuhita, Noriko Tabayashi

Arroscience Research Laboratories, Hokusan Co. Ltd, Kitahiroshima, Hokkaido, Japan

Potato cyst nematode (PCN) is considered a pest that causes enormous damage to potato production, and various experiments are being conducted to develop a control method. In general experiments, field soil contaminated with PCN is used. However, the transport of PCN-contaminated soil is restricted by law, and there is a concern of leakage to the outside when handling PCN in a greenhouse under a license. In addition, many cysts from the field are contaminated with bacteria and fungi, which hinders experimenting. Therefore, we examined a laboratory breeding system that can reduce contaminated eggs while containing PCN. First, a fungicide with little effect on the growth of PCN was screened, and then a cultured seedling support for inoculation with PCN was examined. When the selected fungicide (tebuconazole) is irrigated to the seedling support and inoculated with hatched juveniles, cyst formation was observed after 1 month of inoculation. Moreover, the eggs in the obtained cyst showed a markedly reduced contamination rate compared to those grown in the greenhouse, and showed stable activity against hatching activation substances. Furthermore, when the cysts formed in this propagation system were subcultured, formation of next-generation cysts and stable activity toward hatching activation substances could be confirmed. This work was supported by Smart Cell Project from the New Energy and Industrial Technology Development Organization (NEDO) of Japan.

Keywords: Potato Cyst Nematode - Laboratory growth system - Hatching - Egg parasites.

References:

- [1] Marion A. Foot 1977. New Zealand Journal Zoology, Vol.4,183-186
- [2] Zlatka et al., 2003. Journal of Agricultural Sciences.Vol.48,No1,103-110

Evaluating potential trap crops for control of *Globodera pallida.*

Paige Hickman (phickman@uidaho.edu), Louise-Marie Dandurand

Entomology, Plant Pathology & Nematology, University of Idaho, Moscow, ID, United States

The potato cyst nematode (PCN), *Globodera pallida*, causes substantial yield loss and is a serious threat to potato production worldwide. In Idaho, the goal is to contain and eradicate this quarantine pest so that infested fields can return to potato production. Due to recent regulations and environmental concerns with soil fumigants and lack of resistant varieties suitable for Idaho growers, control options for PCN are fairly limited. PCN also poses a challenge as its eggs remain viable in cysts that can lie dormant in the soil for up to 30 years until in the presence of a hatching stimulus. Trap crops that stimulate egg hatch but prevent development and reproduction of PCN have proven to be an effective management tool. In particular, there has been success with *Solanum sisymbriifolium* (litchi tomato) as a trap crop. However, litchi tomato has no economic value in rotation and there is concern it may become invasive, therefore, it is worthwhile to investigate alternatives. Numerous *Solanum* species have been suggested as potential trap crops. This study seeks to test several species that are of interest due to preliminary data supporting nonhost status and potential for commercial value including: *Solanum qutioense*, *Solanum retroflexum*, *Solanum stipuloideum*, *Solanum aethiopicum*, *Solanum macrocarpon*, and *Solanum douglasii*. Two experiments are being conducted to determine whether these species meet the trap crop criteria of being nonhosts that stimulate hatch. In the first experiment, seedlings are inoculated with cysts and grown in greenhouse conditions for 12 weeks. Any newly produced cysts are extracted from the roots and counted. In the second experiment, root diffusate is collected for use in an assay to assess egg hatch stimulatory effects. In both experiments, the *Solanum spp.* of interest are being compared to susceptible potato cultivars 'Desiree' and 'Russet Burbank' as well as a bare soil negative control. The results of these experiments will reveal whether these crops are worth further pursuit as trap crops for the eradication of PCN. Another implication of these findings is that resistant solanaceous plants may provide a source of R genes in breeding efforts for a resistant potato cultivar. Ultimately, the data obtained from this study will serve to broaden our knowledge of species useful in the control of potato cyst nematodes.

Keywords: Trap crops - Potato cyst nematode - PCN.

Development and application of a rapid loop-mediated isothermal amplication for detection of cereal cyst nematodes.

Peng Huan (hpeng83@126.com), Xu Xiaoqing, Huang Wenku, Kong Linan, Peng Deliang

Institute of Plant Protection, CAAS, Beijing, China

The cereal cyst nematodes (CCN) *Heterodera avenae* and *H. filipjevi* are regulated pest nematodes of great economic importance and infect the root of several cereal crop worldwide. In the present, the Loop-mediated isothermal amplification (LAMP), a novel DNA amplification technique, was established to diagnose *Heterodera avenae* and *H. Filipjevi* directly from soil and infected wheat roots based on the PRAD fragment of them. Specificity of the LAMP primers was confirmed using DNA from various cyst nematode species. The experimental results suggested that the detection limitation of the LAMP assay was as low as 10^{-4} of single nematode DNA, and the detection sensitivity of the LAMP method for CCN DNA was 100 times higher than normal PCR-based detection methods. Moreover, the LAMP assay was successfully performed in total DNA from infected wheat roots using FTA card and the sensitivity was as low as one nematode per 1g roots. Because the LAMP assay does not require expensive equipment or specialized techniques, this LAMP-based diagnostic method is practical and useful under field conditions. This Research was supported by Chinese Special R & D Fund for Public Benefit Agriculture(201503114).

Keywords: Cereal cyst nematodes - LAMP - Rapid detection.

Species diagnosis, hosts and distribution of cyst nematodes of the genus *Heterodera* (Tylenchida: Heteroderidae) with a focus on species of concern for Australia

Daniel C. Huston (manda.khudhir@csiro.au), Manda Khudhir, Mike Hodda

Insect & Nematode Biosecurity, CSIRO National Research Collections, Canberra, ACT, Australia

Cyst nematodes of the genus *Heterodera* include around 80 species of plant-parasites, a number of which are significant pests of important crops. Many species of *Heterodera* are differentiated by subtle morphological differences making diagnoses difficult. Here, we discuss diagnostics of these nematodes and describe our ongoing studies on the species richness, distribution, and host range of cyst nematodes in Australia, which includes putative endemics, potentially undescribed natives, and quarantine exotics. Because of the complexities of cyst nematode taxonomy, diagnostics is becoming increasingly reliant on molecular data. However, around 40% of species have no molecular data available, and sequences of some of the more commonly used gene regions routinely fail to distinguish between some species. We will discuss our analysis of publicly available barcoding data and the utility of these data for species diagnoses. We will also report on several new records of cyst nematodes for Australia, updated distributions for known species and our efforts to recollect a putative endemic. Lastly, we will report on our efforts to build a new reference collection for research on cyst nematodes. Although multiple species are listed as priority plant pests in Australia, few specimens are held in Australian collections. Many species of *Heterodera* will only be distinguishable with more data, especially regarding intraspecific and interspecific morphological and molecular variation, so we are seeking collaborators for collecting specimens to facilitate acquisition of said data, and ultimately to better define what is likely to make a diagnosable species in the genus.

Keywords: Species diagnosis - Evolution - Cyst nematodes - *Heterodera*.

Response of soil nematode community to increased plant species diversity in an intensively managed grassland.

Israel Ikoyi (israelonoja.ikoyi@gmail.com), Guylain Grange, John Finn, David Wall, Fiona Brennan

Department of Environment, Soil and Land Use, Teagasc, The Irish Agriculture and Food Development Authority, Johnstown Castle, Wexford, Ireland

Plant productivity, decomposition and nutrient cycling are important processes that are controlled by plant-soil biota interactions [1,2]. Studies have shown that increasing plant species diversity results in increased biomass yields in intensively managed grasslands [3]. However, little is known about the response of soil nematodes (the most abundant metazoans in the soil) [4] to increasing plant species diversity in intensively managed grasslands. In this study, we investigated how increased plant species diversity affect the soil nematode community and their associated indices in an intensively managed grassland with the aim of studying aboveground-belowground interactions. Nematodes were extracted from 66 experimental plots differing in plant species diversity comprising 1-3 functional groups (FGs) using combinations of six plant species within the three FGs. The FGs included grasses: ryegrass (*Lolium perenne* L.) and timothy (*Phleum pratense* L.); legumes: red clover (*Trifolium pratense* L.) and white clover (*Trifolium repens* L.); herbs: chicory (*Cichorium intybus* L.) and plantain (*Plantago lanceolata* L.). Our results showed that the composition and diversity of the soil nematode community differed significantly among the levels of plant species diversity. Specifically, plots with all six-plant species had a significantly higher nematode diversity, maturity index and proportion of sensitive taxa (omnivore and predators) than the others. In addition, the abundance of plant-feeding nematodes (herbivores) were significantly lower in the six species plots than in the monocultures. Our results support the hypothesis that increasing plant diversity in intensively managed grasslands will result in a positive effect on the belowground soil biota. This is in addition to increased biomass yield obtained from the high-diversity plant communities. Overall, our findings contribute to knowledge of the positive effects of increased aboveground plant diversity on the belowground diversity of soil biota. This creates a pathway towards sustainable production with reduced fertilizer inputs.

Keywords: Plant-nematode interactions - Plant diversity - Intensive grasslands - Nutrient cycling - Sustainable grasslands.

References:

- [1] Neher, 2010. Annu Rev Phytopathol. 48:371–394
- [2] van der Heijden et al., 2008. Ecol Lett. 11:296–310.
- [3] Grange et al., 2021. J. Appl. Ecol. 58(9): 864-1875
- [4] van den Hoogen et al., 2019. Nature 572:194–198.

Post-embryonic development of the K-strategic nematode *Labronema baqrii* Khan, Jairajpuri and Ahmad, 1989; analysis of structural evolution and stability.

Zarrin Imran (zarrin.imran@gmail.com), Wasim Ahmad

Department of Zoology, Aligarh Muslim University, Aligarh, Uttar Pradesh, India

Dorylaims are k-strategists, have slow growth rates, significant taxonomic diversity, and play key roles in soil-food webs. Their post-embryonic development was rarely documented and if so then mainly with reference to odontostyle development. Temporal sampling of cattle manure for two years from greenhouse cultures yielded abundant populations of *Labronema baqrii* representing both adult and all juvenile stages. Here we present the morphological and morphometric data of ninetysix juveniles and forty adult specimens which are useful in determining the adaptive change, evolution, reliability and stability of weighted taxonomic characters. Juveniles of *L. baqrii* exhibit a continuum of character development. Changes were observed in the nature of the guiding ring; *i.e* single to double, increase in the robustness of odontostyle, increase in percentage of expanded part of pharynx, shift in DN and S1N1 and a posterior shift in primordial midpoint position from J1→J4 to adult which represents evolutionary modification in the weighted taxonomic character rather than the developmental plasticity. In the absence of fossil records, their developmental study helps in understanding the successional change (from J1→J4 to adult) in their structure.

Keywords: Developmental plasticity - K-strategist - *Labronema baqrii* - Successional change - Taxonomic characters.

References:

- Khan et al., 1989. Indian J. of Nematology 19, 194–189

DNA profile, species-specific primers for detection, and temperature for development of *Heterodera schachtii* in Japan.

Hideaki Iwahori[1] (iwahori@agr.ryukoku.ac.jp), Akihiro Hayano[2], Noritaka Nakamura[3], Hiroshi Yoshida[4], Satoshi Kitabayashi[5]

[1] *Faculty of Agriculture, Ryukoku University, Ohtsu, Shiga, Japan*; [2] *River Basin Research Center, Gifu University, Gifu, Japan*; [3] *Department of Mushroom Science and Forest Microbiology, Forestry and Forest Products Research Institute, Tsukuba, Ibaraki, Japan*; [4] *Graduate school of Agriculture, Kyoto University, Kyoto, Japan*; [5] *Crop and Flower Research Center, Nagano Prefecture, Shiojiri, Nagano, Japan*

The sugar beet cyst nematode, *Heterodera schachtii*, was found in Japan for the first time in September 2017. The initial infested area was only approximately 35 ha in one extremely narrow area. However, this year a new outbreak area has been found, and the infested area is thought to be expanding. The main control method is fumigation using 1,3-dichloropropene. Several studies have been conducted previously to characterize *H. schachtii* in Japan. **DNA profile**: The mitochondrial COI gene (COI), ribosomal DNA/ITS region (ITS), and LSU/D2-D3 (D2-D3) region were sequenced to determine the origin of *H. schachtii* in Japan. No clear regional differences were observed in the ITS and D2-D3 regions, but COI gene sequencing revealed that the population that invaded Japan was the same as that found in Korea, Hawaii, and the Netherlands, and different from that in Israel and Spain. **Species-specific primers**: Species-specific primers for *H. schachtii* were developed from the DNA sequence of the ITS region. These primers amplify a small DNA fragment of 159 bp that can be used for real-time PCR. DNA extracted from 1, 3, 10, 30, and 100 individuals closely matched with our calibration curve. It was possible to detect *H. schachtii* by DNA extracted from *H. schachtii*-infested soil using bead-based lysis method with a commercial DNA extraction kit. Nonetheless, further investigation is required to determine detection sensitivity. **Temperature for development**: Chinese cabbage was inoculated with *H. schachtii*, cultivated at 18°C, 20°C, 24°C, and 28°C, and the generation time (in days) at each temperature was investigated. Based on these data, the extrapolated zero development temperature of the Japanese *H. schachtii* population was 13.1°C, and the associated cumulative temperature required for one generation was 419 degree (°C) days (average of two experiments). This effective cumulative temperature is almost the same as that of the southern root-knot nematode, *Meloidogyne incognita*, in Japan. Since no abnormal growth was observed, even at 28°C, we inferred that *H. schachtii* could infest almost any region of Japan.

Keywords: Sugar beet cyst nematode - *Heterodera schachtii* - DNA profile - Species-specific primers - Temperature for development.

MiMsp40 effector of *Meloidogyne incognita* interacts with a transcription repressor of Arabidopsis and promotes parasitism.

Heng Jian[1] (hengjian@cau.edu.cn), Pei Liu[2], Yongpan Chen[2], Junhai Niu[3], Qian Liu[1], Jianlong Zhao[1], Shanshan Yang[1], Xianqi Hu[4]

[1] *Department of Plant Pathology, China Agricultural University, Beijing, China*; [2] *China Agricultural University, Beijing, China*; [3] *CATAS, Tropical Crops Genetic Resources Inst., Danzhou, China*; [4] *Yunnan Agricultural University, Kunming, China*

Root-knot nematodes (RKNs) are biotrophic parasites that invade plant roots and cause serious damage to their hosts. The nematode secrets effectors which have immunosuppressing activity and play essential roles in successful parasitism. A novel effector, MiMsp40 from *Meloidogyne incognita*, is known to suppress PTI and ETI of plants, thereby promoting the nematode parasitism. But its mechanism remains largely unknown. Immunolocalization analysis showed that MiMsp40 is produced in nematode gland cells and secreted into plant cells. Transcriptomic analysis revealed that the *LOX3/LOX4, AOS, AOC*, and *OPR3* genes were down-regulated in *MiMsp40* transgenic Arabidopsis compared to wild-type. Transgenic plants produced less endogenous jasmonic acid than wild-type according to phytohormone determinations. Conversely, foliar application of jasmonic acid abolished the susceptibility of transgenic plants to nematodes. Both BiFC and Co-IP assays confirmed MiMsp40 interacts with STZ, an Arabidopsis transcription repressor. In *stz* mutants of Arabidopsis, expression levels of *LOX3/LOX4, AOS, AOC,* and *OPR3* were up-regulated, and jasmonic acid content was also higher, resulting in less susceptibility of the plant to nematodes. STZ could bind to the *LOX3/LOX4* promoters and negatively regulate their respective transcription. Interestingly, MiMsp40 promoted the accumulation of STZ, which may enhance STZ's regulating ability to *LOX3/LOX4*. Our results uncovered a novel mechanism by which the nematode effector MiMsp40 interacts with plant transcription repressor STZ to suppress the expression of key jasmonate biosynthesis genes, thereby diminishing the endogenous jasmonic acid of plants to promote the nematode parasitism.

Keywords: Plant-nematode interactions - Jasmonate biosynthesis pathway - *Meloidogyne incognita* - MiMsp40 - Transcription repressor.

International perspectives on the United States Department of Agriculture Nematode Collection, past, current and future.

Mihail Kantor (mihail.kantor@ars.usda.gov), Zafar Handoo, Lynn Carta, Andrea Skantar, Stephan Rogers

MNGDBL, USDA-ARS, Beltsville, MD, United States

The United States Department of Agriculture Nematode Collection (USDANC) is one of the largest and most valuable nematode collections in the world which includes millions of nematode specimens. The collection was established in 1960 by A. Morgan Golden [1,2,3]. Over the years, nematode samples from 149 countries were incorporated into the collection. Moreover, if we take into consideration all the territories and former countries that were split in different countries, the total number of depositions increased to 172 countries and territories. Deposition of specimens to the USDANC were made from collaborators from all over the world as well as from U.S. point of entries. Today the USDANC is comprised of several collections, which include the Type Collection, General, Thorne Steiner Mermithid, Mass, Gates and a Demonstration Collection. The revised list of the type specimens added to the USDANC since 1998 was published in 2018 [3]. The collection database is accessible online at: https://nt.ars-grin.gov/nematodes/. The database also provides a detailed list of specimens that are deposited in the USDANC available to interested scientists for loan for limited periods of time. Currently, efforts are being made to digitize the images of all the specimens deposited in the collection. To expand the collection, we encourage all nematologists and scientists throughout the world to enrich our collection by depositing valuable type and other specimens in the USDANC for future generations.

Keywords: Collection - Deposition - Nematode slides - Type Collection - USDANC.

References:

- [1] Golden. 1962. Nematologica 8:84-5
- [2] Handoo et al. 1998. Journal of Nematology 30108–158
- [3] Handoo et al. 2018. Journal of Nematology 50(1):51-68

A new aphelenchoidid insect parasite from a tenebrionid beetle, *Uloma marseuli.*

Natsumi Kanzaki (nkanzaki@ffpri.affrc.go.jp), Keiko Hamaguchi

Kansai Research Center, Forestry and Forest Products Res. Inst., Kyoto, Japan

Within five phylogenetic groups of the family Aphelenchoididae, clade 3 is composed of five predatory and entomoparasitic subfamilies. Because of the shortage of available materials and difficulty in culturing, the subfamilies belonging to this clade are not clearly characterized morphologically and phylogenetically. During a recent survey of entomophilic nematodes in Japan, an unidentified aphelenchoidid parasite was isolated from a tenebrionid beetle, *Uloma marseuli*. The isolated species parasitized the body cavity of the host beetle, and it was not successfully cultured, suggesting the species is an obligate parasite of the beetle. Phylogenetically, the species is close to several *Ektaphelenchoides* spp. (Ektaphelenchinae) and *Peraphelenchus orientalis* (Entaphelenchinae). The species is typologically characterized by its lip structure, without clear constriction between the lip and the rest of the body, typical aphelenchoidid-shaped (thorn-shaped) male spicule, two pairs of male genital papillae (with P2 and P3 pairs, and lacking P1 and P4/5 pairs), lack of post-uterine sac of female. However, the species did not fit any described clade 3 genera, suggesting the species belongs to an unidentified genus close to either Ektaphelenchinae or Entaphelenchinae. Using several typological characters, we tentatively place this new species (genus) in the subfamily Ektaphelenchoidinae. However, more material collections and examination of biological characters are necessary for further characterization of the species/genus.

Keywords: Tenebrionidae - Ektaphelenchinae - Entaphelenchinae - New genus - Systematics.

Novel bacteria for biological control of root-knot nematode, *Meloidogyne incognita*.

Rami Kassam[1] (ramikassam9@gmail.com), Aditi Kundu[2], Annapurna K.[3], Haritha Bollinedi[4], Uma Rao[1]

[1] *Division of Nematology, Indian Agricultural Research Institute, New Delhi, Delhi, India*; [2] *Division of Agricultural Chemicals, Indian Agricultural Research Institute, New Delhi, Delhi, India*; [3] *Division of Microbiology, Indian Agricultural Research Institute, New Delhi, Delhi, India*; [4] *Division of Genetics, Indian Agricultural Research Institute, New Delhi, Delhi, India*

Among microorganisms that parasitize nematodes or reduce nematode populations by their antagonistic behaviour, bacteria hold important position and some of them have shown great potential as biocontrol agents. The objective of this study has been to reinvestigate the local bacterial diversity across selected states of India that could be playing a key role in maintaining the natural balance of nematode population *vis-a-vis* exploiting the potential ones for their management. In the present study, soil samples were collected from 13 Indian states from rhizosphere of perennial trees, pulses, cereals, fruits and vegetables and used for isolating potential bacteria employing *Meloidogyne incognita* and *Caenorhabditis elegans* as baits in water agar. Isolation and purification of various bacteria revealed the presence of some predominant genera belonging to both plant pathogenic and non-pathogenic groups: *Alcaligenes faecalis, A. aquatilis, A. endophyticus, Burkholderia cepacia, B. ambifaria, Pseudomonas entomophila, P. mosselii* and *Pseudochrobactruma saccharolyticum.* All the bacteria were identified using 16S molecular markers. Evaluation of different dilutions of culture filtrates (CF) of various bacteria against *M*. *incognita* revealed their potential to kill juveniles and also inhibit egg hatching. GC-MS analysis of CF revealed the presence of nematicidal compounds such as Heneicosane, Myristynoyl pantetheine and Dimethyl disulphide in most of the bacteria. Further evaluation of these bacteria against *M. incognita* infecting tomato plants under green house conditions shown that some of them are highly promising to contain the nematode infection. Additional evaluation of these promising bacteria under field conditions will support commercial exploitation of promising ones in future.

Keywords: Biological control - Nematode - *Pseudomonas* - *Meloidogyne incognita* - *Burkholderia.*

Nematicidal activity of South African botanical plant extracts against Meloidogyne species under vitro conditions.

Mbokota Candy Khosa[1] (mbokota@arc.agric.za), Odwa Gonyela[2], Fannie Van Heerden[2]

[1] *Crop Protection, ARC-Tropical and Subtropical Crops, Mbombela, South Africa*; [2] *School of Chemistry and Physics, University of KwaZulu-Natal, Pietermaritzburg, South Africa*

Plant-parasitic nematodes are detrimental to agriculture resulting in estimated global crop losses of 17% per annum [1]. Chemical soil fumigants and nematicides have been used for years to control plant-parasitic nematodes in crop production systems but due to environmental and human health concerns, several fumigants and nematicides were withdrawn from the market. Consequently, cost-effective and environmentally friendly nematode strategies are needed to address the pest problem. The main aim of the study was to evaluate extracts of *Cassia abbreviata* (Sjambok pod), *Gomphocarpus tomentosus, Lippia javanica, Maerua angolensis* (Bead-bean), *Merwilla plumbea, Pappea capensis, Senegalia mellifera* and *Senna italica* for nematicidal effects on second-stage juveniles (J2) mortality of the root-knot nematode (Meloidogyne species) after 24, 48 and 72 h exposure. Each trial consisted of four extraction methods for treatment extracts, viz. dried crude extract, dried dichloromethane extract, dried water extract, untreated control and 1.8 mg/ml salicylic acid as standard positive control. Each extract was prepared at concentrations from 0.1-1 mg mL^{-1}, arranged in a randomized complete block design with four replications. Each trial was repeated trice. After 72 h exposure, the highest J2 mortality was observed in dried dichloromethane (75-88%) and dried water extract (70-81%) of M. angolensis which was comparable to salicylic acid and differed significantly from untreated control. This was followed by dried crude extract (68-69%) and dried dichloromethane (58-78%) of *C. abbreviata* and dried crude extract (67-69%) and dried dichloromethane (60-63%) of *G. tomentosus* which gave similar results and were significantly different from untreated control. The dried crude extracts from *M. plumbea*, L. javanica and *S. italica* were the only effective extracts with 61-67%, 56-66% and 58-61% mortality, respectively. Results indicate that these plants can potentially serve as alternatives to synthetic nematicides due to their potent nematicidal activity on plant-parasitic nematodes.

Keywords: Concentration - Extracts - Mortality - Root-knot nematode.

References:

- [1] Jones et al., 2013.Mol Plant Pathol 14: 946–961.

Transcriptomic and microscopic evaluations of fungal antagonism of soybean cyst nematode (H*eterodera glycines*) eggs.

Dong-gyu Kim[1] (kimx4617@umn.edu), Deepak Haarith[1], Kathryn Bushley[2]

[1] *Department of Plant Pathology, University of Minnesota - Twin Cities, St. Paul, MN, United States*; [2] *Dept. of Plant and Microbial Biology, University of Minnesota - Twin Cities, St. Paul, MN, United States*

Diverse soil fungi can antagonize the soybean cyst nematode (SCN; *Heterodera glycines*) at various stages of its life cycle through direct parasitism, the secretion of nematostatic and/or nematotoxic secondary metabolites, or a combination of both. Fungal biocontrol candidates offer an alternative to current environmentally damaging or economically unfavorable SCN management practices. Furthermore, fungal secondary metabolites are a potential source of highly specific nematicides. Characterizing the dynamics and mechanisms of fungal antagonism towards SCN eggs may help formulate effective fungal biocontrol agents or biopesticides against this sedentary nematode target. Here we characterize the dynamics and mechanisms of fungal antagonism towards SCN eggs, as well as investigate the nematodes' response to fungal antagonism. The fungi used in this study were the biocontrol egg-parasitic fungus *Pochonia chlamydosporia* isolate 123 (ATCC® MYA-4875TM), and a field isolate demonstrating high *in vitro* nematotoxicity. Axenic SCN eggs were treated with antibiotics to procure clean SCN eggs. Bioassays were conducted by placing clean SCN eggs on fungal colonies, and inoculating SCN eggs or second-stage juveniles in filter-sterilized fungal liquid culture filtrates. Antagonized SCN eggs were harvested at several predetermined time points after inoculation for RNA extraction, and labeled with the non-specific cellulose and chitin stain Calcofluor White M2R and propidium iodide to stain nematode cells undergoing cell death. Our results demonstrate distinct modes of SCN egg antagonism by fungal isolates and identify structural features of successful fungal antagonism through microscopy techniques. We also conducted transcriptomics to elucidate mechanisms of fungal infection and identify how the nematode may detect, evade, or mount an immune response against the fungus. This research will further our understanding of fungus-nematode interactions and has potential to identify putative molecular targets to control this soybean pathogen.

Thaumatin-like protein as a virulence factor of the pine wood nematode, *Bursaphelenchus xylophilus.*

Haru Kirino[1] (harukirino74@gmail.com), Kohki Yoshimoto[1], Ken-ichi Konagaya[2], Ryoji Shinya[1]

[1] *Meiji university, Kawasaki-shi, Japan*; [2] *Forestry and Forest Products Research Institute, Hitachi-shi, Japan*

The causal pathogen of the pine wilt disease is the pine wood nematode (PWN), *Bursaphelenchus xylophilus*. Recently, a highly sensitive proteome analysis identified numerous secreted proteins of PWN. However, the roles of these proteins during the onset of parasitism have not yet been elucidated. In this study, we used a leaf-disk assay based on transient overexpression in *Nicotiana benthamiana* to allow functional screening of 14 candidate pathogenic proteins secreted by PWNs. We demonstrated that five molecules induced statistically significant cell death in tobacco plants. Subsequently, we established a novel functional analysis method for susceptible black pine (*Pinus thunbergii*) seed embryos using transient overexpression by the *Apple latent spherical virus* (ALSV) vector, and investigated five secreted proteins of *B. xylophilus* causing cell death in tobacco to determine whether they induce hypersensitive responses in pine. We found that three of five molecules induced significantly higher expression in pathogenesis-related (PR) genes ($P < 0.05$), indicating hypersensitive response in pine seed embryos compared with mock and green fluorescence protein controls. Among them, Bx-TH2 encoding thaumatin-like protein specifically induced high expression of PR genes at all time points. Our study suggested that the Bx-TH2 likely make important contributions to inducing hypersensitive responses in host pine tree and is the virulence factor of PWN.

Keywords: Pine wilt disease - Exogeneous gene expression - Hypersensitive responses - Functional analysis - Pathogenic proteins.

References:

- Kirino et al., 2020. Plos one. 15(10): e0241613.
- Shinya et al., 2021. Front. Plant Sci. 12:640459.

Host range and geographical distribution of *Sphaerularia vespae*, the nematode parasite of queen hornets.

Hajime Kosaka[1] (hkosaka@ffpri.affrc.go.jp), Katsuhiko Sayama[2], Natsumi Kanzaki[3], Yoshihiro Takahata[2], Shun'ichi Makino[1]

[1] *FFPRI, Tsukuba, Ibaraki, Japan*; [2] *Kyushu, FFPRI, Kumamoto, Japan*; [3] *Kansai, FFPRI, Fushimi, Kyoto, Japan*

A parasitic nematode, *Sphaerularia vespae* was first found from the hornet *Vespa simillima* in Japan [1,2]. This nematode causes parasitic sterilization [1] and manipulates host behavior for the infection of the next generation [3]. The life cycles of the hornet and the nematode are as follows. Unparasitized queens after hibernation found their colonies in spring. Workers produced in the colonies maintain their colonies until late summer, then males and new queens (gynes) are produced. Only new queens hibernate after mating. Nematodes infect hibernating queens and lay eggs in the queens after hibernation. Parasitized queens cannot found colonies since they are sterilized by the parasites. These parasitized queens frequently visit potential hibernating sites such as rotten trees and deposit juvenile nematodes hatched in their body for next infection. To learn its host range and geographical distribution, various hornets were collected from several places in Japan. *S. vespae* was found thriving and laying eggs in the body of *V. simillima*, *V. mandarinia*, *V. dybowskii*, *V. crabro* and *V. ducalis* among the 6 hornets species (*Vespa* spp.) native to Japan. The nematode was found only from queens during or after hibernation, not from workers or males. For the other native hornet, *V. analis*, no post-hibernation queens had the nematode though several hibernating queens had poorly developed nematodes. It is thus revealed that effective hosts of *S. vespae* are queens of the 5 native hornet species other than *V. analis*. Multi-year collections conducted in the northern, central and southern parts of Japan indicate that *S. vespae* utilizes locally dominant hornet species as main hosts in the respective regions. It is considered that *S. vespae* potentially inhabits all over the Japanese mainland as far as the host hornets occur.

Keywords: Ecology - Host specificity - Host use - Social wasps - Sterilization.

References:

- [1] Sayama et al. 2007. Insect Soc. 54: 53-55.
- [2] Kanzaki et al. 2007. Zool Sci. 24: 1134-1142.
- [3] Sayama et al. 2013. Insect Soc. 60: 383-388.

Identification and characterization of Nematode chemosensory GPCRs (NemChRs) in *Heterorhabditis bacteriophora.*

Artha Kundu[1] (arthakundu18@gmail.com), Gitanjali Tandon[2], Nisha Jaiswal[3], Uma Rao[1], Mir Asif Iquebal[2], Vishal Singh Somvanshi[1]

[1] *Nematology, ICAR-Indian Agricultural Research Institute, New Delhi, Delhi, India*; [2] *Centre for Agricultural Bioinformatics, ICAR-Indian Agricultural Statistics Research Institute, New Delhi, Delhi, India*; [3] *Nematology, ICAR-Indian Agricultural Statistics Research Institute, New Delhi, Delhi, India*

The infective juveniles (IJs) of entomopathogenic nematode (EPN) *Heterorhabditis bacteriophora* find and infect their host insects in a heterogeneous soil ecosystem. Chemoreception (esp. olfaction) plays a major role in host finding, as the IJs locate insects by sensing a universal host cue (CO2) or insect/plant-derived odorants, which bind to various sensory receptors, including G-protein-coupled receptors (GPCRs). GPCRs are the largest family and most diverse group of membrane receptors in eukaryotes, including EPNs. NemChRs bind to a diverse set of ligands, including odor molecules. However, there is a lack of information on the GPCRs involved in odor- and chemo-reception in EPNs. Here we investigated GPCRs in the *H. bacteriophora* genome [1]. Primarily, a length-based filter was employed, and only 7613 proteins with >200 amino acid residues were retained. Of these, 69 sequences were found to contain 7-TM domains based on four different transmembrane detectors. Ultimately, we fetched 21 GPCRs out of these 7-TM sequences using various GPCR predictors. The GPCR detection pipeline was validated by reciprocal BLAST, Interproscan, GPCR-CA, NCBI CDD search etc. A Pfam search was made to classify these predicted GPCRs based on their functions, where four chemosensory GPCRs were identified. In addition, GPCRs were classified into various families based on the reciprocal BLAST approach into a frizzled type, a secretin type and 19 rhodopsin types GPCRs. The predicted Local Distance Difference Test (plDDT) scores for 3-D models of all the GPCRs generated by the Alphafold structure prediction server were >90, signifying high accuracy. Models were refined by the GalaxyRefine2 server, and molecular dynamics simulation analyses were done at 100ns in GROMACS using fully hydrated POPC bilayer. We are conducting molecular docking analyses of these GPCRs with selected attractant and repellent molecules in addition to profiling their expression. This knowledge will find applications for developing novel insect-pest management strategies by tweaking EPN IJ behaviour and will also be applicable to animal- and plant-parasitic nematodes to discover novel drug targets for their management.

Keywords: Entomopathogenic nematodes - *Heterorhabditis bacteriophora* - G-protein-coupled receptors (GPCRs) - Chemoreception.

References:

- [1] Bai et al., 2013. Plos One. 77(1):49-58.

Comparative genomics of free-living nematodes.

Yi-Chien Lee[1, 2] (ycl1220@smail.nchu.edu.tw), Hsin-Han Lee[1], Huei-Mien Ke[1], Wei-An Liu[1], Isheng Jason Tsai[1]

[1] *Biodiversity Research Center, Academia Sinica, Taipei, Taiwan*; [2] *Biodiversity Program,Taiwan International Graduate Program, National Taiwan Normal University, Taipei, Taiwan*

Phylogenetics is the inference of evolutionary relationships between species. With advances in sequencing, phylogenomics or inferring species phylogenies using thousands of genes from multiple genomes is considered to be advantageous over conventional approach of only using one or a few marker genes[1]. In nematodes, other than the model *Caenorhabditis elegans*, most species with genome sequence available are parasites because of their impacts on animal health and agricultural productivity. In contrast, genome resources of free-living nematodes are limited, especially nematodes in basal lineages[2]. Resolving their evolutionary relationship as well the genes they contain in the genomes of these most abundant animals on earth provide an opportunity to understand their origin, genomic bases of their diversity and evolutionary histories such as transition of parasitism[3,4]. Hence, in our research, we have focused on trying to isolate and sequence genomes and transcriptomes of free-living nematodes from Dorylaimia, Enoplia and early-branching species in Chromadoria. I will present our efforts in an attempt to alleviate the challenges of culturing these nematodes by sequencing the genomes and transcriptomes using amplification techniques. By comparing the genome and the transcriptome of the free-living nematodes, we aim to gain insight into the early evolution of the nematodes.

Keywords: Free-living nematode - Phylogenomics - Comparative genomics.

References:

- Yokono, M., Satoh, S., & Tanaka, A. 2018. Comparative analyses of whole-genome protein sequences from multiple organisms. Sci Rep, 8(1),1-10
- Bik, H.M., Lambshead, P.J., Thomas, W.K. and Lunt, D.H. 2010. Moving towards a complete molecular framework of the Nematoda: a focus on the Enoplida and early-branching clades. BMC Evol Biol, 10, 353.
- Viney, M. 2017. How Can We Understand the Genomic Basis of Nematode Parasitism?. Trends Parasitol, 33(6), 444-452.
- De Ley, P. 2006. A quick tour of nematode diversity and the backbone of nematode phylogeny. WormBook, 1-8.

Evaluation of galling index on previous crop as a reliable method for correct positioning of nematode control trials (*Meloidogyne* genus).

Salvatore Leocata (sleocata@studiotecnicoasa.it)

Studio Tecnico ASA, Catania, Sicily, Italy

A working methodology, based on root galling index assessment was evaluated in 91 experimental trials during 2011-2018. It was based on the assessment of Galling Severity Index (GSI) (Zeck scale, modified 0-10) on the previous crops, to ascertain evenness of distribution and infestation severity of the root-knot nematodes, *Meloidogyne* spp.. Trials carried out according to the EPPO guidelines, with randomized experimental blocks design with 4-5 replications. GSI was detected on a large number of plants, at the end of the cultivation cycle, to select the greenhouses in which to operate and a map representing the relative distribution was drawn. The percentage of infected roots and of related GSI were then calculated in the greenhouse, in which a trial with nematicides would be later positioned and these values were later compared with the same parameters detected at the end of the crop cycle in the untreated plots of the test crop. In 94% of cases (82 trials) a frequency of 90-100% infected roots on the previous crop confirmed on untreated plots of the trial a frequency of 90-100% of symptomatic roots. With regard to GSI an average value in the previous crop equal to or higher than 5 (49 trials) confirmed in 76% of cases, average values higher than 5 in the check plots and in 90% of cases average values of the GSI higher than 4. In 66 trials, in which a percentage of infected roots of 90-100% had been detected with an average GSI equal to or higher than 4 on the previous crop, in 88% of the cases 90-100% of infected roots with an average GSI equal to or higher than 4 was found on the untreated plots of the test crop. Higher rates of GSI were clearly found during the summer cycle compared with the autumn-winter cycle. No direct relationship was found between number of J2 stages counted before transplanting and GSI assessed at trial end on UTC plots. For the summer cycle, 1÷25, 51÷105/106÷600 J2 larvae on 100 cc of soil caused 100% of GSI > 4 and >5 respectively on UTC plots while for autumn cycle 1÷25 and 51÷105 J2 caused respectively 71,4% of GSI > 5 and 85,7% of GSI > 4. It was observed that if the test crop belongs to a different family compared with the previous crop a reduction of GSI (even severe) could occur on the test crop, and mainly during the autumnal cycle. Correlations with the length of crop cycle and the soil texture were also observed. With very low uncertainty, the adopted methodology showed a very high reliability.

Keywords: Root-knot nematodes - Protected crops - Nematicide efficacy trials.

References:

- Bridge J. & S. L. J. Page (1980). Estimation of Root-knot Nematode Infestation Levels on Roots Using a Rating Chart, Tropical Pest Management, 26:3, 296-298,
- Colombo, A. (2002). Le problematiche nematologiche delle colture ortive in Sicilia. Nematologia Mediterranea (Suppl.) 30: 17-20.
- S. Leocata, G.Pirruccio, A.Myrta, E.Medico, N.Greco (2014). Dimethyl disulfide (DMDS): A new soil fumigant to control root-knot nematodes, Meloidogyne spp., in protected crops in Sicily, Italy. Proceedings of the VIIIth IS on Chemical and Non-Chemical Soil and Substrate Disinfestation. Acta Horticolturae, 1044, 415–420.
- Caroline Djian-Caporalino et al. (2011) .The reproductive potential of the root-knot nematode Meloidogyne incognita is affected by selection for virulence against major resistance genes from tomato and pepper. Eur J Plant Pathol (2011) 131:431–440
- Zeck W.M., (1971). A rating scheme for field evaluation of root-knot infestations. Pflanzenschutz Nachrichten Bayer AG, 24, 141-144.

Interactive effects of biochar type and rate on tomato plant growth and nematode suppression.

Morakene Letsoalo[1] (lambert.letsoalo@ump.ac.za), Dineo Hope Mcunu[1], Zakheleni Palane Dube[2], Mbokota Candy Khosa[3]

[1] *School of Agriculture, University of Mpumalaga, Mbombela, South Africa*; [2] *School of Biology, University of Mpumalaga, Mbombela, South Africa*; [3] *Tropical and Subtropical Crops, Agricultural Research Council, Mbombela, South Africa*

Root-knot nematodes pose a huge threat to vegetable production. The international withdrawal of synthetic chemical nematicides due to their negative environmental and human health impacts has left a void in nematode management. Biochar has been reported to be highly effective in promoting plant growth and controlling some plant diseases. Its use in managing root-knot nematodes has not been reported. The study was conducted to investigate the interactive effect of two biochar types at four rates on Meloidogyne incognita and growth of tomato plants. A 2 x 4 factorial experiment comprising poultry and acacia biochar as first factor and biochar rates of 0, 5, 10 and 20 t/ha as second factor. There was a significant ($P \leq 0.05$) interaction of biochar type x biochar rates on root gall index and root mass, but the interaction was not significant for plant height, stem diameter and shoot mass of tomato plants. Biochar type as main factor had a significant effect on stem diameter and root mass, whereas, biochar rate as main factor had significant effects on root mass and shoot mass. Poultry biochar amendment increased plant growth variables at all application rates with the highest increase observed at 10 and 20t/ha, whereas, acacia biochar increased plant growth variables only at 20t/ha. Under poultry biochar amendments plant nematode infections were not affected, whereas, under acacia biochar a significant nematode increase was observed. In conclusion, the response of tomato plants infested with nematode differ based on type of biochar and rate at which the biochar is applied.

Keywords: Acacia biochar - Botanicals - *Meloidogyne incognita* - Phytonematicides - Poultry biochar.

Present situation of pine wood nematode *Bursaphelenchus xylophilus* in China.

Hongmei Li[1] (lihm@njau.edu.cn), Xue Qing[1], Xinrong Wang[2], Jianfeng Gu[3]

[1] *Department of Plant Pathology, Nanjing Agricultural University, Nanjing, Jiangsu, China*; [2] *College of Plant Protection, South China Agricultural University, Guangzhou, Guangdong, China*; [3] *Technical Centre, Ningbo Customs, Ningbo, Zhejiang, China*

Bursaphelenchus xylophilus, is the causal agent of pine wilt disease and is considered as destructive quarantine pest, producing severe environmental and economic losses worldwide. In China, PWN was first detected in 1982 on *Pinus thunbergii* at the Purple Mountain of Nanjing. Since the first detection PWN remains problematic, as PWN is constantly spreading. In 2020, The National Forestry and Grassland Administration of China reported 667 districts and counties from 18 provinces as pine wilt disease epidemic areas. Despite several governmental activities aimed to controlling PWN and vector beetles *Monochamus* spp., the pine wilt disease is spreading gradually into other regions of the country. Statistical studies based on the existing governmental data have revealed that PWN damage led to a remarkable ecological crisis in forest areas affected by pine wilt disease. On PWN diseased areas, management programs are conducted and aimed to reduce sources of nematode and beetle dispersion. The plant quarantine measures for prevention of spreading PWN remains the basis for management programs. The Chinese quarantine authorities followed the specifications indicated at the International Standard Phytosanitary Measures (ISPM-15) to prevent the introduction of PWN into the country *via* international trades. Regularly inspections on imported logs and wood packaging materials are performed. So far quarantine authorities have recorded more than 2000 interceptions where PWN was detected. In forests where pine wilt disease are identified, cleaning up infected pines are performed. To prevent transmission of PWN through beetles' maturation feeding on healthy trees, traps with insect attractants, and spraying aerial insecticides are used. To suppress nematode infection, nematicides are injected into the pine trunk. In order to decrease environmental effects of the chemicals used in the management programs, several national projects are in progress as breeding programs for *Pinus* with resistance to pine wilt disease, and the use of biological control options for nematodes and their vectors. An appraised review on the occurrence of PWN in the country and the control measures that have been applied over the last decades, their success or limitations on pine wilt disease will be discussed.

Keywords: *Bursaphelenchus xylophilus* - China - Management - *Monochamus* spp. - Occurrence.

Proteomic profiling of *Steinernema carpocapsae* and *Heterorhabditis megidis* infective juveniles stored at 20°C and 9°C.

Peter Lillis[1] (peter.lillis@mu.ie), Christine Griffin[1], James Carolan[2]

[1] *Behavioural Ecology, Maynooth University, Maynooth, Co. Kildare, Ireland*; [2] *Applied Proteomics, Maynooth University, Maynooth, Co. Kildare, Ireland*

Many nematode species have a dauer or infective juvenile (IJ) stage in their life cycle. This non-feeding stage is long lived, stress resistant and seeks out fresh resources. Temperature can have profound effects on behaviour and lifespan of this stage, but the molecular basis for this is not well understood. We examined the somatic proteome of IJs of two distantly related species in response to storage temperature and time. *Steinernema carpocapsae* and *Heterorhabditis megidis* are both entomopathogenic nematodes whose IJs seek insect hosts. The somatic proteome of fresh IJs (week 0) was compared with that of IJs stored for up to 9 weeks at 9°C or up to 6 weeks 20°C. Proteins were identified using label-free, quantitative mass spectrometry based proteomics. Overall, twice as many proteins were detected in the IJs of S. carpocapsae in comparison to H. megidis (1290 versus 645). The two species differed dramatically in their response to storage. The proteome of *H. megidis* changed in response to both storage temperature and time, while that of *S. carpocapsae* changed largely in response to temperature, with less marked change over time. Both species also differed in the scale of change relative to time 0: the maximum change in abundance identified for any protein in H. megidis was 16-fold, while in S. carpocapsae several proteins increased hundred-fold. Notable changes in the *H. megidis* proteome were in proteins associated with autophagy, stress, and cell signalling, and multiple ribosomal proteins which were decreased in abundance over time. In contrast, ribosomal proteins were largely unchanged in *S. carpocapsae*, and the most differently abundant proteins in this species were chaperone proteins. These included LEA proteins, which are known to enhance IJ survival in response to stress, and which increased in abundance by up to 360 times at 9°C, but showed only minor changes at 20°C. Proteins associated with stress, and DNA and mRNA binding proteins, decreased significantly (~64 relative fold change) in both temperatures relative to week 0. Taken together, our findings indicate that the proteomes of these two species respond differently to temperature, and that species-specific stress responses may exist. The results of our study provide insight into the molecular effects of temperature on nematode dauers/IJs and may lead to the development of methods that may enhance their performance as biocontrol agents.

Keywords: Entomopathogenic - Nematodes - Proteomics - Infective Juveniles - Temperature.

Identification of WRKY transcription factors responsive to root-knot nematode in tomato roots by RNAseq analysis.

Xiuhong Lu (447597587@qq.com), Jinling Huang, Yu Zhang, Zhiming Liu

Institute of Plant Protection, Guangxi Academy of Agricultural Sciences, Nanning, China

WRKY transcription factors (WRKYs) are one of the largest families of transcriptional regulators in plants. They have diverse biological functions in plant disease resistance, abiotic stress responses, nutrient deprivation, senescence, seed and trichome development, embryogenesis, as well as additional development and hormone-controlled processes[1]. The tomato genome contains 81 WRKYs[2]. Root-knot nematodes (RKNs,*Meloidogyne incognita*) are the most damaging plant parasites causing severe losses to tomato. Recent studies indicate WRKYs are involved in plant responses to RKNs[3]. Therefore RNAseq analysis technology was used for Genome-wide identification of WRKYs responsive to root-knot nematode in tomato. In this study, we elaborated a comprehensive RNAseq analysis of resistant tomato roots at 6, 12, 24 and 48h after inoculation with the RKN *M.incognita*, a total of 12 WRKYs changed significantly, which differentially expressed were validated by qRT-PCR.After 6 hours, 3 (2 up-regulated, 1down-regulated) WRKYs changed significantly. After 12 hours, 2 WRKYs up-regulated. After 24 hours, 3 WRKYs down-regulated. After 24 hours,10 (8 up-regulated,2 down-regulated) WRKYs were differentially expressed. Genome-wide WRKY expression analysis of tomato by RNAseq analysis helped our understanding of WRKY function in response to RKNs in tomato. A set of differentially expressed WRKYs could pave the way for developing novel treatment and control strategies.

Keywords: WRKY transcription factors - Root-knot nematodes - Responsive.

References:

- [1]Madhunita Bakshi et al.,Plant Signaling & Behavior,9.e27700:1-18.
- [2]Shengxiong Huang et al.,Molecular Genetics and Genomics,287(6):495-513.
- [3]Bharathiraja Chinnapandi et al.,Plant Signaling□Behavior,12e1356530.

Transcriptional changes of *Meloidogyne luci* second-stage juveniles after exposure to 1,4-naphthoquinone.

Joana Cardoso[1] (joana.cardoso@uc.pt), Ivânia Esteves[1], Conceição Egas[2, 3], Mara E.M. Braga[4], Hermínio C. de Sousa[4], Isabel Abrantes[1], Carla Maleita[1, 4]

[1] *Centre for Functional Ecology – Science for People & the Planet, Department of Life Sciences, University of Coimbra, Coimbra, Portugal*; [2] *Center for Neuroscience and Cell Biology, University of Coimbra, Coimbra, Portugal*; [3] *Biocant – Transfer Technology Association, BiocantPark, Cantanhede, Portugal*; [4] *Chemical Process Engineering and Forest Products Research Centre, Department of Chemical Engineering, University of Coimbra, Coimbra, Portugal*

The root-knot nematode *Meloidogyne luci* was added to the European Plant Protection Organization Alert list in 2017 as it represents a threat to the production of several important crops in several parts of the world. The scarce availability of efficient nematicides to control this plant-parasitic nematode and the phasing out of nematicides from the market has intensified the search for alternatives, such as phytochemicals with bionematicidal properties. The 1,4-naphthoquinone (1,4-NTQ), which can be extracted from walnut processing residues, displayed strong nematicidal activity against *M. luci*, affecting mortality, hatching, penetration and reproduction. However, knowledge on potential mode(s) of action of 1,4-NTQ is still scarce. Preliminary studies have been carried out through the assessment of reactive oxygen species generation, but more studies focusing the biological mechanisms of action of these bioactive compounds in RKN are needed. In this study, *M. luci* second-stage juvenile (J2) transcriptome profile after exposure to 1,4-NTQ was achieved to identify genes and pathways that might be involved in its mode of action. *Meloidogyne luci* J2 were incubated at 22°C for 3 days to 1,4-NTQ at 20 ppm. Nematodes incubated in water and Tween® 80 (1,4-NTQ solvent) were used as controls. After exposure, J2 were washed and stored at -80°C, until RNA extraction. Total RNA was extracted from treated and control samples and library construction and sequencing were performed at Genoinseq, Biocant (Portugal), using the Illumina NextSeq platform. *De novo*transcriptome assembly of *M. luci* was obtained and transcript abundance determined. A large set of differentially expressed genes (DEGs) were found among the three conditions, reflecting the antagonist effect of 1,4-NTQ on *M. luci*. Gene annotation of DEGs is being conducted to identify the nematode gene networks and metabolic pathways affected by this promising bionematicide.

Funding: FCT/FEDER projects UIDB/00102/2021, PTDC/ASP-PLA/29392/2017, CENTRO-01-0145-FEDER-000007, CEECIND/02082/2017 and Instituto Ambiente Tecnologia e Vida.

Keywords: Bionematicide - Management – Root-knot nematodes - Transcriptome.

Demographic history and co-evolution study of nematodes parasitizing millipedes of the Western Caucasus.

Svetlana Malysheva (malysheva24@gmail.com), Boris Efeykin, Anastasia Teterina

Centre of Parasitology SIEE RAS, Moscow, Russia

Nematodes Severianoia pachyiuli n.sp. (Oxyuridomorpha: Thelastomatoidea) inhabit the hind gut of millipedes Pachyiulus krivolutskyi Golovatch, 1977 (Diplopoda: Julidae) which is an endemic species in the Western Caucasus. All studied millipedes are infected with a single species that is common for thelastomatids (Sinnott et. al., 2015), which in turn makes them attractive in the light of population study. Specimens of millipedes were collected from nine localities situated on the Southern and Northern slopes of the Caucasus Range. One point in Abkhazia near Lake Ritz, three collection points near the village of Nickel (i.e. Golden streams, river Suk and river Gruzinki), Mamedova crack in the Big Sochi, villages Loo, Merculaevka, Bezymyanni and Jelobnaya. The average size of millipede individuals varied within 5–9 cm; the average number of collected individuals at a separate collection point was 15–20 individuals. DNA sequences were obtained for two markers for millipedes (COI and D2D3 segment 28S rDNA) and for three markers for nematodes (COI, EF1 and D2D3 segment 28S rDNA). Millipede sequencing results have shown that the COI gene in this species is very conservative; sequenses vary in 3-5 nuclotides for 550 bp alignment. Thus, populations located on the eastern side of the Greater Caucasus Mountain Range show almost no differences; differences between eastern and western side populations are insignificant. According to the D2D3 fragment, the differences are more pronounced; on the diagrams and phylogenetic trees, one can see that there are even differences between the populations of millipedes located in neighboring but isolated gorges. The nematode COI gene has proven to be more variable and suitable for intraspecific phylogenetic studies. The average number of nucleotide differences varies for 35-37 nucleotides (alignment length 550 bp). Using the BEAST software package, we performed a median estimate of the effective population size based on the obtained sequences. For millipedes, Pachyiulus krivolutskii it is 6841 specimens, for nematodes Severianoia pachyiuli n.sp. - 8342 specimens. The number is stable for about 3,000 and 3,500 generations, respectively. Current analysis was performed on the basis of one mitochondrial marker. The analysis of two markers (COI and 28S) confirms the hypothesis of co-evolution of nematodes and millipedes (Support: RFBR 18-04-00256-a)

Keywords: Nematoda - Oxyuridomorpha - phylogeny - population - Severianoia.

References:

- Sinnott et. al., 2015. J. Parasitol. 101(4), 445-457.

The use of soil biological indicators to improve soil health.

Tvisha Martin (martin.3325@buckeyemail.osu.edu)

Environment and Natural Resources, The Ohio State University, Wooster, Ohio, United States

U.S. Mid-western agriculture has led to hypoxia in the Gulf of Mexico and directly contributes to algal blooms in Lake Erie, two key bodies of water in North America. Improving soil health could help agriculture increase ecosystem services through carbon stabilization and nutrient retention, while optimizing yield (Anderson, 2017). However, the biological mechanisms that underpin soil health are largely unknown. It is well known that nematode communities, enzyme activity, and microbial communities serve as indicators of soil food web structure (Acosta-Martinez et al.,2003; Banerjee et al.,2019; Ferris et al., 2001). What is less well known is how soil food webs can serve as an early indicator of soil health and influence belowground C dynamics, and root turnover. This study takes place on two identical long-term trials in northern and southern Ohio with three replicates of a no-tillage and conventional tillage system and planting consisting of a corn-soybean rotation and continuous corn. This study compares soil food webs in a shrink-swell clay and a silty loam. Nematodes were filtered via elutriation, and identified to family, microbial communities were determined using metabarcoding, fine root production was sieved to 2mm, washed, dried, weighed, and analyzed for C:N, and enzyme activities were analyzed using a microplate reader, soil health indicators include tests that reflect the labile soil C and N pool. Nematode indices showed increased nematode structure, and channel indices in sustainable agriculture, while the basal index was higher under conventional management. This demonstrates that soil food webs are better maintained in systems with greater biological N fixation and lower disturbance.

Keywords: Free-living nematodes - No-till - Rotational diversity - Soil health.

References:

- Acosta-Martinez, V., Zobeck, T.M., Gill, T.E., Kennedy, A.C., 2003. Enzyme activities and microbial community structure in semiarid agricultural soils. Biol. Fertil. Soils 38, 216–227.
- Anderson, R.L., 2017. Improving resource-use-efficiency with no-till and crop diversity. Renew. Agric. Food Syst. Camb. 32, 105–108
- Banerjee, S., Walder, F., Büchi, L., Meyer, M., Held, A.Y., Gattinger, A., Keller, T., Charles, R., van der Heijden, M.G.A., 2019. Agricultural intensification reduces microbial network complexity and the abundance of keystone taxa in roots. ISME J. 13, 1722–1736
- Ferris, H., Bongers, T., de Goede, R.G.M., 2001. A framework for soil food web diagnostics: extension of the nematode faunal analysis concept. Appl. Soil Ecol. 18, 13–29

Responses of total protein in *Meloidogyne incognita* juveniles to Nemafric-BL phytonematicide.

PW Mashela[1] (phatu.mashela@ul.ac.za), Ebrahim Shokoohi[2], KM Pofu[2]

[1] *University Of Limpopo, Polokwane, Limpopo, South Africa*; [2] *University Of Limpopo, Polokwane, South Africa*

After hatch, second-stage juveniles (J2) of *Meloidogyne* species nematodes could spend at least 12 weeks in soil searching for penetration sites of suitable host plants. The cuticle of nematodes comprises distinct sublayers containing lipids, glycoproteins, collagens (soluble proteins) and cuticulins (insoluble proteins). Generally, cucurbitacins are lipophilic, but there is scant information on how these complex tetrapernoids relate to cuticular proteins in nematodes. A study was conducted to investigate the nature and extent of damage post-exposure of J2 to a wide range of Nemafric-BL phytonematicide concentrations. *Meloidogyne incognita* J2 were prepared and exposed to seven concentrations of Nemafric-BL phytonematicides, with each concentration replicated three times. After dilution in distilled water, approximately 1000 J2 in distilled water were weighed into a tin foil cup and then placed into an automated sample loader of TruSpec CHNS Macro (Leco, St. Joseph, MI, USA) instrument. The final results were calculated from a calibration curve plotted using ethylenediaminetetraacetic acid (EDTA) as the nitrogen calibration standard. Total protein (%) versus Nemafric-BL phytonematicide exhibited negative quadratic relations, with the model explained by 97% association. The minimum total protein was reached at 4.82% phytonematicide concentration. In conclusion, the nature and extent of damage suggested that Nemafric-BL phytonematicide was highly nematicidal as opposed to being nematostatic, thereby explaining its potent suppressive effects on nematode population densities.

Keywords: Biocontrol - Phytonematicide - Protein - Root-knot.

Fast-tracking the development of efficacy and toxicity data for registration of phytonematicides.

PW Mashela[1] (phatu.mashela@ul.ac.za), Ebrahim Shokoohi[2], KM Pofu[2]

[1] *University Of Limpopo, Polokwane, Limpopo, South Africa*; [2] *University Of Limpopo, Polokwane, South Africa*

The main challenge for innovators of phytonematicides is that the registration authorities hardly prescribe procedures for generating pre-registration data. However, the authorities insist that the submitted data for registration should have the attributes of being “fit-for-purpose” with “scientific merit”. Basically, the concept “fit-for-purpose” describes claims by innovators and encompasses efficacy against the target entity and avoiding toxicity to the protected host. Technically, in registration linguistic, “scientific methods” relate to the un-biasness of the protocols used to generate the “fit-for-purpose” data. In this study, we build on the Curve-fitting Allelochemical Response Dose (CARD) model for closing existing gaps in the development of data with «scientific merit” for registration of phytonematicides.

Keywords: Nemafric-BL - Nemarioc-AL - Phytonematicide.

Gene copy number expansions and contractions in the genomes of plant-parasitic nematodes.

Rick Masonbrink[1] (remkv6@iastate.edu), Thomas Maier[2], Parijat Juvale[2], Melissa Mitchum[3], Andrew Severin[1], Thomas Baum[2]

[1] *Genome Informatics Facility, Iowa State University, Ames, IA, United States*; [2] *Plant Pathology, Iowa State University, Ames, IA, United States*; [3] *Plant Pathology, University of Georgia, Athens, GA, United States*

Parasites survive and reproduce by harvesting nutrients from their hosts, thereby exerting a toll on host fecundity and prompting host selection. Eventually, the host develops resistance, and these same selective forces are mirrored in the parasite until resistance is overcome. These ongoing cycles of reciprocal adaptation between host and parasite have a genomic signature in the form of gene duplications, which are frequently concurrent with these evolutionary outcomes. We studied this phenomenon via 11 publicly available genomes in the Tylenchomorpha, a clade of economically damaging plant-parasitic nematodes. Using gene orthology and phylogenetics we discerned gene expansions from three phylogenetic nodes: i) the Tylenchomorpha, a basal node segregating migratory plant-parasitic nematodes from the sedentary root-knot and cyst nematodes; ii) the Tylenchoidea, root-knot and cyst nematodes, which have different modes of infection with similar consequences; and iii) the Heteroderidae, the divergence of cyst nematode species into *Heterodera* and *Globodera* genera. We then advanced the understanding of secretory gene evolution of *Heterodera glycines* by leveraging published expression datasets to characterize preparasitic and parasitic stages with differing aspects of host compatibility. Altogether, using a particular emphasis on genes with conserved secretion signals in multiple species, we examined the expansion of gene families across these clades to discern conserved mechanisms of host cell manipulation and parasitism in *H. glycines*. Using these data, we provide a resource for understanding nematode parasitism and virulence.

Keywords: Expansion - Gene - Soybean - Cyst - Effector.

Patterns of changes of soil nematode communities in relation to biocenosis type and vegetation features.

Elizaveta Matveeva (elizaveta.matveeva@gmail.com), Anna Sushchuk, Daria Kalinkina

Institute of Biology, Karelian Research Centre of Russian Academy of Sciences, Petrozavodsk, Republic of Karelia, Russian Federation

Soil organisms are closely related to the composition and structure of plant communities. To reveal the patterns of formation of pedobiont communities in undisturbed biocenoses, soil nematode fauna was studied and the features of their communities were analysed at the dependence on biocenosis type and vegetation peculiarities. Tundra biocenoses, coniferous (pine and spruce) and deciduous (small- and broadleaf) forests and meadows were investigated in the North-West of Russia (61°-68°N). Nematode diversity, population density and eco-trophic community structure were studied. The lowest values of nematode numbers were found in tundra biocenoses and highest ones – in spruce forests. The distribution of soil nematode density at the regional scale, in contrast to the global scale [1], is characterized by lower abundance in tundra, similar values in broadleaf forest and much higher density in boreal forests. The nematode taxonomic diversity was also lowest in tundra, and increased in the series «pine forest – spruce forest – small-leaved forest – broadleaf deciduous forest». Analysis of nematode community structure showed that bacterial feeders dominated in soil of all types of biocenoses. Nematodes associated with plants were subdominant in the tundra, fungal feeders – in the coniferous forests. And both groups were subdominant in deciduous forests and meadows with similar values of relative abundance. Plant-parasitic nematodes were presented in lowest numbers in the community of tundra and coniferous forests. In deciduous forests the proportion of plant parasites increased and reached a maximum in the meadows. The values of ecological indices showed that the level of soil enrichment with organic matter (Enrichment index) and the pathway of its decomposition in the soil (Channel index) were connected with the vegetation type. Based on these parameters two groups of biocenoses were distinguished: in deciduous forests and meadows there were found high EI and low CI values, and in tundra and coniferous forests were observed the inverse ratio of indices. Structure index had high values in all biocenoses. It was established that the total nematode abundance, fauna diversity, relative abundance of plant parasites, EI and CI indices were the nematological parameters which closely related to the biocenosis type. Study was carried out under state order (FMEN-2022-0005).

Keywords: Soil nematodes - Vegetation - Distribution - Population density - Community structure.

References:

- [1] van den Hoogen et al., 2019. Nature. 572: 194-198.

Spatio-temporal changes of soil nematode communities in northern meadows.

Elizaveta Matveeva (elizaveta.matveeva@gmail.com), Anna Sushchuk, Daria Kalinkina, Maria Yurkevich, Valeria Sidorova

Institute of Biology, Karelian Research Centre of Russian Academy of Sciences, Petrozavodsk, Republic of Karelia, Russian Federation

Research on the dynamics of plant communities, soil cover and nematode fauna, as a numerous and diverse group of pedobionts in the North, provides information on the functioning of natural ecosystems and agrocenoses during their post-agrogenic restoration. We studied abundance, diversity and soil nematode community structure [1] of five meadow biocenoses (Republic of Karelia, Russia, 62°13'41.41»N; 34°2'17,09»E), differing in origin, vegetation [2], soil type, degree of moisture and land use history. Based on archival data there were estimated the changes in soil nematode communities during long (50 yrs) meadow succession. Modern plant communities were presented with dominated species: *Alopecurus pratensis* L. (site 1), *Dactylis glomerata* L. (site 2), *Carex* sp. (site 3), *Deschampsia cespitosa* L. (site 4), *Filipendula ulmaria* L. (site 5). Site 1 was subjected to agricultural use (arable land and hayfield), and site 5 had never been plowed and was primary meadow. It was established that differences in use intensity and plant composition determined the multidirectional changes: some of sites developed according to the meadow type, others gradually grew into forests. Analysis of nematological parameters revealed two groups of sites: in the first one (sites 1, 2) the abundance of bacterial feeders (Ba) increased, SI index decreased; in the second one (sites 3-5 that were forming under high soil moisture and longer post-agrogenic development) nematode communities possessed parameters characterised for stable undisturbed biocenoses; in their structure the total Ba abundance decreased, and, at the same time, the number of Ba1 increased that caused an increase in EI index. SI enlarged due to the enhanced K-strategist's contribution to the nematode fauna, which can be interpreted as an indicator of the "naturalization" of biocenoses: the further course of succession will possibly end with deciduous forest formation. At the present stage, the nematode community of site 5 is already very similar to forest biocenoses (bacterial-, fungal feeders and nematodes, associated with plants were numerous and plant parasites (Pp) were low in numbers); communities of sites 1, 2 have a structure typical of the meadows of South Karelia (Ba and Pp predominated). Study was carried out under state order (FMEN-2022-0005).

Keywords: Soil nematodes - Succession - Long-term changes - Community structure - Ecological indices.

References:

- [1] Yeates et al., 1993. J. of Nematology. 25(3): 315-331
- [2] Raabe, Brandes. 1988. Phytocoenologia. 16(2): 225-258

Potato plant responses to a combination of different abiotic and biotic stresses.

Elizaveta Matveeva (elizaveta.matveeva@gmail.com), Elena Sherudilo

Institute of Biology, Karelian Research Centre of Russian Academy of Sciences, Petrozavodsk, Republic of Karelia, Russian Federation

In the North plants are routinely subjected to a wide range of combination of abiotic (temperature fluctuation during day-cycle, long photoperiod or continuous lighting) and biotic (pathogens and pests) stresses. It is known that plants subjected simultaneously to several stresses exibit stress-specific adaptive responses as well as responses which protected plants from more than one environmental stress [1]. The aim of the study was to investigate potato plant responses to a daily temperature fluctuation and continuous lighting combined with infestation of potato cyst-forming nematode (PCN) *Globodera rostochiensis* Woll. Potato genotypes differing in freezing-tolerance derived from the wild species *Solanum commersonii* Dun. and potato cultivars *S. tuberosum,* resistant or susceptible to nematode, were grown at temperature of 23°C, photoperiod 16 h or continuous (24 h) light, and treated with a temperature drop from 23°C to 5°C for 2 h or constant temperature of 5°C during 6 days. After that plants were infested by PCN (10 cysts per plant) and grown under 23°C and 16 h or 24 h photoperiods for 1 month. During the experiment cold resistance of plants and infestation intensity by PCN (number of cysts on the roots) were estimated. It was shown that temperature fluctuation and continuous light increased cold resistance in all potato genotypes and cultivars. Infestation by PCN after each treatment led to an additional increment of plant cold resistance independently of potato genotypes and cultivars, which remained at high levels for three weeks. Combination of biotic and abiotic factors (infestation by PCN after temperature and light-treatment) also led to an increase in cold resistance of plants. Increased plant cold resistance was accompanied by increased resistance to PCN, depending on potato genotype and cultivar and treatment: freezing-susceptible genotypes and cultivars demonstrated higher resistance to the nematode infestation after temperature drop and continuous light. Possible mechanisms of plant cross-adaptation to biotic and abiotic stresses are to be discussed. Research was supported by the Russian Foundation for Basic Research (project N 20-016-00033).

Keywords: Cold resistance - *Globodera rostochiensis* - Low temperature treatment - Infestation intensity - Light-treatment.

References:

- [1] Chinnusamy et al., 2004. J. Exp. Bot. 55:225–236.

Potato cyst-forming nematode in open data network on invasive alien species between Finland and Russia: Citizen Science.

Elizaveta Matveeva (elizaveta.matveeva@gmail.com), Anna Sushchuk, Nadezhda Mikhailova

Karelian Research Centre of Russian Academy of Sciences, Petrozavodsk, Republic of Karelia, Russian Federation

In 2019-2021 the project "Collaborative Data and Information Exchange Network for Managing Invasive Alien Species" was conducted in Karelian Research Centre of RAS. Invasive alien species (IAS) are species whose introduction or spread outside their natural distribution threatens biological diversity by out-competing native species. They can also cause other impacts in their new environment: health problems, spreading diseases or parasites to humans and farm animals; the species can also be pests to cultivated crops or forests. The aim of this project was to present IAS' distribution, including plant-parasitic nematodes, on the Karelian Internet portal; to help local officials, researchers and citizens on both sides of the border to create joint risk assessments for most important species, communicate on future potential risk of species and create management guidelines. Among nematodes the closest attention was focused on potato-cyst nematode (PCN) *Globodera rostochiensis* Woll., a quarantine obiect. The nematode was described, its distribution in the Republic of Karelia during 45 years shown, with recommendations to the local people on the available methods for detection and regulation of the potato parasite. PCN was introduced by humans into the soil of agrocenoses with seed material or agricultural machinery. As a highly specialized parasite, PCN quickly multiplied in the agrocenoses and went from aggregated distribution in the field (foci) to very high population density, a process that can take only a few years. PCN establishment in agrocenoses may result in a change in the structure of soil nematode's communities, which in turn will affect the functioning of the soil ecosystem and soil fertility in general. At the framework of project, a series of events was arranged aimed to involve volunteers in the Citizen Science' project for closer cooperation with scientists, to increase awareness of local people with potato pests, including educational activities, preparation of information leaflets on the PCN (general information, parasite' detection methods, simple and available control measures) and carrying out best practices on the private fields. The project was supported by ENI CBC Karelia Programme (KA-5046).

Keywords: Potato cyst-forming nematode - Control measures - Best practices - Educational activities.

Search for soil amendments affecting potato cyst nematode: possibilities for parasite control.

Elizaveta Matveeva (elizaveta.matveeva@gmail.com), Maria Yurkevich, Valeria Sidorova, Inna Dubrovina

Institute of Biology, Karelian Research Centre, Russian Academy of Sciences, Petrozavodsk, Republic of Karelia, Russian Federation

The potato cyst nematode *Globodera rostochiensis* Woll., pathotype Ro1 (PCN) is one of the most serious pests of potato. Despite quarantine measures, the phytoparasite has spread progressively throughout the territory of the Russian Federation. The aim of this study was to estimate the effect on PCN populations of 3 soil amendments: 1) biochar (5% in two fractions), 2) schungite (in doses of 5 and 10%), and 3) seaweed (*Fucus vesiculosus* L. as dry powder and water extract). Experiments were conducted in the laboratory using 10 PCN cysts per plant of *Solanum tuberosum* L., cv. "Nevsky" and in field trials. Although biochar did not affect the number of cysts on the plant roots compared to numbers on untreated plants, it increased the percentage of dead eggs inside the newly formed cysts (23-33% versus 13%). By contrast, shungite added to soil decreased the number of new cysts by 23–25% while having no effect on the viability of the eggs and larvae inside cysts compared to those of controls. Addition of *F. vesiculosus* to soil inhibited nematode development due to adverse effects of the material on the plants. The results indicate the potential for using soil amendments to control PCN populations, either by direct chemical effects or indirectly by their influence on plants or on soil properties. Additional research is merited. This study was supported by state order (№0218-2019-0079, 0218-2019-0075).

Keywords: Potato cyst-forming nematode - Ameliorants - Population reproduction - Control measures.

Response of soil nematodes to anthropic gradient in the arganeraie biosphere.

El Hassan Mayad[1] (e.mayad@uiz.ac.ma), Amine Idhmida[2], Amina Braimi[2], Ilyas Fillali Alaoui[2], Mohamed Ait Hamza[2], Thierry Mateille[3], James N. Furze[4], Zahra Ferji[5], Abdelhamid El Mousadik[6]

[1] *Laboratory of Biotechnologie and Valorization of Natural Ressources, Faculty of Sciences, Ibn Zohr Univer, Agadir, Souss Massa, Morocco*; [2] *Laboratory of Biotechnologie and Valorization of Natural Ressources, Faculty of Sciences, Ibn Zohr University, Agadir, Souss Massa, Morocco*; [3] *UMR- Centre de Biologie pour la Gestion des Populations, IRD, Montpellier, France*; [4] *Royal Geographical Society, Londre, United Kingdom*; [5] *Laboratory of Nematology / Crop protection Department, IAV Hassan II, Campus d'Agadir, Agadir, Morocco*; [6] *Laboratory of Biotechnologie and Valorization of Natural Ressources, Ibn Zohr University, Agadir, Morocco*

Soil biodiversity plays a substantial role in several ecosystem functioning. At the context of environmental change and anthropic impact on soil biodiversity in extreme terrestrial ecosystems, this work highlighted the change affecting nematode communities under different managements of agricultural lands to assess the anthropic impact on soil diversity in Arganeraie biosphere. Soil samples for nematodes and phyiscochemical analysis were taken in native soil, organic farms, cereal fields and conventional system. The forest soils were characterized by a rich and diverse nematofauna showing more than 38 genera belonging to 25 families represented by the occurrence of the five trophic groups. Bacterivores nematodes were the most frequent and abundant (Cephalobidae) followed by Herbivores. Predators had an intermediate proportion between Fungivores and omnivores, which were frequent but were the least abundant. However, Significant reduction in taxonomic and functional diversity of nematodes was observed in arable soil especially under continuous monocropping. Soils from conventional systems were characterized by low nematode diversity but with high population densities of endoparasitic plant parasites. More than 60% of taxonomic biodiversity was lost with a complete elimination of some high level trophic groups in most of the prospected soil. This dominant mode of production, increases the potential risk that phytoparasitic nematodes pose to growing crops in conventionally cultivated lands in the Argan biosphere especially by *Meloidogyne spp. and Pratylenchus spp.*

Keywords: Nematodes - Soil ecology - Bioiversity - Land use - Argan biosphere.

Toxicity of essential oils to beneficial nematodes.

Kerry-Lyn McDonald-Howard[1] (k.mcdonaldhoward@2018.ljmu.ac.uk), Archita Barua[1], Rory McDonnell[2], Hayley Jones[3], Christopher Williams[1], Robbie Rae[1]

[1] *School of Biological and Environmental Sciences, Liverpool John Moores University, Liverpool, United Kingdom*; [2] *Department of Crop and Soil Science, Oregon State University, Corvallis, Oregon, United States*; [3] *Garden Wisley, Royal Horticultural Society, Woking, United Kingdom*

Due to the harmful effects caused by chemical pesticides on the environment and to human and animal health [1;2], there has been a movement towards alternative control methods which are less toxic and more sustainable. Using essential oils in bio-rational management of pest species has gained attention as some oils have been shown to have the different constituents, such as terpenes, alcohols, aldehydes, ethers, ketones, phenols and oxides [3] which are effective against various pests including insects, mites, fungi, gastropods and nematodes [4;5]. These studies have demonstrated good levels of control of pestiferous insects, mites nematodes and molluscs while having no phytotoxic effect on the plants. They also show no harmful effects to humans. However, the impact essential oils have on naturally occurring beneficial nematodes has not been considered. Here we exposed three commercially available beneficial nematode products (*Phasmarhabditis hermaphrodita*, *Steinernema feltiae* and *Heterorhabditis bacteriophora*) to 13 essential oils (1% concentration) (plus controls of Tween 80 and water) and assessed thrashing behaviour and survival. Mortality results showed an “all or nothing” response with only three oils – pine oil, peppermint and lemongrass– displaying intermediate mortalities. Thrashing assays confirmed the toxic effects of certain oils. We recommend that the effects of essential oils be tested on beneficial organisms, such as nematodes, in ecosystems before use.

Keywords: Essential oils - Bio-rational management - Toxicity - Beneficial nematodes.

References:

- [1] Van der Werf, 1996. AGR ECOSYST ENVIRON. (60): 81-96.
- [2] Bailey, 2002.CABI Publishing: 33–54.
- [3] Baser and Buchbauer., 2009.CRC Press LLC: 121-149.
- [4] Isman, 2000. Crop Prot. (19): 603-608.
- [5] Barua et al., 2017.Proc. Zool. Soc.(70): 92–96.

The effect of soil type and temperature on the survival of the slug parasitic nematode *Phasmarhabditis hermaphrodita.*

Kerry-Lyn McDonald-Howard[1] (k.mcdonaldhoward@2018.ljmu.ac.uk), Rory McDonnell[2], Hayley Jones[3], Christopher Williams[1], Robbie Rae[1]

[1] *School of Biological and Environmental Sciences, Liverpool John Moores University, Liverpool, United Kingdom*; [2] *Department of Crop and Soil Science, Oregon State University, Corvallis, Oregon, United States*; [3] *Garden Wisley, Royal Horticultural Society, Woking, United Kingdom*

Slugs are highly pestiferous and are usually controlled by molluscicide pellets [1]. However, they can be toxic to non-target organisms [2]. An alternative is the parasitic nematode of slugs, *Phasmarhabditis hermaphrodita* which has been shown to provide an equivalent level of slug protection to crops [3]. It is also an environmentally friendly biological control as it is not harmful to non-target organisms [4]. This has led to the development of the biopesticide product Nemaslug® formulated by Becker Underwood and presently sold and produced by BASF [5]. However, there have been reports of varying efficacy in gardens from members of the Royal Horticultural Society. There is limited research into how abiotic factors impact the survival of the nematode [5]. Our aim was to investigate whether soil type and temperature affected the survival of *P. hermaphrodita*. Soils (garden bed soil and soil from underneath turf) were collected from RHS Gardens, Wisley and Harlow Carr. Compost with peat and compost without was commercially bought from garden centres. 2000 nematodes were applied to Petri dishes with each soil and incubated at 5°C, 10°C, or 15 °C for 3, 6, 12, 24 and 48 days. Nematode survival was quantified at each time point. In most of the soils there were over 500 nematodes on day 24, however, there was a significant difference between survival of *P.hermaphrodita* in garden bed and turf soil and at each temperature across the time series. Interestingly, on day 12 it was observed that the nematodes had progressed from the dauer (infective) stage and transitioned to juveniles and adult stages (non- infective). The number of non-infective nematodes varied with soil type and could severely affect the efficacy of *P. hermaphrodita.*

Keywords*: Phasmarhabditis hermaphrodita* - Abiotic - Survival - Biopesticide - Molluscs.

References:

- [1] Bailey, 2002.CABI Publishing: 33–54.
- [2] Berny, 2007. Vet. Pharmacol. Ther. (30): 93-100.
- [3] Wilson et al., 1993. Biocontrol Science and Technol. (3): 503–511.
- [4] Wilson et al., 2000. Pest Management.(56): 711–716.
- [5] Rae et al., 2007. Pest Management. (63): 1153-64.

Analysis of nematicidal activity of bacterial strains from collection of Institute of Biological Plant Protection.

Varvara Migunova[1] (barbarusha@rambler.ru), Angela Asaturova[2], Alena Konrat[3], Natalya Tomashevich[2]

[1] *Institute of Ecology and Evolution, Moscow, Russia*; [2] *Institute of Biological Plant Protection, Krasnodar, Russia*; [3] *ARSRJP, Moscow, Russia*

Plant parasitic nematodes are wide spread pests causing severe yield losses to numerous agricultural crops. The influence of nematodes on crop damage may be direct or indirect. Nematodes can transmit viral, fungal and bacterial infections. Yield losses in complex plant infections can reach 100%. Endoparasitic nematodes of the genera *Meloidogyne* and *Heterodera* are one of the most economically important. Recently there is a tendency to use microbiological preparations against plant parasitic nematodes. Bacteria of the genera *Bacillus* and *Pseudomonas* can be considered as potential biological control agents. The aim of this research was to analyze the nematicidal activity of 35 bacterial strains from the collection of the Federal State Budgetary Scientific Institution «All-Russian Research Institute of Biological Plant Protection» (VNIIBZR). Bacterial cultures were grown up in liquid nutrient medium. Bacterial suspensions were added to nematode suspensions. Mortality of nematodes was determined after 24 hours. The nematicidal activity ranged from 0% to 100%. Many strains of *Bacillus* demonstrated high levels of nematicidal activity (higher than 50%). *Pseudomonas* species did not show high nematicidal activity. The maximum mortality for *Pseudomonas* was 16%. The influence of dilution of bacterial suspension on plant parasitic nematodes mortality was also studied. Suspensions of *Bacillus siamensis* BZR 86 (10^7 CFU/ml) lost their nematicidal effect when diluted 100 times. The *in vitro* tests with bacterial cultures from the collection of VNIIBZR is the first step to analyze the possibility of using antagonistic bacteria in the struggle against plant diseases caused by plant parasitic nematodes.

Keywords: Antagonistic bacteria - Bacillus - *Pseudomonas* - Nematodes - Nematicidal activity.

Circadian rhythms inform the outcome of a plant-nematode interaction.

Shova Mishra (shovamishra@ufl.edu), Peter DiGennaro

University of Florida, Gainesville, FL, United States

Root-knot nematodes (RKN; *Meloidogyne* spp.) are obligate plant-parasites that constantly communicate with their host to establish and maintain specialized feeding cells. RKN likely regulate this interaction by monitoring host biology and behavior. As plant processes follow tightly regulated circadian patterns, RKN may use similar cues to regulate aspects of this intimate interaction. We investigated RKN biology within the context of host circadian rhythms throughout nematode development. At 24-hours post-inoculation, RKN penetrated host roots significantly more when inoculated during the night compared to the day. This phenomenon is also observed under inverted light conditions when we inoculated plants in inverted light cycles. However, when plants were allowed to adjust their light-driven circadian clock under constant light for one week, the variation in penetration was abolished. Furthermore, egg hatch and infective juvenile mobility did not follow rhythmic patterns. Taken together, it appeared that nematode host-seeking and penetration were at least partially influenced by daily changes in plant signaling and RKN did not display its own circadian clock. To define possible molecular mechanisms behind plant-nematode communication in relation to host circadian rhythms, we profiled root-knot nematode feeding site transcriptomes and identified over 1,500 differentially expressed genes (DEG) which we minimally binned into 6 basal expression profiles. More than 80% of nematode DEG occurred in the middle of the night compared to the middle of the day. Interestingly, plant genes involved in defense response pathways and nematode genes involved in establishing feeding sites were highly upregulated at night, suggesting a strong host-nematode interaction at night. To better characterize these pathways we developed coexpression networks of DEG between nematode and plant genes. Understanding the role and origin of circadian rhythms in the plant-nematode interaction will likely underscore the importance of exploiting basal plant biology to develop novel control methods for these pathogens.

Keywords: Circadian rhythms - Root-knot nematode - Plant-nematode interaction.

Genomics and evolutionary analysis of a new *Trichoderma* species with potential to antagonize plant-parasitic nematodes.

Tulio Morgan[1] (tmorgan@ufmg.br), Angelica Gouveia[2], Thalita Monteiro[3], Leandro Freitas[3], Tiago Mendes[2]

[1] *Laboratório de Bioinformática, Departamento de Bioquímica e Biologia Molecular, Universidade Federal de Viçosa, Viçosa, Minas Gerais, Brazil*; [2] *Departamento de Bioquímica e Biologia Molecular, Universidade Federal de Viçosa, Viçosa, Brazil*; [3] *Departamento de Fitopatologia, Universidade Federal de Viçosa, Viçosa, Brazil*

This study aimed to characterize the genomic repertoire and evolutionary patterns of a new *Trichoderma* species with potential for biocontrol of plant-parasitic nematodes. Also, to raise hypotheses of alternative mechanisms employed by fungi as part of the toolbox to antagonize such plant pathogens. Whole-genome sequencing was conducted using Illumina short-reads (150 bp paired-end reads). After quality control and data filtering, *de novo* genome assembly was carried out using the de Bruijn graph approach. The prediction of protein-coding genes was performed using *ab initio* and similarity-based methods. To predict functional domains of proteins related to biocontrol of plant-parasitic nematodes, an in-depth InterPro analysis was conducted. For genome evolution insights, we used a parsimonious scenario of gene gain and loss based on Dollo's theory. Here we report the genomic features of a newly described *Trichoderma* species, named here *Trichoderma* sp. J7, especially regarding its biocontrol-related enzymes (chitinases, proteases) and secondary metabolite biosynthetic gene clusters. Comparative genomic analysis and the profiles of gene gain and loss throughout the evolutionary trajectory of *Trichoderma* sp. J7 pointed contractions in major families of genes that confer biocontrol abilities against plant-parasitic nematodes. Also, many other genes related to environmental responses, such as transmembrane transporters and transcriptional factors were lost. Despite having a relatively streamlined genomic repertoire, this fungus shows potential for biocontrol of plant-parasitic nematodes. The genomic repertoire and evolutionary patterns of the newly described *Trichoderma* sp. J7 indicates elusive biological processes involved in antagonism against plant-parasitic nematodes. Functional genomics analysis and proteomics will be conducted to obtain data on a global scale and improve our understanding of the biological process in fungus-nematode interactions.

Keywords: *Trichoderma* - Whole-genome sequencing - Genomic analysis - Biocontrol.

Suppressive effect of *Solanum palinacanthum* on root-knot nematodes (*Meloidogyne* spp.).

Gaku Murata[1] (gmurata@affrc.go.jp), Kenta Uesugi[1], Taketo Uehara[2], Kie Kumaishi[3], Yasunori Ichihashi[3], Takeo Saito[4], Yoshimi Shinmura[4]

[1] *NARO, Koshi, Kumamoto, Japan*; [2] *NARO, Tsukuba, Ibaraki, Japan*; [3] *RIKEN, Tsukuba, Ibaraki, Japan*; [4] *NARO, Tsu, Mie, Japan*

Resistant *Solanum* cultivars have been developed for management of root-knot nematodes (RKNs, *Meloidogyne* spp.), such as *Mi*-carrying tomato cultivars and *Solanum torvum*, which is used as rootstock of eggplant in Japan. However, those plants are sometimes inefective against some virulent *Meloidogyne* spp. [1, 2, 3, 4], and thus the demand for new genetic resources with broad-spectrum resistance is increasing worldwide. In this study, we carried out a screening test for RKN-resistance in 200 or more accessions of a *Solanum* spp. collection in NARO and found a promising wild *Solanum* sp. The plant was identified as *S. palinacanthum* from morphological features and by molecular phylogenetic analysis. *S. palinacanthum* has been less studied for utilization in agriculture than for medical or pharmaceutical use [5]. To test the spectrum of RKN-resistance, *S. palinacanthum* in pot culture was inoculated with second-stage Juveniles (J2s) of *M. incognita*, *M. arenaria* genotype A2-J and A2-O, *M. javanica*, and *M. hapla*, which are common species in Japan, revealing plant suppression of reproduction by all the RKNs. In evaluation of nematode invasiveness and developmental features on pot cultured plants inoculated with *M. incognita*, number of nematodes observed within roots and the degree of nematode growth were significantly different between *S. palinacanthum* and susceptible plants (tomato, and eggplant) at 10, and 21 days after inoculation (dai), but not at 3 dai. This result indicates that the suppressive effect of *S. palinacanthum* on RKNs comes from some sort of inhibitory mechanism to nematode development triggered after invasion. To test the robustness of the resistance, we also conducted field trials in 2016. Cultivation of *S. palinacanthum* significantly suppressed the population densities of RKNs in both field trials, confirming the suppressive effect on natural populations of RKNs. The above data show that *S. palinacanthum* could be a useful resource for management of RKNs even in fields infested by multiple *Meloidogyne* spp.

Keywords: Solanaceae - Resistance - Nematode management.

References:

- [1] Brito et al., 2004. Journal of Nematology 36: 232–240.
- [2] Liu and Williamson, 2006. Journal of Nematology 38: 158–164.
- [3] Öçal et al., 2018. Journal of Plant Diseases and Protection 125: 577–580.
- [4] Uehara et al., 2017. Journal of Phytopathology 165: 575–579.
- [5] Matias et al., 2016. Brazilian Journal of Pharmacognosy 26: 147–160.

Effect of deep application of non-fumigant nematicides on *Meloidogyne incognita* in a tomato plasticulture system.

Chinaza Nnamdi[1] (chinaza.nnamdi@uga.edu), Abolfazl Hajihassani[2]

[1] *Department of Plant Pathology, University of Georgia, Tifton, GA, United States*; [2] *Department of Entomology and Nematology, University of Florida, Davie, FL, United States*

Meloidogyne incognita is detrimental to vegetable crops due to the damage it causes. To manage this nematode, placement of a single or double drip tape at a depth of 2.54 cm below the plastic mulch to aid in the delivery of nematicide is a common practice in vegetable plasticulture systems in Southeastern United States. However, nematicide application at this depth might offer little or no control of *M. incognita* populations that exist deep in the soil profile which may move upward and infect host crops later in the season. Field trials were conducted in the spring of 2019 and 2020 and summer of 2021 to evaluate the best combination of drip tapes [Surface and Sub-surface Drip Tapes (SSDT) vs only Surface Drip Tape (SDT)] and nematicides (fluenosulfone – 5.84 liter/ha, fluazaindolizine – 2.24 liter/ha, and fluopyram – 0.47 liter/ha) for *M. incognita* control in two tomato cultivars. Tomato cv. Red Bounty was used in 2019 and 2020 trials, while tomato cv. Roadster was used in 2021 trials. Surface and sub-surface drip tapes were placed at a depth of 5.08 and 30.48 cm below the soil surface, respectively. In 2019, the three nematicides reduced both root galling and *M. incognita* populations compared with 2020, where no effect was observed. Root galling in 2021 was only significantly reduced by fluensulfone and fluazaindolizine treatments, while the nematode population was only reduced by fluensulfone treatments. There was no drip tape effect or drip tape by nematicide effect on the root galling and the nematode population density at the end of the season in almost all the trials. Tomato yield was not impacted by the drip tape placement, nematicide treatments and drip tape by nematicide effect. This study shows that the drip tape utilized for nematicide application plays no role in the control of *M. incognita* in a single cropping system. However, this technique might be effective in multi-cropping plasticulture systems and requires further investigation.

Keywords: Root-knot nematode - Plasticulture - Non-fumigant nematicides - Drip tape - Deep application.

Integration of biocontrol agents and microorganisms found in organic soil amendments to control Potato Early Die.

Luisa Maria Parrado Guevara (parradog@msu.edu), Emilie Cole, Marisol Quintanilla-Tornel

Applied Nematology Lab, Michigan State University, East Lansing, MI, United States

Between 2017 and 2019 numerous trials were conducted both in the lab and in the field to control *Pratylenchus penetrans* and *Verticillium dahliae*. These two organisms can interact, and together cause the Potato Early Die complex (PED) which, when present, can reduce potato quality and yield by 30%-50%. Our initial field and lab trials indicate that chicken manure and Layer Ash Blend (LAB), which is composed of chicken + cattle manure, are most effective in controlling *P. penetrans* and producing greatest yields. As for the biocontrol products commercially available, in 2018, MeloCon (*Paecilomyces lilacinus*) did not control *P. penetrans* populations under a field setting. However, literature suggests that when combined with organic amendments or integrated with another nematode antagonist like *Bacillus firmus,* a nematocidal effect is maximized. The results obtained from these three years are promising and encourages the search for biocontrol strategies, but the exact mechanism behind the effect on root-lesion nematodes is still unknown. Based on literature, the mechanism starts with the enrichment of nutrients that benefits microorganisms that may have an antagonistic effect by the production of metabolites. To determine if this is what happening in our trials, we are going to determine the presence of microbial communities in the poultry manure and LAB and demonstrate their biocontrol potential. Laboratory trials will be conducted to isolate these microorganisms and their pathogenicity and antagonism against *P. penetrans* and *V. dahliae* will be tested. Once these communities are identified, the effect will be then tested under greenhouse conditions. Although our experiments are still ongoing, based on previous literature we hypothesize that known antagonistic communities like *Bacillus* spp., *Pseudomonas* spp. and *Streptomyces* spp will be present. Regarding the integration of biocontrol strategies, the nematicide effect of MeloCon combined with poultry manure and LAB and with Nortica 10WP (*B. firmus*) is being tested under a lab setting. So far, we have recorded results about the potential of these biocontrol products to gain a better understanding of their role in nematode control; however, we will continue to investigate these products under field conditions. Currently, we are still working on the antagonistic effect of the microorganisms found in the poultry manure and LAB.

Keywords: Potato Early Die - Poultry Manure - Layer Ash Blend - Biocontrol - Microorganisms.

References:

- Abd-Elgawad et al., 2018. Egyptian Journal Of Biological Pest Control, 28(1): 2-5
- Anastasiadis et al., 2008. Crop Protection, 27(3-5): 352-36
- Molina et al., 2014. American Journal Of Potato Research, 91(4): 414-428
- Uppal et al., 2008. Biological Control, 44(1): 90-100

Seasonal fluctuations attenuate stimulatory or inhibitory impacts of colonial birds on soil biota.

Stanislav Pen-Mouratov (stanislav_pen_m@yahoo.com)

School of Zoology, George S. Wise Faculty of Life Sciences, and The Steinhardt Museum of Natural History, Tel Aviv University, Tel-Aviv, Israel

The soil biota, including soil microorganisms and free-living nematodes, was seasonally investigated in association with the nesting and roosting habitats of the following piscivorous and omnivorous colonial birds: black kite (*Milvus migrans*), great cormorant (*Phalacrocorax carbo*), black-crowned night heron (*Nycticorax nycticorax*) and little egret (*Egretta garzetta*), in Israel's Mediterranean region.
Abiotic variables, abundance, trophic structure, sex ratio and genus diversity of soil free-living nematodes, and total abundance of bacteria and fungi, were measured during the wet and dry study periods. The observed soil properties were important drivers influencing the soil biota structure with a tight dependence on season of the year. Presence of the most crucial nutrients for plants and soil organisms, such as N and P which limit primary productivity in many ecosystems worldwide, was strongly dependent on the diet of the compared piscivorous and omnivorous bird colonies.
The applied ecological indices indicated that the different species of colonial birds can have different (stimulatory or inhibitory) impacts on abundance and diversity of the soil biota, affecting the structure of the soil free-living nematode population at the generic, trophic and sexual levels. Seasonal fluctuations could change the effect of bird activity on the abundance, structure and diversity of the soil communities.

Keywords: Bird roosting area - Soil biota - Soil nematode diversity - Bird dropping - Seasonal variation.

A novel pore-forming toxin triggers *Ditylenchus destructor* apoptotic cell death via a C2-domain containing protein.

Donghai Peng (donghaipeng@mail.hzau.edu.cn), Feng Chen, Yang Geng, Ling Chen, Mengci Xu, Jinshui Zheng, Ming Sun

Huazhong Agricultural University, Wuhan, China

Apoptosis, or programmed cell death, is a conserved process among multicellular organisms, which plays a central role in eliminating damaged cells that prevent pathologic conditions and maintains tissue homeostasis. In chordates, apoptosis can be triggered by extrinsic pathwaya (death receptor pathway) or intrinsic pathways (mitochondria pathway). In contrast, studies in *Caenorhabditis elegans* (*C. elegans*) indicated that apoptosis in nematode depends on four key genes (*egl-1*, *ced-9*,*ced-4*,*ced-3*). However, genome analyzing of plant parasitic nematodes showed that *egl-1*, *ced-9* and *ced-4* were not conserved in plant parasitic nematodes. So does apoptosis exist in plant parasitic nematodes and can it be used as a target for control? In our research, we used *Ditylenchus destructor* (*D. destructor*) and Cry55Aa which is produced by *Bacillus thuringiensis* (Bt) as materials. We found that Cry55Aa exhibit high toxicity to PPN such as root-knot nematode, cyst nematode and *D. destructor* but not to *C. elegans*. By monitoring the phenotype of lysosomes, mitochondria, calcium ions, caspase activity as well as the transcription level of key genes of apoptosis and necrosis after toxin treatment, we confirmed that Cry55Aa could induce the apoptotic-cell death in nematodes. Subsequently, through yeast two-hybrid screen of a *D.destructor* AD-DNA library, 9 proteins were identified that could interact with Cry55Aa. Using RNAi to reduce the transcription of those 9 gens, we found that a C2 domain containing protein could significantly reduce the toxicity of the toxin and alleviate the phenotype of apoptosis. Our study confirmed that the mitochondria apoptosis pathway is conserved in plant-parasitic nematodes and can be induced by external pressure, and we also provide a safe target for the control of plant parasitic nematodes.

Keywords: Apoptosis - Ditylenchus destructor - Cry55Aa - Apoptotic-cell death - C2 domain.

Host suitability of biofortified *Fusarium*-resistant sweet potato lines to *Meloidogyne* species in South Africa.

Kgabo Pofu[1] (pofukgabo@gmail.com), Sunette Laurie[2], Phatu William Mashela[1]

[1] *Green Biotechnologies Research Centre of Excellence, University of Limpopo, Polokwane, South Africa*; [2] *Plant Breeding, Agricultural Research Council-Vegetable and Ornamental Plants, Pretoria, South Africa*

Generally, genes that confer insect resistance in plants usually do not confer nematode resistance, with limited information on host-status of fungus-resistant genotypes to *Meloidogyne* species. Eighteen *Fusarium*-resistant biofortified sweet potato lines were each inoculated with 3000 eggs + juveniles of *M. incognita* race 2, *M. incognita* race 4 and *M. arenaria* in parallel pot experiments. The South African cv. 'Blesbok' and the USA cv. 'Beauregard' were used as nematode-susceptible standards. At 56 days after inoculation, eggs and juveniles were extracted from total root systems, which were expressed as reproductive potential (RP = Pf/g fresh root), where Pf constituted eggs + juveniles. In *M. javanica* experiment, FS-10-14, FS-10-19 and FS-10-25 lines had RP less than 1, whereas on all other lines RP was greater than unity. In *M. incognita* race 2, RP values were the lowest on FS-10-14 and FS-10-19 lines, although the RP values were above one. In contrast, in *M. incognita* race 4, all lines had RP values greater than unity. The experiments were validated once, with the RP responses showing similar trends to those in the first trials. In conclusion, although results of this study suggested lack of nematode-resistance genotypes in fusarium-resistant lines, FS-10-14, FS-10-19 and FS-10-25 lines versus *M. javanica* could provide some light on genes responsible to nematode resistance in sweet potato *Fusarium*-resistant lines.

Keywords: Fusarium-resistant - Biofortification - Blesbok - Meloidogyne species - Nematode susceptible.

Potential new targets for entomopathogenic nematodes.

Vladimír Půža (vpuza@seznam.cz)

Institute of Entomology, Biology Centre of the AS CR, Ceske Budejovice, Czech Republic

Entomopathogenic nematodes from the families Steinernematidae and Heterorhabditidae are among the most promising biocontrol agents. At the beginning of the year 2020, 103 species of steinernematids and 16 species of heterorhabditids were known, but only about 10 species were used in biocontrol. Although these nematodes are morphologically quite uniform they adopt various life history strategies, ranging from active search for the host to sit and wait strategy. This fact increases the spectrum of potential EPN targets. Traditionally, EPN applications have been focused on insects that are present in the soil environment. Later, foliar pests and pests in cryptic habitats were successfully targeted by EPNs. It can be expected, that this spectrum will further grow in the future. Factors that will drive this growth are the advances in EPN formulation, application methods, and also policy changes. These factors and potential new target insects for EPNs will be discussed, and special emphasis will be devoted to the potential of EPNs against non-insect pests.

A novel *Meloidogyne incognita* effector Mi-ISC-1 promotes parasitism by disrupting salicylic acid biosynthesis in host plants.

Xin Qin (1832725309@qq.com), Cong Chen, Xiang He, Xuan Wang, Hongmei Li

Department of Plant Pathology, Nanjing Agricultural University, Nanjing, China

As an obligate biotrophic parasite, root-knot nematode *Meloidogyne incognita*, is one of the most important plant parasitic nematodes worldwide. This parasite can suppress host defense responses and facilitate infection by secreting effectors, which are typically identified based on the presence of signal peptides and expression in esophageal glands of nematodes. Herein we described a novel effector of *M. incognita*, named Mi-ISC-1, which is a member of the isochorismatase (ISC) protein family. The transcripts of Mi-ISC-1 were exclusively accumulated in the esophageal glands and up-regulated in parasitic-stage juveniles. Although Mi-ISC-1 lacks a classical secreted signal peptide at N-terminal, the yeast invertase secretion assay confirmed its secretion activity. The subcellular localization assays showed that Mi-ISC-1 was localized in cytoplasm and nucleus of the transfected *Nicotiana benthamiana* leaf cells. Meanwhile, the transient expression of Mi-ISC-1 in *N. benthamiana* reduced the expression of *PR1* gene and the levels of the salicylic acid (SA), which consequently promoted the infection of *Phytophthora capsici*. In addition, *Tobacco rattle virus*-induced gene silencing targeted the *Mi-isc-1* attenuated *M. incognita* parasitism. *In vitro* enzyme activity assays confirmed that Mi-ISC-1 can catalyse hydrolysis of isochorismate into 2,3-dihydro-2,3-dihydroxybenzoate. Moreover, Mi-ISC-1 impairs the reconstitution of *de novo* SA biosynthesis in *N.benthamiana* leaves. These results demonstrated that the unconventionally secreted effector Mi-ISC-1 can promote nematode parasitism, most likely via disrupting SA biosynthesis and suppressing plant innate immunity.

Keywords: *Meloidogyne incognita* - Isochorismatase - Secretion - Salicylic acid - Plant innate immunity.

References:

- [1] Liu et al., 2014. Nature Commun. 5: 4686.
- [2] Wang et al., 2018. Plant Pathol. 67(2): 1436-1448.

Pathotypic variation between isolates of *Meloidogyne enterolobii* collected from sweetpotato in North and South Coralina.

William Rutter[1] (William.Rutter@usda.gov), Phillip Wadl[1], John Mueller[2], Paula Agudelo[3]

[1] *United States Vegetable Laboratory, USDA-ARS, Charleston, SC, United States*; [2] *Department of Plant and Environmental Sciences, EREC, Clemson University, Blackville, SC, United States*; [3] *Department of Plant and Environmental Sciences, Clemson University, Clemson, SC, United States*

Meloidogyne enterolobii (syn. *mayaquensis*) has been slowly spreading across the southeastern United States over the past 20 years. Serious concerns have been raised by growers and regulatory agencies because of its high-level of virulence on *M. incognita*-resistant sweetpotato clones such as 'Covington'. It is also now widely accepted that sweetpotato is effectively vectoring the spread of this nematode through the movement of infected 'seed' roots used for propagation. Our ongoing screening efforts have focused on fresh market sweetpotatoes, and we have identified and cultured multiple isolates of *M. enterolobii* on roots originating from different producers. Despite their close geographic origin and relatively recent introduction to the region, we have observed that these isolates display distinct virulence profiles on different host crops. We will present data on *M. enterolobii* resistance in sweetpotato and pepper germplasm, and present a new high throughput method for detecting the presence of *M. enterolobii* in batches of sweetpotato storage roots. This new method could help to monitor and slow the spread of *M. enterolobii* to new regions.

Keywords: Meloidogyne enterolobii - Plant resistance - Vegetables - Sweetpotato - Pepper.

Coffee selection based on molecular characterization and identification of coffee resistant to *Meloidogyne paranaensis*.

Sonia Salgado[1] (soniaepamig@gmail.com), Alexsandra Correia[2], Bárbhara Fatobene[3], Flaminia Ferreira[3], Eveline Caixeta[4]

[1] *EPAMIG Sul, Empresa de Pesquisa Agropecuária de Minas Gerais, Lavras, Minas Gerais, Brazil*; [2] *BIOAGRO-Café, Universidade Federal de Viçosa, Viçosa, Minas Gerais, Brazil*; [3] *EPAMIG SUL, Laboratório de Nematologia, Consórcio Pesquisa Café -EPAMIG, Lavras, Minas Gerais, Brazil*; [4] *BIOAGRO-Café, EMBRAPA Café, Viçosa, Minas Gerais, Brazil*

The objective of this work was to characterize the molecular diversity of breeding populations and nematode resistance to *Meloidogyne paranaensis,* the root-knot nematode most destructive to coffee in Brazil. The SSR markers were initially analyzed in the genotypes Amphillo Collection 1 (AC1), Amphillo Collection 2 (AC2) and Amphillo Collection 3 (AC3) from the AGB –IAC/Brazil, and in the cultivar Catuaí Vermelho IAC144. These coffee plants are considered the potential parental of the resistant progenies. The F4 progenies: MG 0185 R2-1 (T1), MG 0179 R1-5 (T2), MG 0179 R1-1(T3), MG 0179 R1-3 (T4). Phenotyping was realized in 40 plant of each progenies and CV144 (susceptible control). DNA extraction was performed before infection with 1600 eggs of *M. paranaensis* (EST P1). Reproduction Factor (RF=Final population/initial population) was calculated 180 days postinfeccion. Among the 16 SSR markers analyzed, five that showed polymorphism (CaEST 022; CaEST 045; CaEST 072; CaEST 002; CaEST 058) among the parents were evaluated in the entire F4 progenies. T1 and T3 progenies showed genetic resistance segregation to *M. paranaensis* indicating that the resistance is conferred by a single dominant gene (3R: 1S). No segregation was observed in T2 and T4 genotypes, being 100% resistant. The molecular analyses also showed that T2 and T4 have low diversity, and T1 and T3 high diversity. The highest diversity was found for T3 progeny. Based on the dendrogram, the coffee plants were clustered in four groups. Group 1 was formed by the cultivar CV144, Amphillo AC3-R2 and seven plants of theT3 progenies. Group 2 consisted of 21 T3, one T1 and three T2. Group 3 had only the parental AC3-R1. Amphillo AC1 and AC2 and the most plants of the T1, T2 and T4 was clustered in group 4, which was sub grouped in 4a, 4b, 4c and 4d. Resistant plants were found in all groups. Group 1 contains the coffees genetically closest to the commercial cultivar Catuaí, and four of them are resistant to *M. paranaensis*. This result shows that these coffees (7T3, 8T3, 16T3 and 40T3) should be selected for the next generation in the breeding program, as they are resistant to nematode and are genetically close to the commercial cultivar. In this sense, 11 plants (3T3, 4T3, 9T3, 10T3, 11T3, 13T3, 17T3, 21T3, 23T3, 25T3 and 31T3), although resistant, are genetically close to the wild parent (Amphillo) and should be avoided in the breeding program. Financial Support: CNPq, Consórcio Pesquisa Café.

Keywords: *Meloidogyne paranaensis* - Resistant - Coffee arabica - SSR markers - Breeding.

References:

- Santos et al. 2017. Plant Pathology, Doi: 10.1111/ppa.12718
- Pereira et al.2016 Genetics and Molecular Research 15 (3): gmr.15038054
- ALVES et al. 2019. Nematology 0 (2019) 1-12. DOI 10.1163/15685411-00003254

Antagonistic effect of fungal filtrates on hatching, mobility and mortality of *Meloidogyne incognita*.

Débora Cristina Santiago (santiago@uel.br), Fernanda Pereira de Sousa, Gabriel Ribeiro de Lima, Maria Isabel Balbi-Peña

Department of Agronomy / Center of Agrarian Sciences, State University of Londrina - UEL, Londrina, Paraná, Brazil

The effects of filtrates from the fungi *Memnoniella levispora* (MEL), *Myrothecium* sp. (MYR), *Phialomyces macrosporus* (PHM) and *Purpureocillium lilacinum* (PAE 1112, PAE 1105, PAE 1104 and PAE 1101) were evaluated on the hatching, mobility and mortality (MM test) of second-stage juveniles (J2) of *Meloidogyne incognita* in laboratory conditions. Fungi were cultured in potato-dextrose (0.05 gL^{-1} of streptomycin) at 25°C for 10 days + two days at 5°C. Filtrates were obtained by vacuum filtration, stored in the dark at 5°C and used at 50% and 100%. For use in the MM test, J2 were obtained from soybean roots infested by *M. incognita* that were incubated at 26°C for three days in hatching chambers. J2 suspension was adjusted to 85 $J2.mL^{-1}$. For the hatching test, a 300 $eggs.mL^{-1}$ suspension from previously separated *M. incognita* egg masses was used. Two mL of the filtrates were added to 2 mL of these J2/egg suspensions. As a control, sterile water was used. Experimental design was completely randomized with five replicates. For the hatching test, J2/egg suspensions with filtrates or water were incubated at 26 °C for 16 days, while for the MM test the incubation time was one day. After this period, the suspension content was passed through 200 and 500 mesh sieves. Eggs and J2 from the last sieve were counted to determine unhatched J2 (UHJ2) and immobile J2 (IMJ2), respectively. After the first evaluation of the MM test, suspensions remained for another seven days at 26ºC for determination of dead J2 (DEJ2). The weighted average was calculated considering the total of eggs and/or J2 found. In the hatching test MEL, MYR and PAE 1104 at 100%, PHM and PAE 1105 at 50% showed 74-100 % of UHJ2 comparing with 44% in the control. In the MM test most treatments showed higher values of IMJ2 than the control (53%), except for MEL 50% and PAE 1105 50%. However, in the subsequent evaluation, only PHM, MYR and PAE 1105 at 50% obtained DEJ2 superior to the control (76%), characterizing the nematicidal effect of these treatments. Thus, treatments that resulted in IMJ2 superior to the control and DEJ2 inferior to the control have nematostatic effect. It can be concluded that the MYR showed satisfactory results in both tests, being the best in the MM test with 93% of DEJ2 in the concentrations used, followed by PHM 50% with 84%. In the hatching test MEL 50% and 100% resulted in 100% and 99% of UHJ2, respectively.

Keywords: Biological control - Root-knot nematode - Biological nematicidal - Purpureocillium lilacinum.

Molecular insights into an interaction of the resistant plant *Solanum torvum* and virulent/avirulent root-knot nematodes.

Kazuki Sato[1] (kazuki.sato@riken.jp), Yasuhiro Kadota[1], Pamela Gan[1], Taketo Uehara[2], Takahiro Bino[3], Katsushi Yamaguchi[3], Yasunori Ichihashi[4], Hideaki Iwahori[5], Noriko Maki[1], Shuji Shigenobu[3], Takamasa Suzuki[6], Bruno Favery[7], Shahid M. Mukhtar[8], Ken Shirasu[1]

[1] *RIKEN CSRS, Yokohama, Japan*; [2] *CARC, NARO, Tsukuba, Japan*; [3] *NIBB Core Research Facilities, NIBB, Okazaki, Japan*; [4] *RIKEN BRC, Tsukuba, Japan*; [5] *Department of Agriculture, Ryukoku University, Otsu, Japan*; [6] *Department of Biological Chemistry, Chubu University, Kasugai, Japan*; [7] *INRAe, Université Côte d'Azur, CNRS, ISA, Sophia Antipolis, France*; [8] *Department of Biology, University of Alabama at Birmingham, Birmingham, AL, United States*

Plant-parasitic nematodes, such as root-knot nematodes (RKNs), are among the most devastating pests in agriculture. *Solanum torvum*, which is horticulturally used as a rootstock for eggplants, confers resistance against RKNs such as *Meloidogyne incognita* and *Meloidogyne arenaria* Okinawa isolate (*Ma* Okinawa). However, *Ma* Japanese Main island isolate (*Ma* Main island) can infect *S. torvum* [1]. The infection with *Ma* Okinawa *in vitro* induces the accumulation of brown pigments at the site of infection in *S. torvum*, while the infection with *Ma* Main island induces the formation of gall-like structure. To understand the molecular basis of plant immunity and virulence of RKNs, we performed dual RNA-sequencing of *S. torvum* root tissues invaded by *Ma* Okinawa and *Ma* Main island, as well as genomic sequencing of these two *Ma* isolates using long-read sequencer PacBio. We revealed that *S. torvum* infected with *Ma* Okinawa, but not with *Ma* Main island, dramatically up-regulated the expression of genes involved in defense responses such as lignin biosynthesis within a few days. We confirmed that lignin accumulated at the site of *Ma* Okinawa infection in *S. torvum*. These results suggest that *S. torvum* reinforces the cell wall by the accumulation of lignin and may prevent the progression of *Ma* Okinawa and their secondary infection. To understand the molecular basis of virulence and avirulence of *Ma,* we have screened virulent and avirulent effector candidate genes based on genomic and transcriptomic information. So far, we found 95 effector candidates from *Ma* Okinawa and 63 effector candidates from *Ma* Main island, whose expressions are dramatically upregulated within a few days after infection with *S. torvum*. We have performed functional analyses of these effector candidates by expression in *S. torvum* and *Nicotiana benthamiana*, and found that some effector candidates have the ability to induce plant cell death and others can suppress the production of reactive oxygen species, which is one of the most characteristic immune responses in plants. To reveal their plant targets, we performed yeast-two-hybrid screening against plant root cDNA library. We found novel target proteins as well as known effector-target proteins such as CSN5a, a subunit of the COP9 signalosome which is involved in jasmonic acid-dependent defense against RKN [2]. We will discuss the commonalities and differences of effector candidates of the two *Ma* isolates as well as their possible plant targets.

Keywords: Root-knot nematode - Resistant plant - Plant immunity - Effector - Virulence.

References:

- [1] Uehara et al., 2017. J Phytopathol. 165:575–579.
- [2] Shang et al., 2019. Front Plant Sci. 10: 1223.

Nematode vertical distribution in peanut-cotton cropping systems.

Lesley Schumacher[1] (Lesley.Schumacher@usda.gov), Zane Grabau[2], David Wright[3]

[1] *Crop Genetics Research Unit, USDA-ARS, Jackson, TN, United States*; [2] *Department of Entomology and Nematology, University of Florida, Gainesville, FL, United States*; [3] *North Florida Research and Education Center, University of Florida, Quincy, FL, United States*

Reniform nematode (*Rotylenchulus reniformis*) is a pathogen of cotton (*Gossypium hirsutum*) capable of reducing lint yields and crop rotation is commonly used for its management. One specific rotation system is a sod-based rotation, which uses two years of bahiagrass (*Paspalum notatum*) followed by one year each of peanut (*Arachis hypogaea*) and cotton, versus a conventional peanut-cotton-cotton rotation. Peanut and bahiagrass are poor hosts of reniform nematode but are good hosts for ring (*Mesocriconema ornatum*) and spiral (*Helicotylenchus dihystera*) nematodes, respectively. Additionally, reniform nematode is present deep in the soil profile, but not much is known about other nematodes at deeper depths. Our study aimed to investigate both plant-parasitic and free-living nematodes (fungivores, bacterivores, omnivores, and predators) at different soil depths in sod-based and conventional rotation with or without irrigation. Soil samples were collected to a depth of 120 cm before planting, after harvest, and in the winter of 2017-2018 using a hydraulic probe. Nematode abundances were analyzed in 30 cm-sections. There were no significant effects of irrigation on nematode abundances (P>0.05). All nematode trophic groups were present up to 120 cm deep in the soil profile except for predators, which were only found from 0 to 30 cm. There were significantly greater abundances of reniform and omnivore nematodes in the conventional rotation than the sod-based rotation. Conversely, ring, spiral, bacterivore, and fungivore nematode abundances were significantly greater in the sod-based than conventional rotation. There were significant crop by depth interactions for omnivore abundances for all sampling dates, but only in harvest sampling dates for reniform nematode abundances. There were no differences in omnivore abundances at the 0-30 cm depth, but from 30-120 cm depths abundances were greater following two years of conventional cotton. Sod-based rotation reduced reniform nematode abundances at all depths compared with conventional rotation for post-harvest sampling dates. Fungivore abundances were greater following two years of bahiagrass at the 0-30 cm depth while at deeper depths there were no differences between the rotations. Overall, nematode trophic group abundances varied by crop rotation system and depth, with the majority of nematodes present from 0 to 30 cm deep in the soil profile.

Keywords: Crop rotation - Free-living nematodes - Nematode management - Reniform nematode - Vertical distribution.

First report of the apple root-knot nematode, *Meloidogyne mali*, in Korea.

Jongmin Seo[1] (whdals5135@pusan.ac.kr), Sohee Park[1], Namsook Park[2], Donggeun Kim[2], Insoo Choi[3]

[1] *Plant Bioscience, Pusan National University, Miryang, South Korea*; [2] *Nematode Research Center, Pusan National University, Miryang, South Korea*; [3] *Plant Bioscience & Nematode Research Center, Pusan National University, Miryang, South Korea*

In September 2019, root-knot nematode was found on maple tree (*Acer palmatum* Thunb.) in Jeolla province, Korea. Identification of *Meloidogyne* sp. was conducted based on morphology and molecular data. The perineal patterns of females were oval shaped with low dorsal arch, fine interval and smooth striae, large phasmids. There were one or two lateral incisures which were narrow and broken. The morphological measurements of females(n=10), second-stage juveniles(n=20) were similar to those of *M. mali*. Molecular identification of *Meloidogyne* sp. was based on PCR-RFLP and BLAST. Polymerase Chain Reaction (PCR) amplification of the region between COII and 16S rRNA of the mitochondrial DNA produced a single fragment approximately 570bp. Amplified PCR product was not digested with *Hinf* I. Digestion with *Alu* I generated 470bp and 110bp fragments. Amplified PCR product of the 28S region using D2A and D3B primers was approximately 770bp and the ITS region using TW81 and AB28 primers was approximately 670bp. COII, ITS, 28S region sequences confirmed it as *M. mali*. *M. mali* has a very broad host range parasitizing about 44 different plant species including *Acer palmatum*. To our knowledge, this is the first report of *M. mali* in Korea.

Damage potential of *Heterodera sojae* to soybean.

Jongmin Seo[1] (whdals5135@pusan.ac.kr), Heonil Kang[1], Sohee Park[1], Namsook Park[2], Donggeun Kim[2], Insoo Choi[3]

[1] *Plant Bioscience, Pusan National University, Miryang, South Korea*; [2] *Nematode Research Center, Pusan National University, Miryang, South Korea*; [3] *Plant Bioscience & Nematode Research Center, Pusan National University, Miryang, South Korea*

Heterodera sojae, white soybean cyst nematode, was found in 2016 and its damage potential is not studied yet. Soybean was seeded in 30-cm-d clay pots filled with steam sterilized soil and placed in the greenhouse. The pots were inoculated with five initial population densities (Pi) (0 to 1,000 eggs/100 cm^3 soil) of *H. sojae*. Soybean plant weight, number of branches, seed yields, and number of nematodes per root system (Pf) were determined after five months. At the highest Pi (1,000 eggs), *H. sojae* caused yield losses up to 32% while lowest Pi (1 egg) caused yield loss of 7%. The Seinhorst equation was used to describe the relation between soybean yield and Pi and to calculate the damage threshold density (T). Pf increased with increasing Pi of *H. sojae*. The reproduction rate (Pf/Pi) of *H. sojae* was highest at the lowest Pi.

First genetic mapping of resistance to root-lesion nematode (*Pratylenchus thornei*) in chickpea (*Cicer arietinum*).

Jason Sheedy[1] (jason.sheedy@usq.edu.au), Kelvin Khoo[2], Janine Croser[3], Julie Hayes[2], Julian Taylor[2], Tim Sutton[4], John Thompson[1], Diane Mather[2]

[1] *Centre for Crop Health, University of Southern Queensland, Toowoomba, Queensland, Australia*; [2] *Waite Research Institute, University of Adelaide, Glen Osmond, South Australia, Australia*; [3] *School of Agriculture and Environment, University of Western Australia, Perth, Western Australia, Australia*; [4] *SARDI/University of Adelaide, Adelaide, South Australia, Australia*

Chickpea (*Cicer arietinum*) is a globally important cool-season legume that fixes atmospheric nitrogen and is the primary source of dietary protein for ~20% of the world's population. The root-lesion nematode, *Pratylenchus thornei*, infests cereal and legume crops globally. The use of genetic resistance is considered to be the most effective strategy for management of *P. thornei* in commercial farming systems. In chickpea, yield losses of up to 58% have been attributed to *P. thornei*. In Australia, *P. thornei*-resistant chickpea varieties are needed to support farm profitability and to maintain stability of supply for a >$900 M/yr export market. Numerous accessions of chickpea and its genetically-compatible relatives (*C. reticulatum*; *C. echinospermum*) have been identified as resistant or moderately resistant to *P. thornei*, but little is known about the genetics of this resistance. A population of 225 recombinant inbred lines were developed by accelerated single seed descent from a cross between the Australian chickpea varieties PBA HatTrick (moderately resistant to *P. thornei*) and Kyabra (susceptible). The population was subjected to genotyping-by-sequencing and a linkage map was constructed. Resistance to *P. thornei* was evaluated for 205 lines in the F6 generation and 218 lines in the F7 generation. Based on data from the F6 lines, two quantitative trait loci (QTL) were detected and mapped: one on chromosome Ca4 (LOD=2.6) and another on chromosome Ca7 (LOD=6.0). They explained 17% and 36% of the phenotypic variation, respectively. At both loci, resistance came from PBA HatTrick. The estimated QTL positions were anchored to the Kabuli V2.6.3 genome assembly at ~37.6 Mbp on Ca4 and ~22.7 MBp on Ca7. KASP™ assays were developed for 75 single-nucleotide polymorphisms linked to these QTL. The two novel QTL mapped here and the marker assays developed here offer the opportunity to include this trait in marker-assisted selection in chickpea breeding programs to improve the efficiency of the selection and release of *P. thornei*-resistant cultivars.

Keywords: *Pratylenchus thornei* - Root-lesion nematode - Quantitative Trait Loci - Breeding for disease resistance - Chickpea.

Mapping resistance to the potato cyst nematode, *Globodera pallida*, in a tetraploid, russet-skinned potato population.

Rocio Silvestre[1] (silv0890@vandals.uidaho.edu), Louise-Marie Dandurand[2], Richard Novy[3], Jonathan Whitworth[3], Joseph Kuhl[1]

[1] *Plant Sciences, University of Idaho, Moscow, Idaho, USA*; [2] *Entomology, Plant Pathology, Nematology, University of Idaho, Moscow, Idaho, USA*; [3] *Agricultural Research Service, US Department of Agriculture, Aberdeen, Idaho, USA*

Globodera pallida is a major quarantine pest of potato in the United States which has had a significant economic impact on the Idaho potato industry. In the western United States, the predominant market class is for processing and is represented by long tuber shape and russet skin which have historically lacked breeding for potato cyst nematode (PCN) resistance. European *G. pallida* is classified into three pathotypes (Pa1-Pa3) based in their virulence reactions on different potato genotypes and the Idaho population has been classified as Pa2/3. Previous studies have identified in the Scottish cultivar Eden resistance locus $GpaIV^{s}_{adg}$ which confers resistance to Pa2 and Pa3 [1,2]. In this study, PCN resistance was analyzed in a tetraploid population derived from PCN resistant Eden and susceptible Western Russet. A total of 245 offspring, four cultivars controls and four PCN differential clones were evaluated. Phenotypic evaluation involved transfer of tissue culture plantlets to pots and inoculated with 5 eggs per gram of soil in three experiments, with an average of 10 replicates per genotype. To map resistance loci the entire population was genotyped with 21,027 SNP markers with the SolCap SNP chip version 3. Quantitative trait linkage (QTL) mapping was performed for seven traits, including cyst and egg-related phenotypes. *Globodera pallida* reproduced well (average number of cysts per plant > 200) in the susceptible genotypes Desiree, Russet Burbank and Western Russet. Moreover, primary QTL associated with PCN resistance were detected on chromosome 4 and 6 with heritability estimates ranging from 0.10 to 0.90. QTL analysis indicated while cyst phenotypes mapped to a single locus on chromosome 4, egg phenotype were located on chromosome 4 and 6. This study will help to further characterize PCN resistance from Eden and identify genetic regions valuable for PCN resistance in oblong, russet-skinned processing potatoes.

Keywords: Potato cyst nematode - Globodera pallida - Russet potato - Quantitative trait loci.

References:

- [1] Bryan et al., 2002. Theor Appl Genet. 105(1): 68-77.
- [2] Moloney et al., 2010. Theor Appl Genet. 120(3): 679-689.

Identification of Quantitative Trait Loci for Resistance to *Pratylenchus neglectus* in Triticale.

Gurminder Singh[1, 2] (g.singh@ndsu.edu), Guiping Yan[2], Zhaohui Liu[2], Justin D. Faris[3]

[1] *Department of Plant Sciences, North Dakota State University, Fargo, ND, United States*; [2] *Department of Plant Pathology, North Dakota State University, Fargo, ND, United States*; [3] *USDA-ARS Cereal Crops Research Unit, Edward T. Schafer Agricultural Research Center, Fargo, ND, United States*

The root-lesion nematode (RLN), *Pratylenchus neglectus*, is an important pathogen of wheat. Utilizing host resistance is the most economical and environment-friendly way to control this nematode. To date, no RLN-resistant commercial wheat cultivar has been reported, and sources of resistance in wheat are scarce. Previous studies have reported high levels of resistance in rye and triticale. Triticale is a hybrid of wheat and rye, and identifying RLN resistance genes in triticale would be the first step to utilizing triticale resistance in wheat. In this study, a triticale population consisting of 141 recombinant inbred lines (RILs) derived from a cross between Siskiyou (susceptible parent) and Villax St. Jose (moderately resistant parent) was evaluated for reaction to *P. neglectus* under greenhouse conditions. Our goal was to identify QTL (quantitative trait loci) associated with resistance to RLN. The population was genotyped using genotyping-by-sequencing (GBS) and SSR markers, and a high-density genetic linkage map was constructed covering all wheat and rye chromosomes. Single trait multiple-interval mapping analysis revealed the presence of a single QTL (LOD = 8.6) on rye chromosome 5R that accounted for approximately 24.6% of the total phenotypic variation. The identified QTL needs to be validated in different genetic backgrounds to confirm its stability. The PCR-based molecular markers associated with this QTL will be useful for fine-mapping and to better understand the genetics of RLN resistance. This work provides a foundation for the introgression of rye-derived *P. neglectus* resistance into wheat germplasm.

Keywords: Root-lesion nematode - Triticale - QTL mapping - Introgression of resistance genes - Genotyping-by-sequencing.

References:

- [1] Elshire et al., 2011. PLOS ONE 6(5): e19379. doi.org/10.1371/journal.pone.0019379
- [2] Farsi et al., 1995. Australian Journal of Experimental Agriculture 35: 597-602
- [3] Wen et al., 2018. Theoretical and Applied Genetics 131: 649-658

New reports and molecular diagnostics of circumfenestrate cyst nematodes in the United States.

Andrea Skantar (andrea.skantar@usda.gov), Maria Hult, Mihail Kantor, Zafar Handoo

Mycology and Nematology Genetic Diversity and Biology Laboratory, USDA ARS, Beltsville, MD, United States

Cyst nematodes are a widespread and important group of plant-parasitic nematodes that limit production of numerous crops throughout the world. Aside from the *Globodera* spp. (and unlike the well-described Heteroderinae), relatively few circumfenestrate species have been characterized molecularly, with those limited mostly to ribosomal markers. We have recently identified several isolates from the subfamily Punctoderinae, including a new occurrence of the corn cyst nematode, *Vittatidera zeaphila,* from Indiana [1], and the cactus cyst nematode, *Cactodera cacti,* from Idaho [2], and a new species of *Punctodera* from Oregon. Here we present molecular characterizations for these and other populations that include ribosomal ITS, 28S, and 18S, as well as mitochondrial (COI) and nuclear Hsp90 genes, and present new phylogenetic information for these cyst nematodes. Expansion of this dataset will continue to strengthen evolutionary analysis of cyst nematodes through integration of multi-gene molecular phylogenies with morphological characters and information on geographical origin and host plant speciation.

Keywords: Cyst - Phylogenetics - Molecular - Hsp90 - rDNA.

References:

- [1] Skantar et al., 2020. Journal of Nematology (in press).
- [2] Skantar et al., 2019. Journal of Nematology 51:1-6.

Understanding the *Heterorhabditis* nematode factors involved in modulating symbiosis with *Photorhabdus* bacteria.

Vishal Singh Somvanshi (vishal.somvanshi@icar.gov.in), Chaitra Ganapati Bhat, Uma Rao

Division of Nematology, ICAR- Indian Agricultural Research Institute, New Delhi, Delhi, India

Entomopathogenic nematodes (EPNs) of the families Heterorhabditidae and Steinernematidae have independently evolved specific symbiotic relationships with entomopathogenic bacteria *Photorhabdus* and *Xenorhabdus*, respectively. This symbiotic relationship is highly specific, i.e., the symbiont bacteria do not colonise nematodes other than thier host but for few exceptions. Although some information is available on bacterial genes and proteins involved in the symbiosis with the nematodes; little is known about the nematode factors that are vital for symbiosis. We used RNA-Seq to understand the *Heterorhabditis bacteriophora* nematode factors involved in symbiosis with *Photorhabdus luminescens*. The transcriptomes of axenic and monoxenic *H. bacteriophora* nematodes were sequenced at the early adult stage (36-48 h after IJ recovery) when the symbiont attachment and biofilm formation takes place in the INT9L and INT9R cells in the posterior nematode intestine. The axenic and monoxenic nematodes were collected, RNA was isolated, quality checked, libraries prepared for 150 x 2 paired-End format and sequenced using Illumina HiSeq 2500 platform in three replicates. A total of 207 million reads were generated for all samples of which approximately 200 million reads were of high quality. All the reads were pooled to generate a 95.7 Mb assembly comprising of 46,599 transcripts of which 72.5% could be annotated. The reads were mapped back to the assembly, and the differential expression analysis was performed using DESeq tool. A total of 207 transcripts were found up-regulated and 547 transcripts down-regulated in axenic nematodes as compared to the symbiont monoxenic nematodes, whereas 13,584 transcripts were found only in monoxenic nematodes and 152 were specific to axenic nematodes. A subset of these differentially expressed transcripts was possibly involved in the regulation of symbiosis with the bacteria. Few of the differentially expressed transcripts were suggested to be involved in cell recognition and adhesion (for example, Plexin-2, membrane components, Contactin-associated protein-like 5); nematode innate immunity pathways (for example, turtle homolog B, few LRR genes, *lec-8*); carbohydrate-binding (C-type lectin domain-containing protein 87). All these processes are important in context of symbiosis with *Photorhabdus*. Efforts are ongoing to validate the role of these genes in nematode-bacterium symbiosis using functional genomic tools.

Keywords: *Heterorhabditis* - *Photorhabdus* - Symbiosis - Transcriptome - RNA-Seq.

Improving the draft genome of rice root-knot nematode *Meloidogyne graminicola* by long-read sequencing.

Vishal Singh Somvanshi (vishal.somvanshi@icar.gov.in), Uma Rao

Division of Nematology, ICAR-Indian Agricultural Research Institute, New Delhi, Delhi, India

The rice root-knot nematode *Meloidogyne graminicola* is a major biotic stress for the rice crop under upland, rain-fed lowland and irrigated rice cultivation conditions [1]. *M. graminicola* infestation is known to kill the rice nurseries and reduces the quality and vigour of rice seed [2]. However, similar to other plant-parasitic nematodes, there is a scarcity of management options for *M. graminicola*. Identification of newer targets using the 'omics' information is a promising approach to develop new nematode management strategies. Previously, we generated the first draft genome for *M. graminicola* using the Illumina GAIIx platform in 2018 [3]. The genome assembly size was 38.18 Mb, and a total of 4,304 contigs were reported with N50 of 20.4 Kb. Genome completeness assessment (BUSCO) showed that out of 303 conserved eukaryote genes, 73.6% were fully present, 15.2% were partially mapped and 11.2% were missing [3]. Here, we used the long-read sequencing approach (PacBio Sequel platform) and improved the draft genome of *M. graminicola*. A total of 7 Gb of raw reads were generated, and the preliminary Falcon assembly size was 55 Mb with 1388 contigs and N50 value of 61.9 kb. Further improvement in the assembly by using Falcon Unzip, purging haplotigs, and two rounds of polishing by using short, and short and long reads, respectively, resulted in the new genome assembly of 36.86 Mb size including 514 contigs, with N50 of 105 kb. The genome completeness (BUSCO) showed complete presence of 82% genes, 6.6% genes as partial and 10.5% as missing. Gene prediction using GenMark ES predicted ~10,500 genes, and 50% of predicted genes could be annotated using *C. elegans* database including 2,675 unique full length homologues of *C. elegans* proteins. We are now doing the *M. graminicola* genome annotation using Ref-Seq protein database, Interpro, KAAS, COG along with the tertiary analysis to find out more about the genes and processes that enable *M. graminicola* to become a successful parasite of rice-wheat cropping system.

Keywords: *Meloidogyne graminicola* - Rice root-knot nematode - Draft genome - PacBio Sequel - Long-read sequencing.

References:

- [1] Mantelin et al., 2017. Mol Plant Pathol. 18(1): 3-15.
- [2] Patil et al., 2014. Nematology. 16(5): 555-564.
- [3] Somvanshi et al., 2018. J Nematol. 50(2): 111-116.

A draft genome for the entomopathogenic nematode *Heterorhabditis indica.*

Vishal Singh Somvanshi (vishal.somvanshi@icar.gov.in), Chaitra Ganapati Bhat, Uma Rao

Division of Nematology, ICAR-Indian Agricultural Research Institute, New Delhi, Delhi, India

Entomopathogenic nematodes (EPNs) of the genus *Heterorhabditis* live in a mono-specific symbiotic relationship with *Photorhabdus* bacteria. Together, they are highly virulent insect-pathogens and are used for the biological control of agricultural insect pests. *Heterorhabditis indica* is the most prevalent strain of the genus in India and is predominantly found in soils of the tropical regions of the world. It is also one of the most available commercial entomopathogenic nematodes. We sequenced the genome of *H. indica* to create a genomic resource to facilitate comparative and functional genomics. An inbred line of *H. indica* strain H-V was developed by inbreeding the nematode for 20 generations and used for genome sequencing. Two genomic libraries with insert sizes of 300 bp and 600 bp were prepared for Illumina paired-end sequencing to generate approximately 50x coverage, and one mate-pair library for 5 kb fragments was prepared to target 30x coverage to achieve a comprehensive assembly. A total of 160 million reads (23 GB) of raw sequence data was generated, of which 155 million reads were of high quality and used for creating the genome assembly by SOAPdenovo. The final *H. indica* genome assembly was 91.26 Mb size, comprising 3,538 scaffolds with the N50 value of 217 Kb. The minimum and maximum scaffold lengths were 501 bp and 2,409 Kb, respectively. The GC content of the assembled genome was 35.31%, and there were 7.3% N's in the assembly. The genome completeness assessment by BUSCO showed that out of 303 conserved eukaryote genes, 87.78% were fully present, 4.95% were partially mapped and 7.26% were missing, suggesting that the draft genome is of high quality. SNAP, Augustus, Genemark, and Maker tools were used to predict 10,974 genes from the assembly, of which 8,758 could be annotated using the RefSeq database. Further tertiary analysis of the genome is ongoing. This study would facilitate genomic exploration, and functional and comparative genomic studies in *Heterorhabditis* nematodes leading to insights on nematode-bacterium symbiosis and specificity, as well as other interesting aspects of entomopathogenic nematode biology.

Keywords: Entomopathogenic nematodes - *Heterorhabditis indica* - Genome - Resource - Illumina.

A *Meloidogyne graminicola* effector interacts with a novel Cu metallochaperone protein to improve Cu/Zn-SOD activities and suppress plant immunity.

Handa Song (jlliao@scau.edu.cn), Borong Lin, Qiuling Huang, Jiansong Chen, Lili Hu, Longhua Sun, Kan Zhuo, Jinling Liao

Laboratory of Plant Nematology, South China Agricultural University, Guangzhou, China

Effectors originating from the esophageal gland cells of root-knot nematodes can suppress the host immune responses essential for parasitism by nematodes, but the detailed molecular mechanisms involved in plant immunity suppression of nematode effectors are still to be discovered in the most cases. In this study, we functionally characterize a novel effector MgMO289 from *Meloidogyne graminicola*, which is exclusively expressed in the dorsal esophageal gland cell and markedly up-regulated in parasitic third/fourth-stages juveniles (7 days post-infection). *In planta* RNAi targeting *MgMO289* attenuated the parasitic ability of *M. graminicola.* Meanwhile, transgenic rice lines expressing *MgMO289* showed more susceptible to *M. graminicola*. Yeast two-hybrid and bimolecular fluorescent complementation (BIFC) showed that MgMO289 interacts with the rice heavy metal associated domain containing protein (OsHPP04). The expression of *OsHPP04* is induced by the infection of *M. graminicola* by the β-glucuronidase (GUS) staining. OsHPP04 was considered to be a metallochaperone protein involved in metal ions binding, the yeast expressing *OsHPP04* assay in this study confirm that OsHPP04 is indeed involved in Cu binding. Interestingly the copper/zinc superoxide dismutase (Cu/Zn-SOD) activity is enhanced in *MgMO289* transgenic lines compared to WT plants under Cu stress condition. Moreover MgMO289 suppresses host basal immunity and cell death induced by Gpa2/RBP-1, and O2- content are significantly lower in transgenic lines expressing *MgMO289* by nitroblue tetrazolium (NBT) staining. Considering that metallochaperones can traffic metal ions to the metalloenzymes including Cu/Zn-SOD, activating metalloenzymes, and SOD plays an important role in protecting cells from oxidative damage by scavenging reactive oxygen species (ROS), it is summarized that the effector MgMO289 promotes the parasitism of *M. graminicola* probably by interacting with OsHPP04, causing a Cu binding and trafficking, then enhancing the activity of Cu/Zn-SOD and promoting ROS degradation, and suppressing host immunity.

Exploring the Role of RanGAP2 in Recognition by the Potato CC-NB-LRR immune receptors Rx1 and Gpa2.

Octavina Sukarta (octavina.sukarta@wur.nl), Amalia Diaz-Granados, Erik Slootweg, Jaap Bakker, Geert Smant, Aska Goverse

Laboratory of Nematology, Wageningen University, Wageningen, The Netherlands

The RanGAP GTPase activating protein 2 (RanGAP2) has long been established as a co-factor of the potato CC-NB-LRR immune receptor Rx1. However, its role in defence by Rx1 remains to be fully elucidated. Artificial tethering studies of RanGAP2 to the nematode effector of GpRbp-1 was shown previously to enhance the cell death response of Gpa2, which is a closely related immune receptor of Rx1 that also interacts with RanGAP2 as a co-factor (Sacco et al., 2009). Such findings hint that RanGAP2 may contribute to immunity by facilitating effector recognition. Here, we expand further on this model using a combination of structure-informed approaches, including co-immunoprecipitation assays and cellular imaging studies. We show that RanGAP2 can in fact form complexes *in planta* with the cognate effector of Rx1 and Gpa2, the coat protein of PVX (PVX-CP) and secretory protein GpRbp-1 respectively. Interestingly, this interaction was observed for both the eliciting and non-eliciting variants of PVX-CP and Gp-Rbp1 suggesting that the association of RanGAP2 and the effectors alone may not be sufficient to confer recognition. This is in line with existing data demonstrating that the C-terminal region of the Rx1 and Gpa2 LRR domain is required for successful perception of the pathogen (Slootweg et al., 2013). Currently, we are performing structural and biochemical analysis to explore a model whereby RanGAP2 acts as a bait that facilitates the specific recognition of PVX-CP and Gp-Rbp1 either by bringing the effector in closer proximity to the cognate LRR domain or by enhancing the binding affinity of the LRR/effector interaction. This model would also explain the bifurcation of pathogen recognition specificities of these two closely related plant immune receptors during evolution.

Keywords: NB-LRR - Virulence target - Recognition - RanGAP2.

References:

- Sacco et al., 2009. PLoS Pathogens. 5(8):e1000564.
- Slootweg et al., 2013. Plant Physiology. 162(3):1510-28

Association of *Caenorhabditis inopinata* with the wasp and bacteria in the fig ecosystem.

Ryusei Tanaka[1] (caenorhabditisjaponica@gmail.com), Tanzila Afrin[2], Taisei Kikuchi[1]

[1] *Parasitology laboratory, Miyazaki university, Miyazaki, Japan*; [2] *Department of pathology and molecular medicine, Queen's University, Kingston, Ontario, Canada*

Caenorhabditis inopinata is a new *Caenorhabditis* species recently discovered from syconia of the fig *Ficus septica* in Ishigaki island Japan. DNA sequence analyses revealed that *C. inopinata* is the most-closely-related sister species of *C. elegans*, but the new nematode shows ecological features very distinct from *C. elegans*. For example, the main proliferating niche of *C. inopinata* is fresh fig syconia and the nematodes are likely vectored by fig wasps from old to new syconia, whereas *C. elegans* is mainly found in leaf litter, rotten fruits, decomposing plant stems and only occasionally found in insects. Fig trees are well known for their obligate mutualistic relationship with the species-specific pollinating wasps. In this study, we aimed to clarify *C. inopinata* population dynamics in the well-established fig ecosystem. We found that *C. inopinata* proliferates mainly in the early stages of male fig syconia. Ratio of the dauer stage increased in the late stage of male syconia when the wasps develop to adults. The dauer nematodes were detected from flying female wasps that escaped from old fig syconia but not frequently from the male wasps. We also investigated bacteria community structure in the fig syconia using 16S rRNA-based microbiota. Subsequent changes in the microbiota was observed along the fig stage development. The PCoA analysis clusters young stage of fig syconium microbiota distant from the other stages. The wasp and the late stage of fig syconia share several bacteria taxa that were not found in the early stage, suggesting that the wasp plays vector roles for bacteria as well as for nematodes. Moreover, overgrowth of specific bacterial taxa such as *Rhodobacter* sp. was found in the late stage of syconia, which may be related to the emergence of dauer nematodes in those syconia. These results taken together suggest that *C. inopinata* has a commensalistic relationship with the fig-wasp ecosystem.

Keywords: Vector - Commensal - Fig - Caenorhabditis - Bacteria.

A Quick Dip: Alternatives to hot-water dipping to control *Meloidogyne hapla* in daylily (*Hemerocallis)* production.

Emilie Cole (coleemi1@msu.edu), Kristin Poley, Marsiol Quintanilla, Rambika Thapa

Entomology, Michigan State University, East Lansing, Michigan, USA

In bare-root ornamental daylily (*Hemerocallis*) production, the northern root knot nematode (*Meloidogyne hapla*; NRKN) is of great economic importance and can be difficult to manage due to being grown under field conditions. New plantings of daylily are established from divisions of mature plants, and since these mature plants have been under field conditions, they frequently have moderate NRKN infestation. To prevent further spread of the nematode, a hot water dip pre-plant procedure is often utilized. The current standard involves heating the roots from 88F to 107.5F over the course of 1.5 hours after which they are dipped twice more at 55F for 20 minutes and another 10 minutes. Although this method is effective in reducing NRKN root populations it can result in upwards of 30% plant mortality. Therefore, our objective was to evaluate alternative methods to hot water dipping to reduce NRKN and prevent dip related mortality. Under greenhouse conditions, eleven treatments were tested alongside an untreated control on two daylily varieties, “Rose Katherine” and “Stolen Treasure”. Treatments included the standard hot water dip, 4 hot water dip modifications, standard dip with bleach, two ultra-violet light alternatives, and two chemical nematicide dips. For each variety, treatments were replicated eight times, and the plants were monitored for plant growth, mortality, and damage over the course of 8 weeks. At the end of the experiment roots were collected from each plant and stained to determine NRKN populations. Plant height was greatest for both varieties when treated with the Standard Dip, followed by Velum, and UV Treatment #1. Nematode populations in the “Rose Katherine” variety were significantly lower than the control when treated with bleach, UV treatment #2 and the Standard Dip. For “Stolen Treasure” none of the treatments significantly differed from the control however UV treatment #2 was numerically the lowest in nematode counts. In conclusion, the standard dip worked well in reducing nematodes, however utilizing an alternative method such as UV light or Velum may be equally as effective. The next steps will be to test these treatments in a commercial setting to determine if they’re not only effective but easily executed.

Keywords: Root-knot nematode - Ornamental - Daylily - Hot-water dip.

Genomic features of the rice root-knot nematode *Meloidogyne graminicola.*

Thi Ngan Phan[1] (ngan.phan-thi@ird.fr), Julie Orjuela[1], Laetitia Perfus-Barbeoch[2], Djampa Kozlowski[2], Etienne Danchin[2], Christophe Klopp[3], D. Georgios Koutsovoulos[2], Celine Lopez-Roques[3], Olivier Bouchez[3], Guillaume Besnard[4], Bellafiore Stephane[1]

[1] *IPME, IRD, Cirad, University of Montpellier II, Montpellier, France*; [2] *Institut Sophia Agrobiotech, INRAe, Université Côte d'Azur, CNRS, Antibes, France*; [3] *Plateforme Génomique, US 1426 GeT-PlaGe, INRAe, Toulouse, France*; [4] *UMR5174, EDB,, CNRS-UPS-IRD, Toulouse, France*

Meloidogyne graminicola (*Mg*) is a root-knot nematode (RKN) species which is considered to be a major threat to rice production. Yet, this pest has only been recently discovered, and its origin, genomic structure, and intraspecific diversity are still poorly understood. Although a short-read based *Mg* genome assembly has been recently released, it is incomplete and fragmented. In this study, sequencing data from both Oxford Nanopore and Illumina technologies were combined to offer a new genome assembly with a mean sequencing depth of 160×, and considerable improvement in scaffold metrics (*i.e.* 306 scaffolds, 42.1 Mb). . Our genome assembly obtained the largest N50 scaffold (294 Kb) and highest CEGMA completeness score among *Meloidogyne* spp. up to date. In addition, flow cytometry indicated that the total nuclei DNA of *Mg* genome is of ca. 94 ± 10 Mb, more than twice our consensus assembly. The *k*-mer analysis on the nuclear genome sequences and the detection of divergent homologous genomic sequences indicate that the *Mg* genome is highly heterozygous (> 1%), and this has never been reported in facultative meiotic parthenogenetic RKNs. We predict 10,307 protein-coding genes bearing the *pfam* domain (spanning 74.6% of the genome assembly). Among them, 67 genes are identified as putative horizontal gene transfers from bacteria and fungi, which supposedly contribute to the nematode's adaptation including the plant cell wall degradation, nutrient progress, and plant defense manipulation. In addition, 585 transposable elements (TE) belonging to seven specific TE orders are detected. Annotated TEs occupy 2.69% of the genome length and could promote genomic reorganizations putatively related to the evolution of plant parasitism in *M. graminicola*. The high-quality *Mg* genome assembly reported here will be a valuable molecular resource for future genetic studies of *Meloidogyne* species, especially to deepen comparative genomic studies, and more specifically, to retrace the evolutionary history of *M. graminicola* and its closest relatives.

Keywords: Genome reference - Root-knot nematode - Horizontal gene transfers - Transposable elements - Flow cytometry.

References:

- Besnard et al., 2019. Genes. 10(2):175.
- Blanc-Mathieu et al., 2017. PLOS Genetics. 13(6):e1006777.
- Somvanshi et al., 2018. Journal of Nematology. 50(2):111:116
- Haegeman et al., 2011. Mol Plant Microbe Interact. 24(8):879-87

Influence of root exudates on suppression of root-knot nematodes by *Pasteuria penetrans*.

Chang Liu[1] (c.liu@ufl.edu), Patricia Timper[2]

[1] *Entomology and Nematology Department, University of Florida, Gainesville, Florida, United States*; [2] *USDA ARS, Tifton, Georgia, United States*

Pasteuria penetrans is an endospore-forming bacterium parasitic on *Meloidogyne* spp. Spores of *P. penetrans* adhere to the surface coat of second-stage juveniles (J2). Previously, we showed that past exposure of J2 to root exudates reduced attachment of *P. penetrans* spores, and this reduction persisted at least 24 hours following exposure, suggesting that the surface coat of the nematode was altered by the exudates [1]. These results have implications for biological control: J2 encountering spores within the rhizosphere may be more resistant to *P. penetrans* than J2 encountering spores outside the rhizosphere. We hypothesized that seed applications with *P. penetrans* spores will be less effective than in furrow applications in reducing *Meloidogyne arenaria* egg production. The experiment was conducted in the greenhouse with three treatments: control (no spores), in furrow application of spores, and seed application (spores applied to the planting hole after seed placement). Soil was infested with *M. arenaria* by mixing root pieces containing egg masses into soil prior to planting peanut seed. Each treatment was replicated seven times and the experiment was conducted twice. Eggs of *M. arenaria* were counted 30 days after seedling emergence. Compared to the control, both the seed and in furrow application of *P. penetrans* reduced egg production by *M. arenaria*; however, significantly fewer eggs were produced in the furrow than in the seed application (51% vs 37% reduction compared to the control). The in-furrow application of *P. penetrans* spores may be more effective in suppressing *M. arenaria* because more spores are distributed outside the rhizosphere than in the seed application.

Keywords: Biological control - Pasteuria penetrans - Root exudates - Root-knot nematodes.

References:

- [1] Liu, et al., 2017. J. Nematol. 49:304-310

Interplay between plant circadian clock and biotic stresses: from *Arabidopsis* to Soybean.

Wang Xingwei[1] (pkuwangxw@pku.edu.cn), Cao Lijun[2], Maier Tom[2], Masonbrink Rich[3], Li Meina[2], Severin Andrew[3], Zhou Mian[4], Baum Thomas[2], Wang Wei[1]

[1] *School of Life Sciences, Peking University, Beijing, China*; [2] *Plant Pathology and Microbiology, Iowa State University, Iowa, United States*; [3] *Genome Informatics Facility, Iowa State University, Iowa, United States*; [4] *College of Life Sciences, Capital Normal University, Beijing, China*

The plant circadian clock has definitely become an integrative hub that coordinates plant immunity and other biological processes for growth and development. Recently, the plant circadian clock has showed its crucial relationship with plant pathogens, but all the discoveries regarding the interactions were made through studies on leaf pathogens only. Virtually nothing is known about the relationship between the root circadian clock and root pathogens. However, soil is where an extraordinary diversity of microbes actually resides, and roots are at the very battle front against many soil pathogens and pests. To fill this critical knowledge gap, we initiated a comprehensive study on the bidirectional interactions between soybean root circadian clock and soybean cyst nematodes (SCN). Through establishment and application of the soybean molecular timetable, we found that SCN may specifically perturb soybean circadian rhythm in syncytia. We validated this computational prediction and further pinpointed soybean clock genes targeted by SCN using transgenic soybean hairy root harboring circadian clock gene reporters. While SCN was shown to perturb soybean circadian rhythm, soybean circadian clock also has profound impact on SCN. Through diurnal and circadian time-course infection assay, we found that soybean's resistance against SCN showed both diurnal and circadian rhythm. Using a soybean clock mutant, we showed that these rhythms are dependent on soybean circadian clock. To further dissect the interplay between soybean root circadian clock and SCN, we performed a large-scale time-course circadian transcriptomics analysis. Through this comprehensive transcriptomics analysis, we have uncovered the detailed transcriptome changes of both soybean and SCN during their interaction. Taken together, our study suggests an intimate interplay between soybean circadian clock and SCN.

Keywords: Plant circadian clock - Soybean cyst nematode - Transcriptomics - Bioinformatics.

Variation in biological and molecular responses of *Meloidogyne* spp. to post-plant nematicides.

Catherine Wram[1] (wramc@oregonstate.edu), Inga Zasada[2]

[1] *Oregon State University, Corvallis, OR, United States*; [2] *USDA ARS HCRU, Corvallis, OR, United States*

Despite being a pervasive and economically important pest, there are a limited number of methods to control plant-parasitic nematodes (PPN). Chemical controls are the primary means of managing PPN, with more than $1 billion a year of the nematicide market spent on suppressing *Meloidogyne* species. Recently, several new nematicides have entered the market including fluazaindolizine, fluopyram and fluensulfone. There is limited research on their effects on *Meloidogyne* species. To ensure proper stewardship of these compound, knowledge of how *Meloidogyne* spp. respond to these compounds at the biological and molecular levels is required. In this study, a microwell assay system was used to generate 24-hr dose-response curves for *M. incognita* second stage juveniles (J2) to post-plant nematicides (fluazaindolizine, fluopyram, fluensulfone, and oxamyl). The 24-hr effective doses (ED) that resulted in the immobility of 50% and 100% of *M. incognita* J2 were then used to perform a gene expression study with the post-plant nematicides and a water control. Additionally, the response of three *Meloidogyne* species (*M. incognita, M. chitwoodi,* and *M. hapla*) and multiple populations within a species to fluazaindolizine were determined in laboratory and greenhouse studies. Greater than 7,000 genes were differentially expressed in post-plant nematicide treated *M. incognita* J2. Treatments with fluazaindolizine had >2,000 more differently expressed genes when compared with the other nematicide treatments. *Meloidogyne incognita* treated with fluensulfone and fluazaindolizine had the most differently expressed genes in common, with 1,400 and 3,600 genes in the ED50 and ED100 treatment, respectively. Oxamyl treated *M. incognita* J2 had enrichment for genes involved in ion channel activity and neural calcium censors. Fluopyram treated J2 had an enrichment of differently expressed genes with GO terms that were integral membrane components. For fluazaindolizine, enrichment of genes involved in oxidation-reduction and oxidoreductase activity was observed. While fluazaindolizine suppressed all of the *Meloidogyne* species and populations, there was variation in response. Of the *Meloidogyne* species examined*, M. incognita* was the most sensitive to fluazaindolizine, with an ED50 2X lower than for *M. chitwoodi*, and 3X lower than that for *M. hapla*. Higher rates of fluazaindolizine were required to suppress reproduction of *M. hapla* and *M. chitwoodi* on tomato compared to that of *M. incognita*.

Keywords: Nematicides - RNAseq - Root-knot nematode.

Diversity of nematodes in the family Tripylidae de Man, 1876 in Shanxi Province, North China.

Yu Mei Xu[1] (ymxu0310@163.com), Xiaohua Hao[1], Meina Liu[1], Zengqi Zhao[2], Jianming Wang[1]

[1] *Laboratory of Nematology, Shanxi Agricultural University, Taigu county, Shanxi Province, China*; [2] *Manaaki Whenua - Landcare Research, Auckland, New Zealand*

The family Tripylidae de Man, 1876 is a predominantly aquatic group of nematodes, with species found on land, continental water and occasionally in sea. Traditionally the family includes five genera: *Tripyla* Bastian, 1865 (=*Promononchus* Micoletzky, 1923, *Paratripyla* Brzeski, 1963), *Tripylella* Brzeski & Winiszewska-Ślipińska, 1993, *Tripylina* Brzeski, 1963, *Trischistoma* Cobb, 1913 and *Tobrilia* Andrássy, 1967 [1]. To date, there are 34 valid species in *Tripyla*, 11 species in *Tripylella*, 22 species in *Tripylina,* 17 species in *Trischistoma* and two species in *Tobrilus*. Since March 2011, a total of 930 soil and litter mixture samples from native forests, agricultural land, fruit orchards and grassland have been collected and examined from 20 locations in Shanxi province, China. The nematodes were obtained from the samples using the Whitehead and Hemming [2] tray method. For morphological study, nematodes were killed and fixed using hot, 3% formaldehyde, and left to harden for at least two weeks. All nematodes were processed to glycerol, and mounted on glass slides, as described by Southey [3] and modified by Davies and Giblin-Davis [4]. DNA extraction, PCR, sequencing and phylogenetic analysis were conducted as described in Zhao & Buckley [5]. Based on morphological and molecular studies, six species in three genera were found from Shanxi Province: *Tripyla aquatica* Brzeski & Winiszewska-Ślipińska, 1993, *T. setifera* Bütschli, 1873, *Tripylina puxianensis Xu et al.,* 2013, *Tr. zhejiangensis* Pham *et al.,* 2013, *Trischistoma taiguensis* Xu *et al.,* 2015 and *Tri. Pellucidum* Cobb, 1913. Among these, *Tripylina zhejiangensis* was the predominant species of Tripylidae. The natural habitat includes fruit trees, herbs and garden plants. Our molecular phylogenetic results further indicated that the genera *Tripylina* and *Trischistoma* are sisters to*Trefusia* de Man, 1893 and is more closely related to Enoplida than to Triplonchida.

Keywords: Tripylidae - *Tripylina* - *Tripyla* - *Trischistoma* - *Tripylella*.

References:

- [1] Andrássy 2007. Hungarian Natural History Museum and Systematic Zoology Research Group of the Hungarian Academy of Sciences: 419-423.
- [2] Whitehead & Hemming, 1965. Annals of Applied Biology. 55: 25-38.
- [3] Southey 1986. MAAF Reference Book 402. 59-80.
- [4] Davies & Giblin-Davis 2004. 18(3): 291-319.
- [5] Zhao & Buckley 2009. Zootaxa 2238: 25-32.

A novel venom allergen-like protein from *Meloidogyne incognita* suppresses plant defenses and promotes parasitism.

Jiarong Yu (786365782@qq.com), Renpeng Zhou, Qing Yuan, Xuan Wang, Hongmei Li

Department of Plant Pathology, Nanjing Agricultural University, Nanjing, China

Meloidogyne incognita infects almost all vascular plants, and is generally considered to be one of the most widespread and damaging plant-parasitic nematodes. It can secret effector proteins into host cells to facilitate successful parasitism. The venom allergen-like protein (VAP) family is demonstrated to be involved in nematode infection. We identified a novel VAP effector from *M. incognita*, named Mi901, which was expressed in the nematode esophageal gland cells and up-regulated in the late parasitic stages. *Tobacco rattle virus*-mediated gene silencing of *Mi901* significantly reduced *M. incognita* infection, indicating Mi901 may play an important role in plant-nematode interaction. In addition, ectopic expression of Mi901 in Arabidopisis suppressed the burst of reactive oxygen species (ROS) and the expression levels of defense-related genes triggered by the flg22, and subsequently promoted the infection of *Botrytis cinerea* in Arabidopsis leaves. The yeast two-hybrid assays revealed an interaction between Mi901 and an Arabidopsis papain-like cysteine protease(PLCP) RD21A and this was further confirmed by split-luciferase complementary assay in *N. benthamiana*. Moreover, we found that Mi901 inhibits the proteolytic activity of RD21A. Our results suggest that the VAP effector Mi901 may be able to enhance plant susceptibility by targeting a host PLCP to modulate plant immune responses.

Keywords: *Meloidogyne incognita* - Venom allergen-like protein - Secretion - Papain-like cysteine protease - Plant immune responses.

References:

- [1] Lozano-Torres et al., 2014. PLoS Pathog. 10(12): e1004569.
- [2] Misas-Villamil et al., 2016. New Phytol. 212(4): 902-907.

Identification the mechanisms of *Trichoderma longibrachiatum* T6 against *Heterodera avenae* by metabonomics and transcriptome.

Shuwu Zhang (zhangsw704@126.com), Bingliang Xu, Jia Liu

College of Plant Protection, Gansu Agricultural University, Lanzhou, Gansu, China

Heterodera avenae is a class of widely distributed plant parasitic nematodes in soil, and also is an important pathogen causing plant diseases, affecting the growth, yield and quality of crops which result in economic losses. Chemical nematicides are currently considered the main method to control nematode infection in agricultural crops. However, some chemical nematicides are deregistered due to toxicity to the environment and human. In view of the limitation of chemical control and the requirements of healthy crops, we discovered a strain of *Trichoderma longibrachiatum* T6 (TL-6) which had the ability to control *H. avenae*. Microscopic observation and chemical characterization found that TL-6 had strong parasitic and lethal effects on the cysts of *H. avenae*, including increased chitinase, glucanase and caseinase activity which served as the probable mechanism. Meanwhile, the interaction mechanism of TL-6 with cysts was analyzed and exploited by transcriptome sequencing. Our results indicated that TL-6 can lead to a number of differently expressed genes down-regulated in cysts, whereas the cysts can induce a number of differently expressed genes up-regulated in TL-6. The relative expression was highest at initial stage, as were the up-regulated genes that can code the activity of protease and chitinase, and increase the ability of TL-6 to dissolve the cyst body wall. Moreover, an efficient genome editing CRISPR/Cas9 system was constructed for TL-6 to identify the nematicidal genes function. In addition, six species of nematicidal compounds were isolated and identified from TL-6 by the methods of column chromatography, gas chromatography-mass spectrometry (GC-MS) and nuclear magnetic resonance (NMR). Our results clarified the parasitic and lethal mechanisms of TL-6 against *H. avenae* at the level of metabonomics and transcriptome. Furthermore, our results indicate the significance in accelerating the functional genomics research in *Trichoderma* spp., and improving the strain's function. Theyprovide a theoretical basis for developing new types of highly efficient microbial nematicidal agents in the future.

Keywords: *Trichoderma* spp. - *Heterodera avenae* - Parasitic and lethal effects - Nematicidal compounds and genes - CRISPR/Cas9 system.

References:

- Suarez et al., 2004. Applied Microbiology and Biotechnology. 65(1): 46-55.
- Yang et al., 2012. Journal of Asia-Pacific Entomology. 15: 647-650.
- Lora et al., 1995. Molecular Genetics and Genomics. 247(5): 639-645.
- Liu et al., 2015. Cell Discovery. 1: 15007.

The root-knot nematode MiPDI1 effector targets a zinc finger protein to establish disease in *Solanaceae* and Arabidopsis.

Zhao Jianlong[1] (zhaojianlong@caas.cn), Mejias Joffrey[2], Favery Bruno[2], Quentin Michaël[2], Chen Yongpan[3], Mao Zhenchuan[1], Jian Heng[3], Abad Pierre[2], Xie Bingyan[1]

[1] *Institute of Vegetables and Flowers, Beijing, China*; [2] *INRAe, Université Côte d'Azur, CNRS, ISA, Nice, France*; [3] *Department of Plant Pathology, China Agricultural University, Beijing, China*

Plant-parasitic nematodes (PPNs) are a great threat to agriculture. In recent decades, researches showed that PPNs secreted a large quantity of effectors from esophageal gland to promote parasitism, but their molecular mechanism is largely unknown. In this study, we characterized a novel effector, protein disulfide isomerase (PDI) -like protein from the plant-parasitic nematode *Melodogyne incognita* (MiPDI1), which assisted nematode parasitism. *In situ* hybridization showed MiPDI1 was expressed specifically in the subventral glands of *M. incognita* and significantly up-regulated in the parasitic stages. Immunolocalization demonstrated MiPDI1 secretion in giant cells. *In planta* RNA interference (RNAi) of *MiPDI1* affected nematode infection ability, while MiPDI1 expressing in Arabidopsis increased susceptibility to *M. incognita*, providing evidence that MiPDI1 played pivotal roles in *M. incognita* parasitism. Moreover, MiPDI1 expression in Arabidopsis affected defense-associated genes expression. Yeast two-hybrid, BiFC and Co-IP confirmed that MiPDI1 interacted with a tomato zinc finger protein, orthologous to an Arabidopsis zinc finger protein that played important roles in plant abiotic and biotic stress responses. Our results suggested that MiPDI1 act as a pathogenicity factor promoting disease by fine-tuning with zinc finger protein-mediated responses at the interface of defense and stress acclimation.

Keywords: *Melodogyne incognita* - Effector - Protein disulfide isomerase - Zinc finger protein - Parasitism.

Bursaphelenchus eggersi or *B. hildegardae* (Nematoda: Parasitaphelenchidae)? Does it really matter to New Zealand?

Zengqi Zhao[1] (zhaoz@landcareresearch.co.nz), Michael Surrey[2], Wellcome Ho[2], Milen Marinov[3], Carolyn Bleach[3], Brett Alexander[2]

[1] *Systematics Team, Manaaki Whenua-Landcare Research, Auckland, New Zealand*; [2] *Plant Health & Environment Laboratory, Ministry for Primary Industries, Auckland, New Zealand*; [3] *Biosecurity Surveillance & Incursion, Ministry for Primary Industries, Auckland, New Zealand*

An unexplained mortality of Pinus radiata trees was observed in Kaingaroa Forest (KF) in the central North Island of New Zealand (NZ) during a forest health assessment survey conducted in March 2019. Close examination failed to determine the cause with no obvious fungal disease symptoms or insect damage. Subsequently, a group of three trees and a single dead tree were sectioned (cut into disks) and examined for nematodes. Nematodes were isolated from the wood disks and identified as *Bursaphelenchus hildegardae* Braasch et al., 2006. Before this finding, only two *Bursaphelenchus* spp. had been reported from NZ: *B. eggersi* Ruhm, 1956 and *B. fungivorus* Franklin & Hooper, 1962. In April 2019, the Ministry for Primary Industries (MPI) initiated a biosecurity investigation and a delimiting survey within KF to establish the area of spread and hosts for *B. hildegardae*. In total, more than 158 wood discs, 50 beetles and one soil sample were collected. Nematode identification, DNA extraction, PCR and sequencing protocols were conducted as described in Zhao & Buckley (1). In addition to fresh material, previously collected dried Hylaster ater Paykull, 1800 specimens were obtained from Scion New Zealand collections and analysed for the presence of nematodes to determine if any earlier records of *B. hildegardae* could be found. *Bursaphelenchus hildegardae* was isolated from both, wood samples and H. ater specimens, and identified based on morphology and molecular diagnostics. The dauer larvae of *Bursaphelenchus* sp. cf. *hildegardae* were also found from the preserved specimens collected in 2002. Since *B. hildegardae* and *B. eggersi* are morphologically similar and belong to the eggersi-group (2), it is questioned if the *B. eggersi* specimens from NZ that were identified by Dale (3) and *B. hildegardae* specimens from the present survey were conspecific. As *B. hildegardae* was only described in 2006, it is likely that Dale had isolated *B. hildegardae* in 1967 but identified these nematodes as the closest organism known at the time - *B. eggersi*. Dale isolated *B. eggersi* from the bark beetle H. aster in eight of 14 sites across the North and South islands of NZ. In the current study, *B. eggersi* was not found from the pine samples or *H. ater* specimens, suggesting that *B. hildegardae* may have been well established in NZ for a long time but previously misidentified. So far, the nematode has not been recorded to cause problems to pine trees.

Keywords: *Pinus radiata* - Disease - Taxomony - *B. hildegardae* - *B. eggersi*.

References:

- [1] Zhao & Buckley, 2009. Zootaxa. 2238, 25-32.
- [2] Braasch, 2001. Bulletin OEPP/EPPO. 31, 127-124.
- [3] Dale, 1967. New Zealand Journal of Science.10, 222-234.

7th International Congress of Nematology
ICN 2022
Antibes Juan-les-Pins - France

"Crossing borders: a world of nematode diversity and impact to discover"

European Society of Nematologists

LIST OF POSTERS

Posters

e-Posters online viewing

LIST OF POSTERS

S1. Plant-nematode interactions

SESSION 1 - Plant resistance and nematode virulence

Last name	First name	Title of poster	# poster
LETIA	Sharon	Deciphering the mechanism of ascarosides perception in plants	S1-PF1
OLAJIDE	Emmanuel	Screening of plantain and banana cultivars for resistance against *Radopholus similis* with prospects of using macropropagation plantlets for selection	S1-PF2
CHANNALE	Sonal	Identification of candidate resistance genes in chickpea *(Cicer arietinum)* against *Pratylenchus thornei* using GWAS	S1-PF3

SESSION 5 - Plant resistance and nematode virulence

Last name	First name	Title of poster	# poster
STORELLI	Alan	Investigation of resistance against *Ditylenchus dipsaci* on sugar beet	S5-PF1
MANTELIN	Sophie	Prospective identification through DNA-capture technologies of a rice resistance gene to control *Meloidogyne graminicola*	S5-PF2
KONIGANAHALLI GOPAL	Hemanth Kumar	Host status of Crop plants to *Meloidogyne enterolobii* populations	S5-PF3

SESSION 21 - Nematode-plant interactions

Last name	First name	Title of poster	# poster
COOMER	Alison	Trade-offs between virulence and breaking resistance in root-knot nematodes	S21-PF1
HASAN	M. Shamim	Parasitic worms redirect host metabolism via NADPH oxidase-mediated ROS to promote infection	S21-PF2
GUIMARAES	Patricia M.	Meta-analysis of wild *Arachis* transcriptome data unravels candidate genes for combined nematode and drought resistance	S21-PF3

SESSION 31 - Effectors in plant parasitic nematodes

Last name	First name	Title of poster	# poster
STOJILKOVIC	Boris	The root-knot nematode effector Mj-NEROSS suppresses plant immunity by interfering with the ROS production in plastids	S31-PF1
KUMAR	Anil	Studying root-knot nematode *Meloidogyne javanica* MAP-1 variation and their implication in parasitism.	S31-PF2
CHEN	Yujin	The *M. javanica* effector Mj-10A08 downregulates ethylene receptors via protein-protein interactions to facilitate tomato parasitism	S31-PF3

Last name	First name	Title of poster	# poster
ANTIPOLIS	Inra	Daf16like and Skn1like genes are reliable targets to develop biotechnological tools for control of Meloidogyne incognita	S1-P01
BOZBUGA	Refik	Determining genes conferring resistance against nematodes in Capsicum spp and relationship with capsaicin	S1-P02
BRITO	Janete	Vertical movement of Meloidogyne enterolobii as influenced by temperature and plant stimuli	S1-P03
CARTA	Lynn	Microscopy and organismal associations of Litylenchus crenatae in beech leaf disease of Fagus grandifolia in Ohio, USA	S1-P04
CASEY	Adam	Characterising tolerance to root knot nematodes in robusta coffee (Coffea canephora)	S1-P05
CLAUDIUS-COLE	Abiodun	Nodal vine cutting technique for assessing nematode resistance in yams	S1-P06
COPELAND	Rhys	Determining the distribution of Pratylenchus quasitereoides and Pratylenchus curvicauda in the WA Wheatbelt and understanding how they find host roots	S1-P07
COSTA	Sofia R.	HANDLER - Host adaptation and root-knot nematode selection by partially-resistant tomato rootstocks	S1-P08
DAHLIN	Paul	Biochemical aspects of plant sterols in plant nematode interaction	S1-P09

DE ALMEIDA ENGLER	Janice	Molecular markers for cell damage induced by root-knot nematodes	**S1-P10**
DESAEGER	Johan	A Multi-State Effort to Contain and Manage Meloidogyne enterolobii in Vegetable Crops	**S1-P11**
DUBE	Zakheleni	Acacia biochar promotes tolerance of tomato plants' to root-knot nematodes	**S1-P12**
EBRAHIM	Awol Seid	Heat stability of resistance in tomato breeding lines to M. incognita and M. javanica populations under soil temperature	**S1-P13**
ELEKCIOĞLU	Ibrahim Halil	The development of root lesion nematodes (Pratylenchus thornei, P. neglectus, P. penetrans) on some chickpea varieties	**S1-P14**
FARIA	Jorge	Element levels in wood from maritime pine infected by *Bursaphelenchus xylophilus* in field conditions	**S1-P15**
GARTNER	Ulrike	Virulence in UK Globodera *pallida* populations in relation to resistance and durability	**S1-P16**
GOMES	Cesar	Pathogenicity of Aphelenchoides besseyi populations from rice (Oryza sativa) to soybean (Glycine max) plants	**S1-P17**
GOVERSE	Aska	Bifurcation of pathogen recognition specifity and convergence of immune responses by the potato R genes Rx1 and Gpa2	**S1-P18**
GUARNERI	Nina	Root architecture plasticity in response to endoparasitic cyst nematodes is mediated by damage signaling	**S1-P19**
GUIMARAES	Patricia	Small RNAs (miRNAs) involved in wild Arachis resistance against Meloidogyne arenaria	**S1-P20**
HAYES	Ashley	The Plant Secretory Pathway During Cyst Nematode Infection	**S1-P21**
JONES	John	Uncovering the role of the syncytia-forming nematode core effector GrGLAND11 in *Globodera rostochiensis*	**S1-P23**
KRANSE	Olaf	Delivery of macro molecules to plant-parasitic nematodes	**S1-P25**
KUD	Joanna	An Insight into Host-Specific Behavior of Globodera spp. Hatched in Root Exudates from Potato and Its Wild Relative, Solanum sisymbriifolium	**S1-P26**
LOPEZ-NICORA	Horacio	Effect of Radopholus similis and Meloidogyne spp. on plant growth and yield of Musa AAB 'Dominico Hartón	**S1-P28**
MATTOS	Vanessa	Multi-resistant reactions of new resistant sources of Oryza spp. to root-knot nematodes from the graminis group	**S1-P29**
MAULANA	Muhammad Iqbal	The link between genetic diversity and pathogenicity variation in temperate root-knot nematode Meloidogyne hapla	**S1-P30**
MEIJER	Anikó	Inherited memories: Preparing the next generation for battle	**S1-P31**
MEYER	Susan	Host status of Salvia hispanica (chia) to Meloidogyne incognita and nematotoxic activity of root and shoot extracts	**S1-P32**
MIAMOTO	Angelica	Extract of Macrotyloma axillare 'Java' on hatching, penetration and development of Meloidogyne javanica in soybean plants	**S1-P33**
MIKAIA	Nona	Efficacy of Aphelenchoides sp. against Spruce bark beetle Ips typographus from Bakuriani (Georgia)	**S1-P34**
NOSKOV	Ilya	Distribution and damage potential of plant parasitic nematodes on medicinal and aromatic plants in Germany	**S1-P35**
OLAJIDE	Emmanuel	Characterization of plantain cv Agbagba infection with Radopholus similis and Meloidogyne spp. using macropropagated plantlets	**S1-P36**
PRETORIUS	Mathys C.	Characterization of T. semipenetrans in South African citrus orchards and baseline data for beneficial nematode communities in Citrus tree rhizospheres	**S1-P37**
RANTY-ROBY	Sarah	Identifying molecular plant functions targeted by root knot nematode nuclear effectors	**S1-P38**
ROBERTFARIA	Denner	Physiological and biochemical defense response of soybean cultivars parasitized by Pratylenchus brachyurus	**S1-P39**
ROS IBÁÑEZ	Caridad	Pathogeneicity of Meloidogyne incognita populations from pepper crops in southeast Spain to resistant pepper	**S1-P40**
SCHAVELING	Arno	Unravelling the genetic and molecular basis of North-West European Globodera pallida populations overcoming resistance in potato	**S1-P41**

SILVA	Santino Aleandro	What is the best inoculum density of Meloidogyne incognita to evaluate its reproduction factor in cotton?	**S1-P42**
SOULÉ	Salomé	Functional characterization of root-knot nematode effectors and their host targets during giant feeding cell ontogenesis	**S1-P43**
TOMAZ	Juarez	Arabidopsis thaliana phenotyping to Meloidogyne paranaensis interaction and identification of SNPs linked this trait	**S1-P44**
VAN GHELDER	Cyril	TIR-NBS-LRR genes in Prunus spp. offer a fantastic resource to decipher the molecular determinants involved in root-knot nematode resistance	**S1-P45**
VARANDAS	Raquel	Identification of promising genes on the resistance to Potato Cyst Nematodes	**S1-P46**
WANG	Xizhuo	Study on Pathogenicity of Bursaphelenchus xylophilus in Seedlings of Larix olgensis	**S1-P47**
WEI	Lihui	miRcn1 targets ethylene responsive transcription factor 4 (ERF4) to enhance resistance to root-knot nematodes in tomato	**S1-P48**
WILLIG	Jaap-Jan	The Arabidopsis transcription factor TCP9 regulates root growth response during cyst nematode infections via ROS-mediated signalling	**S1-P49**
XIAO	Fangming	Manipulation of host proteins and processes by potato cyst nematode effector RHA1B	**S1-P50**
PUTKER	Vera	Activation of Gpa2 by Globodera pallida RBP(P) variants is suppressed by RBP (S) effector variants: a molecular interplay between different RBP family members	**S1-P51**
CROW	William	Population dynamics and diagnosis of Hoplolaimus galeatus on hybrid bermudagrass	**S1-P52**
MENDY	Badou	A glycolytic enzyme from plant parasitic nematode induce defence responses in plants	**S1-P53**
NAFARI	Reza	An investigation of organophosphate nematicides' effects on Pratylenchus loosi, a root lesion nematode in tea plantations	**S1-P54**

S2. Systematics, phylogeny and phylogeography

Last name	First name	Title of poster	# poster
SESSION 6 - Phylogenetics/Phylogenomics: the latest updates on the Phylum Nematoda			
MATTOS	Vanessa	Genetic and phylogenetic characterization of different populations of *Meloidogyne izalcoensis*, a new species from coffee in Brazil	**S6-PF1**
KHAWLA	Mehalaine	Molecular identification and phylogenetic diversity of cereal cyst nematode (*Heterodera* spp.) populations from Algeria	**S6-PF3**
SESSION 24 - New challenges in nematodes taxonomy and evolution			
SINGH	P. Rolish	Morphological and molecular characterization of several *Paratylenchus* spp. from Belgium	**S24-PF1**
FADAKAR	Samira	An update to the identification compendium of *Aphelenchoides* Fischer, 1894 (Aphelenchoidea)	**S24-PF2**
TE MOLDER	Dennie	Unravelling the race complex: A first look Into the population genetics of the stem nematode *Ditylenchus dipsaci*	**S24-PF3**

Last name	First name	Title of poster	# poster
BEHMAND	TOHID	Occurrence and population dynamics of Ditylenchus dipsaci (stem and bulb nematode) on chickpea fields in Turkey	**S2-P01**
BLAXTER	Mark	A catalogue of nematode karyotypes	**S2-P02**
CAMACHO	Maria João	Epidemiological approach to potato cyst nematodes in Portugal, with special reference to Globodera pallida	**S2-P03**
CEDANO	Carolina	Determination of cyst nematodes affecting commercial fields of potato in julcan, la libertad, Peru	**S2-P04**
DA SILVA MATTOS	Vanessa	Integrative taxonomy of Meloidogye ottersoni (Thorne,1969) Franklin, 1971 parasitizing flooded rice in Bazil	**S2-P05**
ELEKCIOGLU	Ibrahim Halil	Molecular and biochemical identification of Meloidogyne spp. in East-Mediterranean Region of Turkey	**S2-P06**

FANELLI	Elena	Comparative analysis and evolutionary implications of exon-intron-structure of the hsp-90 gene in nematodes	**S2-P07**
GOMES	Cesar	Variability of Mesocriconema populations associated with Grapevine Decline Disease in South Brazil	**S2-P08**
HALLMANN	Johannes	Reproductive fitness, pathogenicity, morphometric and genetic variability among geographic isolates of Pratylenchus penetrans	**S2-P09**
HEYDARI	Fariba	Ektaphelenchus sp. (Rhabditida: Ektaphelenchinae), a tentative new member of the genus, from dead wood in north Iran	**S2-P10**
HUMPHREYS PEREIRA	Danny	Plant-parasitic nematodes associated with the Solanaceae family in Costa Rica	**S2-P11**
HUMPHREYS PEREIRA	Danny	New records of Globodera pallida in Costa Rica	**S2-P12**
JAHANSHAHI AFSHAR	Farahnaz	Ten populations of the genus Hoplolaimus Daday, 1905 (Hoplolaimidae), associated with date palm in south of Kerman province, Iran	**S2-P13**
KIDANE	Selamawit	Characterization of nematode pests of enset (Ensete ventricosum welw. Cheesman) and their management	**S2-P14**
KIDANE	Selamawit	Occurrence of plant-parasitic nematodes on Ensete ventricosum in Ethiopia with focus on Pratylenchus goodeyi	**S2-P15**
MARQUEZ	Josiah	Identification of Meloidogyne floridensis populations and their virulence on vegetables in Georgia, USA	**S2-P16**
MENNAN	Sevilhan	Detection of Root-Knot Nematodes in Northern Iraq	**S2-P17**
NJEŽIĆ	Branimir	Morphological characterization of Race A and Race B of Meloidogyne hapla	**S2-P18**
NÚÑEZ RODRÍGUEZ	Lester	Morphological and molecular characterization of Heterodera trifolii in Costa Rica, and pathogenicity on three plants species	**S2-P19**
PALOMARES-RIUS	Juan Emilio	Synonymization of Rotylenchulus macrosoma Dasgupta et al., 1968 with R. borealis Loof & Oostenbrink, 1962 and its ecology and phylogeography	**S2-P20**
PEDRAM	Maid	Proposal for a new Xiphinema Cobb, 1913 (Longidoridae) species from Iran belonging to the X. americanum-group	**S2-P21**
PEDRAM	Majid	Proposal for a new species of the genus Sphaerularia from Iran	**S2-P22**
PEDRAM	Majid	An unknown species of the genus Longidorus Mikoletzky, 1922, associated with beech trees in natural forests of northern Iran	**S2-P23**
PEDRAM	Majid	Proposal for a new species of the genus Sphaerularia Dufour, 1837 (Sphaerulariidae) from Iran	**S2-P24**
RANA	Aasha	Morphological and molecular characterization of Acrobeloides saeedi Siddiqi, De Ley and Khan, 1992 (Rhabditida, Cephalobidae) from India	**S2-P25**
RIASCOS ORTIZ	Donald Heberth	Plant-parasitic nematodes associated with Musa spp. crops in Colombia	**S2-P26**
ROLISH SINGH	Phougeishangbam	Uncovering diversity in plant-parasitic nematodes using morphological, molecular and phylogenetic approaches	**S2-P27**
SILVA	Santino A.	Morphological, morphometrical and oogenesis aspects of Tubixaba tuxaua	**S2-P28**
SUBBOTIN	Sergei	Recombinase polymerase amplification assay for rapid detection of the root-knot nematodes	**S2-P29**
TROCCOLI	Alberto	Occurrence of Pratylenchus species on raspberries in North Italy with morpho-molecular characterization of a new species	**S2-P30**
UYSAL	Gulsum	Plant parasitic nematodes associated with anise in Turkey	**S2-P31**
WEN	Yanhua	A new cyst nematode, Heterodera Luodingensis n. sp. (Nematoda: Heteroderidae) on Rice from Guangdong province, China	**S2-P32**
XIE	Chuanshuai	Comparative genomics reveals genome architecture and evolutionary adaptation in Meloidogyne	**S2-P33**
HUSTON	Daniel C.	Species diagnosis, hosts and distribution of cyst nematodes of the genus *Heterodera* (Tylenchida: Heteroderidae) with a focus on species of concern for Australia	**S2-P34**
AHMED	Mohammed	Addressing the challenges of High-throughput nematode identification using metabarcoding	**S2-P35**

S3. Biodiversity and ecology

Last name	First name	Title of poster	# poster
SESSION 3 - Ecology of free living nematodes			
MENNAN	Sevilhan	The relations between glyphosate and soil health based on nematode trophic groups in Turkish hazelnut orchards	S3-PF1
SHEPHERD	Rachel M.	Soil nematode communities around gopher tortoise burrows in native and degraded ecosystems	S3-PF2
HAN	Ziduan	CRISPR and mutagenesis experiments reveal mechanisms of integration of horizontally acquired cellulases in Pristionchus	S3-PF3
MEJÍA-MADRID	Hugo H.	Natural ecosystem diversity and functioning of nematode communities in a semi-desert ecosystem in Mexico	S3-PF4
SESSION 7 - Biodiversity of aquatic nematodes			
GATTONI	Kaitlin	Tobrilidae communities in western Nebraska sandhill lakes are driven by alkalinity and biotici interactions	S7-PF1
POWERS	Kirsten	Nematode predators catalyze an increase of chloroviruses by foraging on the symbiotic hosts of zoochlorellae	S7-PF2
VIEIRA	Soraia	Spatial distribution patterns of microbiome and free-living benthic nematodes in response to sediment ecological conditions in Sado estuary, Portugal	S7-PF3
SESSION 18 - Nematodes as bioindicators			
KAKOULI-DUARTE	Thomaé	A study on the ecological impact of recycling derived fertilisers (RDFs) using nematodes as environmental bioindicators	S18-PF1
HÖSS	Sebastian	Evaluating the environmental risks of microplastics using nematodes as bioindicators	S18-PF2
BRAIMI	Amina	Nematodes of argan biosphere: Biodiversity and assessment of soil quality	S18-PF3
SESSION 28 - Interactions of nematodes with micro-organisms			
MARCHENKO	Polina	Soil-born endophytic fungi antagonize plant-parasitic root-knot nematodes in tomato	S28-PF1
MCQUEEN	J. Parr	External and internal microbiomes of Antarctic dry valley nematodes are distinct, but more similar to each other than the surrounding environment	S28-PF2
TAÑAN	Veronica	Characterization of nematode-bacteria associations with gastropods and their virulence towards crop pests in northern Mindanao, Philippines	S28-PF3
SESSION 30 - Nematode community assemblies			
AKSAN	Muhammad	Nematode communities in organic and conventional rice production in Indonesia: a morphological and metabarcoding approach	S30-PF1
KNOETZE	Rinus	Association of beneficial terrestrial nematodes with glyphosate-tolerant and conventional soybean-based cropping system	S30-PF2
NGUYEN	Thi A. Duong	Research on free-living terrestrial nematodes (Order Dorylaimida) from tropical rain forest in Vietnam	S30-PF3

Last name	First name	Title of poster	# poster
ADDIS	Temesgen	Interaction of the bacteria Photorhabdus luminescens with life history traits and virulence of Heterorhabditis bacteriophora	S3-P01
AL BANNA	Luma	Anatomical Alterations in Aleppo Pine roots Induced by the dagger nematode Xiphinema vuittenezi	S3-P02
BASTIDAS	Brynelly	Community composition and metabolic footprints of soil nematodes in fruit systems in Mediterranean areas	S3-P03
BATCHELDER	Ellen	Nematode Parasite Loads on Ruminant Grazing Sites at Allen Island, Maine, USA	S3-P04

BELLO	Tesleem	Free-living nematode assemblages in the rhizosphere of watermelon plants in Nigeria: a baseline study	**S3-P05**
BELLO	Tesleem	Meloidogyne species and other plant-parasitic nematodes associated with watermelon in Nigeria	**S3-P06**
BETT	Silas Kipsang	Diversity of plant-parasitic nematodes associated with Crocus sativus L. in Morocco: relationships with edaphic factors	**S3-P07**
CAMACHO	Maria João	MaisSolo: nematodes as bioindicators of soil status	**S3-P08**
CHAUVIN	Camille	Nematode communities as bio-indicators of constructed soil: a case of study in a Mediterranean city, Montpellier	**S3-P09**
CHINNASRI	Buncha	Rice-Root Nematodes (Hirschmanniella spp.) and Rice Root-Knot Nematodes (Meloidogyne graminicola) in Takeo and Prey Veng Provinces, Cambodia	**S3-P10**
CLAVERO CAMACHO	Ilenia	Pin nematodes (Paratylenchus spp.) parasitizing Prunus in Spain: distribution, ecological factors and specific PCR for major species identification	**S3-P11**
CONCEIÇÃO	Isabel	Potato-cyst nematodes, Globodera spp., and root-knot nematodes, Meloidogyne spp., on potato in Portugal	**S3-P12**
DALAN	Loel	First report of Caenorhabditis brenneri (Nematoda: Rhabditida) association with the terrestrial slug Philippinella moellendorffi from the Philippines	**S3-P13**
DE OLIVEIRA	Clemen	Selective feeding and reproductive activities of a facultative plant-parasitic nematode (*Aphelenchoides besseyi*) and a fungal feeder (*A. pseudogoodeyi*) on isolates of fungi pathogenic and non-pathogenic to strawberry	**S3-P14**
DE SMIDT	Anke	Nematode survey of grassland habitats in two nature reserves of the Free State Province, South Africa	**S3-P15**
DIANO	Michelle Anne	Isolation and identification of bacteria associated with nematodes from Achatina fulica: an unexpected occurrence of anaerobic Clostridium lundense	**S3-P16**
DIAS-ARIEIRA	Claudia	Survival of Meloidogyne graminicola in soil under different moisture conditions	**S3-P17**
GANSFORT	Birgit	Should I stay, or should I go? Triggers of nematode dispersal	**S3-P18**
HAMMAM	Mostafa	Phytoparasitic nematodes of strawberry and their management in Egypt	**S3-P20**
HIGGINS	Rebecca	Linking barcodes, images and metadata to construct nematode reference libraries in BOLD, the Barcode of Life Database	**S3-P21**
JACKSON	Abigail	Tracing patterns of glacial refugia with scottnema lindsayae nematode	**S3-P22**
KATOOLI	Nafiseh	Morphological and molecular identification of species of Meloidogyne and distribution in pomegranate orchards of Iran	**S3-P23**
KNOETZE	Rinus	Glyphosate-tolerant and conventional soybean cultivars and the plant-parasitic nematodes associated with its rhizosphere	**S3-P24**
LAZNIK	Ziga	Chemotactic response and motility of mollusc parasitic nematode Phasmarhabditis papillosa toward mucus from different slug species	**S3-P25**
MBURU	Harrison	Potato Cyst Nematode Diversity and Adaptation in the Tropics: A case of Kenyan cropping system	**S3-P26**
PRETORIUS	Marné	In vitro and in vivo evaluation of fungal strains with biocontrol characteristics for their effects on the motility and reproduction of Meloidogyne incognita	**S3-P27**
PUŠKARIĆ	Josipa	Nematode biodiversity as a soil health indicator in agroforestry ecosystems	**S3-P28**
RUSINQUE	Leidy	Root-Knot Nematode Species Associated with Horticultural Crops in the Island of Azores, Portugal	**S3-P29**
RUSINQUE	Leidy	Assessment of plant-parasitic nematodes in Portuguese rice agro-systems: preliminary findings	**S3-P30**
SIRENGO	David Kihoro	Mitochondrial DNA-based identification of Meloidogyne spp. from pineapple roots and cutivated soils in Kenya	**S3-P31**
TRAP	Jean	Multiple-nutrient limitation of soil free-living nematodes in Ferralsols from natural grasslands in Madagascar	**S3-P32**
VILLENAVE	Cécile	Nematofauna analysis and ecotoxicological bioassay using C. elegans applied to soil toxicity assessment of three polluted sites	**S3-P33**

OLAORE	Deborah	Evaluation of carbofuran metabolism in three different soil types and effect of the metabolites on nematode population	S3-P34
PTATSCHECK	Christoph	Gone with the wind: The passive dispersal of nematodes	S3-P35
STOCK	S. Patricia	Nematode community patterns along an elevational gradient in the Santa Catalina Sky Islands in Arizona	S3-P36
GOVINDASAMY	Kavitha	Documentation and characterization of nematode biodiversity in Nilgiri forests of India for their functional role in soil health	S3-P37

S5. Integrated nematode management

Last name	First name	Title of poster	# poster
SESSION 2 - Soil suppressiveness and nematode control using cover crops			
HALLMANN	Johannes	Host status of different cover crops for three *Pratylenchus* species	S2-PF1
KOMBRINK	Anja	Using nematode community analysis to assess the resilience of agricultural soils in the Netherlands	S2-PF2
VAN HIMBEECK	Robbert	Characterization and steering of the native, soil microbiome-based suppression of plant-parasitic nematodes	S2-PF3
SESSION 8 - Nematode management in tropical conditions			
RUSINQUE	Leidy	EUPHRESCO – MELORISK: Preventing *Meloidogyne graminicola* spread in European rice paddies	S8-PF1
ASYIAH	Iis Nur	Dominance index of soil nematodes on a coffee plantation after cost-effective bionematicide application	S8-PF2
TORRINI	Giulia	Control of the rice root-knot nematode *Meloidogyne graminicola* using rice plants as trap crops	S8-PF3
SESSION 12 - Chemical control of nematodes			
STAMATAS	Yannis	Salibro™ (Reklemel™ active): A novel nematicide for the control of *Meloidogyne* spp. in key annual and perennial crops in North America & Mexico	S12-PF1
KNOX	Jessica	Discovery and characterization of bioactivated nematicides for selective control of parasitic nematodes	S12-PF2
SESSION 17 - Integrated nematode management			
LILLO	Paula	Synergies between climate change impacts and conservation tillage practices on agricultural soils functionality	S17-PF1
WILES	John A.	A new mode of action classification scheme for nematode control agents ("nematicides") - nematode working group of IRAC	S17-PF2
TEKLU	Misghina G.	Damage threshold, population dynamics and host-status of *Meloidogyne chitwoodi* on five selected crops	S17-PF3
SESSION 23 - Integrated nematode management			
GÓDOR	Anita	Case studies of root-knot nematode (*Meloidogyne* spp.) control in protected vegetables in Hungary	S23-PF1
ROS IBÁÑEZ	Caridad	Ozone treatments for the management of *Meloidogyne* sp. in greenhouse tomato cultivation in southeastern Spain	S23-PF2
BUCKI	Patricia	Interaction between *Fusarium* spp and root lesion nematode *Pratylenchus capsici* on pepper crops in the Arava (Israel)	S23-PF3
DA ROCHA	Mara Rubia	Reaction of *Phaseolus vulgaris* accessions from EMBRAPA core collection as to resistance to *Heterodera glycines*	S23-PF4
SESSION 27 - Next-generation nematicides			
BURNS	Andrew R.	Selective control of parasitic nematodes using bioactivated nematicides	S27-PF1
TROCCOLI	Alberto	The effect of 1-Octen-3-ol and 3-Octanone on plant parasitic nematodes	S27-PF2
KAMMERER	Christian	Impacts of long-term SDHI nematicide use on turfgrass	S27-PF3

Last name	First name	Title of poster	# poster
BAČIĆ	Jasmina	First report of Meloidogyne arenaria on calla (Zantedeschia aethiopica) in Serbia	**S5-P01**
BUDHATHOKI	Sabina	Vertical distribution of nematodes in peanut-cotton cropping systems	**S5-P02**
CABRERA	Alfonso	Management of plant parasitic nematodes in California almond production with fluopyram (Velum™ One)	**S5-P03**
CALANDRELLI	Angelica	Sorghum genotypes reaction to Meloidogyne javanica	**S5-P04**
CUNHA	Maria José	Effects of Solanum linnaeanum and S. sisymbriifolium on Globodera pallida hatching and mortality	**S5-P05**
DE WAAL	Jeanne	Corteva nematicides' compatibility with soil applied biologicals for nematode, insect and disease management	**S5-P07**
DIAS-ARIEIRA	Claudia	Castor bean cake extracts: nematicidal potential and chemical composition	**S5-P08**
DJIAN CAPORALINO	Caroline	Design and assessment of innovative Mediterranean vegetable cropping systems to manage root-knot nematodes	**S5-P09**
ELEKCIOĞLU	İbrahim Halil	Investigations of mix infections of root lesion nematodes (Pratylenchus thornei and Pratylenchus neglectus) and cereal cyst nematodes (Heterodera avenae and Heterodera latipons) on wheat	**S5-P10**
EMERSON	Michael	Field Performance of Several Maturity Group IV and V Soybean Cultivars in a Southern Root-Knot Nematode Infested Field	**S5-P11**
FONTANA	Lais	Ultrasound-assisted extraction of nematicidal compounds from Ricinus communis and its potential against Meloidogyne javanica	**S5-P12**
FONTES	Maria Geane	Detection of the root-knot nematode Meloidogyne luci parasitizing tomatoes in Sacatepéquez Province, Guatemala	**S5-P13**
GABRIEL	Marcia	Assessment of the resistance spectrum of the tomato Mi-1.2 gene/locus against fifteen Meloidogyne species	**S5-P14**
GALHANO	Cristina	Can Arbutus unedo L. leaves be used as a biological alternative to control Meloidogyne javanica?	**S5-P15**
GALHANO	Cristina	Could an agri-food be converted into a valuable environmentally-friendly nematicide?	**S5-P16**
GARTNER	Ulrike	Mapping a new resistance to the potato cyst nematode Globodera pallida from the wild potato Solanum spegazzinii	**S5-P17**
GODINHO DE ARAÚJO	Fernando	Genetic diversity of Soybean Cyst Nematode (SCN) Heterodera glycines populations in southeastern Goiás state, Brasil	**S5-P18**
GONÇALVES FRANCO SILVA	José Paulo	Nematicide and ovicide effect of thiophanate-methyl and fluazinam (Certeza N®) against nematodes	**S5-P19**
GORDON	Kara	Management strategies utilizing fertilizers and nematicides to reduce Rotylenchulus reniformis induced damage on cotton	**S5-P20**
HAFEZ	Saad	Effect of fluopyram, spirotetramat, and oxamyl on the cereal cyst nematodes, Heterodera avenae in Idaho	**S5-P21**
HALLMANN	Johannes	Development of Heterodera schachtii in sugar beet genotypes with varying levels of resistance	**S5-P22**
HOLDEN-DYE	Lindy	'Magic Bullets' for Plant Parasitic Nematodes	**S5-P23**
KNOETZE	Rinus	Host status of cover crops for root lesion nematode species (Pratylenchus spp.) associated with apple in South Africa	**S5-P24**
LE ROUX	Anne-Claire	Risk analysis, waste disinfestation methods and rotation plants as tools for a management of the risks associated with nematodes.	**S5-P25**
LUFF	Kelly	Managing causal pathogens of the potato early die complex with a new chemistry, fluopyram	**S5-P26**
MACGUIDWIN	Ann	Premature and sudden death complexes: How and why both are important questions	**S5-P27**
MADAURE	Jacqueline Tinashe	Compatibility of Nemafric-BL phytonematicide and biocontrol agents for the management of Meloidogyne species	**S5-P28**

MAŇASOVÁ	Marie	Influence of the length of sugar beet sludge storage on the amount and viability of beet cyst nematode embryos (Heterodera schachtii A.Schmidt, 1871)	**S5-P29**
MASSON	Anne-Sophie	Depicting the drivers of the root-associated microbiome in fields infected by root-knot nematodes in Cambodia	**S5-P30**
MEDINA	Karla	Performance of a commercial heat-killed Burkholderia rinojensis bio-based product in agricultural crops	**S5-P31**
MUNERA URIBE	Gladis Emilia	Meloidogyne-Fusarium interaction for the management of vascular wilt of Physalis peruviana plants in Colombia	**S5-P32**
MWANGI	Grace Nyambura	Potential of biofumigant cover crops (Brassica spp.) for suppression of stubby root nematodes (Trichodorus and Paratrichodorus spp.), associated with Docking disorder in sugar beet (Beta vulgaris)	**S5-P33**
NOLING	Joseph	Vertical management zones for enhancing yield and nematode control in Florida strawberry	**S5-P35**
ORLANDO	Valeria	Invasion and reproduction of P. penetrans on 'Maris Peer' potatoes	**S5-P36**
PUERARI	Heriksen	Influence of Acibenzolar-S-methyl application on the penetration and development of Pratylenchus brachyurus in maize	**S5-P37**
ROS IBÁÑEZ	Caridad	Management of pepper varieties resistant to Meloidogyne spp. for nematode control in greenhouse pepper crops	**S5-P38**
RUTHES	Andrea	Biogas digestate as potential source for nematicides	**S5-P39**
SHEPHERD	Rachel	Potential chemical control options for Aphelenchoides besseyi in ornamental plants	**S5-P40**
SILVA	Monique	Reaction of sorghum genotypes to Pratylenchus brachyurus	**S5-P41**
SIMMONS	Jeffrey	Measurement of Soil Mobility of TymiriumR Nematicide Using Three Different Types of Diffusion Assays	**S5-P42**
SIMMONS	Jeffrey	Evaluate Plant Effects, Efficacy and Yield Benefits of Tymiriam® against Meloidogyne incognita on Potato	**S5-P43**
SITHOLE	Nokuthula	Synergistic interaction of plant biomass and rhizobacteria for the management of Meloidogyne sp. on Solanum lycopersicum	**S5-P44**
THAPA	Sita	An integrated approach to manage soybean cyst nematode: rotation of the resistant sources, compost, and cover crops	**S5-P45**
THODEN	Tim	Soil health & nematicides: considerations for integrated nematode management	**S5-P46**
THODEN	Tim	Compatibility of SalibroTM and Vydate® with Pasteuria penetrans spore attachment to Meloidogyne javanica and M. incognita	**S5-P47**
UYSAL	Gulsum	Host status of lavender and lavandin cultivars to Meloidogyne incognita	**S5-P48**
VAZ MOREIRA	Valdeir Junio	RNAi-mediated Minc03328 gene silencing for the management of Meloidogyne incognita in transgenic Arabidopsis thaliana	**S5-P49**
VILLENAVE	Cécile	ELISOL environnement: a private French structure specialized in nematology R&D: soil bio-indication and crop protection.	**S5-P50**
VISSER	Johnny	Inundation: an effective method to control the root knot nematode Meloidogyne chitwoodi	**S5-P51**
WESEMAEL	Wim	Damage threshold and host-plant status of spinach (Spinacia oleracea) for Meloidogyne chitwoodi and Pratylenchus penetrans	**S5-P52**
WESTERDAHL	Becky	Field evaluation of sugarbeet varieties resistant to sugarbeet cyst nematode	**S5-P53**
ZOUHAR	Miloslav	Sugar beet field storage, a possible source of sugar beet cyst nematode (Heterodera schachtii A.Schmidt, 1871)	**S5-P54**
GRABAU	Zane	Evaluating new commercial cotton (Gossypium hirsutum) cultivars for resistance to Rotylenchulus reniformis and Meloidogyne incognita	**S5-P55**
MOLENDIJK	Leendert	Potato as a catch crop in late summer to control Potato Cyst Nematodes	**S5-P56**
LUANGKHOT	Justin	Root preservation in epoxy resin to highlight in-season treatment response in potato	**S5-P57**
MATLALA	Francinah L.	Nematode population dynamics in tomato nethouses over a three year period	**S5-P58**

S6. Legal and regulatory aspects of nematode management

Last name	First name	Title of poster	# poster
SESSION 14 - Social impact of nematode management			
NTIDI	Nancy	Abundance and diversity of plant-parasitic nematodes in the rhizospheres of maize cultivars grown by commercial farmers in rural areas of South Africa	S14-PF1
COYNE	Danny	Banana fibre paper: effectively delivering ultra-low nematicide dosages for more acceptable nematode management	S14-PF2
HOWLAND	Amanda	Best management practices for root-knot nematode *(Meloidogyne hapla)* in daylily (*Hemerocallis* spp.) production	S14-PF3
SESSION 15 - Advances in nematode detection and identification: instrumentation and applications			
DAUB	Matthias	A novel approach for applying machine learning for detection and phenotyping of cyst nematodes in soil extracts	S15-PF1
KAGIMU	Nicholas	ATR-FTIR spectroscopy and hyperspectral imaging in determining quality of formulated entomopathogenic nematodes	S15-PF2
ORLANDO	Valeria	Rapid detection and quantification of plant-parasitic nematodes from large volumes of soil	S15-PF3
SESSION 29 - Trade and market access implications of plant parasitic nematodes			
ALAKE	Gideon	Negative binomial modeling of nematode count data yield more accurate mean and variance estimates	S29-PF1
VIAENE	Nicole	FAGUSTAT: Investigating Beech Leaf Disease, a threat to beech trees and forests in Europe	S29-PF2
NGUYEN	Huu Tien	Plant-parasitic nematode: a potential threat to medicinal plants in Vietnam	S29-PF3

Last name	First name	Title of poster	# poster
GAMEL	Sylvie	Method validations for reliable plant-parasitic nematode diagnosis: the example of Heterodera glycines identification	S6-P01
HEYDARI	Fariba	Study of Cyclobutrifluram mode of action using on Caenorhabditis elegans	S6-P02
KÖNIG	Stephan	Development of appropriated measures and methods to close pathways for the distribution of cyst nematodes	S6-P03
MOORE	Scott	An experimental design optimized for rate-profiling of plant parasitic nematodes in row crops	S6-P04
ORLANDO	Valeria	Detection and distribution of Pratylenchus spp. in UK potato fields.	S6-P05
QUINTERO	Tonia	USDA regulations, decisions and operations for nematode management in the United States	S6-P06
RIVA	Gabrieli	Molecular detection and distribution of root-knot nematode species in Florida	S6-P07
TALAVERA	Miguel	A cost-benefit and efficacy analysis of Meloidogyne management strategies in Mediterranean intensive horticulture	S6-P08
VAN HEESE	Evelyn	Influence of relative humidity during drying on viability of Globodera cysts	S6-P09

S7. Biological control of nematodes

Last name	First name	Title of poster	# poster
SESSION 20 - Natural Products as nematicides			
NJEKETE	Cliven	Nematicidal plants for root-knot nematode management in tomato agrosystems	S20-PF1
ELEKCIOĞLU	İbrahim Halil	Investigation on the effectiveness of some plant extractions against *Ditylenchus dipsaci* and *Meloidogyne incognita*	S20-PF2

Last name	First name	Title of poster	# poster
ANDIKA	Balawara	Biological control of Meloidogyne graminicola in rice plant by cost-effective bionematicide formula	S7-P01
BALOG	Luca Eszter	Parasitic nematodes of the harlequin ladybird, Harmonia axyridis in Hungary	S7-P02
BEHMAND	TOHID	The effects of root lesion nematodes (Pratylenchus thornei) on chickpea plant and rhizobium bacteria	S7-P03
CANAGUIER	Renaud	Arbuscular Mycorrhizal Fungus Mobilization against Root-Knot Nematodes of Tomato and Pepper Roots	S7-P04
CASTANEDA ALVAREZ	Carlos	Symbiotic bacteria of entomopathogenic nematodes for the biocontrol of dagger nematode Xiphinema index	S7-P05
CHEN	Feng	Interkingdom cooperation between rhizosphere bacteria and nematodes modulates the infectivity of plant-nematodes	S7-P06
CHEN	Hanqiao	New member of the Cytolysin A family: Cry6Aa controls nematocidal activity through glycosphingolipids	S7-P07
CONCEIÇÃO	Isabel	Effects of Trichoderma secondary metabolites on the fitness of Root Knot Nematodes	S7-P08
COSTA	Sofia R.	Compatibility of selected pesticides with Pochonia chlamydosporia	S7-P09
DA ROCHA	Mara Rubia	Trichoderma spp. isolates as potential resistance inducers and biocontrol agents of Meloidogyne javanica on banana	S7-P10
DOUDA	Ondřej	Alternative management of Sugarbeet nematode (Heterodera schachti)	S7-P11
FABIYI	Oluwatoyin	Application of starch citrate biopolymer for controlled release of carbofuran for Meloidogyne incognita management	S7-P12
FOURIE	Hendrika	The biocontrol link between rhizosphere microorganism communities and Meloidogyne populations	S7-P13
FREITAS DE ALMEIDA	Sheila	Trichoderma spp., a growth promoter of tomato roots and Meloidogyne enterolobii populations	S7-P14
GERIČ STARE	Barbara	Two nematicidal Bacillus strains revealed a wide range of possible virulence factors	S7-P15
HAJIHASSANI	Abolfazl	Effects of application timing on the efficacy of Xenorhabdus and Photorhabdus metabolites for control of Meloidogyne incognita	S7-P16
HUSSAIN	Manzoor	Virulence and microbial activity of different nematophagous fungi and chemicals against root-knot nematodes, Meloidogyne incognita, on tomato	S7-P18
KAKOULI-DUARTE	Thomais	Potential of plant growth promoting bacteria as biocontrol agents against the root knot nematode Meloidogyne javanica	S7-P19
KAKOULI-DUARTE	Thomais	Effects of an Alltech® soil health product on entomopathogenic and plant parasitic nematodes in invitro bioassays	S7-P20
KUDJORDJIE	Enoch Narh	Tomato rhizosphere under RKN attack - Deciphering the Meloidogyne incognita pathobiome	S7-P21
LAPEYRE	Laurent	Effects of a chicken manure fertilizer on beneficial nematode communities in vineyard	S7-P22
MARIETTE	Nicolas	Hatching of cyst nematodes in soil drenched with root exudates under controlled environmental conditions	S7-P23
MCPEAK	Sloane	An evaluation of small grain cover crops to reduce Meloidogyne incognita population density in cotton fields	S7-P24
MOGOLLON ORTIZ	Angela Maria	Soil actinobacteria with biocontrol potential against Meloidogyne javanica	S7-P25
MORALES MONTERO	Patricia	Evaluation of bacterial extracts of Xenorhabdus, Photorhabdus, and rhizobacteria to control Meloidogyne ethiopica	S7-P26
OWADA	Kyoko	Free-living nematodes and microorganisms in soil improvement materials for plant parasitic nematode control	S7-P27
PETRIKOVSZKI	Renáta	The combined use of Metarhizium anisopliae and Trichoderma asperellum bioeffectors in the control of Meloidogyne incognita	S7-P28
PRADANA	Ankardiansyah Pandu	Efficacy of a cost-effective bionematicide to control Pratylenchus coffeae on robusta coffee	S7-P29

ROBERT FARIA	Denner	Orchid mycorrhizal fungus Waitea circinata on the control of Meloidogyne enterolobii in tomato crop	S7-P30
SCHLEKER	A. Sylvia S.	Microbial rhamnolipids as a powerful tool in modern agriculture nematode control	S7-P31
SILVA VALENZUELA	Manuel	Endophytic fungi: a biological alternative for the management of root-knot nematodes	S7-P32
SUSIČ	Nik	Nematicidal and plant growth-promoting effects of Bacillus cf. firmus in white-fruited strawberries	S7-P33
VERONICO	Pasqua	Evaluation of different isolates of Trichoderma spp. for antagonistic activity against Meloidogyne incognita	S7-P34
DEGROOTE	Eva	Cold water extract of Cucurbitaceae as basis for future nematode control agents	S7-P35
MANZANILLA-LÓPEZ	Rosa Helena	Pochonia chlamydosporia var. mexicana response to physicochemical factors, rhizosphere colonization and egg parasitism	S7-P36
KISITU	Joseph	Towards high throughput phenotyping of banana for nematode resistance	S7-P37

S8. Nematode omics, metabolism and physiology

Last name	First name	Title of poster	# poster
SESSIONS 9 - 'Omics' in nematology			
MAQSOOD	Maria	In search of a common target for control of nematode and aphid pests	S9-PF1
BELLIARDO	Carole	Metagenomics mining improves analysis of horizontal gene transfers involved in parasitic function in plant-parasitic nematode	S9-PF2
GENDRON	Eli	Development of mitometagenomics protocols for the enhancement of nematode identification and biodiversity study	S9-PF3
SESSIONS 13 - 'Omics' in nematology			
XIANG	Hui	Mining new nematode effectors interacting with plant transcription factors by Cr-Y2H	S13-PF1
PIJNACKER	Anna	SMART UP – Spatial Mapping of Root Transcriptomes Upon Nematode Parasitism	S13-PF2
DANCHIN	Etienne	The strange chromosome ends of root-knot nematodes	S13-PF3

Last name	First name	Title of poster	# poster
ANDERSON	Scott	Manipulating lipid metabolism in plants as a novel plant parasitic nematode control measure	S8-P01
BOURNAUD	Caroline	The search for "parasitism-genes" readers in the world's most damaging plant-parasitic nematode, Meloidogyne incognita	S8-P02
DA ROCHA	Martine	A bioinformatics pipeline for the characterization of small RNAs involved in the plant-root knot nematode interaction	S8-P03
DALZELL	Johnathan	Early life stress promotes aggression and inhibits learning in male nematodes	S8-P04
DALZELL	Johnathan	Temperature modulates tomato gene expression networks, root exudate composition and parasite interactions	S8-P05
DANCHIN	Etienne	Parasitic success without sexual reproduction: what more than 10 years of root-knot nematode genomics revealed?	S8-P06
EVES-VAN DEN AKKER	Sebastian	The transcriptional regulation of plant-nematode parasitism	S8-P07
HASSANALY GOULAMHOUSSEN	Rahim	Chromatin Landscape Dynamics in the Early Development of the Plant Parasitic Nematode Meloidogyne incognita	S8-P08
LEE	Wan-Chun	The roles of neuropeptide genes flp16 and flp18 in Pratylenchus vulnus	S8-P09
MIRGHASEMI	Seyedeh Negin	Molecular characterization and functional importance of b-1,4-endoglucanases from the root-lesion nematode Pratylenchus loosi	S8-P11
MITREVA	Makedonka	Comparative genomics of parasitic nematodes	S8-P12
O'CONNOR	Vincent	Elucidating the mode of action of a novel nematicide, fluensulfone, using plant parasitic nematode, *Globodera pallida*	S8-P13

PETTRICH	Laura	Unravelling the demographic history of a Pleistocene nematode	**S8-P14**
ROBINSON	Colin	Temperature Response of Metabolic Activity of an Antarctic Nematode	**S8-P15**
SAGAWA	Marika	A newly identified volatile sex pheromone of *Caenorhabditis* elegans	**S8-P16**
SCHIFFER	Philipp	Beyond omics: establishing new nematode model systems to study the evolution of parthenogenesis (and development)	**S8-P17**
VAN STEENBRUGGE	Joris	Highly Polymorphic Regions in the Genome of Meloidogyne chitwoodi Reveal Potential Effectors	**S8-P18**
VERMA	Anju	Functional characterization of a highly expanded superfamily of dorsal gland effector proteins in cyst nematodes	**S8-P19**
RODRIGUES	Jules	Growing the tree: an update of the Onchocercidae evolutionary history with a multi-locus phylogeny	**S8-P20**
VICENTE	Cláudia	Silencing a new female-specific multi-gene family of Pratylenchus penetrans can reduce nematode propagation	**S8-P21**

S9. Entomopathogenic nematodes

Last name	First name	Title of poster	# poster

SESSION 4 - EPN ecology and biology

CHACON	Andrea	War in the darkness: the use of volatile organic compounds and entomopathogenic nematodes to control wireworms	**S4-PF1**
COCKX	Bram	Mass spectrometry-driven discovery of neuropeptidergic systems regulating nictation in free-living and parasitic nematodess	**S4-PF2**
DRITSOULAS	Alexandros	Standardized surveys confirm greater EPN presence and diversity in a subtropical compared to Mediterranean citrus orchards	**S4-PF3**

SESSION 26 - EPN commercialization and application

VANDENBOSSCHE	Bart	Efficacy of species mixtures of entomopathogenic nematodes against different larval stages of cockchafers	**S26-PF1**
SHEHATA	Ibrahim	Early season use of *Heterorhabditis bacteriophora* increases strawberry yield in fields infested by the white grub *Temnorhynchus baal*	**S26-PF2**
GARRIGA	Anna	The undetectable killer: *Steinernema carpocapsae* avoid recognition when infecting *Drosophila suzukii* larvae.	**S26-PF3**

Last name	First name	Title of poster	# poster
BALOG	Luca Eszter	Diversity of entomoparasitic nematodes in the rose chafer, Cetonia aurata grub	**S9-P01**
BETANZO	Nicolás	Host-seeking behavior of the native entomopathogenic nematode Steinernema unicornum and its phylogenetic relationship with the exotic strains marketed in Chile	**S9-P02**
BHAT	Aashaq Hussain	Steinernema siamkayai (Rhabditida: Steinernematidae): notes on its morphology, bionomy and distribution from the Indian subcontinent	**S9-P03**
BLANCO-PÉREZ	Ruben	Impact of differentiated farming practices on the native entomopathogenic nematodes in DOCa Rioja vineyards (Northern Spain)	**S9-P04**
CHAUBEY	Ashok	Steinernema abbasi (isolate CS2)-Xenorhabdus indica complex and Helicoverpa armigera immune response	**S9-P05**
DANSO	Yaw	Natural populations of entomopathogenic nematodes on sweetpotato farms in southern Ghana	**S9-P06**
DUNCAN	Larry	Arthropod community responses reveal potential predators and prey of entomopathogenic nematodes in a citrus orchard	**S9-P07**
DUNN	Murray David	Protein source impact on the recovery and yield of entomopathogenic nematodes, using in vitro liquid culture	**S9-P08**
DUNN	Murray David	In vitro liquid mass production of Steinernema jeffreyense, using a designer desktop bioreactor	**S9-P09**

EHLERS	Ralf-Udo	Heterorhabditis bacteriophora: An excellent model for genetic improvement of biocontrol traits	**S9-P10**
ELIÁŠ	Sara	Bioactive molecules produced by Heterorhabditis bacteriophora affects the phenoloxidase system of Galleria mellonella	**S9-P11**
GALEANO	Magda	The successful story of Entomopathogenic Nematodes against foliar pests: our silver bullet	**S9-P12**
GALEANO	Magda	How to Successfully Apply Entomopathogenic Nematodes in an IPM System?	**S9-P13**
GONZÁLEZ-PAZ	Lenin	Metformin as enhancer of entomopathogenic nematode performance	**S9-P14**
GRUBIŠIĆ	Dinka	Isolation of entomopathogenic nematodes on agricultural land in Croatia	**S9-P15**
MADAURE	Jacqueline Tinashe	Morphological responses of Steinernema feltiae exposed to purified active ingredient of Nemarioc-AL phytonematicide	**S9-P16**
MALAN	Antoinette	In vitro-cultured entomopathogenic nematodes to control the false codling moth, Thaumatotibia leucotreta	**S9-P17**
NAVAREZ	Mara Louisa	Isolation, characterization, and virulence of entomopathogenic nematodes in Davao del Sur, Philippines against superworm Zophobas morio	**S9-P18**
NAVARRO	Patricia	Steinernema australe display chemotaxis towards volatiles identified from blueberry roots	**S9-P19**
PETRIKOVSZKI	Renáta	The effect of aqueous extracts of mulch materials on entomopathogenic, slug-parasitic and root-knot nematodes	**S9-P20**
SEBUMPAN	REA	Co-cultivation of entomopathogenic fungi and entomopathogenic nematodes in search of improved biocontrol against Spodoptera litura	**S9-P22**
STOKWE	Nomakholwa	Potential of local entomopathogenic nematodes for control of the vine mealybug, Planococcus ficus	**S9-P26**
SUAN	Mayvel	Distribution, characterization and virulence of the isolated entomopathogenic and entomophilic nematodes in selected vegetable and root crop farms in Bukidnon province, Philippines against cotton cutworm (Spodoptera litura)	**S9-P27**
SUMAYA	Nanette Hope	Isolation and biocontrol potential of entomopathogenic and entomophilic nematodes from Talakag, Bukidnon and Claveria, Misamis Oriental, Philippines	**S9-P28**
VANDENBOSSCHE	Bart	Breeding for improved virulence and post-application longevity of Heterorhabditis bacteriophora dauer juveniles	**S9-P30**
VICENTE DÍEZ	Ignacio	Plasticity in the use of Xenorhabdus nematophila and Photorhabdus laumondii against Botrytis cinerea	**S9-P31**
WU	Sheng-Yen	Concomitant species of entomopathogenic nematodes alter dispersal behavior and increase insecticidal efficiency	**S9-P32**
WANG	Zhen	Broad phenotyping of DJ-recovery in Heterorhabditis bacteriophora using highly homozygous mutants and WT-inbred lines	**S9-P33**

S10. Future of nematology, education and training

Last name	First name	Title of poster	# poster
INACIO	Maria	Nematology lab of INIAV: a 10-year overview of research, training and services	**S10-P01**
KAKOULI-DUARTE	Thomais	Nematology and the environment: a worm's tail	**S10-P02**
ROMERO MOYA	Lizzete Dayana	Thirty years of plant-parasitic nematode research in America	**S10-P03**
SHAPIRO-ILAN	David	The Journal of Nematology.	**S10-P04**
VAN HEESE	Evelyn	The nematode collections at the NPPO, Wageningen, The Netherlands	**S10-P05**
LOPES	Carina	Preliminary development of an automated system for identification and quantification of nematodes	**S10-P06**

LIST OF E-POSTERS ONLINE VIEWING

A - F

Last name	First name	Title of poster
ABRANTES	Isabel	PineWALL Project: Linking pine cell wall composition and structure to pinewood nematode resistance under climate change.
ABRANTES	Isabel	*Bursaphelenchus* spp. inhabitants of a centennial *Pinus pinea* tree of public interest.
AFFOKPON	Antoine	Assessing the influence of yam genotype on the performance of abamectin-treated banana paper to manage nematodes.
AHMED	Mohammed	Addressing the challenges of high-throughput nematode identification using metabarcoding.
ALAKE	Gideon	Negative binomial modeling of nematode count data yield more accurate mean and variance estimates.
ALMEIDA M.	Teresa M.	Plant-parasitic nematodes in potato crops in Portugal: patterns and influencing factors.
ANIH	Agatha	Diversity and geographical distribution of plant parasitic nematodes of the federal capital territory Abuja, Nigeria.
ANJAM	Muhammad Shahzad	*In silico* analyses of genes responsible for somatic sex determination in plant-parasitic nematode, Heterodera glycines.
ANJAM	Muhammad Shahzad	Understanding damage signals propagation required to build resilient cell walls in plant roots.
ARYAL	Sitaram	Diversity of the entomopathogenic nematodes in Australia and their potential for the control of Queensland fruit fly.
ASAMIZU	Erika	Identification of soil factors associated with root-knot nematode density in green manure-applied fields.
ASHRAFI	Samad	Novel endophytic nematode antagonistic fungi- potential for nematode biocontrol.
BARBOSA	Pedro	Root lesion nematode biocontrol in potato.
BUDHATHOKI	Sabina	Strategies to enhance efficacy of entomopathogenic nematodes for management of diamondback moth and imported cabbageworm.
CARES	Juvenil E.	Histopatholoy of cotton resistant line CNPA 17-26 B2RF to *Meloidogyne incognita* obtained by marker-assisted selection.
CHENG	Xinyue	Comparative and evolutionary analysis of RNAi pathways in plant parasitic nematodes.
COULIBALY	Laëtitia	Occurrence and abundance of parasitic nematodes of papaya (*Carica papaya* Linnaeus) in western region of Burkina Faso.
CUI	Jiangkuan	Pathotype and resistance classification of Heterodera avenae and H. *filipjevi* in Huanghuaihai Valley of China.
DAI	Dadong	Widespread DNA N6-methyladenine plays a crucial role in parasitic nematodes.
DASH	Manoranjan	Transcriptome analysis of *Meloidogyne graminicola* infected roots of resistant mutant rice.
DE OLIVEIRA ROCHA	Raquel	Identification of new esophageal gland effector candidates from adult females of the root-knot nematode.
DING	Zhong	Molecular and functional characterization of the *Ditylenchus destructor* voltage-gated calcium channel α1 subunits.
DOSHI	Pratik	Comparative study of neem-derived pesticides on *Meloidogyne incognita* under *in vitro* and pot trials in a greenhouse.
DYER	David	Effect of *Meloidogyne incognita* and *Fusarium oxysporum* f. sp. *vasinfectum* resistance traits in cotton.
DYER	Steven	*Ethylene Response Factor* genes modulate plant root exudate composition and the attraction of plant parasitic nematodes.
EKINO	Taisuke	Variation of recognition mechanism of predator species to other nematodes.

ESTEVES	Ivânia	Advancing knowledge of the root-knot nematode *Meloidogyne luci*.
ESTEVES	Ivânia	Nematicidal activity of naphthoquinones – nematode generation of reactive oxygen species.
FEIXUE	Cheng	Control of rice root knot nematode by flooding.
FLEMING	Thomas	Plant parasitic nematode biosecurity threats from a Northern Ireland and UK perspective.
FORGE	Thomas	Population dynamics of *Mesocriconema xenoplax* parasitizing sweet cherry trees in British Columbia, Canada.

To see the e-posters, click here

G - L

Last name	First name	Title of poster
GHAVAMISHIREHJIN	Nazanin	Survey and Development of Molecular Soil Test for Soybean Cyst Nematode in Manitoba.
GODJO	Tognisse Anique	Ability of indigenous EPN isolates to control *Bactrocera dorsalis* and their formulations for mango fruit fly biocontrol in Benin.
GORNY	Adrienne	Impact and management of *Meloidogyne enterolobii* in sweetpotato production in North Carolina, United States.
GOUVEIA	Angelica	Metabolic profile of tomato infected by *Meloidogyne javanica* in the presence of the fungus *Pochonia chlamydosporia*.
GRAY	Amanda	Efficacy of *Solanum sisymbriifolium* to control *Globodera pallida*.
GUESMI-MZOUGHI	Ilhem	Integrative characterisation of eight plant-parasitic nematode species on olive trees in central Tunisia.
GUESMI-MZOUGHI	Ilhem	Plant-parasitic nematode communities associated with olive trees in Central Tunisia.
HANDOO	Zafar	Plant-parasitic nematodes associated with medical hemp in Maryland.
HAYASHI	Kotomi	Establishment of laboratory growth system for potato cyst nematode.
HICKMAN	Paige	Evaluating Potential Trap Crops for Control of *Globodera pallida*.
HUAN	Peng	Development and application of a rapid Loop-mediated Isothermal Amplication for detection of Cereal cyst nematodes.
HUSTON	Daniel C.	Species diagnosis, hosts and distribution of cyst nematodes of the genus *Heterodera* (Tylenchida: Heteroderidae) with a focus on species of concern for Australia
IKOYI	Israel	Response of soil nematode community to increased plant species diversity in an intensively managed grassland.
IMRAN	Zarrin	Post-embryonic development of K-strategic nematode *Labronema baqrii* Khan, Jairajpuri and Ahmad, 1989; analysis of structural evolution and stability.
IWAHORI	Hideaki	DNA profile, species-specific primers for detection, and temperature for development of *Heterodera schachtii* in Japan.
JIAN	Heng	MiMsp40 effector of *Meloidogyne incognita* interacts with a transcription repressor of Arabidopsis and promotes parasitism.
KANTOR	Mihail	International perspectives on the United States Department of Agriculture Nematode Collection, past, current and future.
KANZAKI	Natsumi	A new aphelenchoidid insect parasite from a tenebrionid beetle, *Uloma marseuli*.
KASSAM	Rami	Novel bacteria for biological control of root-knot nematode, *Meloidogyne incognita*.
KHOSA	Mbokota C.	Nematicidal activity of South African botanical plant extracts against *Meloidogyne species* under vitro conditions.
KIM	Dong-gyu	Transcriptomic and microscopic evaluations fungal antagonism of soybean cyst nematode (*Heterodera glycines*) eggs.
KIRINO	Haru	Thaumatin-like protein as a virulence factor of the pine wood nematode, *Bursaphelenchus xylophilus*.
KOSAKA	Hajime	Host range and geographical distribution of *Sphaerularia vespae*, the nematode parasite of queen hornets.

KUNDU	Artha	Identification and characterization of Nematode chemosensory GPCRs (NemChRs) in *Heterorhabditis bacteriophora.*
LEE	YiChien	Comparative genomics of free-living nematodes.
LEOCATA	Salvatore	Evaluation of galling index on previous crop as a reliable method for correct positioning of nematode control trials (*Meloidogyne Genus*).
LETSOALO	Morakene	Interactive effects of biochar type and rate on tomato plant growth and nematode suppression.
LI	Hongmei	Present situation of pine wood nematode *Bursaphelenchus xylophilus* in China.
LILLIS	Peter	Proteomic Profiling of Steinernema carpocapsae and *Heterorhabditis megidis* Infective Juveniles stored at 20°C and 9°C.
LU	Xiuhong	Identification of WRKY transcription factors responsive to root-knot nematode in tomato roots by RNAseq analysis

To see the e-posters, click here

M - P

Last name	First name	Title of poster
MALEITA	Carla	Transcriptional changes of *Meloidogyne luci* second-stage juveniles after exposure to 1,4-naphthoquinone.
MALYSHEVA	Татьяна	Demographic history and co-evolution study of nematodes parasitizing in millipedes of the Western Caucasus.
MARTIN	Tvisha	The Use of Soil Biological Indicators to Improve Soil Health.
MASHELA	PW	Responses of total protein in *Meloidogyne incognita* juveniles to Nemafric-BL phytonematicide.
MASHELA	PW	Fast-tracking the development of efficacy and toxicity data for registration of phytonematicides.
MASONBRINK	Rick	Gene copy number expansions and contractions in the genomes of plant-parasitic nematodes.
MATVEEVA	Elizaveta	Patterns of changes of soil nematode communities in relation to biocenosis type and vegetation features.
MATVEEVA	Elizaveta	Spatio-temporal changes of soil nematode communities in northern meadows.
MATVEEVA	Elizaveta	Potato plant responses to a combination of different abiotic and biotic stresses.
MATVEEVA	Elizaveta	Potato cyst-forming nematode in open data network on invasive alien species between Finland and Russia: Citizen Science.
MATVEEVA	Elizaveta	Search for soil amendments affecting potato cyst nematode: possibilities for parasite control.
MAYAD	El Hassan	Response of soil nematodes to anthropic gradient in the arganeraie biosphere.
MCDONALD-HOWARD	Kerry-Lyn	Toxicity of essential oils to beneficial nematodes.
MCDONALD-HOWARD	Kerry-Lyn	The effect of soil type and temperature on the survival of the slug parasitic nematode *Phasmarhabditis hermaphrodita.*
MIGUNOVA	Varvara	Analysis of nematicidal activity of bacterial strains from collection of Institute of Biological Plant Protection.
MISHRA	Shova	Circadian rhythms inform the outcome of a plant-nematode interaction.
MORGAN	Túlio	Genomics and evolutionary analysis of a new *Trichoderma* species with potential to antagonize plant-parasitic nematodes.
MURATA	Gaku	Suppressive effect of *Solanum palinacanthum* on root-knot nematodes (*Meloidogyne spp.*).
NNAMDI	Chinaza	Effect of deep application of non-fumigant nematicides on *Meloidogyne incognita* in a tomato plasticulture system.
PARRADO GUEVARA	Luisa Maria	Integration of biocontrol agents and microorganisms found in organic soil amendments to control Potato Early Die.
PEN-MOURATOV	Stanislav	Seasonal fluctuations attenuate stimulatory or inhibitory impacts of colonial birds on soil biota.
PENG	Donghai	A novel pore-forming toxin triggers *Ditylenchus destructor* apoptotic cell death via a C2-domain containing protein.

POFU	Kgabo	Host suitability of biofortified fusarium-resistant sweet potato lines to Meloidogyne species in South Africa.
PŮŽA	Vladimír	Potential new targets for entomopathogenic nematodes

To see the e-posters, click here

Q - Z

Last name	First name	Title of poster
QIN	Xin	A novel *Meloidogyne incognita* effector Mi-ISC-1 promotes parasitism by disrupting salicylic acid biosynthesis in host plants.
RUTTER	William	Pathotypic variation between isolates of Meloidogyne enterolobii collected from Sweetpotato in North and South Coralina.
SALGADO	Sonia	Coffee selection based on molecular characterization and identification of coffee resistant to Meloidogyne paranaensis.
SANTIAGO	Débora C.	Antagonistic effect of fungal filtrates on hatching, mobility and mortality of Meloidogyne incognita.
SATO	Kazuki	Molecular insights into an interaction of resistant plant *Solanum torvum* and virulent/avirulent root-knot nematodes.
SCHUMACHER	Lesley	Nematode vertical distribution in peanut-cotton cropping systems.
SEO	Jongmin	First report of the apple root-knot nematode, Meloidogyne mali, in Korea.
SEO	Jongmin	Damage potential of Heterodera sojae to soybean.
SHEEDY	Jason	First genetic mapping of resistance to root-lesion nematode (*Pratylenchus thornei*) in chickpea (*Cicer arietinum*).
SILVESTRE	Rocio	Mapping resistance to the Potato Cyst Nematode, *Globodera pallida*, in a tetraploid, russet-skinned potato population.
SINGH	Gurminder	Identification of Quantitative Trait Loci for Resistance to *Pratylenchus neglectus* in Triticale.
SKANTAR	Andrea	New reports and molecular diagnostics of circumfenestrate cyst nematodes in the United States.
SOMVANSHI	Vishal Singh	Understanding the *Heterorhabditis* nematode factors involved in modulating symbiosis with *Photorhabdus* bacteria.
SOMVANSHI	Vishal Singh	Improving the draft genome of rice root-knot nematode *Meloidogyne graminicola* by l ong-read sequencing.
SOMVANSHI	Vishal Singh	A draft genome for the entomopathogenic nematode *Heterorhabditis indica.*
SONG	Handa	A *Meloidogyne graminicola* effector interacts with a novel Cu metallochaperone protein to improve Cu/Zn-SOD activities and suppress plant immunity.
SUKARTA	Octavina	Exploring the Role of RanGAP2 in Recognition by the Potato CC-NB-LRR immune Receptors Rx1 and Gpa2.
TANAKA	Ryusei	Association of *Caenorhabditis inopinata* with the wasp and bacteria in the fig ecosystem.
THAPA	Rambika	A Quick Dip: Alternatives to hot-water dipping to control *Meloidogyne hapla* in daylily (Hemerocallis) production.
THI NGAN	Phan	Genomic features of the rice root-knot nematode *Meloidogyne graminicola*.
TIMPER	Patricia	Influence of root exudates on suppression of root-knot nematodes by *Pasteuria penetrans.*
WANG	Xingwei	Interplay between plant circadian clock and biotic stresses: from *Arabidopsis* to Soybean.
WRAM	Catherine	Variation in biological and molecular responses of *Meloidogyne* spp. to post-plant nematicides.
XU	Yu Mei	Diversity of nematodes in the family Tripylidae de Man, 1876 in Shanxi Province, North China.
YU	Jiarong	A novel venom allergen-like protein from *Meloidogyne incognita* suppresses plant defenses and promotes parasitism.
ZHANG	Shuwu	Identification the mechanisms of *Trichoderma longibrachiatum* T6 against *Heterodera avenae* by metabonomics and transcriptome.

ZHAO	Jianlong	The root-knot nematode MiPDI1 effector targets a zinc finger protein to establish disease in Solanaceae and Arabidopsis.
ZHAO	Zengqi	Bursaphelenchus eggersi or B. hildegardae (Nematoda: Parasitaphelenchidae)? Does it really matter to New Zealand?

To see the e-posters, click here

7th International Congress of Nematology
ICN 2022
Antibes Juan-les-Pins - France

"Crossing borders: a world of nematode diversity and impact to discover"

European Society of Nematologists

LIST OF PARTICIPANTS

AASHA RANA Aasha
Nematology Laboratory - Dep. of Zoology
Chaudhary Charan Singh University
MEERUT – INDIA
+7 248 537 339
aasha.aasharana@ymail.com

ABAD Pierre
UMR IBSV
INRAe
SOPHIA ANTIPOLIS – FRANCE
+33 4 92 38 64 00
pierre.abad@inrae.fr

ABD-ELGAWAD Mahfouz
Plant Pathology Department
National Research Centre
DOKKI – EGYPT
+02 35460355
mahfouzian2000@yahoo.com

ABRANTES Isabel
Department of Life Sciences - Nemato-lab
University of Coimbra
COIMBRA – PORTUGAL
+351 239 240 772
isabel.abrantes@uc.pt

ADAMS Byron
Biology
Brigham Young University
PROVO – USA
+1 80 14 22 31 32
byron_adams@byu.edu

ADÃO Helena
MARE - University of Évora
ÉVORA – PORTUGAL
+351 266 760881
hadao@uevora.pt

AGUDELO Paula
Plant and Environmental Sciences
Clemson University
CLEMSON – USA
+1 864 656 2810
pagudel@clemson.edu

AKSAN Muhammad
Nematology Research Unit
Ghent University
GHENT – BELGIUM
+32 926 452 23
muhammad.aksan@ugent.be

AL BANNA Luma
Plant Protection
University of Jordan
AMMAN – JORDAN
lalbanna@ju.edu.jo

AMORA Deisy
Plant Health
Chr Hansen
TAASTRUP – DENMARK
+45 238 430 18
dkdeam@chr-hansen.com

ANDERSON Scott
Plant Pathology
Washington State University
PULLMAN – USA
+1 42 55 01 41 00
scott.d.anderson@wsu.edu

ANDIKA Balawara
Biology Education
University of Jember
JEMBER – INDONESIA
+62 331 334 270
180210103042@mail.unej.ac.id

APOLÔNIO SILVA DE OLIVEIRA Daniel
Nematology / Molecular Biology
NPPO NL National Reference Center
WAGENINGEN – THE NETHERLANDS
+31 6 21981007
d.apolonio@nvwa.nl

ARCHIDONA YUSTE Antonio
Department of Ecological Modelling
UFZ
LEIPZIG – GERMANY
+49 341 235 1714
antonioj.archidona@juntadeandalucia.es

ASYIAH Iis Nur
Biology Education Study Program
University of Jember
JEMBER – INDONESIA
085 7 47 30 76 92
iisnaza.fkip@unej.ac.id

BACIC Jasmina
Plant Nematology Laboratory
PSS Institut Tami?
PANCEVO – SERBIA
+381 13 313 092
jasminabacic4691@gmail.com

BALOG Luca Eszter
Department Systematic Zoology and Ecology
Eötvös Loránd University
BUDAPEST – HUNGARY
+36 1 38 12 19 3
baloglucaeszter@gmail.com

BAMUDO Yisihak Alemu
Ghent University
GHENT – BELGIUM
yisualpha3@gmail.com

BARBARY Arnaud
Bretagne Plants Innovation
HANVEC – FRANCE
+33 3 21 60 46 68
a.barbary@plantsdebretagne.com

BARBU Corentin
UMR Agronomie
INRAe
THIVERVAL-GRIGNON – FRANCE
+33 1 30 81 52 26
Corentin.barbu@inrae.fr

BATCHELDER Ellen
School of Biodiversity Conservation
Unity College
UNITY – USA
+1 207 509 7268
ebatchelder@unity.edu

BAXTER Jeffrey
Bayer CropScience
USA
+1 512-317-4743
jeffrey.baxter@bayer.com

BEHMAND Tohid
Plant Protection Department
Agriculture Faculty
ADANA – TURKEY
+90 534 960 2112
tohid.behmand63@gmail.com

BELL Christopher
Faculty of Biological Sciences
University of Leeds
LEEDS – UK
+44 113 343 5664
C.A.Bell@leeds.ac.uk

BELLIARDO Carole
INRAe
SOPHIA ANTIPOLIS – FRANCE
+33 4 92 38 64 00
carole.belliardo@inrae.fr

BELLO Tesleem
Unit of Environmental Science and Management
North-West University
POTCHEFSTROOM – SOUTH AFRICA
+27 803 576 9499
tesleembello@yahoo.com

BELLOF Svenja
Research & Development, Crop Science
Bayer AG
MONHEIM AM RHEIN – GERMANY
svenja.bellof@bayer.com

BELTMAN Marieke
ClearDetections B.V.
WAGENINGEN – THE NETHERLANDS
+31 61 50 63 020
Marieke.Beltman@cleardetections.com

BENT Andrew
Plant Pathology
University of Wisconsin
MADISON – USA
+1 608 265 3034
afbent@wisc.edu

BERRY Shaun
BASF
DURHAM – USA
shaun.berry@basf.com

BERT Wim
Nematology Research Unit
Ghent University
GHENT – BELGIUM
++3292645223
wim.bert@ugent.be

BERTRAN André
Laboratory of Nematology
Wageningen University & Research
WAGENINGEN – THE NETHERLANDS
+31 3 17 48 21 97
andrebertran@hotmail.com

BETANZO FUENTES Nicolás
Biocontrol Laboratory
Universidad de La Frontera
TEMUCO – CHILE
+56 45 259 6855
n.betanzo01@ufromail.cl

BETT Silas Kipsang
Ghent University
GHENT – BELGIUM
silazkaybett@hotmail.com

BHAT Aashaq Hussain
Department of Zoology
Chaudhary Charan Singh University
MEERUT – INDIA
+91 9027447214
aashiqhussainbhat10@gmail.com

BIRD David
Department of Entomology and Plant Pathology
North Carolina State University
RALEIGH – USA
+1 919 637 0075
bird@ncsu.edu

BLANCO-PÉREZ Rubén
ICVV
CSIC
LOGROÑO – SPAIN
+34 9 41 89 49 80
ruben.blanco@icvv.es

BLAXTER Mark
Tree of Life
Wellcome Sanger Institute
CAMBRIDGE – UK
01223494902
mb35@sanger.ac.uk

BOERNERT Klara
Seedcare
Syngenta
BASEL – SWITZERLAND
+41 6 13 23 97 46
klara.boernert@syngenta.com

BOISEN Morten
Global Marketing
Adama Agricultural Solutions
CHRISTIANSFELD – DENMARK
+45 74 56 85 85
morten.boisen@adama.com

BONAVENTURE Kenou Gnimavo
Ghent University
GHENT – BELGIUM
kenoubonaventure@gmail.com

BONNET Gregori
Vegetable
Syngenta Seeds
SARRIANS – FRANCE
+33 621761714
gregori.bonnet@syngenta.com

BORGMEIER Abigail
Dept. of Plant Pathology
University of Nebraska-Lincoln
LINCOLN – USA
+1 402 472 5700
aborgmeier2@huskers.unl.edu

BOROS Gergely
Dept. of Zoology and Animal Ecology
Szent István University
GÖDÖLLO – HUNGARY
+36 28 522 000
henlea@gmail.com

BOURNAUD Caroline
Department of Plant Sciences
University of Cambridge
CAMBRIDGE – UK
+33 1 22 37 48 96 9
bournaudcaroline8@gmail.com

BOZBUGA Refik
Eskisehir Osmangazi University
ESKISEHIR – TURKEY
+903 223 441 784
rbozbuga@yahoo.com

BOZINÉ PULLAI Krisztina
Plant Protection Institute
Szent István University
GÖDÖLLO – HUNGARY
+36 28 522 000
kriszti.pullai@gmail.com

BRAIMI Amina
Lab of Biotech & Valorization of Natural Resources
Ibn Zohr University
AGADIR – MOROCCO
+212 655 758 056
aminabraimi7075@gmail.com

BRASILEIRO Ana C M
Genetic Resources and Biotechnology
Embrapa
BRASÍLIA – BRAZIL
+55 6 13 44 84 90 1
ana.brasileiro@embrapa.br

BRAUN MIYARA Sigal
Entomology and Nematology Department
Agricultural Research Organization Volcani C
RISHON LEZION – ISRAEL
+972 506 220 084
sigalhor@volcani.agri.gov.il

BRINKMAN Pella
Field Crops
WUR
LELYSTAD – THE NETHERLANDS
+31 3 20 29 32 66
pella.brinkman@wur.nl

BRITO Janete
Division of Plant industry
GAINESVILLE – USA
+1 352 395 4758
janete.brito@fdacs.gov

BUCKI Patricia
Plant Protection Department
ARO - Volcani Center
RISHON LEZION – ISRAEL
+972 50 6220525
pbucki@agri.gov.il

BUDHATHOKI Sabina
University of Florida
GAINESVILLE – USA
+1 808 517 2144
sbudhathoki@ufl.edu

BUISSON Mireille
Gautier Semences
EYRAGUES – FRANCE
+33 4 60 24 02 65
mireille.buisson@gautiersemences.com

BULGHERESI Silvia
Environmental Cell Biology
University of Vienna
VIENNA – AUSTRIA
+431427776514
silvia.bulgheresi@univie.ac.at

BURNS Andrew
Molecular Genetics
University of Toronto
TORONTO – CANADA
+1 416 946 8437
andy.burns@utoronto.ca

CABRERA Alfonso
Field Solutions North America
Bayer LEAD Sponsor
FRESNO – USA
+1 55 98 40 70 36
alfonso.cabrera@bayer.com

CALANDRELLI Angelica
Postgraduate Program in Agronomy
State University of Maringa
MARINGA – BRAZIL
+55 (43)99965-5715
a.calandrelli@hotmail.com

CALCAGNO Vincent
Institut Sophia Agrobiotech
INRAe
SOPHIA ANTIPOLIS – FRANCE
+33 492386485
vincent.calcagno@inrae.fr

CAMACHO Maria João
Biologia - NemaLab - IIFA
Universidad de Évora
ÉVORA – PORTUGAL
+351 266 760 848
mjoao.camacho@iniav.pt

CAMPOS-HERRERA Raquel
ICVV
CSIC
LOGROÑO – SPAIN
+34 941 894 980
raquel.campos@icvv.es

CANAGUIER Renaud
NIXE
Hello Nature
VALBONNE – FRANCE
+33 6 82 20 43 37
canaguier@nixe.fr

CARAVEL Claire
INRAe
FRANCE
Claire.caravel@inrae.fr

CARTA Lynn
MNGDBL
USDA Agricultural Research Service
BELTSVILLE – USA
+1 301 504 8787
lynn.carta@usda.gov

CASEY Adam
Faculty of Biological Sciences
University of Leeds
LEEDS – UK
+44 113 343 3035
bsacas@leeds.ac.uk

CASSELLS Maria
Department of Biology
Maynooth University
MAYNOOTH – IRELAND
Maria.Cassells@mu.ie

CASTAGNONE Philippe
UMR1301 IBSV
INRAe
SOPHIA ANTIPOLIS – FRANCE
+33 4 92 38 64 39
philippe.castagnone@inrae.fr

CASTANEDA-ALVAREZ Carlos
Dept. Crop Protection - Laboratory of Nematology
Universidad de Chile
SANTIAGO – CHILE
+56229785821
carlos.castaneda@uchile.cl

CEDANO Carolina
Phytopathology
Universidad Nacional de Trujillo
TRUJILLO – PERU
+51 949 016 952
carolina_cedano@hotmail.com

CHACON Andrea
Functional and Evolutionary Entomology
Gembloux Agro-Bio Tech - TERRA (ULiège)
GEMBLOUX – BELGIUM
+32 081 62 26 63
achacon@uliege.be

CHANNALE Sonal
Centre for Crop Health
University of Southern Queensland
TOOWOOMBA – AUSTRALIA
+61 470 298 365
sonal.channale@usq.edu.au

CHARLOT Bernard
WISSOUS – FRANCE
+33 9 69 20 74 73
agrcharlot@aol.com

CHATWIN Jennifer
Syngenta Crop Protection AG
BASEL – SWITZERLAND
klara.boernert@syngenta.com

CHAUBEY Ashok Kumar
Nematology Laboratory, Department of Zoology
Chaudhary Charan Singh University
MEERUT – INDIA
+91 902 705 0266
akc.nema@gmail.com

CHAUVIN Camille
ELISOL environnement
CONGÉNIES – FRANCE
+33 4 66 71 92 59
camille.chauvin@elisol-environnement.fr

CHAVAN Satish Namdeo
Epigenetics and defence group, Dept. of Biotech
Ghent University
GHENT – BELGIUM
092646234
satish.namdeochavan@ugent.be

CHAVERRI Fabio
IRET
Universidad Nacional
HEREDIA – COSTA RICA
+506 227 73584
fabio.chaverri@una.cr

CHEN Feng
Huazhong Agricultural University
WUHAN – PEOPLE'S REPUBLIC OF CHINA
+86 027 8728 3455
fengmy@webmail.hzau.edu.cn

CHEN Hanqiao
Huazhong Agricultural University
WUHAN – PEOPLE'S REPUBLIC OF CHINA
+86 027 8728 3455
chenhanqiao@webmail.hzau.edu.cn

CHEN Yujin
Molecular Biotechnology
Ghent University
GHENT – BELGIUM
+32 926 462 34
yujin.chen@ugent.be

CHINNASRI Buncha
Dept. of Plant Pathology - Faculty of Agriculture
Kasetsart University
BANGKOK – THAILAND
+66 2579 1026
agrbcc@ku.ac.th

CIANCIO Aurelio
Instituto Protezione Piante
Consiglio Nazionale delle Ricerche
BARI – ITALY
+39 080 5929 221
a.ciancio@ba.ipp.cnr.it

CLARKE Megan
University of Leeds
LEEDS – UK
+44 113 343 5664
bsmgc@leeds.ac.uk

CLAUDIUS-COLE Biodun
Crop Protection and Environmental Biology
University of Ibadan
IBADAN – NIGERIA
+234 802 338 7891
bi_cole@yahoo.com

CLAVERO CAMACHO Ilenia
Plant Protection
Institute for Sustainable Agriculture
CÓRDOBA – SPAIN
957499225
ileniaclavero@gmail.com

COCKX Bram
Department of Biology
KULeuven
LEUVEN – BELGIUM
+32 16 32 75 55
bram.cockx@kuleuven.be

COKE Mirela
Faculty of Biological Sciences
University of Leeds
LEEDS – UK
+44 113 343 5664
M.C.Coke@leeds.ac.uk

COLAGIERO Mariantonietta
Istituto per la Protezione Sostenibile delle Piant
Consiglio Nazionale delle Ricerche
BARI – ITALY
+39 0805 929 222
mariantonietta.colagiero@ipsp.cnr.it

COLLETT Raymond Lesley
Unit for environmental sciences and management
North-West University
POTCHEFSTROOM – SOUTH AFRICA
+27 769640560
pi.collett.rc@gmail.com

COLLIENNE Ursel
R&D
Bayer AG
MONHEIM AM RHEIN – GERMANY
ursel.collienne@bayer.com

CONCEIÇÃO Isabel
Department of Life Sciences
University of Coimbra
COIMBRA – PORTUGAL
+351 239 240 759
luci@zoo.uc.pt

CONSOLI Erika
Crop Protection, Phytopathology and Zoology group
Montana State University
BOZEMAN – USA
+1 4069947844
erikaconsoli@montana.edu

COOLMAN Hilde
HLB B.V.
WIJSTER – THE NETHERLANDS
+31 (0)593 582 828
h.coolman@hlbbv.nl

COOMER Alison
Plant Pathology - Siddique Lab
University of California
DAVIS – USA
+16 3 63 57 37 43
alcoomer@ucdavis.edu

COPELAND Rhys
Crop Biotechnology Research Group
Murdoch University
PERTH – AUSTRALIA
+61 431 315 573
31553633@student.murdoch.edu.au

CORDIER-DEMISSY Marion
Gautier Semences
EYRAGUES – FRANCE
+33 4 90 24 01 86
marion.cordier@gautiersemences.com

COSTA Sofia
Departamento de Biologia
CBMA - University of Minho
BRAGA – PORTUGAL
+351253601500
sofia.costa@bio.uminho.pt

COUVREUR Marjolein
Biology - Nematology research unit
UGent
GHENT – BELGIUM
+32 926 487 40
marjolein.couvreur@ugent.be

COYNE Danny
East Africa
IITA
NAIROBI – KENYA
+254 20 863 2932
d.coyne@cgiar.org

CROW William
Entomology and Nematology Dept.
University of Florida
GAINESVILLE – USA
+1 352 273 3941
wtcr@ufl.edu

CUNHA Maria José
Ciências Agrárias e Tecnologias
Politécnico de Coimbra - CERNAS
COIMBRA – PORTUGAL
+351 239 802 940
mjcunha@esac.pt

DA ROCHA Martine
Institut Sophia Antipolis
INRAe
SOPHIA ANTIPOLIS – FRANCE
+33 4 92 38 66 30
martine.da-rocha@inrae.fr

DA SILVA MATTOS Vanessa
Embrapa
BRASÍLIA – BRAZIL
+55 613 4484 931
vsmattos.agro@gmail.com

DAHLIN Paul
Plant Protection
Agroscope
WÄDENSWIL – SWITZERLAND
+41 58 480 84 87
paul.dahlin@agroscope.admin.ch

DALAN Loel
Nematology Research Unit
Ghent University
GHENT – BELGIUM
+63 632 214 050
loel.dalan@g.msuiit.edu.ph

DALZELL Johnathan
Queen's University Belfast
BELFAST – UK
+44 2890 975 875
jjdalzell@gmail.com

DAMME Nicole
Plant
ILVO
MERELBEKE – BELGIUM
+32 9 27 22 90 0
nicole.damme@ilvo.vlaanderen.be

DANCHIN Etienne
Institut Sophia Agrobiotech
SOPHIA ANTIPOLIS – FRANCE
+33 492 386 402
etienne.danchin@inrae.fr

DANEEL Mieke
Crop Protection
ARC - Tropical and Subtropical Crops
NELSPRUIT – SOUTH AFRICA
+27 137 537 000
mieke@arc.agric.za

DANSO Yaw
Nematology Lab
Crops Research Institute
KUMASI – GHANA
abiacea@yahoo.co.uk

DAUB Matthias
Plant Protection in Field Crops and Grassland
Julius Kuehn Institute
ELSDORF – GERMANY
+49 227 464 46
matthias.daub@julius-kuehn.de

DAUM Eve
Syngenta
BASEL – SWITZERLAND
eve.daum@syngenta.com

DAUMANN Manuel
Professional & Specialty Solutions
BASF SE
LIMBURGERHOF – GERMANY
+49 621 60-74283
manuel.daumann@basf.com

DAVIS Eric
Department of Entomology & Plant Pathology
North Carolina State University
RALEIGH – USA
+1 919 515 6130
eric_davis@ncsu.edu

DE ALMEIDA ENGLER Janice
INRAe
SOPHIA ANTIPOLIS – FRANCE
+33 4 92 38 64 59
janice.de-almeida@inra.fr

DE GOEDE Ron
Soil Biology Group
Wageningen University & Research
WAGENINGEN – THE NETHERLANDS
+31 3 17 48 50 48
ron.degoede@wur.nl

DE LUCA Francesca
Nematology
Institute for Sustainable Plant Protection
BARI – ITALY
+39 080 592 9227
francesca.deluca@ipsp.cnr.it

DE MELLO Sueli Correa Marques
Phytopathology Laboratory
Embrapa Genetic Resources and Biotechnology
BRASÍLIA – BRAZIL
+55 61 344 84776
sueli.mello@embrapa.br

DE OLIVEIRA Clemen
University of Florida
WIMAUMA – USA
+1 81 34 19 65 82
clemendeoliveira@ufl.edu

DE SMIDT Anke
North-West University
SOUTH AFRICA
ankedesmidt@gmail.com

DE WAAL Jeanne
Field Science
Corteva AgriScience
STELLENBOSCH – SOUTH AFRICA
0636946877
jeanne.dewaal@corteva.com

DEBENEDET Erica
Hello Nature
ITALY
erica.debenedetti@hello-nature.com

DECRAEMER Wilfrida
Nematology Research Unit
Ghent University
GHENT – BELGIUM
+32 09 023 50 66
wilfrida.decraemer@ugent.be

DEGROOTE Eva
Ghent University
GHENT – BELGIUM
Eva.Degroote@UGent.be

DEHENNIN Inge
Biology - Nematology Research Unit
Ghent University
GHENT – BELGIUM
+32 926 452 16
inge.dehennin@ugent.be

DEL VALLE Sylvia
Pathologie
Takii France
EYRAGUES – FRANCE
+33 4 90 92 81 86
s.chaneac@takii.fr

DELGADO Martín Augusto
Phytopathology
Universidad Privada Antenor Orrego
TRUJILLO – PERU
+51 994 214 001
mdelgadoj@upao.edu.pe

DEMANGEAT Gérard
UMR SVQV, Team Virology and Vection
INRAe
COLMAR – FRANCE
+33 3 89 89 22 49 76
gerard.demangeat@inrae.fr

DESAEGER Johan
University of Florida
WIMAUMA – USA
+1 813 419 6592
jad@ufl.edu

DESPOT-SLADE Evelin
Divison of Molecular Biology
Ruder Boskovic Institute
ZAGREB – CROATIA
Evelin.Despot.Slade@irb.hr

DESTA Temesgen Addis
e-nema GmbH
SCHWENTINENTAL – GERMANY
+49 4307 8295 278
t.addis@e-nema.de

DIANO Michelle Anne
IMaNema program
Ghent University
GENT – BELGIUM
michelleanne.diano@g.msuiit.edu.ph

DIAS-ARIEIRA Claudia
State University of Maringa
UMUARAMA – BRAZIL
+55 4 49 84 03 31 98
crdarieira@uem.br

DICKSON Donald
Entomology and Nematology
University of Florida
GAINESVILLE – USA
+1 352 273 3938
dwd@ufl.edu

DIGENNARO Peter
Entomology and Nematology
University of Florida
GAINESVILLE – USA
+1 352 273 3904
pdigennaro@ufl.edu

DILLMAN Adler
University of California
RIVERSIDE – USA
+1 95 18 27 39 12
adlerd@ucr.edu

DING Serena
Genes and Behaviour
Max Planck Institute of Animal Behaviour
KONSTANZ – GERMANY
+49 7 53 18 85 90 4
serena.ding@ab.mpg.de

DJIAN-CAPORALINO Caroline
SPE - UMR IBSV
INRAe
SOPHIA ANTIPOLIS – FRANCE
+33 4 92 38 64 58
caroline.caporalino@inrae.fr

DONG Lemeng
Plant Hormone Biology group - SILS
University of Amsterdam
AMSTERDAM – THE NETHERLANDS
+31 6 16 43 17 99
l.dong2@uva.nl

DOS SANTOS Maria Amelia
Laboratório de Nematologia
Universidad Federal de Uberlândia
UBERLÂNDIA – BRAZIL
+55 34 25 12 67 25
ameliaufu@gmail.com

DOUDA Ond?ej
Crops Research Institute
RUZYN? – CZECH REPUBLIC
+42 0 23 30 22 49 4
douda@vurv.cz

DRITSOULAS Alexandros
Entomology & Nematology dpt. - Larry Duncan's
Citrus Research and Education Center
LAKE ALFRED – USA
+1 863 956 8822
alexdritsi@gmail.com

DU PREEZ François
Faculty of AgriSciences
Stellenbosch University
STELLENBOSCH – SOUTH AFRICA
fdupreez@sun.ac.za

DU PREEZ Gerhard
Unit for Environmental Sciences and Management
North-West University
POTCHEFSTROOM – SOUTH AFRICA
+27 7417 55407
gerhard.dupreez@nwu.ac.za

DUBE Zakheleni
School of Biological and Environmental School
University of Mpumalanga
MBOMBELA – SOUTH AFRICA
+271 300 202 63
zakheleni.dube@ump.ac.za

DUNCAN Larry
University of Florida
LAKE ALFRED – USA
+1 863 956 8821
lwduncan@ufl.edu

DUNN Murray David
Stellenbosch University
STELLENBOSCH – SOUTH AFRICA
+27 723 583 597
mdunn94@sun.ac.za

EBRAHIM Awol Seid
Plant Sciences
Wollo University
DESSIE – ETHIOPIA
+251 255 530 325/31/54
awolseid07@gmail.com

EHLERS Ralf-Udo
e-nema GmbH
SCHWENTINENTAL – GERMANY
+49 430 782 950
ehlers@e-nema.de

EINARSON Stefan
Brill Publishers
LEIDEN – THE NETHERLANDS
+31 71 535 3500
Einarson@brill.com

ELASHRY Abdelnaser
Genetics and genomics - sugar beet plant breeding
Strube Research
SÖLLINGEN – GERMANY
+49 15 25 48 71 214
a.elashry@strube-research.net

ELBORAI Fahiem
Entomology and Nematology
University of Florida
WIMAUMA – USA
+18 6 34 19 66 12
fahiem@ufl.edu

ELEKCIOGLU Ibrahim Halil
Faculty of Agriculture, Plant Protection Dept
Cukurova University
ADANA – TURKEY
+90 322 338 6727
halile@cu.edu.tr

ELIÁS Sara
Masaryk University
BRNO – CZECH REPUBLIC
+420 549 49 3419
saraelias43@gmail.com

EMERSON Michael
Plant Pathology
University of Arkansas
LONOKE – USA
+1 501 676 3124
memerson@uada.edu

ERDOS Eszter
Plant Protection Institute
Szent István University
GÖDÖLLO – HUNGARY
+36 285 220 00
erdos.esztike@gmail.com

ESCOBAR AVILA Ilia Mariana
Fernando de la Jara
Lab. de Nematología Agrícola
MEXICO – MEXICO
+52 5557296000 ext. 62
mariana_miss140@hotmail.com

ESLAH Shemila Hassen
Ghent University
GHENT – BELGIUM
remuyahasu@gmail.com

ESMENJAUD Daniel
UMR interactions biotiques et santé végétale
INRAe
SOPHIA ANTIPOLIS – FRANCE
+33 4 92 38 64 00
daniel.esmenjaud@inrae.fr

ESPADA Margarida
Mediterranean Institute for Agriculture - MED
University of Évora
ÉVORA – PORTUGAL
+35 1 26 67 40 80 0
mespada@uevora.pt

ESPINO CABALLERO Ricardo Florentino
Crop Protecction Department
Universidad Nacional San Luis Gonzaga
ICA – PERU
+51 56 257444
ricardofespinoc@hotmail.com

EVES-VAN DEN AKKER Sebastian
University of Cambridge
CAMBRIDGE – UK
se389@cam.ac.uk

FABIYI Oluwatoyin
Department of Crop Protection
University of Ilorin
ILORIN – NIGERIA
+234 706 435 6721
fabiyitoyinike@hotmail.com

FADAKAR Samira
Department of Plant Pathology
Tarbiat Modares University
TEHRAN – ISLAMIC REPUBLIC OF IRAN
+98 21 482 922 66
fadakar9473@yahoo.com

FANELLI Elena
Nematology
Institute for Sustainable Plant Protection
BARI – ITALY
+39 080 592 9246
elena.fanelli@ipsp.cnr.it

FARIA Jorge M. S.
Laboratorio de Nematologia
INIAV
OEIRAS – PORTUGAL
(+351) 214 403 500
jorge.faria@iniav.pt

FAVERY Bruno
IPN
INRAe - UCA - CNRS
SOPHIA ANTIPOLIS – FRANCE
+33 4 92 38 64 64
bruno.favery@inra.fr

FÉLIX Marie-Anne
Institut Jacques Monod
PARIS – FRANCE
+33 1 44 32 36 05
felix@bio.ens.psl.eu

FERREIRA Tiarin
Crop Protection Development
Syngenta South Africa
CENTURION – SOUTH AFRICA
tia.ferreira@syngenta.com

FINKERS-TOMCZAK Anna
Vegetable Crop Research
KeyGene
WAGENINGEN – THE NETHERLANDS
+31 317 466 866
ihr@keygene.com

FOLCHER Laurent
Nematology Unit - EURL Plant Parasitic Nematodes
Anses - Plant Health Laboratory
LE RHEU – FRANCE
+33 2 99 30 48 28
laurent.folcher@anses.fr

FONTANA Lais Fernanda
Fitopatology - Nematology
UEM
UMUARAMA – BRAZIL
+55 449 990 73818
laisffontana@hotmail.com

FONTES Maria Geane
East-West Seed Guatemala
SANTA LUCIA MILPAS – GUATEMALA
+502 779 3 52 52
geane.fontes@eastwestseed.com

FOUCHER Justine
GEVES
BEAUCOUZÉ – FRANCE
+33 241228619
justine.foucher@geves.fr

FOURIE Hendrika
Unit for Environmental Sciences and Management
North-West University
POTCHEFSTROOM – SOUTH AFRICA
0182933683
driekie.fourie@nwu.ac.za

FOURNET Sylvain
UMR IGEPP 1349
INRAe
LE RHEU – FRANCE
+33 2 23 48 51 58
sylvain.fournet@inrae.fr

FOURNOL Stephanie
Corteva AgriScience
GLAND – SWITZERLAND
stephanie.fournol@corteva.com

FREITAS DE ALMEIDA Sheila
Fhytopathology Laboratory
University of Brasília
BRASÍLIA – BRAZIL
+55 613 448 4686
sheilafreitas92@hotmail.com

FRETTINGER Patrick
Global Regulatory Affairs
BASF
ECULLY – FRANCE
+33622477200
patrick.frettinger@basf.com

GABERTHÜEL Matthias
Syngenta Crop Protection AG
BASEL – SWITZERLAND
+41 613 237 133
matthias.gaberthueel@syngenta.com

GABL Ines
Department for Healthy Plants in Arable Crops
Austrian Agency for Health and Food Savety
VIENNA – AUSTRIA
+43 505 553 3315
ines.gabl@ages.at

GABRIEL Marcia
Universidad Federal de Santa Maria
SANTA MARIA – BRAZIL
gabriel.marcia@gmail.com

GALEANO Magda
R&D
Koppert Espana
VICAR – SPAIN
+34 9 02 48 99 00
mgaleano@koppert.es

GALHANO Cristina
Department of Environment
ESAC - IPC - CFE
COIMBRA – PORTUGAL
+351 239 802 940
cicgalhano@esac.pt

GAMEL Sylvie
Nematology unit - EURL Plant parasitic nematodes
Anses - Plant Health Laboratory
LE RHEU – FRANCE
+33 2 99 30 92 31
sylvie.gamel@anses.fr

GANSFORT Birgit
Bielefeld University
BIELEFELD – GERMANY
05211062703
birgit.gansfort@uni-bielefeld.de

GARCIA Nathan
Nematology unit
Anses - Plant Health Laboratory
LE RHEU – FRANCE
+33 2 99 30 90 37
nathan.garcia@anses.fr

GARRIGA OLIVERAS Anna
Centro de Biotecnologia dos Açores
Universidad dos Açores
PONTA DELGADA – PORTUGAL
+34 6 61 19 10 68
anna.garriga.oliveras@uab.cat

GARTNER Ulrike
James Hutton Institute
AUSTRIA
ulrike.gartner@sbg.ac.at

GATTONI Kaitlin
University of Florida
GAINESVILLE – USA
+1 201 316 7519
kgattoni@ufl.edu

GENDRON Eli
University of Florida
GAINESVILLE – USA
+ 505 948 8029
egendron@ufl.edu

GERBORE Jonathan
R&D
Koppert
CAVAILLON – FRANCE
jgerbore@koppert.fr

GERIC STARE Barbara
Plant Protection Departmen
Kmetijski institut Slovenije
LJUBLJANA – SLOVENIA
+386 1 280 52 62
barbara.geric@kis.si

GHEYSEN Godelieve
Ghent University
GHENT – BELGIUM
+32 9 26 45 88 8
godelieve.gheysen@ugent.be

GITHINJI Zipporah
Ghent University
GHENT – BELGIUM
Zipporah.Githinji@UGent.be

GLEASON Cynthia
Washington State University
PULLMAN – USA
+1 509 335 3742
cynthia.gleason@wsu.edu

GODDARD Tom
BASF
LITTLEHAMPTON – UK
+44 1903 736 928
thomas.goddard@basf.com

GODINHO DE ARAÚJO Fernando
Campus Urutaí
Instituto Federal Goiano
PIRES DO RIO – BRAZIL
+55 643 465 1973
fernando.godinho@ifgoiano.edu.br

GÓDOR Anita
Plant Protection Institute
Szent István University
GÖDÖLLO – HUNGARY
+36 2 04 92 60 34
agodor@gmail.com

GOMES Cesar
Fitopatology and Nematology
Embrapa Clima Temperado
PELOTAS – BRAZIL
+53 3275 8152
cbauergbr@gmail.com

GONZÁLEZ-PAZ Lenin
University of Zulia
MARACAIBO – VENEZUELA, RB
+58 424 6201 133
lgonzalezpaz@gmail.com

GORDON Kara
Auburn University
AUBURN – USA
+1 281 910 2981
klg0037@auburn.edu

GOVERSE Aska
Laboratory of Nematology
Wageningen University & Research
WAGENINGEN – THE NETHERLANDS
+31 3 17 48 50 86
aska.goverse@wur.nl

GOVINDASAMY Kavitha
Tamil Nadu Agricultural University
COIMBATORE – INDIA
08056506038
kavitha_nem@yahoo.com

GRABAU Zane
Entomology & Nematology
University of Florida
GAINESVILLE – USA
+1 352 273 3983
zgrabau@ufl.edu

GREMINGER Jennifer
Bayer
SAINT-LOUIS – USA
+1 314 694 9189
jennifer.greminger@bayer.com

GRENIER Eric
UMR IGEPP
INRAe
LE RHEU – FRANCE
+33 2 23 48 51 73
eric.grenier@inrae.fr

GROSSI DE SA Maira
Nematology Unit - EURL Plant parasitic nematodes
Anses - Plant Health Laboratory
LE RHEU – FRANCE
+33 2 99 30 48 28
maira.grossidesa@anses.fr

GROSSI-DE-SA Maria Fatima
Biotechnology
Embrapa Genetic Resources and Biotechnology
BRASÍLIA – BRAZIL
+55 6 13 44 84 902
fatima.grossi@embrapa.br

GRUBISIC Dinka
Department of Agricultural Zoology
University of Zagreb Faculty of Agriculture
ZAGREB – CROATIA
+38 51 23 93 967
djelinic@agr.hr

GRUNDLER Florian
Molecular Phytomedicine
University Bonn
BONN – GERMANY
+49 228 731 675
grundler@uni-bonn.de

GUARNERI Nina
Wageningen University & Research
WAGENINGEN – THE NETHERLANDS
+313 17 48 55 37
nina.guarneri@wur.nl

GUDEN Rodgee Mae
Marine Biology Lab
Ghent University
GHENT – BELGIUM
+32 470 751 100
rodgeemae.guden@ugent.be

GUERRA Mayamaru
Universidad Tecnológica de Bolívar
CARTAGENA – COLOMBIA
+57 300 321 8812
mayamaru.guerra@gmail.com

GUIMARAES Patricia
Biotechnology
Embrapa - Cenargen
BRASÍLIA – BRAZIL
+55 6 13 44 84 787
patricia.guimaraes@embrapa.br

GUTIÉRREZ-GUTIÉRREZ Carlos
NemaLab
ICAAM - Universidade de Évora
ÉVORA – PORTUGAL
+35 266 740 800
carlosg@uevora.pt

H. VON REUSS Stephan
Laboratory for Bioanalytical Chemistry
University of Neuchatel
NEUCHATEL – SWITZERLAND
+41 327182510
stephan.vonreuss@unine.ch

HAFEZ Saad
University of Idaho
PARMA – USA
+1 208 880 5005
shafez@uidaho.edu

HAJIHASSANI Abolfazl
Plant Pathology
University of Florida
DAVIS – USA
+1 954 5 77 63 33
ahajihassani@ufl.edu

HALLMANN Johannes
Institute of Epidemiology and Pathogen Diagnostics
Julius Kuehn Institute
BRAUNSCHWEIG – GERMANY
+49 531 2993700
johannes.hallmann@julius-kuehn.de

HAMMAM Mostafa
Plant pathology
National Research Center
CAIRO – EGYPT
mmahammam@yahoo.com

HAN Ziduan
Department 4
Max Planck Institute
TÜBINGEN – GERMANY
01637164906
ziduan.han@tuebingen.mpg.de

HARRINGTON Sean
Pharmacology & Toxicology
University of Toronto
TORONTO – CANADA
+1 416 946 8437
sean.harrington@mail.utoronto.ca

HARTMANN-WITTULSKY Sebastian
Crop Strategy & Portfolio Management
Bayer
MONHEIM AM RHEIN – GERMANY
+49 2173 38 7343
sebastian.wittulsky@bayer.com

HASAN Md Shamim
Molekulare Phytomedizin
University of Bonn
BONN – GERMANY
+49 228 73 1764
shamim.hasan@uni-bonn.de

HASSAN Ahmed
Faculty of Biological Sciences
University of Leeds
LEEDS – UK
+33 113 343 5664
bsarhh@leeds.ac.uk

HASSANALY GOULAMHOUSSEN Rahim
Sophia Agrobiotech Institute
SOPHIA ANTIPOLIS – FRANCE
+33601482820
rahim.hassanaly-goulamhoussen@inrae.fr

HAUKELAND Solveig
Plant Health
ICIPE
NAIROBI – KENYA
shaukeland@icipe.org

HAYES Ashley
Faculty of Biological Sciences
University of Leeds
LEEDS – UK
+44 113 343 5664
bsaha@leeds.ac.uk

HEITMANN Carolina
Syngenta Crop Protection AG
BASEL – SWITZERLAND
klara.boernert@syngenta.com

HELDER Johannes
Lab Nematology
Wageningen University & Research
WAGENINGEN – THE NETHERLANDS
+31 317 483 136
hans.helder@wur.nl

HERNANDEZ-OCHANDÍA Dainé
Nematology Laboratory
National Center of Animal and Plant Health
LA HABANA – CUBA
47849148
daineh@censa.edu.cu

HEUER Holger
Julius Kuehn Institute
BRAUNSCHWEIG – GERMANY
+49 531 299 37 17
holger.heuer@julius-kuehn.de

HEYDARI Fariba
University of Fribourg
FRIBOURG – SWITZERLAND
026 300 71 11
fariba.heydari@unifr.ch

HILTL Clarissa
INRES - Molecular Phytomedicine
University of Bonn
BONN – GERMANY
+49 228 73 16 70
clarissa.hiltl@uni-bonn.de

HILTPOLD Ivan
Entomology
Agroscope
NYON – SWITZERLAND
+41 58 48 49 281
ivan.hiltpold@agroscope.admin.ch

HODDA Michael
National Research Collections
CSIRO
CANBERRA – AUSTRALIA
+61 2 6246 4371
mike.hodda@csiro.au

HOLDEN-DYE Lindy
School of Biological Sciences
University of Southampton
SOUTHAMPTON – UK
+44 238 059 9006
lmhd@soton.ac.uk

HOLGADO Ricardo
Division of Biotechnology and Plant Health
Norwegian Institute of Bioeconomy Research
ÅS – NORWAY
+47 916 82 703
ricardo.holgado@nibio.no

HOLMES Will
Syngenta Crop Protection AG
BASEL – SWITZERLAND
klara.boernert@syngenta.com

HOLOVACHOV Oleksandr
Department of Zoology
Swedish Museum of Natural History
STOCKHOLM – SWEDEN
+46 733 402 568
oleksandr.holovachov@nrm.se

HÖSS Sebastian
Ecossa
STARNBERG – GERMANY
+4981515509172
hoess@ecossa.de

HUMPHREYS-PEREIRA Danny
Laboratory of Nematology - CIPROC
University of Costa Rica
MONTES DE OCA – COSTA RICA
+506 251 15421
danny.humphreys@ucr.ac.cr

HUNT Vicky
Department of Biology & Biosciences
University of Bath, UK
BATH – UK
+44 1225 383703
v.l.hunt@bath.ac.uk

INACIO Maria L.
Plant Health/ Nematology Lab
INIAV
OEIRAS – PORTUGAL
+35 916 626 641
lurdes.inacio@iniav.pt

IQBAL Mudassir
Forest Mycology and Plant Pathology
Swedish University of Agricultural Sciences
UPPSALA – SWEDEN
+46 76 749 0413
mudassir.iqbal@slu.se

JACKSON Abigail
Department of Biology
Brigham Young University
PROVO – USA
ajackson260@gmail.com

JAHANSHAHI AFSHAR Farahnaz
Department of Nematology
Iranian Research Institute of Plant Protect.
TEHRAN – ISLAMIC REPUBLIC OF IRAN
+98 212 240 3012
afshar_fj@yahoo.com

JANSEN VAN RENSBURG Candice
Zoology and Entomology
University of the Free State
BLOEMFONTEIN – SOUTH AFRICA
+27 514 019 357
jvrensc@ufs.ac.za

JAUBERT-POSSAMAI Stéphanie
INRAe
SOPHIA ANTIPOLIS – FRANCE
+33492386464
stephanie.jaubert@inrae.fr

JENSSEN Jared
AgBiome
RESEARCH TRIANGLE PARK – USA
6025022628
jjensen@agbiome.com

JINDAPUNNAPAT Kansiree
Khon Kaen University
MUEANG – THAILAND
+66 43 34 31 14
kansji@kku.ac.th

JONES John
Cell and Molecular Sciences
The James Hutton Institute
DUNDEE – UK
+44 1382 568 828
john.jones@hutton.ac.uk

JORNA Jesse
Dep. of Biology, Nematode Evolution Lab
Brigham Young University
PROVO – USA
+1 801 422 8723
jjgjorna@outlook.com

JUNIOR KIKA Lusu
Department of Biology
Ghent University
GHENT – BELGIUM
kikalusu@gmail.com

JUSTUS Aisu
Ghent University
GHENT – BELGIUM
ayso.gasts2015@alexu.edu.eg

KAGIMU Nicholas
FABI - Zoology and Entomology
Future Africa - University of Pretoria
HILCREST – SOUTH AFRICA
0603312122
nalwogaf@gmail.com

KAKOULI-DUARTE Thomaé
enviroCORE, Department of Science and Health
Institute of Technology
CARLOW – IRELAND
+353 59 9175545
thomae.kakouli@itcarlow.ie

KALOSHIAN Isgouhi
Nematology
University of California
RIVERSIDE – USA
+1 95 18 27 39 13
isgouhi.kaloshian@ucr.edu

KAMMERER Christian
University of Florida
GAINESVILLE – USA
kammech@ufl.edu

KANFRA Xorla
EP
Julius Kuehn Institute
BRAUNSCHWEIG – GERMANY
+49 53 12 22 38 26
xorla.kanfra@julius-kuehn.de

KAPLAN Fatma
Pheronym Inc.
DAVIS – USA
+13522836967
fkaplan@pheronym.com

KARPINSKA Anna
Science and Health
Institute of Technology
CARLOW – IRELAND
0851778669
anna.karpinska@itcarlow.ie

KATOOLI Nafiseh
Plant Protection Department
Ferdowsi University of Mashhad
MASHHAD – ISLAMIC REPUBLIC OF IRAN
+98 513 880 2415
n.katooli@gmail.com

KIDANE Selamawit
International Inst. of Tropical Agriculture
NAIROBI – KENYA
+25 4 20 86 54 00 0
sa.kidane@cgiar.org

KIEWNICK Sebastian
Inst. A. - Nematology
Julius Kuehn Institute
BRAUNSCHWEIG – GERMANY
+49 531 229 3929
sebastian.kiewnick@julius-kuehn.de

KIM Chami
Syngenta
SAINT-SAUVEUR – FRANCE
chami.kim@syngenta.com

KING Erna
Wellcome Sanger Institute
CAMBRIDGE – UK
+44 12 2 38 34 24 4
ek5@sanger.ac.uk

KISITU Joseph
Nematology
International Inst. of Tropical Agriculture
KAMPALA – UGANDA
+25 6 70 49 93 34 9
jspkst@gmail.com

KNOETZE Rinus
Plant Protection
Agricultural Research Council
STELLENBOSCH – SOUTH AFRICA
+27 21 8093461
knoetzer@arc.agric.za

KNOX Jessica
Department of Molecular Genetics
University of Toronto
TORONTO – CANADA
+1 416 946 8437
jess.knox@utoronto.ca

KOHN CARNEIRO Aline
C214
INRAe
SOPHIA ANTIPOLIS – FRANCE
+33 0492386465
alinekohn@hotmail.com

KOMBRINK Anja
HLB B.V.
WIJSTER – THE NETHERLANDS
+31 615 496 140
a.kombrink@hlbbv.nl

KONG Pengfei
Cell Press
AMSTERDAM – THE NETHERLANDS
+31 6 51 64 37 17
pkong@cell.com

KÖNIG Dr. Stephan
Institute for National and International Plant Hea
Julius Kuehn Institute
BRAUNSCHWEIG – GERMANY
+49 (0) 531 2994332
stephan.koenig@julius-kuehn.de

KONIGANAHALLI GOPAL Hemanth Kumar
Inst. for Plant Protec. in Field Crops & Grassland
Julius Kuehn Institute
BRAUNSCHWEIG – GERMANY
+49 5 31 29 93 93 2
hemanth.gopal@julius-kuehn.de

KONOPICKÁ Jana
Institute of Entomology
Biology Centre CAS
CESKÉ BUDEJOVICE – CZECH REPUBLIC
+42 038 777 5245
jkonopicka@seznam.cz

KOUTSOVOULOS Georgios
INRAe
SOPHIA ANTIPOLIS – FRANCE
+33492646538
georgios.koutsovoulos@inra.fr

KRANSE Olaf
Plant Science Department
University of Cambridge
CAMBRIDGE – UK
ok297@cam.ac.uk

KREUZINGER- JANIK Bianca
Bielefeld University
BIELEFELD – GERMANY
+495211062703
bianca.kreuzinger-janik@uni-bielefeld.de

KUD Joanna
EPPN
University of Idaho
MOSCOW – USA
jkud@uidaho.edu

KUDJORDJIE Enoch Narh
Department of Agroecology
Aarhus University
SLAGELSE – DENMARK
+45 24 21 72 33
enochnarh@agro.au.dk

KULYA Cosimasi
Ghent University
GHENT – BELGIUM
kulyacosmas@gmail.com

KUMAR Anil
Dept. of Entomology, Nematology and Chemistry
Agricultural Research Organization
RISHON LEZION – ISRAEL
+97 2 39 68 33 53
anilmalik58@gmail.com

KURTZ Benedikt
SC Insecticides & Nematicides
Syngenta Crop Protection
STEIN – SWITZERLAND
+41 628 66 03 27
benedikt.kurtz@syngenta.com

LANGENDOEN Daan
Phytopathology
Averis Seeds
VALTHERMOND – THE NETHERLANDS
+31 5 98 66 21 02
daan.langendoen@avebe.com

LAPEYRE Laurent
NIXE Laboratoire
SOPHIA ANTIPOLIS – FRANCE
+33 4 92 38 64 52
lapeyre@nixe.fr

LAWRENCE Kathy
Entomology & Plant Pathology
Auburn University
AUBURN – USA
+1 513 787 7605
lawrekk@auburn.edu

LAZNIK Ziga
University of Ljubljana, Biotechnical facult
LJUBLJANA – SLOVENIA
040758095
ziga.laznik@bf.uni-lj.si

LE ROUX Anne-Claire
INRAe - UMT InnoPlant
LE RHEU – FRANCE
+33 2 23 48 51 86
anneclaire.leroux@inov3pt.fr

LEE Wan-Chun
Plant Pathology and Microbiology
National Taiwan University
TAIPEI – REPUBLIC OF CHINA (TAIWAN)
+886 233 664 601
r06633006@ntu.edu.tw

LEOCATA Salvatore
Studio Tecnico ASA
CATANIA – ITALY
+39 335 1242 837
sleocata@studiotecnicoasa.it

LETIA Sharon
Molecular Phytomedicine
University of Bonn
BONN – GERMANY
015 7 34 72 21 19
s7shleti@uni-bonn.de

LILLO Paula
Department of Environment and Agronomy
INIA - CSIC
MADRID – SPAIN
+34677039377
paula.lillo@inia.es

LOMBARD Lorenzo
St. NAK
EMMELOORD – THE NETHERLANDS
l.lombard@nak.nl

LONDONO Diana
Global R&D Biologicals
BASF
DURHAM – USA
+1 919 547 2496
diana.londono@basf.com

LOPES Carina Mariani
Nematology
Nemafito
LUIS EDUARDO MAGALHAES – BRAZIL
5577999470381
carina@nemafito.com

LOPEZ Juan Pablo
Syngenta Crop Protection AG
BASEL – SWITZERLAND
juan_pablo.lopez@syngenta.com

LOPEZ-NICORA Horacio
Department of Plant Pathology
The Ohio State University
COLUMBUS, OH – USA
614-688-0528
lopez-nicora.1@osu.edu

LOURENÇO-TESSUTTI Isabela
Limpp
Embrapa
BRASÍLIA – BRAZIL
+556134484926
isabela.lourenco@embrapa.br

LOVATO Bruno
Global Development
Adama Agricultural Solutions
SCHAFFHAUSEN – SWITZERLAND
+41 79 56 86 909
bruno.lovato@adama.com

LOZANO-TORRES Jose L.
Laboratory of Nematology
Wageningen University & Research
WAGENINGEN – THE NETHERLANDS
+31 317 485 254
jose.lozano@wur.nl

LUFF Kelly
CropScience Division
Bayer
KIMBERLY – USA
+1 208 423 6366
kelly.luff@bayer.com

MA Xinyuan
Plant and Environmental Sciences
Clemson University
CLEMSON – USA
+1 864 650 1438
xm@g.clemson.edu

MACGUIDWIN Ann
Plant Pathology
University of Wisconsin
MADISON – USA
+1 608 843 1635
aem@plantpath.wisc.edu

MADAURE Jacqueline Tinashe
Plant Production
University of Limpopo
MANKWENG – SOUTH AFRICA
+27 6 78 05 16 34
jacquelinemadaure@yahoo.com

MAGELEUW Clinton Mbokeh
Ghent University
GHENT – BELGIUM
mbohkeclintonmageleuw@gmail.com

MAGNUSSON Christer
Norwegian Institute of Bioeconomy Research
ÅS – NORWAY
+47 952 053 04
christer.magnusson@nibio.no

MAJDI Nabil
Animal Ecology
University of Bielefeld
BIELEFELD – GERMANY
+49 521 106 2703
nabil.majdi@uni-bielefeld.de

MAJI Ivana
University of Osijek
OSIJEK – CROATIA
+38531554905
imajic@fazos.hr

MALAN Antoinette
Conservation Ecology and Nematology
Stellenbosch University
STELLENBOSCH – SOUTH AFRICA
+27 21 808 2921
apm@sun.ac.za

MANASOVÁ Marie
Department of Plant Protection
Czech University of Life Sciences
PRAGUE – CZECH REPUBLIC
+420724037696
manasova@af.czu.cz

MANCEAU Florian
RD3PT
ACHICOURT – FRANCE
florian.manceau@inov3pt.fr

MANTELIN Sophie
CMS
The James Hutton Institute
DUNDEE – UK
+44 3449 285 428
sophie.mantelin@inrae.fr

MANZANILLA-LÓPEZ Rosa Helena
CEPROBI
Instituto Politécnico Nacional de México
HARPENDEN – UK
+44 1582 769 201
rosa.manzanilla@gmail.com

MARCHENKO Polina
University of Natural Resources
TULLN AN DER DONAU – AUSTRIA
+43 1 47 654 95306
polina.marchenko@students.boku.ac.at

MARIETTE Nicolas
Unité de nématologie
Anses - Plant Health Laboratory
LE RHEU – FRANCE
+33 2 23 48 51 73
nicolas.mariette@anses.fr

MARQUEZ Josiah
Plant pathology
University of Georgia
TIFTON – USA
+1 808 391 7159
josiah.marquez@uga.edu

MARTÍNEZ MORA Celia
IMIDA
LA ALBERCA - MURCIA – SPAIN
+34 9 68 39 45 59
celia.martinez@carm.es

MASSON Anne-Sophie
IPME
IRD
MONTPELLIER – FRANCE
annesophie.masson@ird.fr

MATLALA Francinah Lesego
North-West University
POTCHEFSTROOM – SOUTH AFRICA
+27 838 602 761
u10238663@tuks.co.za

MAULANA Muhammad Iqbal
Laboratory of Nematology
Wageningen University & Research
WAGENINGEN – THE NETHERLANDS
0610325147
muhammad.maulana@wur.nl

MAUSHE Dorothy
Institute of Plant Sciences
University of Bern
BERN – SWITZERLAND
+41 31631 31 55
dorothy.maushe@ips.unibe.ch

MBURU Harrison
East Africa
IITA - Icipe
NAIROBI – KENYA
+254 208 632 932
h.mburu@gmail.com

MCGAWLEY Edward
Plant Pathology
Louisiana State University
BATON ROUGE – USA
+1 225 769 0748
ecmcgawley@yahoo.com

MCPEAK Sloane
Auburn University
AUBURN – USA
+1 281 910 2981
sem0070@auburn.edu

MCQUEEN Parr
Department of Entomology and Nematology
University of Florida
GAINESVILLE – USA
+1 352 273 2936
jparr.mcqueen@ufl.edu

MEDINA Karla
Certis Biologicals
WIMAUMA – USA
+1 614 441 3379
kmedina@certisbio.com

MEIJER Anikó
Ghent University
GHENT – BELGIUM
+32 9 22 65 96 7
aniko.meijer@ugent.be

MEJIA Hugo
Ecology - Nematology
UNAM UNA 290722 7Y5
MEXICO – MEXICO
+52 555 622 4902
hugo_mejia_madrid@ciencias.unam.mx

MELAKEBERHAN Haddish
Department of Horticulture
Michigan State University
EAST LANSING – USA
+1 517 388 4833
melakebe@msu.edu

MENDY Badou
Molecular Phytomedicine
University of Bonn - INRES
BONN – GERMANY
02 28 73 30 69
bmendy@uni-bonn.de

MENNAN Sevilhan
Plant protection department
Ondokuz Mayıs University
SAMSUN – TURKEY
+903 623 121 919
smennan@omu.edu.tr

MEYER Susan
Mycology, Nematology Genetic Diversity and Biology
USDA Agricultural Research Service
BELTSVILLE – USA
+1 301 504 5091
susan.l.meyer@usda.gov

MIAMOTO Angelica
State University of Maringa
MARINGA – BRAZIL
+55 44 999 0 44 320
angelicamiamoto@gmail.com

MILLER Robert
Cell Biology
Universidad de Brasília - CAPES PrInt
BRASÍLIA – BRAZIL
+55 61 31 07 30 48
robertmiller@unb.br

MIRGHASEMI Seyedeh Negin
Plant pathology
University of Guilan
LAHIJAN – ISLAMIC REPUBLIC OF IRAN
+98 0134 222 8908
neginmirghasemi@yahoo.com

MITREVA Makedonka
Washington University School of Medicine
SAINT-LOUIS – USA
+1 31 428 620 05
mmitreva@wustl.edu

MOENS Maurice
Plants and Crops
University Ghent
GHENT – BELGIUM
09 368 00 98
maurice.moens@ugent.be

MOENS Tom
Biology Department, Marine Biology Lab
Ghent University
GHENT – BELGIUM
+32 9 2648522
tom.moens@ugent.be

MOERMAN Tirza
Faculty of Envir. Sciences & Natural Resource
Norwegian University of Life Sciences
ÅS – NORWAY
tirza.moerman@nmbu.no

MOGOLLON ORTIZ Angela Maria
Fungal Molecular Genetics Laboratory
Universidad Federal de Viçosa
VIÇOSA – BRAZIL
+55 31 3899 2553
ammogollon@gmail.com

MOISAN Kay
Koppert Biological Systems
BERKEL EN RODENRIJS – THE NETHERLANDS
+31 6 20 61 15 90
KMoisan@koppert.nl

MOLENDIJK Leendert
WUR Field Crops
Wageningen University & Research
LELYSTAD – THE NETHERLANDS
+31 320 291 644
leendert.molendijk@wur.nl

MOLINA Carlos
e-nema GmbH
SCHWENTINENTAL – GERMANY
+49 430 782 952 81
c.molina@e-nema.de

MONJE Almendra
Entomology
INIA Carillanca
VILCUN – CHILE
+56 45 229 7241
almendra.monje@inia.cl

MONTARRY Josselin
UMR1349 IGEPP
INRAe
LE RHEU – FRANCE
+33 2 23 48 51 73
josselin.montarry@inrae.fr

MONTCHATRE Olivier
Corteva AgriScience
CASTELNEAU LE LEZ – FRANCE
olivier.montchatre@corteva.com

MONTEIRO Luana
Marine Biology Research Unit
Ghent University
GHENT – BELGIUM
+32 926 485 23
luana.dacostamonteiro@ugent.be

MOORE Scott
Syngenta
WEST MONROE – USA
+1 318 282 6552
scott.moore@syngenta.com

MORALES-MONTERO Patricia
Ciencias vegetales - Fitopatología molecular
Pontificia Universidad Catolica de Chile
SANTIAGO – CHILE
+56935739006
pzmorales@uc.cl

MOREIRA David
Nematology
University of Florida
WIMAUMA – USA
daanmoca94@ufl.edu

MOTA Manuel
Nemalab - MED
University of Évora
ÉVORA – PORTUGAL
+351 266 760 800
mmota@uevora.pt

MUIGAI Onesmus Mwaura
Ghent University
GHENT – BELGIUM
onesmusmwaura@gmail.com

MULET Karine
INRAe
FRANCE
karine.mulet@inrae.fr

MUNERA URIBE Gladis Emilia
Agrosavia
RIONEGRO – COLOMBIA
+57 4 537 1490
gladismunera@yahoo.com

MWANGI Grace
NEWPORT, SHROPSHIRE – UK
+254 716 911 858
rycmwangi@gmail.com

NAFARI Reza
Shiraz University
MARVDASHT – ISLAMIC REPUBLIC OF IRAN
989 3 74 45 04 15
rezanafari1987@gmail.com

NAGY Péter
Department of Zoology and Animal Ecology
Szent István University
GÖDÖLLO – HUNGARY
+36 705 560 546
nagy.peter@mkk.szie.hu

NAVAREZ Mara Louisa
Biological Sciences
MSU - Iligan Institute of Technology
ILIGAN – PHILIPPINES
+63 948 684 8943
maralouisa.navarez@g.msuiit.edu.ph

NAVARRO Patricia
Entomology
INIA
VILCUN – CHILE
+56 45 2 297 108
patricia.navarro@inia.cl

NAVIA GINE Pablo A.
Product Development
Adama U.S.
WELLINGTON – USA
+1 229 256 7762
pablo.navia@adama.com

NDAYISENGA Marc
Ghent University
GHENT – BELGIUM
marc1ndayisenga1986@gmail.com

NEHER Deborah
Dept of Plant & Soil Science
University of Vermont
BURLINGTON – USA
+1 802 656 0474
dneher@uvm.edu

NERMUT Jirí
Institute of Entomology
Biology Centre CAS
CESKÉ BUDEJOVICE – CZECH REPUBLIC
+42 038 777 5245
nermut@entu.cas.cz

NEVEUX Marie-Sophie
RD3PT
LE RHEU – FRANCE
+33 2 23 48 70 28
mariesophie.neveux@inov3pt.fr

NGALA Bruno Massa
Organisme Nuisible de Quarantaine et Émergence
FN3PT - RD3PT
ACHICOURT – FRANCE
bruno.ngala@inov3pt.fr

NGANGANU Alvine Asonglefa
Ghent University
GHENT – BELGIUM
alvineasong@gmail.com

NGUEMA-ONA Eric
Lab Nutrition végétale Pôle Stress Biotique
Centre Mondial de l'Innovation Roullier
SAINT-MALO – FRANCE
eric.nguemaona@roullier.com

NGUYEN Huu Tien
Nematology Research Unit
Ghent University
GHENT – BELGIUM
+32 9 264 52 16
huutien.nguyen@ugent.be

NGUYEN Van Chung
Division of Pathology & Phyto-imunology
Plant Protection Research Institute
HA NOI – VIET-NAM
0915624268
nchung.ppri@gmail.com

NIEBES Jean-Francois
AG Division CS
Bayer
MONHEIM AM RHEIN – GERMANY
jean-francois.niebes@bayer.com

NJEKETE Cliven
INRAe
FRANCE
cliven.njekete@inrae.fr

NJEZIC Branimir
Faculty of Agriculture
University of Banja Luka
BANJA LUKA – BOSNIA AND HERZEGOVINA
+38 7 51 33 09 92
branimir.njezic@agro.unibl.org

NKENGEH AJONGLEFAC Fabrice
Ghent University
GHENT – BELGIUM
fabricenkengeh@gmail.com

NOLING Joseph
Entomology - Nematology Department
University of Florida
LAKE ALFRED – USA
+1 863 956 1151
jnoling@ufl.edu

NOSKOV Ilya
Inst. for Epidemiology and Pathogen Diagnostics
Julius Kuehn Institute
BRAUNSCHWEIG – GERMANY
+49 531 299 3828
ilya.noskov@julius-kuehn.de

NOUREDDINE Yara
Institut Sophia Agrobiotech, INRAe
SOPHIA ANTIPOLIS – FRANCE
yara.noureddine@inrae.fr

NTALLI Nikoletta
Department of Agriculture
University of Thessaly
NEA IONIA MAGNISIAS – GREECE
+30 210 818 0343
nntalli@uth.gr

NTIDI Nancy
Nematology
Agricultural Research Council
POTCHEFSTROOM – SOUTH AFRICA
+27 18 299 6341
ntidin@arc.agric.za

NÚÑEZ RODRÍGUEZ Lester
Laboratory of nematology - CIPROC
University of Costa Rica
SAN PEDRO – COSTA RICA
+506 863 70979
lesnr92@gmail.com

O'CONNOR Vincent
School of Biological Sciences
University of Southampton
SOUTHAMPTON – UK
V.M.OConnor@soton.ac.uk

OCHIENG' Effie
Nematology
IITA
NAIROBI – KENYA
+254 716 170 263
lilefy@gmail.com

OCHOLA Juliet
East Africa
IITA - Icipe
NAIROBI – KENYA
+254 20 8632932
jltochola@gmail.com

OGAYA Christopher
e-nema
SCHWENTINENTAL – GERMANY
+49 4 30 78 29 50
c.ogaya@e-nema.de

OKA Yuji
Nematology Unit
Agricultural Research Organization
M. P. NEGEV – ISRAEL
+972 899 28659
okayuji@volcani.agri.gov.il

OKUMURA Etsuko
Teisotoyoka
NAGOYA – JAPAN
+81 528 918 311
etsuko8okumura@gmail.com

OLAJIDE Emmanuel
Nematology Research Unit
Ghent University
GHENT – BELGIUM
olajide.o.emmanuel@gmail.com

OLAORE Deborah
Department of Crop Protection
University of Ilorin
ILORIN – NIGERIA
+23 4 80 63 25 57 62
princessayomide297@gmail.com

OLIVEIRA Cleiton
Technical Marketing
Lallemand Plant Care
PATOS DE MINAS – BRAZIL
+55 34 38 26 04 00
coliveira@lallemand.com

ORLANDO Valeria
Crop & Environment Sciences
Fera Science Ltd
YORK – UK
+44 1 90 44 62 04
Valeria.Orlando@fera.co.uk

OUYEDER Sumeja
Crop Strategy & Portfolio Management
Bayer
MONHEIM AM RHEIN – GERMANY
+49 2173 38 7343
sumeja.ouyeder@bayer.com

OWADA Kyoko
Dept. of Chemistry and Materials Science
National Institute of Tech - Gunma College
MAEBASHI – JAPAN
+81 272 549 215
kyokoowa@chem.gunma-ct.ac.jp

PAJAR Jessil Ann
MPI-Chemical Ecology, Jena/iDiv
LEIPZIG – GERMANY
+49 341 973 32 25
jessil_ann.pajar@idiv.de

PALOMARES-RIUS Juan Emilio
Plant Protection
IAS - CSIC
CÓRDOBA – SPAIN
+34 9 57 49 91 69
palomaresje@ias.csic.es

PATEL Mithunkumar Bhupendrabha
Agronomy
Gujarat Co. Op. Growers Limited
ANAND – INDIA
+91 2 69 22 33 78 9
mithun.gtfd@gmail.com

PEDRAM Majid
Department of Plant Pathology
Faculty of Agriculture, Tarbiat Modares Univ
TEHRAN – ISLAMIC REPUBLIC OF IRAN
+98 21 44 80 2697
mj_pedram@yahoo.com

PELLEGRIN Clement
University of Cambridge
CAMBRIDGE – UK
+44 1223 748 969
cp678@cam.ac.uk

PENTIMONE Isabella
Istituto per la Protezione Sostenibile delle Piant
Consiglio Nazionale delle Ricerche
BARI – ITALY
+39 080 5929 222
isabella.pentimone@ipsp.cnr.it

PERRY Roland
Department of Biological & Environmental Sciences
School of Life and Medical Sciences
HATFIELD – UK
r.perry2@herts.ac.uk

PETERS Rik
Field Crops
Wageningen University & Research
LELYSTAD – THE NETHERLANDS
+31 3 20 29 11 76
rik.peters@wur.nl

PETRIKOVSZKI Renáta
Szent István University
GÖDÖLLO – HUNGARY
+36 285 220 00
petrencsi@gmail.com

PETTER Françoise
EPPO
PARIS – FRANCE
+33 1 45 20 77 94
petter@eppo.int

PETTRICH Laura
Departement for Biology - Institute of Zoology
University of Cologne
COLOGNE – GERMANY
+49 2 21 47 08 80 4
l.pettrich@uni-koeln.de

PEYRONEL Stéphane
Hello Nature
FRANCE
stephane.peyronel@hello-nature.com

PICKUP Jon
SASA
EDINBURGH – UK
44 131 244 8859
jon.pickup@sasa.gov.scot

PIJNACKER Anna
Laboratory of Nematology
Wageningen University & Research
WAGENINGEN – THE NETHERLANDS
+31317482197
anna1.pijnacker@wur.nl

PILI Njira
Moi University
ELDORET – KENYA
0791711637
jirapi1983@yahoo.com

PIRES Andre
School of Life Sciences
University of Warwick
COVENTRY – UK
+44 2476 573 329
andre.pires@warwick.ac.uk

PIRES David
Nematology Lab
INIAV
OEIRAS – PORTUGAL
: (+351) 214 463 700
david.pires@iniav.pt

PLASSARD Claude
UMR Eco & Sols
INRAe
MONTPELLIER – FRANCE
+33 4 99 61 29 79
claude.plassard@inrae.fr

POPOVIC Brigita
Dept. of Agroecology and Environmnet Protection
Faculty of Agrobiotechnical Sciences
OSIJEK – CROATIA
+38 531 554 826
brigita.popovic@fazos.hr

PORAZINSKA Dorota
Department of Entomology and Nematology
University of Florida
GAINESVILLE – USA
+1 352 273 3936
dorotalp@ufl.edu

PORTIER Ulysse
INRAe
FRANCE
ulysse.portier@inrae.fr

POWERS Kirsten
Dept. of Plant Pathology
University of Nebraska
LINCOLN – USA
+1 402 472 5700
kpowers1@unl.edu

POWERS Thomas
Dept. of Plant Pathology
University of Nebraska
LINCOLN – USA
+1 402 472 5726
tpowers1@unl.edu

PRADANA Ankardiansyah Pandu
Plant Protection
University of Jember
JEMBER – INDONESIA
+62 3 31 33 02 24
pandu@unej.ac.id

PRETORIUS Marné
North-West University
POTCHEFSTROOM – SOUTH AFRICA
+27 18 285 3006
marne0911pretorius@gmail.com

PRETORIUS MC
SOUTH AFRICA
MC@cri.co.za

PRICE James
James Hutton Institute
DUNDEE – UK
jp203@st-andrews.ac.uk

PTATSCHECK Christoph
Animal Ecology
Bielefeld University
BIELEFELD – GERMANY
+49 521 106 2705
christoph.ptatscheck@uni-bielefeld.de

PUERARI Heriksen
Escola de Agronomia
Universidad Federal de Goiás
GOIANIA – BRAZIL
+55 62 3521 1531
heriksenhp@hotmail.com

PUISSANT Jérémy
UMR Eco & Sols
Institut de Recherche pour le Développement
MONTPELLIER – FRANCE
jeremypuissant@gmail.com

PUSKARIC Josipa
Department of Phytomedicine
Faculty of Agrobiotechnical Sciences
OSIJEK – CROATIA
+38 595 502 2737
josipa.puskaric@fazos.hr

PUTKER Vera
Lab. of Nematology
Wageningen University & Research
WAGENINGEN – THE NETHERLANDS
+31 3 17 48 21 97
vera.putker@wur.nl

QUENTIN Michaël
INRAe, Université Côte d'Azur, CNRS, ISA
SOPHIA ANTIPOLIS – FRANCE
+33 4 92 38 64 95
michael.quentin@inra.fr

QUINTERO Tonia
United States Department of Agriculture
RALEIGH – USA
+1 91 98 55 76 55
tonia.g.quintero@usda.gov

RADAKOVIC Zoran
P.H. Petersen Saatzucht Lundsgaard GmbH
GRUNDHOF – GERMANY
+4946368954
z.radakovic@phpetersen.com

RANTY-ROBY Sarah
INRAe
FRANCE
sarah.ranty-roby@inrae.fr

RASHIDIFARD Milad
Unit for environmental and management sciences
North-West University
POTCHEFSTROOM – SOUTH AFRICA
+27 766 395 051
milad.rashidifard@nwu.ac.za

RIASCOS Donald
Agronomy faculty
Universidad del Pacífico
BUENAVENTURA – COLOMBIA
+57 2 240 5555
dhriascoso@unal.edu.co

RIBEIRO Rejanne
Technical Marketing
Lallemand Plant Care
PATOS DE MINAS – BRAZIL
+55 34 38 26 04 00
rribeiro@lallemand.com

RIGGS Jennifer
BASF
RTP – USA
919 4 75 61 26
jennifer.riggs@basf.com

RIST Marc
Crop Strategy & Portfolio Management
Bayer
MONHEIM AM RHEIN – GERMANY
+49 2173 38 7343
marc.rist@bayer.com

RIVA Gabrieli
USA
gabrieliriva@ufl.edu

ROBERT FARIA Denner
Genetics and Crop Breeding
Universidad Federal de Goiás
GOIANIA – BRAZIL
denner.robertfaria@gmail.com

ROBERTS Philip
Nematology
University of California
RIVERSIDE – USA
+1 951 827 7332
philip.roberts@ucr.edu

ROBINSON Colin
Brigham Young University
PROVO – USA
+1 385 216 7228
cr464@byu.edu

ROCHA Mara
Escola de Agronomia
Universidad Federal de Goiás
GOIANIA – BRAZIL
+55 62 3521 1531
darochamararubia@gmail.com

RODRIGUES Jules
UMR7245
Museum National d'Histoire Naturelle
PARIS – FRANCE
jules.rodrigues@edu.mnhn.fr

ROMERO MOYA Lizzete Dayana
Institute of Plant Protection
Chinese Academy of Agricultural Sciense
BEIJING – PEOPLE'S REPUBLIC OF CHINA
81936167
ldromerom@unal.edu.co

RONDET Damien
NIXE
FRANCE
rondet@nixe.com

RØNN Regin
Danish Veterinary and Food Administration
RINGSTED – DENMARK
+45 72 27 60 93
remro@fvst.dk

ROS IBÁÑEZ Caridad
Crop protection departament
IMIDA
LA ALBERCA - MURCIA – SPAIN
+34 968 394559
caridad.ros@carm.es

ROSS Jenna
Crop Health & Protection
YORK – UK
jenna.ross@chap-solutions.co.uk

RUSINQUE Leidy
Nematology Lab
INIAV
OEIRAS – PORTUGAL
+35 12 14 40 35 00
leidy.rusinque@iniav.pt

RUTHES Andrea
Agroscope
WÄDENSWIL – SWITZERLAND
+41 58 46 95021
andreacaroline.ruthes@agroscope.admin.ch

SAGAWA Marika
School of Agriculture
Meiji University
KAWASAKI – JAPAN
+81 44 934 7818
jasmine.0915.snow@gmail.com

SAIKAI Kanan
Plant Pathology
University of Wisconsin
MADISON – USA
+1 608 263 6131
kutsuwa@wisc.edu

SALIOU Papa
Ghent University
GHENT – BELGIUM
PapaSaliou.Niang@UGent.be

SALUTE Stefano
Centro Attività Vivaistiche
FAENZA – ITALY
+ 39 0546 47150
salute@cavtebano.it

SAN BLAS Ernesto
Independent researcher
SANTIAGO – CHILE
+56 941 615 295
esanblas@yahoo.com

SANCHEZ-MORENO Sara
INIA
MADRID – SPAIN
+34 9 13 47 69 56
sarasm@inia.es

SAUCET Simon
Institut Sophia Agrobiotech
SOPHIA ANTIPOLIS – FRANCE
+33(0)4 92 38 64 60
simonsaucet@orange.fr

SAUZAY Stéphanie
Bayer CropScience
LYON – FRANCE
+33 4 72 85 48 81
stephanie.sauzay@bayer.com

SCHAVELING Arno
Nematology
Wageningen University & Research
WAGENINGEN – THE NETHERLANDS
+31 3 17 48 21 97
arno.schaveling@wur.nl

SCHENK Janina
Animal Ecology
Bielefeld University
BIELEFELD – GERMANY
+49 52 11 06 24 89
janina.schenk@uni-bielefeld.de

SCHIFFER Philipp
Worm Lab
University of Cologne
KÖLN – GERMANY
+49 221 4703 238
p.schiffer@uni-koeln.de

SCHILLER Karl
Pheronym
WOODLAND – USA
+13522194464
schiller@pheronym.com

SCHLEKER Sylvia
Molecular Phytomedicine
University of Bonn
BONN – GERMANY
+49 2 28 73 39 00
sylvia.schleker@uni-bonn.de

SCHMIDT Jan Henrik
Dept of Ecological Plant Protection
Julius Kühn-Institute
BRAUNSCHWEIG – GERMANY
+49 554 298 1705
jan-henrik.schmidt@julius-kuehn.de

SCOTT Dora
Ghent University
GHENT – BELGIUM
Dora.Scott@ugent.be

SEBUMPAN Rea
MSU - Iligan Institute of Technology
ILIGAN – PHILIPPINES
+63 9 55 97 42 37
rea.sebumpan@g.msuiit.edu.ph

SEMPRUCCI Federica
University of Urbino
URBINO – ITALY
+39 0722 304248
federica.semprucci@uniurb.it

SHAHZAD GONDAL Amjad
Department of Plant Pathology
Bahauddin Zakariya University
MULTAN – PAKISTAN
+92 5 14 31 85 20
amjadshahzad@live.com

SHAKYA Pallavi
Plant Pathology - Siddique Lab
University of California
DAVIS – USA
5307996735
pshakya@ucdavis.edu

SHEEHY Laura
Biological and Environmental Sciences
Liverpool John Moores University
LIVERPOOL – UK
L.Sheehy@ljmu.ac.uk

SHEHATA Ibrahim
National Research Centre
CAIRO – EGYPT
010 9 89 87 07 4
ib_shehata80@yahoo.com

SHEPHERD Jack
BASF
LITTLEHAMPTON – UK
+44 1903 736 937
jack.shepherd@basf.com

SHEPHERD Rachel
University of Florida
GAINESVILLE – USA
r.shepherd@ufl.edu

SHIFERAW Habtamu Kefelegn
Nematology Research Unit
Ghent University
GHENT – BELGIUM
+3292645223
habtamukefelegn@gmail.com

SIDDIQUE Shahid
Entomology and Nematology
UC Davis
DAVIS – USA
+1 530 754 7659
ssiddique@ucdavis.edu

SIKORA Richard
University of Bonn - INRES-Phytomedizin
BONN – GERMANY
+49 228 281423
rsikora@uni-bonn.de

SILVA Jose
Technical Department
IHARA
SOROCABA – BRAZIL
+55 153 2 35 77 00
jose.paulo@ihara.com.br

SILVA Monique
Universidad Estadual de Maringá
MARINGA – BRAZIL
44988393090
moniquetrs@hotmail.com

SILVA Santino Aleandro
Plant Protection
Instituto Agronômico do Paraná
LONDRINA – BRAZIL
+55 43 3376 2337
santinoaleandro@gmail.com

SILVA VALENZUELA Manuel
Fitosanidad - Fitopatología Laboratorio 209
Colegio de Postgraduados
TEXCOCO – MEXICO
+595 95 20 200
manuel.silva@colpos.mx

SIMMONS Jeffrey
Crop Protection - Nematology
Syngenta
VERO BEACH – USA
+1 772 794 7119
jeff.simmons@syngenta.com

SIMÕES Nelson
Universidad dos Açores
PONTA DELGADA – PORTUGAL
(351) 296 650 119
nelson.jo.simoes@uac.pt

SINGH Phougeishangbam Rolish
Nematology Research Unit
Ghent University
GHENT – BELGIUM
+32 9 264 52 23
phougeishangbamrolish.singh@ugent.be

SIRCA Sasa
Agricultural Institute of Slovenia
LJUBLJANA – SLOVENIA
+38 612 805 262
sasa.sirca@kis.si

SIRENGO David Kihoro
Nematology Research Unit
University of Idaho
PARMA – USA
2082302587
davidkihoro9@gmail.com

SITHOLE Nokuthula
Research Centre of Plant Growth and Development
University of KwaZulu-Natal
SCOTTSVILLE – SOUTH AFRICA
+2733 260 5130
nooku.nt@gmail.com

SLAATS Brigitte
Marketing
Syngenta Crop Protection AG
BASEL – SWITZERLAND
+41 786 697 037
brigitte.slaats@syngenta.com

SMANT Geert
Laboratory of Nematology
Wageningen University & Research
WAGENINGEN – THE NETHERLANDS
+31 317 485 254
geert.smant@wur.nl

SMITH Moray
University of Saint Andrews
SAINT ANDREWS – UK
+44 334 928 5428
Moray.Smith@hutton.ac.uk

SOLER Roxina
Koppert Biological Systems
BERKEL EN RODENRIJS – THE NETHERLANDS
+31 6 30 73 54 39
rsoler@koppert.nl

SOMMER Ralf Josef
Dept. for Evolutionary Biology
Max Planck Institute for Developmental Biol.
TÜBINGEN – GERMANY
+49 7 07 16 01 371
ralf.sommer@tuebingen.mpg.de

SONAWALA Unnati
Department of Plant Sciences
University of Cambridge
CAMBRIDGE – UK
+44 1223 748 966
us275@cam.ac.uk

SOULÈ Salomè
INRAe
FRANCE
salome.soule@inrae.fr

STAMATAS Yannis
Corteva AgriScience
HALANDRI – GREECE
+30 2106889744
yannis.stamatas@corteva.com

STEINMETZ Nicole
UC San Diego
LA JOLLA – USA
+1 858 356 8515
nsteinmetz@ucsd.edu

STERKEN Mark
Laboratory of Nematology
Wageningen University & Research
WAGENINGEN – THE NETHERLANDS
+31 317 482 998
mark.sterken@wur.nl

STERZ Thorsten
BASF SE
LIMBURGERHOF – GERMANY
0491733798218
thorsten.sterz@basf.com

STOCK S. Patricia
College of Agriculture
California State University
CHICO – USA
spstock@csuchico.edu

STOJILKOVIC Boris
Faculty of Bioscience Engineering - UGhent
GHENT – BELGIUM
+32 4 98 13 17 24
boris.stojilkovic@ugent.be

STOKWE Nomakholwa
Conservation Ecology and Entomology
Stellenbosch University
STELLENBOSCH – SOUTH AFRICA
+27 218 082 546
nfs@sun.ac.za

STORELLI Alan
HAFL
Bern University of Applied Sciences
ZOLLIKOFEN – SWITZERLAND
+49 055 613 114 43
alan.storelli@bfh.ch

STOREY Sheila
Nemlab
DURBANVILLE – SOUTH AFRICA
+27 21 875 8160
sheila@nemlab.co.za

STUBBS Rebecca
CABI
WALLINGFORD – UK
+44 14 91 82 94 17
r.stubbs@cabi.org

SUAN Mayvel
Department of Biological Sciences
Mindanao State University - Iligan Institute
LANAO DEL NORTE – PHILIPPINES
+63 063 492 1173
mayvel.suan@g.msuiit.edu.ph

SUBBOTIN Sergei
Plant Pest Diagnostic Center
CDFA
SACRAMENTO – USA
+1 916 738 66 96
sergei.a.subbotin@gmail.com

SUMAYA Nanette Hope
Dept of Biological Sciences - PRISM Biodiversity
MSU - Iligan Institute of Technology
ILIGAN – PHILIPPINES
+63 221 4050
nanettehope.sumaya@g.msuiit.edu.ph

SUSIC Nik
Plant Protection Department
Agricultural Institute of Slovenia
LJUBLJANA – SLOVENIA
+38 6 12 80 52 76
nik.susic@kis.si

TALAVERA Miguel
Sustainable Plant Protection
IFAPA
CÓRDOBA – SPAIN
+34 671 532 127
miguelf.talavera@juntadeandalucia.es

TALLA SEH Alieen
Ghent University
GHENT – BELGIUM
Tallaseh@mail.com

TAÑAN Veronica
Biological Sciences
MSU - Iligan Institute of Technology
ILIGAN – PHILIPPINES
+63 955 901 6871
veronicatanan1998@gmail.com

TANGVIK Marte
Viruses, Bacteria & Nematodes in Forestry, Agric
Norwegian Institute of Bioeconomy Research
ÅS – NORWAY
+47 932 539 53
marte.tangvik@nibio.no

TE MOLDER Dennie
Nematology
Wageningen University & Research
WAGENINGEN – THE NETHERLANDS
+31317482197
dennie.temolder@wur.nl

TEFU Grace Mohoanatso
Crop Protection
Agricultural Research Council
NELSPRUIT – SOUTH AFRICA
+27 137 537 000
grace@arc.agric.za

TEKLU Misghina Goitom
Agrosytems research - Nematology
Wageningen University & Research
WAGENINGEN – THE NETHERLANDS
+31 317 481 044
misghina.goitomteklu@wur.nl

THEVENOUX Romain
ELISOL Environnement
CONGÉNIES – FRANCE
+33 4 66 71 92 59
r.thevenoux@hotmail.fr

THODEN Tim
Corteva AgriScience
MUNICH – GERMANY
+49 152 21 41 53 78
tim.thoden@corteva.com

TIMMERMANS Bernd
Ghent University
GHENT – BELGIUM
timmermansbernd.mail@gmail.com

TIRADO Jorge
BASF Agricultural Specialities Ltd.
LITTLEHAMPTON – UK
+441903736927
jorge.tirado@basf.com

TODERI Alice
Hello Nature
VALBONNE – FRANCE
alice.toderi@hello-nature.com

TOMAZ Juarez
LBI
IAPAR
LONDRINA – BRAZIL
+55 43 33 76 21 23
tomaz@iapar.br

TOP Frederic
Crop Strategy & Portfolio Management
Bayer AG - Crop Science Division
MONHEIM AM RHEIN – GERMANY
frederic.top@bayer.com

TOPALOVIC Olivera
Aarhus University
AARHUS – DENMARK
olivera.topalovic.best@gmail.com

TORRINI Giulia
CREA - Plant protection and Certification
FLORENCE – ITALY
+39 055 2492 262
giulia.torrini@crea.gov.it

TOUBARRO Duarte
Biotechnology Center of Azores - Fac. of Sciences
Universidad dos Açores
PONTA DELGADA – PORTUGAL
+351 919260020
duarte.nt.tiago@uac.pt

TRAUNSPURGER Walter
Animal Ecology
Bielefeld University
BIELEFELD – GERMANY
+495211062702
traunspurger@uni-bielefeld.de

TROCCOLI Alberto
CNR
Institute for Sustainable Plant Protection
BARI – ITALY
+39 080 5929 228
alberto.troccoli@ipsp.cnr.it

TUKAHIRWA Benius
Ghent University
GHENT – BELGIUM
benius.tukahirwa@gmail.com

UYSAL Gulsum
Nematology
Batı Akdeniz Agricultural Research Institute
ANTALYA – TURKEY
+902423216797
gulsumuysal.gu@gmail.com

VAN AALST Eveline
Phytopathology Research
Rijk Zwaan Breeding B.V.
DE LIER – THE NETHERLANDS
+31 174 79 42 02
e.van.aalst@rijkzwaan.nl

VAN BRUGGEN Anne Sophie
National Reference Centre Nematology
National Plant Protection Organization
WAGENINGEN – THE NETHERLANDS
+31 8 82 23 04 99
a.s.vanbruggen@nvwa.nl

VAN GHELDER Cyril
INRAe
SOPHIA ANTIPOLIS – FRANCE
+33 4 92 38 64 80
cyril.van-ghelder@inrae.fr

VAN HEESE Evelyn
National Reference Centre, Nematology
NPPO-NL
WAGENINGEN – THE NETHERLANDS
+31 8 82 23 10 73
e.y.j.vanheese@nvwa.nl

VAN HIMBEECK Robbert
Wageningen University & Research
WAGENINGEN – THE NETHERLANDS
+31317482197
robbert.vanhimbeeck@wur.nl

VAN OOSTEN Vivian
Vegetable Seeds - Discovery Pathology
Bayer CropScience
WAGENINGEN – THE NETHERLANDS
+31 317 468 468
vivian.vanoosten@bayer.com

VAN SLUIJS Lisa
Wageningen University & Research
WAGENINGEN – THE NETHERLANDS
+31 317 482 998
lisa.vansluijs@wur.nl

VAN STEENBRUGGE Joris
Laboratory of Nematology
Wageningen University & Research
WAGENINGEN – THE NETHERLANDS
+31 74 855 37
joris.vansteenbrugge@wur.nl

VANDENBOSSCHE Bart
e-nema GmbH
SCHWENTINENTAL – GERMANY
+49 4307 8295 283
b.vandenbossche@e-nema.de

VARANDAS Raquel
University of Coimbra
COIMBRA – PORTUGAL
+351 239 855 760
raquelrv@gmail.com

VAZ MOREIRA Valdeir Junio
Embrapa Genetic Resources and Biotechnology/Labora
Embrapa Genetic Resources and Biotechnology
BRASÍLIA – BRAZIL
+31 998960965
valdeirjmoreira22@gmail.com

VERDUN Cyrille
Koppert Biological Systems
CAVAILLON – FRANCE
+33 4 90 78 30 13
cverdun@koppert.fr

VERHAGE Adriaan
Phytopathology Research
Rijk Zwaan Breeding B.V.
DE LIER – THE NETHERLANDS
+31 1 74 79 42 02
a.verhage@rijkzwaan.nl

VERMA Anju
Plant Pathology
Univeristy of Georgia
ATHENS – USA
+1 70 65 42 72 60
anju.verma@uga.edu

VERONICO Pasqua
CNR
Institute for Sustainable Plant Protection
BARI – ITALY
+39 080 558 3377
pasqua.veronico@ipsp.cnr.it

VESTERGÅRD Mette
Department of Agroecology
Aarhus University
SLAGELSE – DENMARK
+45 871 581 21
mvestergard@agro.au.dk

VIAENE Nicole
Plant
ILVO
MERELBEKE – BELGIUM
+32 9 27 22 90 0
nicole.viaene@ilvo.vlaanderen.be

VICENTE Claudia
MED
ÉVORA – PORTUGAL
+35 121 440 3500
cvicente@uevora.pt

VICENTE DÍEZ Ignacio
Mohammed V University
RABAT – MOROCCO
ignacio.vicente@icvv.es

VIEIRA Soraia
Marine and Environmental Sciences Centre
University of Évora
ÉVORA – PORTUGAL
+351 266 760 881
svvieira@uevora.pt

VIKETOFT Maria
Department of Ecology
Swedish University of Agricultural Sciences
UPPSALA – SWEDEN
+46 18 671 505
maria.viketoft@slu.se

VILLEGAS Laura
Department of Biology
University of Cologne
COLOGNE – GERMANY
+49 2 21 47 04 69 1
lvilleg1@uni-koeln.de

VILLENAVE Cécile
ELISOL environnement
CONGÉNIES – FRANCE
+33 4 66 71 92 59
cecile.villenave@elisol.fr

VISSER Johnny
WUR Field Crops
LELYSTAD – THE NETHERLANDS
+31 3 20 29 16 72
johnny.visser@wur.nl

VOS Christine
Microbiology
Scientia Terrae vzw
SINT-KATELIJNE-WAVER – BELGIUM
+3215305590
cvo@scientiaterrae.org

WANG Xizhuo
Research Institute of Forest Ecology, Environment
Chinese Academy of Forestry
BEIJING – PEOPLE'S REPUBLIC OF CHINA
+86 10 628 892 84
ladydal@163.com

WANG Zhen
The Faculty of Agricultural & Nutritional Science
Christian - Albrechts - Universität zu Kiel
KIEL – GERMANY
+49 (0)4307 8295-286
stu225777@mail.uni-kiel.de

WANJIRU Margaret Wangari
Ghent University
GHENT – BELGIUM
margaretwangari1995@gmail.com

WATSON Tristan
Department of Plant Pathology and Crop Physiology
Louisiana State University AgCenter
BATON ROUGE – USA
+1 941 243 13 97
twatson@agcenter.lsu.edu

WEBSTER John M
Biological Sciences
Simon Fraser University
NORTH VANCOUVER – CANADA
+1 604 980 2296
jwebster@sfu.ca

WEN Yanhua
Plant Pathology Department - Nematology Lab
South China Agricultural University
GUANGZHOU – PEOPLE'S REPUBLIC OF CHINA
+86 20 3829 7416
yhwen@scau.edu.cn

WESEMAEL Wim
ILVO
MERELBEKE – BELGIUM
+32 92 72 24 45
wim.wesemael@ilvo.vlaanderen.be

WESTPHAL Fen
Fresno Pacific University
FRESNO – USA
+1 559 453 2000
fen.westphal@fresno.edu

WIECZOREK Krzysztof
Institute of Plant Protection
TULLN AN DER DONAU – AUSTRIA
+43 1 47654 95306
krzysztof.wieczorek@boku.ac.at

WILES John
Corteva AgriScience
BISHOPS STORTFORD – UK
+44 7810 056 719
john.wiles@corteva.com

WILLIG Jaap-Jan
Laboratory of Nematology
Wageningen University & Research
WAGENINGEN – THE NETHERLANDS
+31 317 482 996
jaap-jan.willig@wur.nl

WITEK Kamil
2 Blades
The Sainsbury Laboratory
NORWICH – UK
+441603450030
kamil.witek@tsl.ac.uk

WU Sheng-Yen
Entomology and Nematology
University of Florida
LAKE ALFRED – USA
+1 352 213 5515
sywu@ufl.edu

XIANG Hui
Bioscience Engineering Department
Biotechnology
Ghent University
GHENT – BELGIUM
+32 94625888
hui.xiang@ugent.be

XIAO Fangming
Plant Sciences
University of Idaho
MOSCOW – USA
+1 208 885 0120
fxiao@uidaho.edu

XIE Chuanshuai
Huazhong Agricultural University
WUHAN – PEOPLE'S REPUBLIC OF CHINA
+86 027 8728 3455
xiesc@webmail.hzau.edu.cn

XU Bingliang
College of Plant Protection
Gansu Agricultural University
YINGMEN – PEOPLE'S REPUBLIC OF CHINA
+86 931 763 1294
zhangsw704@126.com

YENNI Erica
BASF
DURHAM – USA
erica.yenni@basf.com

YIMER Sihen Getachew
Horticulture and Plant Scince
Jimma University
JIMMA – ETHIOPIA
+251471118105
sihengetachew8@gmail.com

ZAMBIASI Tatiane
Nematologia
Agrolab
PRIMAVERA DO LESTE – BRAZIL
+55 66 34 98 74 41
tatiane@agrolab.agr.br

ZAMBONI MACHADO Andressa Cristina
Plant Protection
Instituto Agronômico do Paraná
LONDRINA – BRAZIL
+55 43 3376 2337
andressaczmachado@hotmail.com

ZHANG Gang
Beijing Shoushang Ecological Agriculture Co.
BEIJING – PEOPLE'S REPUBLIC OF CHINA
1523623656
luquan_wang@hotmail.com

ZHENG Qi
Laboratory of Nematology
Wageningen University & Research
WAGENINGEN – THE NETHERLANDS
+31 644 316 217
qi.zheng@wur.nl

ZOTTA MOTA Ana Paula
INRAe
FRANCE
ana-paula.zotta-mota@inrae.fr

ZOUHAR Miloslav
Department of Plant Protection
Czech University of Life Sciences
PRAGUE – CZECH REPUBLIC
+42 060 486 6749
zouharmiloslav@seznam.cz

ZURLETTO Laetitia
ISA, UCA, INRAe 1355, CNRS 7254
Université Côte d'Azur - ISA
SOPHIA ANTIPOLIS – FRANCE
+33 4 92 38 64 94
laetitia.zurletto@inrae.fr

7th International Congress of Nematology
ICN 2022
Antibes Juan-les-Pins - France

"Crossing borders: a world of nematode diversity and impact to discover"

European Society of Nematologists

LIST OF PUBLIC PARTNERS

Antibes Juan-les-Pins
https://www.antibes-juanlespins.com

Communauté d'Aglomération Sophia Antipolis
https://www.agglo-sophiaantipolis.fr

INRAe
https://www.inrae.fr/en

Région Sud Provence-Alpes-Côte d'Azur
https://www.maregionsud.fr

Université Côte d'Azur
https://univ-cotedazur.eu

LIST OF SPONSORS

PICO©
3 Shagaret El Dor St. Zamalek
11211 CAIRO – EGYPT
(+202) 2737-1000 / (+202) 2735-7770
m.ashraf@picoagri.com
https://picoagriculture.com

Rijk Zwaan Breeding B.V.
Burgemeester Crezéelaan 40
PO Box 40
2678 ZG DE LIER – THE NETHERLANDS
+31 174 53 28 62
info@rijkzwaan.com
www.rijkzwaan.com

SESVanderhave
Ferme de l'Ermitage
62121 GOMIECOURT
+33 3 21 24 84 27
contact@sesvanderhave.com
www.sesvanderhave.com

Vilmorin & Cie (Limagrain Group)
CS 20001
Saint-Beauzire
63360 GERZAT – FRANCE
+ 33 (0)4 73 63 40 92
sophie.boulinguez@limagrain.com
www.limagrain.com

7th International Congress of Nematology
ICN 2022
Antibes Juan-les-Pins - France

"Crossing borders: a world of nematode diversity and impact to discover"

European Society of Nematologists

LIST OF EXHIBITORS

Adama Agricultural Solutions

Stand 3

Crop Protection business.
Airport City, Golan Street
7015103 – ISRAEL
+55 43 99102 4216
bruno.lovato@adama.com
www.adama.com

BASF Corporation

Stand 13

At BASF, we create chemistry – and have been doing so for 150 years. Our portfolio ranges from chemicals, plastics, performance products and crop protection products to oil and gas.
26 Davis Drive
27709 DURHAM, RTP – USA
+1 919 547 2400
shaun.berry@basf.com
www.basf.com

Bayer AG

Stand 1

Bayer is the world-leading agriculture enterprise with businesses in seeds, crop protection and nonagricultural pest control.
Alfred-Nobel-Str. 50
40789 MONHEIM – GERMANY
+49 173 318 3817
sumeja.ouyeder@bayer.com
www.bayer.com

Brill

Stand 9

Founded in 1683, Brill is a publishing house with a rich history and a strong international focus. Brill's focus on the Humanities and Social Sciences, International Law and selected Sciences.
Plantijnstraat 2
2321 JC LEIDEN – THE NETHERLANDS
+31 7 15 35 35 00
stefan.einarson@brill.com
www.brill.com

CABI

Stand 11

CABI is an international, inter-governmental, not-for-profit organization that improves people's lives worldwide by providing information and expertise in agriculture and the environment.
Nosworthy Way
OX10 8DE WALLINGFORD – UK
01491 832111
r.stubbs@cabi.org
www.cabi.org

Corteva Agriscience

Stand 4

Corteva Agriscience is a company 100% dedicated to agriculture. Selecting the best seeds, protection crops and designing services: this is Corteva's ambition to ensure profitable and sustainable agriculture for farmers.
Batiment Equinoxe 2
1 bis Avenue du 8 mai 1945
78280 GUYANCOURT – FRANCE
+41 79 597 27 09
stephanie.fournol@corteva.com
www.corteva.fr

Hello Nature

Stand 8

Producer of organic fertilizers, biostimulants of vegetal origin and beneficial micro-organisms.
Les espaces de Sophia
80 route des lucioles
06560 VALBONNE – FRANCE
+33 4 92 96 76 40
hellonature@hello-nature.com
www.hello-nature.com

Koppert BV

Stand 5

Koppert produces and supplies biological solutions against pests ans diseases for horticulture and outdoor crops.
Veilingweg 14
2651BE BERKEL EN RODENRIJS
NETHERLAND
+31 10 514 04 44
info@koppert.nl
www.koppert.com

Marrone Bio Innovations

Stand 6

Development, manufacturing and marketing of bioinsecticides, biofungicides, bio-nematicides and bionutrient plant health products.
7780-420 Brier Creek Parkway
27617 RALEIGH – USA
info@marronebio.com
www.marronebio.com

Roullier

Stand 10

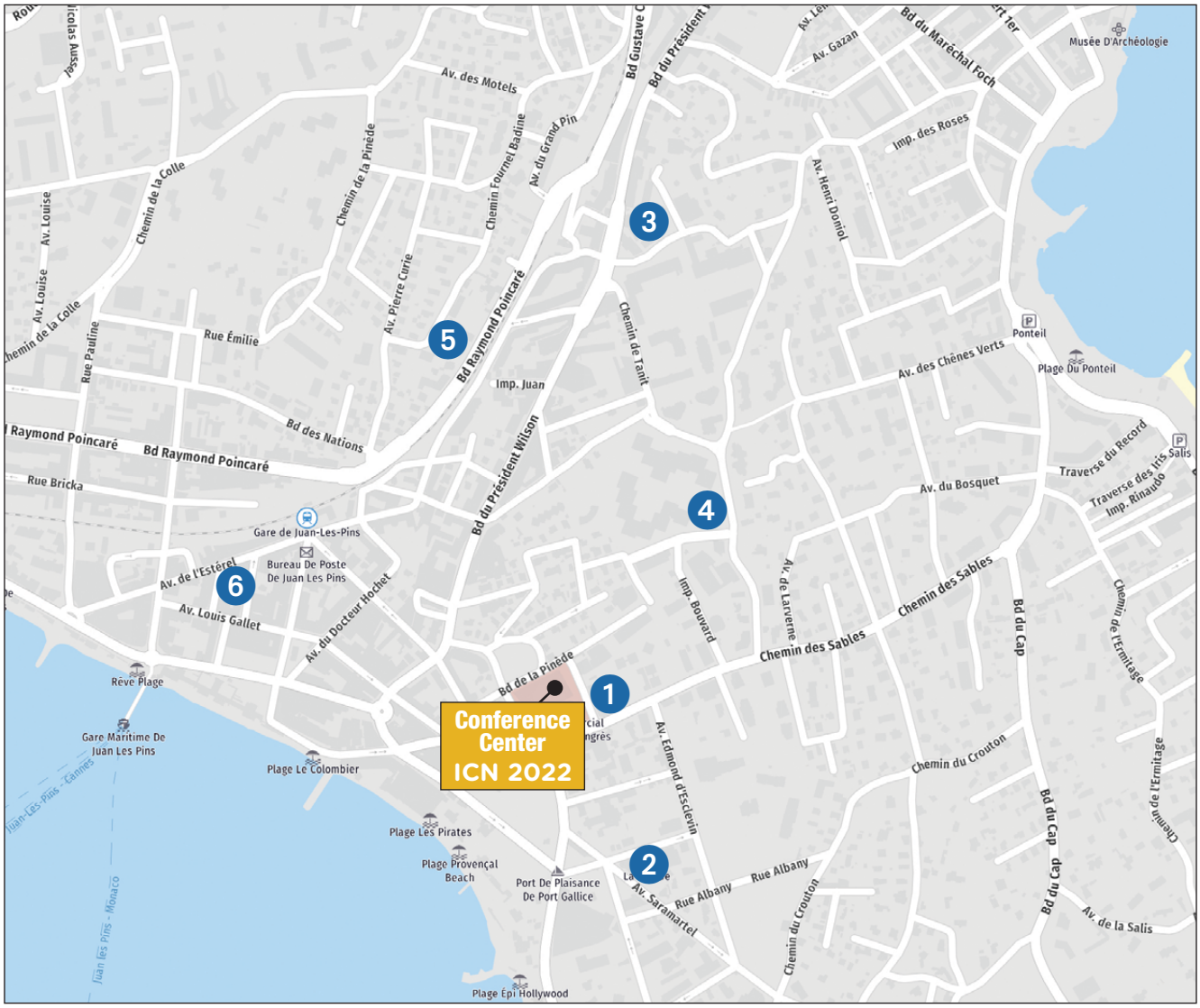
+33 2 99 20 58 30
eric.nguemaona@roullier.com
www.roulier.com

Syngenta Crop Protection AG

Stand 2

Syngenta is a leading science-based agtech company. Syngenta innovates with world-class science to protect crops and improve seeds. Our 2 core businesses support farmers with technologies, knowledge and services so they can sustainably provide the world with better food, feed, fibre, and fuel.
Rosentalstrasse 67
4002 BASEL – SUISSE
+ 41 61 323 1111
klara.nezvalova@syngenta.com
www.syngenta.com

HOTELS LOCATION - ICN 2022 - ANTIBES JUAN-LES-PINS, France



1 AC Hotel Ambassadeur ****

Located 100m from the conference center.

P ❄ 📶 🍸 🍴 🏊 🤿

2 Hotel Le Grand Pavois ***

Located 300m from the conference center.

P ❄ 📶 🍸

3 Hotel Journel ***

Located 950m from the conference center.

P ❄ 📶 🍸

4 Résidence Pierre et Vacances - La rostagne **

Located 650 m from the congress center.

P ❄ 📶 🏊

5 Hotel Astor **

Located 1km from the conference center.

P 📶

6 Hotel La Résidence **

Located 650m from the conference center.

❄ 📶