

Retrotransposons as putative key elements on the genetic variability and regulation of virulence in *Hemileia vastatrix*

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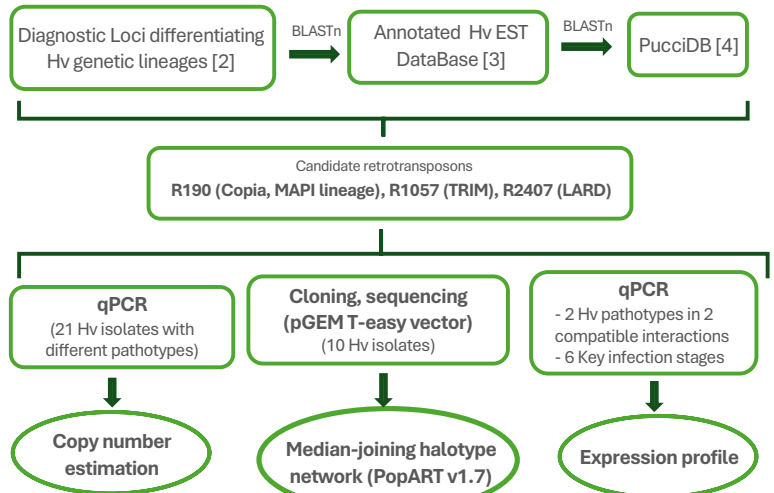
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1. Introduction

Hemileia vastatrix (Hv) is the causal agent of Coffee Leaf Rust, the major constraint to Arabica coffee production. The dynamic co-evolution of the Hv-coffee pathosystem promotes the emerging of new pathotypes with increased virulence profiles, capable of overcoming the resistance mechanisms of the host. The continuous resistance breakdown observed in the field has been providing evidence of the high pathogenic variability in *H. vastatrix*. Unveiling the mechanisms that promotes genetic variability and virulence in this pathogen is important to improve disease control. Retrotransposons have been implicated in genome shaping and expansion events, as well as in virulence increase [1].

To assess the putative involvement of retrotransposons in diversity and virulence profiling of Hv, this study aims to understand the genetic variability (nucleotide sequence and copy number) and relative expression profiles along the course of the infection process of three retrotransposons among Hv contrasting pathotypes.

2. Material and methods



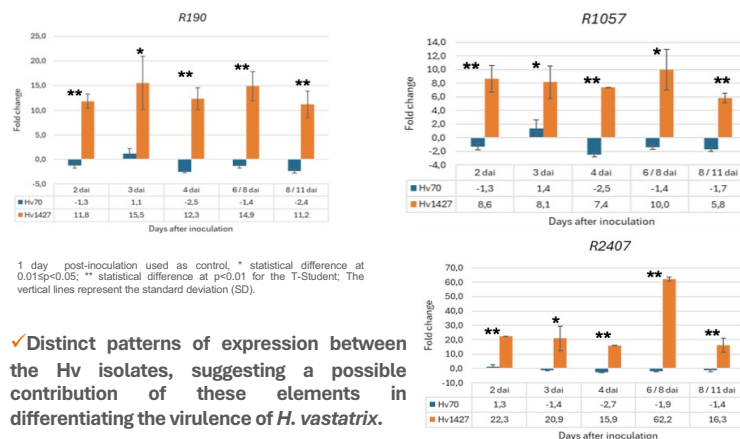
3. Results

Copy number

Hv isolates	Genome size (Mbp)	No copies per genome			Hv isolates	Genome size (Mbp)	No copies per genome		
		RHv00190	RHv01057	RHv02407			RHv00190	RHv01057	RHv02407
Hv999	719	800	1	1216	Hv264	788	1079	41	758
Hv1321	721	1781	3	3377	Hv3302	789	1435	6	649
Hv1427	751	1108	3	578	Hv3305	792	3486	4	4457
Hv741	758	873	4	1949	Hv92	794	495	10	923
Hv999a	771	623	4	304	Hv71	824	1008	24	408
Hv178a	772	934	4	1008	Hv264a	824	1771	39	728
Hv178	774	663	2	1109	Hv535	827	1934	2	1067
Hv166	777	932	2	889	Hv22	834	725	NA	1322
Hv1065	782	1650	103	1515	Hv2191	842	1447	28	3460
Hv995	783	725	3	1144	Hv70	879	1848	2	1486

Copia/MAPI lineage and LARD are the most proliferative elements, reaching over 3000 copies per genome

Expression profile



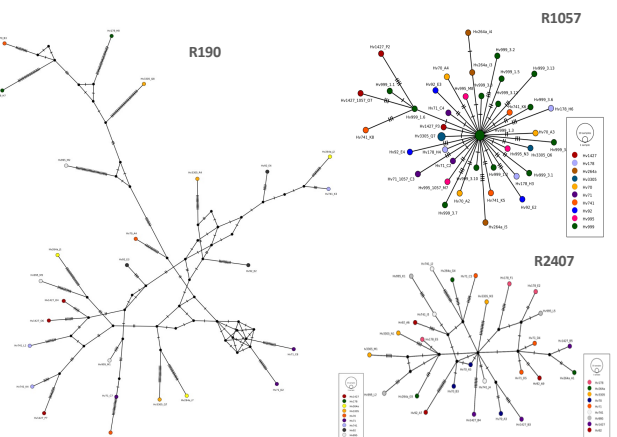
✓ Distinct patterns of expression between the Hv isolates, suggesting a possible contribution of these elements in differentiating the virulence of *H. vastatrix*.

✓ All retrotransposons were upregulated in all time-points for the Hv1427 compatible interaction, and mostly downregulated in the Hv70 compatible interaction

✓ RHv2407 shows a much higher level of expression for at all time-points, in relation to RHv190 and RHv1057 (for Hv1427 compatible interaction)

Funding LEAF 2025

Funging: FCT – Fundação para a Ciência e Tecnologia, I.P. through project reference UID/04129/2025; LEAF research center UID/04129/2025



✓ High level of genetic variation with very divergent haplotypes/copies (>30 SNPs, up to 84 bp Indels), both between isolates and within the same isolate

✓ Analysis of haplotype network topologies: Complex diversification and proliferation patterns without apparent correlation with isolate or pathotype group

✓ The most proliferative retrotransposon seems to have more complex diversification patterns

4. Conclusions / Perspectives

This study reveals new insights about the *Hemileia vastatrix* adaptive evolution, by suggesting that retrotransposons might contribute to the generation of genetic diversity and virulence of this rust fungus

References:

- [1] Lorrain et al. (2021). DOI:10.1016/B978-0-12-819990-9.00042-1.
- [2] Silva et al. (2018). DOI: 10.1111/mpp.12657.
- [3] Talhinhos et al. (2014). DOI: 10.3389/fpls.2014.00088.
- [4] Orozco-Arias, S et al., (2022). DOI: 10.3390/agronomy12071665

Acknowledgements: This work is being co-funded by Lisboa 2020, Portugal 2020 and European Union through FEDER funds (LISBOA-01-0145-FEDER-029189), and FCT through national funds under project PTDC/ASP-PLA/29189/2017.