

# Evaluation of virus-induced gene silencing (VIGS) in coffee plants

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## Introduction

Virus-induced gene silencing (VIGS) is an RNA-mediated reverse genetics technology that has evolved into an indispensable approach for analyzing the function of genes. We tested the tobacco rattle virus (TRV) vector system in coffee to elucidate the functional role of coffee resistance related-candidate genes.

## Materials/Methods

Agroinfiltration of TRV1 and TRV2 vector constructs carrying a *Coffea arabica* (Ca) and *Nicotiana benthamiana* (Nb) phytoene desaturase genes (PDS) were done both in Ca and Nb, accordingly with the experimental design (Fig 1). Different bacterial culture concentrations were used. Agrodrench and root dipping were also tested.

## Conclusion/Perspectives

A first attempt to apply VIGS to coffee is described. The efficacy of the construction was confirmed by agroinfiltration of tobacco resulting in the expected albino phenotype. However, since no systemic silencing of the *PDS* gene in coffee was observed, further experimental conditions and other methods are under study to establish a reliable tool for functional studies in coffee.

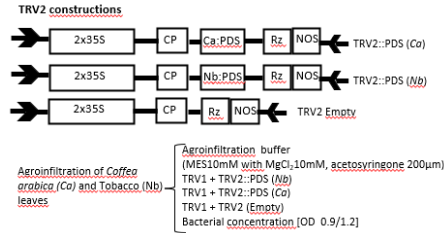


Figure 1: Experimental design



Figure 2: Chlorosis in coffee leaves agro-infiltrated with TRV1 + TRV2::PDS from coffee.

## Results/Discussion

For coffee plants, chlorosis was observed in the infiltrated leaves (Fig 2) but no systemic silencing of the *PDS*



Figure 3: Tobacco leaves with albino phenotype, after agroinfiltration with TRV1 + TRV2::CaPDS (A) and TRV1 + TRV2::NbPDS (B). Positive control.

gene was obtained. Contrasting with the positive control (Fig 3). Although the TRV2::PDS vector was integrated into inoculated plants as shown by PCR assays, it seems it may have been an insufficient downregulation of the gene to affect the phenotype. The different approaches used did not show any improvements. These results corroborate the difficulty of developing a VIGS protocol for *Coffea* spp. plants (Zulfiqar et al. 2023).