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CANDIDATE EFFECTORS FROM THE COFFEE RUST *Hemileia vastatrix* SUPPRESS PAMP-TRIGGERED IMMUNITY / Candidatos a efetores de *Hemileia vastatrix* capazes de suprimir a imunidade desencadeada por PAMP. G. MARIN-RAMIREZ[1]; T. MAIA[1]; D. REZENDE[1]; S. H. BROMMONSCHENKEL[1]. [1]Departamento de Fitopatologia/Instituto de Biotecnologia Aplicada a Agropecuária-BIOAGRO, Universidade Federal de Viçosa, MG, 36570-000, Brazil. Email: thiagomaiaufv@gmail.com

Rust fungi secrete and translocate several effector proteins into the cytoplasm of plant cells to suppress defense responses during the interaction with host plants. In previous studies dozens of *Hemileia vastatrix* candidate effector genes (*HvECs*) with unknown functions were identified. To understand the role of these *HvECs* in the pathogenesis of coffee rust, we carried out functional analysis of these candidate effectors by evaluating their ability to suppress Pathogen Associated Molecular Patterns (PAMPs) responses triggered by the non-pathogenic bacterium *Pseudomonas fluorescens* in *Nicotiana benthamiana*. This immune response, known as PTI (PAMP-Triggered Immunity), is the first line of induced defenses used by plants to resist pathogen infection. Gene sequences encoding the 54 HvECs were individually cloned into pEDV6 vector (without signal peptide). Recombinant plasmids were transferred to *P. fluorescens* EtHAn (Effector to Host Analyser), which has a type three secretion system able to translocate candidate effectors encoded by the *HvEC* genes into the cytoplasm of *N. benthamiana*. Fifteen effector candidates suppressed PTI with high reproducibility in different co-infiltration experiments of EtHAn with the pathogenic bacterium *P. syringae* pv. tomato DC3000. Suppression of PTI has been confirmed by analyzing DC3000 population growth in the infiltrated tissues where PTI was suppressed for all 15 HvECs. In order to verify cell viability in the infiltrated areas, *N. benthamiana* leaves were subjected to the trypan blue staining at 60 hours post-infiltration. Staining of the areas showing hypersensitivity responses was observed. Our results indicate that the transient expression assay using the EtHAn-adapted EDV system in *N. benthamiana* is an effective tool to identiify candidate effectors from *H. vastatrix* capable to suppress plant defense mechanisms.

Key words: *Pseudomonas fluorescens* EtHAn, Effectors biology, Effector delivery vector