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A GENE TRUNCATION STRATEGY GENERATING C-TERMINAL DELETION VARIANTS OF A EFFECTOR PROTEIN FROM *Hemileia vastatrix* for CHARACTERIZATION OF ITS AVIRULENCE ACTIVITY IN COFFEE PLANTS WITH THE S_H1 RUST RESISTANCE GENE / Estratégia de clonagem gênica gerando deleções C-terminal de uma proteína efetora de *Hemileia vastatrix* para caracterização da sua atividade de avirulência em cafeeiros portadores do gene de resistência à ferrugem S_H1. M. N. ARAUJO, T. MAIA, S. H. BROMMONSCHENKEL[1]. [1]Departamento de Fitopatologia/Instituto de Biotecnologia Aplicada a Agropecuária-BIOAGRO, Universidade Federal de Viçosa, MG, 36570-000, Brazil. Email: araujo.n.maicon@gmail.com

A candidate *Avr* gene (*HvEC-016*) has been cloned from *Hemileia vastatrix*. The encoded mature protein is able to trigger an immunity response in coffee plants carrying the S_H1 rust resistance gene. The mature HvEC-016 is a protein of 201 amino acids, containing seven cysteine residues. In order to identify the domain of this protein that is able to activate the immune response mediated by S_H1, we have generated several HvEC-016 truncated sequences encoding seven peptides with size range from 44 to 173 amino acids that have been cloned into the plasmid pEDV6. This vector allows transient expression of encoded proteins and its subsequent translocation into the coffee leaf cells via the type three secretion system of Pseudomonas syringae pv. garcae (Psgc), the causal agent of coffee bacterial blight. pEDV6::HvEC016 truncated constructions were mobilized from Escherichia coli DH5α to Psgc by standard triparental mating using E. coli HB101 as helper strain. Transformed Psgc cells were selected on solid King's B medium containing rifampicin (100 µg ml^{-1}) and gentamicin (25 µg ml^{-1}). DNA sequencing confirmed the frame fusions with the secretion signal of AvrRps4 and integrity for all HvEC-016 C-terminal deletions. Psgc suspension carrying pEDV::HvEC-016 wild type and pEDV::HvEC-016 truncated forms will be infiltrated into leaves from coffee genotypes carrying the S_H1. Based on the presence or absence of bacterial blight symptoms in these genotypes, we expect to identify the domain of HvEC-016 that confers the recognition specificity.

Key words: Plant-pathogen interaction, Avirulence gene, Coffee rust