Proteomic Analysis of Tomato Plants Submitted to *Xanthomonas campestris* pv. *vesicatoria* as a Biotic Elicitor

Martins, M.L.B.[1]; Antunes, P.W.P.[1]; Almeida, H.O.[1]; Games, P.D.[1]; Barbosa, M.O.[1]; Romeiro, R.S.[2]; Baracat-Pereira, M.C.[1]

[1]Departamento de Bioquímica e Biologia Molecular, BIOAGRO, UFV, Viçosa - MG; [2]Departamento de Fitopatologia, UFV, Viçosa - MG.

The natural resistance of plants to pathogens is based on pre-formed and induced mechanisms. After infections, latent defense mechanisms, which confer induced resistance, are activated. This work aims to evaluate the differential synthesis of proteins in tomato (*Solanum lycopersicum*) inoculated with the plant-pathogen *Xanthomonas campestris pv. vesicatoria*. Tomato plant were inoculated with the pathogen by spreading in three different periods, and harvested with 25 days of age, getting four groups (treatments): non-inoculated plants (control) and plants harvested 1, 7, and 14-days after inoculation. Leaves were extracted with Tris-HCl containing protease inhibitors. The extracts were centrifuged and the supernatants called Soluble Extracts (SE). Each precipitate was extracted in LiCl, centrifuged, and the supernatant designated Cell Wall Extract (CWE). CWE and SE were fractionated by ammonium sulfate (30-75% sat.). The precipitates were resuspended and fractionated in MM by ultrafiltration, using membranes with cut-off 30, 10, and 1kDa. SE samples were submitted to anion-exchange chromatography (AEC) in HPLC. CWE samples and the ES1-10 cationic peak (P1 eluted during AEC) were separated in a C18-RP-HPLC. The chromatograms of the four treatments were compared for each SE and CWE, showing differencial protein synthesis, which is observed by the presence, types and intensities of peaks. Selected fractions (peaks) were analyzed by MALDI-TOF/TOF, and 12 molecular masses were obtained (from 1,802.083 to 8,452.687 Da, peptide masses). The identification of differentially synthesized proteins, related to defense or that act directly by inhibiting pathogens, is important to develop new defense techniques for plants.

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