

PS9: 41

The fast identification by MALDI-TOF ICMS of *Fusarium guttiforme* infecting pineapple stem and being antagonised by *Trichoderma asperellum*

J.A. Ventura^{1,3}, C. Santos², H. Costa³, P. Fernandes³, N. Lima²

¹Instituto Capixaba de Pesquisa, Assistência Técnica e Extensão Rural – INCAPER – Vitória - ES, Brazil; ²IBB/Centre of Biological Engineering, University of Minho, Braga, Portugal; ³Núcleo de Biotecnologia, Universidade Federal do Espírito Santo, Vitória - ES, Brazil

Matrix-Assisted Laser Desorption/Ionisation Time-Of-Flight Intact Cell Mass Spectrometry (MALDI-TOF ICMS) is a spectral technique that analyses the chemical cellular composition of microorganisms providing rapid and discriminatory fingerprints for identification. The remarkable reproducibility of this technique is based on the measurement of constantly expressed and highly abundant proteins (Santos *et al.* 2010). The usually observable molecular mass range is between 2000 and 20000 Da, where important ribosomal proteins appear, which is an advantage because these can be easily used as biomarkers. MALDI-TOF ICMS offers advantages over PCR. The method is now used in taxonomic assessments (e.g. bacteria, filamentous fungi, yeast, phages, virus, etc.) once it is capable to identify microorganisms up to level species and, in some cases, up to strain level. The procedure is rapid and in some cases the sample preparation does not need pre-treatment. Time required for the pathogen inactivation is an important determinant of infection-related mortality rates in contaminated crops. Costs associated with pathogen infections in crops could be significantly reduced by employing new rapid identification techniques such as MALDI-TOF ICMS (Santos 2011). In this work pineapple stem was infected with an aqueous suspension of *Fusarium guttiforme* spores and incubated at 25 °C during 5 days. After direct *F. guttiforme* mycelium analysis by MALDI-TOF ICMS the infected pineapple stem was naturally contaminated by *Trichoderma* sp. *Trichoderma* sp. antagonising *F. guttiforme* was directly analysed by MALDI-TOF ICMS. In order to confirm its identity *Trichoderma* sp. was purified, grown in culture plate and analysed by MALDI-TOF ICMS and, macro- and micro-morphology. The results showed that the *F. guttiforme* isolate infecting pineapple stem presented a mass spectrum close to the same isolate grown in Potato Dextrose Agar (PDA) medium and those stored on the SARAMIS™ data base. Additionally, *Trichoderma* sp. spectra (before and after isolate purification) were compared with other spectra stored on the SARAMIS™ data base and identified as *Trichoderma asperellum*. The morphological analyses for the *Trichoderma* isolate corroborate with the MALDI-TOF ICMS results.

Santos *et al.* (2010) *J. App. Microbiol.* 108, 375–385, Santos *et al.* (2011) *Tropical Plant Pathology* 36 (Supplement), August 2011