USE OF MALDI-TOF MASS SPECTROMETRY FOR PLANT GROWTH PROMOTING BACTERIA IDENTIFICATION

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Abstract:

The interest in the use of plant growth promoting bacteria (PGPB) as biofertilizer in agriculture is likely to increase in the coming years, due to the world population growth, higher costs of chemical fertilizers, concerns over pollution and emphasis on sustainable agriculture. The diversity of PGPB isolates from the rhizosphere, plant surface or inner tissues can be vast, and some bacteria are phylogenetically very close. Usually, the main molecular tool for bacteria taxonomic identification and phylogenetic reconstruction is based on 16S rRNA gene sequence. However, the resolving power of this approach is rare at the species level and absent, below it. Recent studies have reported the success of whole-cell mass spectrometry to discriminate bacterial species at the strain level, mainly for bacteria of clinical importance. In this study, we used matrix-assisted laser desorption ionization (MALDI) time-of-flight (TOF) mass spectrometry (MS) to assess the diversity of PGPB isolates from the rhizosphere of maize plants. Total cellular extracts of 77 isolates were analyzed by MALDI-TOF MS and the raw data were converted into a peak list using FlexAnalysis 3.0 software. Hierarchical clustering of mass spectral fingerprint from whole-cell bacteria was performed by SPECLUST. The 16S rRNA gene from bacteria was sequenced using ABI 3500 DNA sequencer. Reads were trimmed for the removal of low quality bases using the Phred program. The Phrap and Consed program were used to view and edit the sequence assembly, respectively. Taxonomic identification based on 16S rRNA gene sequences showed MALDI-TOF clustering of *Pseudomonas fluorescens*, *Luteibacter veojuensis*, *Variovorax sp*. among others, with prevalence of *Burkholderia sp*. Predominant isolates presented a 16S rRNA gene sequence clusterization congruent with the mass spectra one. Moreover, MALDI-TOF whole cell mass profiling allowed a finer discrimination among isolates, suggesting that this technique can be also applied to rapidly identify environmental bacteria.

Keywords: Plant growth promoting bacteria, DNA sequencing, 16S rRNA.

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