

Title: 16S rRNA gene, MALDI-TOF and phenotypic techniques in identification of Gram-negative nonfermenting bacilli recovered from biodegradation process

Authors: Sant' Anna, D. A. S.¹, Sampaio, J. ², Levy, C. E.¹

Institution: ¹Unicamp – Universidade Estadual de Campinas (Rua Tessália Vieira de Camargo, 126 – Cidade Universitária “Zeferino Vaz” – 13083-887 – Campinas – SP), ² Instituto Fleury (Avenida General Valdomiro de Lima, 508 – Jabaquara - São Paulo - SP)

Abstract:

Gram-negative nonfermenting rods are present in the soil and water, being opportunistic pathogens of man, animals and plants. They play a fundamental role in degradation of natural compounds derivated from vegetal and animal substracts and are also important for environmental biodegradation because they can cleave pollutants such as pesticides and detergents. The identification of this bacterial group by phenotypic techniques is often subject to errors, and the genetic sequencing technique is the current “gold standard” for determining a result. This study aims to compare phenotypic procedures by manual and a commercially available system (VITEK®-2) and MALDI-TOF mass spectrometry with 16S rRNA gene sequencing. Eighty six isolates of environmental Gram-negative nonfermentative rods recovered from a biodegradation process were evaluated. For this group of isolates, the 16S rRNA gene sequence analysis showed greater reliability just for the identification at the genus level, with 12 different genus, thus identification at species level for this technique, were not considered. When comparing the results of the three methods, only 39.5% of the isolates agreed at the genus level with the sequencings results, and of these, 41.1% of the isolates agreed with each other at the species level. The results showed greater concordance for the genus *Pseudomonas sp.*: 64.7% of the isolates, 36.3% of them agreeing at species level. On the other hand, 60.4% of the isolates disagreed at genus level with the sequencing results, with the greatest amount of discordant results for the genus *Ochrobactrum sp.*: 28.8% isolates. Although MALDI-TOF be cheaper and fast, still has limitations for this group of bacteria, while the gene sequencing, despite being more accurate, is time consuming, expensive and the 16S rRNA gene not suitable, requiring the use of specific genes for certain genus.

Key-words: 16S rRNA, Gram-negative nonfermentative bacilli, biodegradation, MALDI-TOF, VITEK®-2

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