TITLE: EVALUATION OF THE MALDI-TOF MS SYSTEM AS A SCREENING TOOL FOR THE IDENTIFICATION AND DIFFERENTIATION OF *Burkholderia cepacia* Complex ISOLATES

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ABSTRACT:

Bacteria of Burkholderia cepacia Complex (Bcc) are commonly associated with respiratory tract infection in patients with cystic fibrosis (CF). The treatment of infections by Bcc in these patients is complex and related to a poor prognosis. In fact, Burkholderia cenocepacia, a member of the Bcc, is commonly associated with necrotizing pneumonia. Therefore, an early identification of B. cenocepacia would favor the CF patients prognosis. However, the identification and differentiation of members of Bcc is difficult to be achieved using phenotypic methods. Molecular methods are considered the gold standard for the differentiation of the Bcc members; but these methods require a molecular biology laboratory and skilled labor. The use of mass spectrometry for the identification of microorganisms such as the MALDI-TOF MS system has presented a good accuracy in the identification of Bcc. Due to the need for rapid diagnostics, the objective of this study was to evaluate the performance of the MALDI-TOF MS system as a screening tool in the identification and differentiation of Burkholderia cenocepacia from other Bcc species. A total of 53 colonies suggestive of Bcc were identified by the MALDI-TOF system. We compared two protocols of protein extraction: (A) Direct Method: colonies were transferred to a MALDI-TOF plate and fixed with 1µL of 70% Formic Acid spot; (B) Tube Extraction: colonies were transferred to a microtube and added of 900 µL of ethanol 100%; the ethanol was removed after centrifugation and the pellet was mixed with 25 µL of Formic Acid 70% and 25 µL of Acetonitrile 70%. A volume of 1 µL of the mixture was transferred to a MALDI-TOF target plate. After the colonies were fixed in the plate, in both methods were add 1uL of α-cyano-4-hydroxycinnamic acid and subjected to identification in a Microflex MALDI-TOF equipment. In parallel, all isolates were subjected to molecular diagnosis (PCR with specific primers) to identify species belonging to Bcc and to differentiate B. cenocepacia in genomovar IIIA or IIIB. MALDI-TOF was able to identify 100% (53/53) at the gender level and 94.34% (50/53) at species level using either method of protein extraction (A) and (B). Although both extractions presented reliable results, method (A) is faster and requires fewer reagents. MALDI-TOF can be used as a screening to differentiate all species identified as B. cenocepacia from the other members. Author contact: Fabiana Volpato – fabiana volpato@yahoo.com.br.

Keywords: Burkholderia cepacia complex; MALDI-TOF; Screening tool.

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