TITLE: EVALUATION OF MALDI-TOF MASS SPECTOMETRY FOR DIRECT KPC DETECTION IN ENTEROBACTERALES

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ABSTRACT

Infections caused by carbapenem-resistant *Enterobacterales* are a complex public health challenge worldwide. A rapid and reliable detection of carbapenemase is a subject of major concern, impacting both in patient treatment and infection control purposes. MALDI-TOF (Matrix-Assisted Laser Desorption Ionization Time-of-Flight) has proved to be an accurate tool for rapid bacteria identification and has been evaluated for detection of resistance by using different approaches, such as identifying hydrolytic activity or seeking for specific peaks of these enzymes. According to a recent study, the mass of mature KPC-2 is 28,718.23 Da. However, the m/z is different in KPC subtypes varying from 28,707.84 to 28,730.36 Da. Our study aimed to evaluate the ability of MALDI-TOF to detect the KPC enzyme. Isolates of *Enterobacterales* with characterized carbapenemase genes (*bla*KPC, *bla*NDM and *bla*OXA48-like) were evaluated, as well as non-carbapenemase producers. Bacterial proteins were extracted from Mueller-Hinton agar plates, using formic acid, isopropyl alcohol and water (17:33:50). Samples were prepared with a double layer of synapinic acid. Analyzes were performed in triplicate using MALDI Biotyper CA System (Bruker Daltonics). We evaluated a total of 112 KPC-producing and 104 non-KPC-producing isolates recovered from patients attended in a tertiary hospital in Porto Alegre, South Brazil. After an initial visual analysis of the spectra, KPC-producing isolates showed characteristic peaks between 28,640-28,751, while non-KPC-producing isolates did not show these peaks. Sensitivity and specificity were 100%, proving to be a promising methodology for KPC detection. Further analysis are in progress using ClinProTools 3.0 software (Bruker Daltonics) to normalize and compare the spectra in order to stablish a cutoff to discriminate between KPC-producing and non-producing isolates. In conclusion, besides the detection of hydrolytic activity, the identification of KPC specific peak by MALDI-TOF seem to be an accurate and rapid methodology to detect KPC producing bacteria.

Keywords: carbapenem-resistance, MALDI-TOF MS, KPC

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