

**Title: USE OF MALDI-TOF MASS SPECTROMETRY FOR GROUP C STREPTOCOCCI SPECIES IDENTIFICATION**

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**Summary:**

Group C streptococci (GCS) are appointed as etiological agents of many diseases in humans, especially those caused by *Streptococcus dysgalactiae* subspecies *equisimilis* (SDSE). The incidence of this species may be underestimated, since most cases are not correctly diagnosed or the sample is classified only up to the Lancefield group. A precise clinical diagnostic could provide important data to understand the role of these species as agents of many infections, including invasive. Conventional and miniaturized tests often offer imprecise results. Recently, Matrix-Assisted Laser Desorption/Ionization Time of Flight mass-spectrometry (MALDI-TOF MS) has been appointed as a quick and efficient technique for bacterial identification. This study aimed to evaluate: the ability of MALDI-TOF MS to discriminate species and subspecies of a GCS sample collection, and to compare the results with the identification by conventional methods; the capacity of MALDI-TOF MS to epidemiologically characterize SDSE samples according to their *emm* type. In our study, 157 isolates were analyzed, and only 3 samples showed conflicting identifications when evaluated by MALDI-TOF MS and conventional methods. MALDI-TOF MS identification was later confirmed by rRNA16S sequencing. Most of the tested samples (76.2%) presented scores consistent with a good characterization to the species level, while thirty samples had scores considered highly credible to the species level. A few samples (6.4%) had their identification considered reliable only on a genus level. However, we also observed that this technique could not always determine the subspecies of *S. dysgalactiae* isolates. Regarding the epidemiological characterization of SDSE samples, 18 had their *emm* type determined by sequencing. Analyzing the results of the protein profiles provided by MALDI-TOF MS, we concluded that this technique was not able to epidemiologically relate the samples characterized by *emm* typing. In the present study, we conclude that MALDI-TOF MS is a quick and effective alternative to identify species of the GCS; however it requires the addition of more spectral references for the different species in its database. By doing so, the sensibility and specificity of the identification on a subspecies level would be greatly increased. On the other hand, the analysis of a larger number of samples is needed to properly evaluate the capacity of this technique in the epidemiological characterization of GCS isolates.

**Keywords:** Group C Streptococci, MALDI-TOF MS, Bacterial Identification

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