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MALDI-TOF: driving change in microbiology laboratories

The optimization of the MALDI-TOF MS database for the identification of anaerobic bacteria

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Background: The introduction of Matrix Assisted Laser Desorption Ionization Time-of-Flight Mass Spectrometry (MALDI-TOF MS) is able to identify 70% of the anaerobic bacteria isolated from human clinical specimens and therefore needs optimalisation for the identification of anaerobes. To achieve this goal, the European Network for the Rapid Identification of Anaerobes (ENRIA) has been founded. The ENRIA project is initiated by the ESCMID study groups ESGAI and ESGEM, and consists of 7 expertise laboratories in Europe. The purpose of ENRIA is to produce a minimum of 5 Main Spectral Profiles (MSPs) for each species in the database and to add MSPs of species not yet present in the database.

Material/methods: Each expertise laboratory provided isolates of human clinical anaerobes to the University Medical Center Groningen, The Netherlands. Strains were sub-cultured and identity was confirmed using 16S rRNA gene sequencing. Only strains with a sequence similarity of >98.7% with their closest relative were suitable for MSP creation and addition to the MALDI-TOF MS database. From these strains ethanol suspensions were made, which were sent to Bruker Daltoniks, Bremen, Germany. For the identification of gram-positive anaerobic cocci (GPAC) 111 MSPs representing species underrepresented in the database and species not present in the database were created. The optimized database was validated using 137 clinical isolates of which the identity was confirmed using 16S rRNA gene sequencing.

Results: More than 600 strains were collected, representing 258 different species. The number of species represented by ≥5 MSPs in the database increased from 35 to 166 and 75 species which were not yet present in the database were collected. Analyses of the MSPs of the GPAC strains showed that species form a confined cluster while other species show intra-species variation, resulting in a non-confined cluster. Validation of the optimized database showed that the percentage of reliably identified strains at the species level increased from 56% to 83% and strains only identified at the genus level decreased from 18% to 10%. For 47% of the strains a higher log score was obtained using the optimized database.

Conclusions: With combined efforts of different laboratories within Europe we collected >600 anaerobic strains for the optimalisation of the MALDI-TOF MS database for the identification of anaerobic bacteria. The results of the improved database for the identification of GPAC show what can be achieved when the MALDI-TOF MS database is optimized. Collaborative research by different European laboratories can result in significant improvement of the MALDI-TOF MS database.

Continuing efforts will result in further improvement of this useful diagnostic tool for clinical microbiology laboratories.