COMPARISON OF TWO MALDI-TOF MS SYSTEMS FOR IDENTIFICATION OF ISOLATED BACTERIAL STRAINS FROM IMMUNOBIOLOGICAL PRODUCTION INDUSTRY

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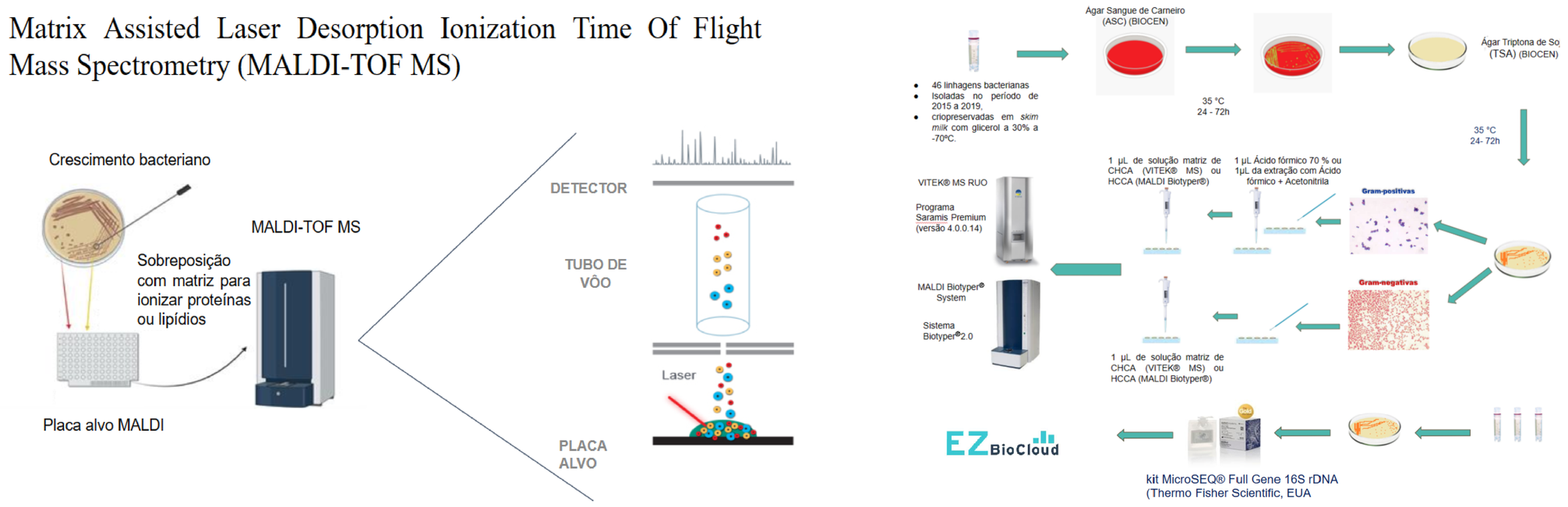
INTRODUCTION

Microbial contamination is one of the main risks associated with drug production. The precise identification of the microbiota detected in the pharmaceutical production environment is essential for investigating sources of contamination, and for carrying out preventive and corrective actions. Studies using the Matrix-Assisted Laser Desorption Ionization-Time of Flight/Mass Spectrometry (MALDI-TOF/MS) methodology, which provides information about the bacterial proteome, have shown good rates in the identification of strains of pharmaceutical origin compared to other phenotypic methodologies, in addition to the high speed in obtaining results. The aim of this study was to compare two MALDI-TOF MS systems for the identification of bacterial strains isolated from an immunobiological manufacturing facility, not previously identified by the semi-automated system VITEK®2.

METHODOLOGY



RESULTS AND CONCLUSIONS

Of the 46 strains analyzed by the VITEK® MS system, 42 (91.3%) weren`t identified and four (8.7%) were identified at the genus level. The 16S rRNA gene sequencing analysis confirmed the genus of the four (100.0%) strains. The MALDI Biotyper® system was able to identify 16 (34.8%) strains, 10 (62.5%) at the genus level and six (37.5%) at the species level. Among the strains identified at the genus level, seven (70.0%) genera were confirmed by 16S rRNA sequencing; two were identified by the basonym of *Sutcliffiella cohnii* (*Bacillus cohnii*) by MALDI Biotyper® ; and one was identified as *Bacillus siralis* by MALDI Biotyper® and *Microbacterium laevaniformans* by sequencing. Of the six strains identified at the species level by MALDI Biotyper® , four (66.6%) species were confirmed by sequencing, and two others were among the possible species showed by EzBioCloud with similarity above 98.7%. One strain identified by sequencing as *Sutcliffiella cohnii* showed a similarity < 98.7%, which may indicate a new species. In conclusion, the MALDI-TOF/MS system was unable to identify most of the strains evaluated. However, MALDI Biotyper® showed higher identification efficiency than VITEK® MS, suggesting that the database of the former presents a greater number of spectra of bacteria species from the pharmaceutical environment. The database customization, based on the introduction of new spectra of environmental strains, becomes a good alternative for improving the MALDITOF/MS ability to obtain better identification results

Tabela 1. Results of bacterial identifications by MALDI-TOF MS and 16S rRNA gene sequencing.

Strains	VITEK® MS (Biomerieux)	MALDI Biotyper® (Bruker)	16S rRNA Gene Sequencing Species (Similarity)
1	Not identified	*Bacillus cohnii* (1.94)	*Sutcliffiella cohnii* (96,37%)
2	*Ureibacillus* spp. (84,60%)	Not identified	*Ureibacillus chungkukjangi* (99,86%); *Ureibacillus sinduriensis* (99%)
3	Not identified	*Agrococcus terreus* (2.19)	*Agrococcus terreus* (99,51%)
4	Not identified	*Bacillus siralis* (1.80)	*Microbacterium laevaniformans* (99,13%)
5	Not identified	*Cupriavidus metallidurans* (2.17)	*Cupriavidus metallidurans* (99,65%); *Cupriavidus pauculus* (99,02%); *Cupriavidus plantarum* (98,81%); *Cupriavidus gilardii* (98,77%)
6	Not identified	*Microbacterium laevaniformans* (1.85)	*Microbacterium laevaniformans* (99,57%); *Microbacterium dextranolyticum* (98,96%)
7	Not identified	*Brachybacterium nesterenkovii* (1.74)	*Brachybacterium nesterenkovii* (99,1%)
8	Not identified	*Exiguobacterium aurantiacum* (2.40)	*Exiguobacterium mexicanum* (99,78%); *Exiguobacterium aquaticum* (99,52%); *Exiguobacterium aurantiacum* (99,32%); *Exiguobacterium alkaliphilum* (98,83%)
9	Not identified	*Paraburkholderia fungorum* (2.31)	*Paraburkholderia insulsa* (100%); *Paraburkholderia fungorum* (99,79%); *Paraburkholderia agricolaris* (99,5%); *Paraburkholderia madseniana* (99,36%); *Paraburkholderia kirstenboschensis* (99,19%); *Paraburkholderia aromaticivorans* (98,93%); *Paraburkholderia strydomiana* (98,84%); *Paraburkholderia phytofirmans* (98,79%); *Paraburkholderia elongata* (98,79%); *Paraburkholderia caledonica* (98,72%); *Paraburkholderia solitsugae* (98,72%)
10	Not identified	*Tsukamurella tyrosinosolvens* (2.27)	*Tsukamurella tyrosinosolvens* (99,93%); *Tsukamurella sputi* (99,93%); *Tsukamurella ocularis* (99,79%); *Tsukamurella hominis* (99,79%); *Tsukamurella conjunctivitidis* (99,78%); *Tsukamurella asaccharolytica* (99,78%); *Tsukamurella pulmonis* (99,65%); *Tsukamurella strandjordii* (99,65%); *Tsukamurella pseudospumae* (99,51%); *Tsukamurella sinensis* (99,5%); *Tsukamurella inchonensis* (99,3%); *Tsukamurella spumae* (99,3%); *Tsukamurella hongkongensis* (99,28%); *Tsukamurella paurometabola* (99,16%)
11	Not identified	*Rhodococcus rhodochrous* (1.95)	*Rhodococcus gordoniae* (99,86%); *Rhodococcus biphenylivorans* (99,29%); *Rhodococcus pyridinivorans* (99,15%)
12	Not identified	*Acinetobacter guillouiae* (1.84)	*Acinetobacter guillouiae* (99,1%); *Acinetobacter bereziniae* (98,76%)
13	Not identified	*Bacillus cohnii* (1.86)	*Sutcliffiella cohnii* (99,53%); *Sutcliffiella catenulatus* (99,53%); *Sutcliffiella zhanjiangensis* (98,91%)
14	Not identified	*Microbacterium laevaniformans* (2.90)	*Microbacterium laevaniformans* (99,48%); *Microbacterium dextranolyticum* (98,79%)
15	Not identified	*Acinetobacter guillouiae* (1.81)	*Acinetobacter guillouiae* (98,94%)
16	Not identified	*Brevibacterium pityocampae* (1.95)	*Brevibacterium pityocampae* (100%)
17	Not identified	*Rhodococcus corynebacterioides* (1.87)	*Rhodococcus corynebacterioides* (99,64%); *Rhodococcus kroppenstedtii* (99,22%)
18	*Brachybacterium* sp. (79,00%)	Not identified	*Brachybacterium paraconglomeratum* (99,93%); *Brachybacterium conglomeratum* (99,64%); *Brachybacterium saurashtrense* (99,14%)
19	*Brachybacterium* sp. (82,00%)	Not identified	*Brachybacterium paraconglomeratum* (99,93%); *Brachybacterium conglomeratum* (99,64%); *Brachybacterium saurashtrense* (99,14%)
20	*Brachybacterium* sp. (75,50%)	Not identified	*Brachybacterium paraconglomeratum* (99,93%); *Brachybacterium conglomeratum* (99,64%); *Brachybacterium saurashtrense* (99,14%)

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