Evaluation Of Automated MALDI – TOF Target Plate Preparation

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ABSTRACT

Microorganism identification by Matrix Assisted Laser Desorption Ionization – Time of Flight (MALDI – TOF) requires target plate spotting with an isolated colony and addition of matrix. The Copan Colibri™ is an FDA cleared instrument that spots the target plate and automatically adds matrix to help automate and standardize these normally manual process. In this study, we performed 218 identifications of bacterial urine Gram–positive and Gram-negative isolates by the Vitek MS (bioMerieux) prepared by Colibri and compared the identifications to those obtained by our manual target spotting/ matrix method. All testing was performed according to manufacturer's instruction.

For Colibri testing, isolated colonies were chosen from digital images of plates on the WASP Webapp image in the Copan WASPLab system. The automatically picked by the Colibri using a disposable pipet tip The target plates were spotted and matrix was added by the Colibri. Target plates were placed onto the VITEK MS for MALDI-TOF. Routine manual spotting was also performed by standard laboratory procedure and the results compared. All plates included QC and E.coli calibrator standard.

A total of 228 organisms, including 210 isolates from 188 culture plates and 18 ATCC QC isolates were tested and evaluated. Only 2 discrepancies were observed. One was Streptococcus agalactiae (manual) and Proteus mirabilis (Colibri) and one Raoultella ornithinolyticus (manual) and Enterobacter complex (Colibri). The accuracy of identification between the two methods was 226/228 (99.1%). No purity plates were examined and repeat testing was not performed to determine root cause of these 2 discrepancies. There were 10/228 (4.4%) organisms that yielded no identification by MALDI-TOF processed by Colibri. These included 4 Enterococcus faecalis, 3 Streptococcus agalactiae, 1 Streptococcus gallolyticus spp., 1 Streptococcus pyogenes and 1 E. coli. By procedure, these isolates with no identification would be repeated with manual spotting and were not considered discrepant results. The Colibri instrumentation with automated spotting and addition of matrix is an accurate method for preparation of MALDI-TOF target plates.

INTRODUCTION

Microorganism identification by Matrix Assisted Laser Desorption Ionization – Time of Flight (MALDI-TOF) requires target plate spotting with an isolated colony and addition of matrix. The Copan Colibri is an FDA cleared instrument that spots the target plate and automatically adds matrix to help automate and standardize these normally manual processes.

The Colibri is an automated device for colony picking and spotting of MALDI-TOF target plates used for microbial identification. The system can accommodate both VITEK® MS and Bruker Biotyper® target plates. The Colibri picks colonies designated by operators from digital images of culture plates viewed on the Copan WASPLab® reading station. Optionally, colonies may be automatically selected by the PhenoMATRIX® TAG. Colonies on plates are automatically picked by a robotic pipettor and organism spotted onto the appropriate target plate. The system adds matrix and optionally adds formic acid. The Colibri also has the capability to prepare microbial suspensions for antimicrobial susceptibility testing (not tested in this study).

METHODS

1. One hundred and eighty-eight (188) positive Urine cultures were obtained from WASPLab system.
2. Two hundred and ten (210) isolated colonies were chosen from digital images of plates on the WASP Webapp image.
3. The plates were loaded onto the Colibri instrument, and the designated colony was automatically picked by the Colibri using a disposable pipet tip, Figure 1.
4. The target plates were spotted, and matrix was added by the Colibri, Figure 2.
5. Target plates were placed onto the VITEK MS for MALDI-TOF identification.
6. Routine manual spotting was performed by standard laboratory procedure and the results were compared.
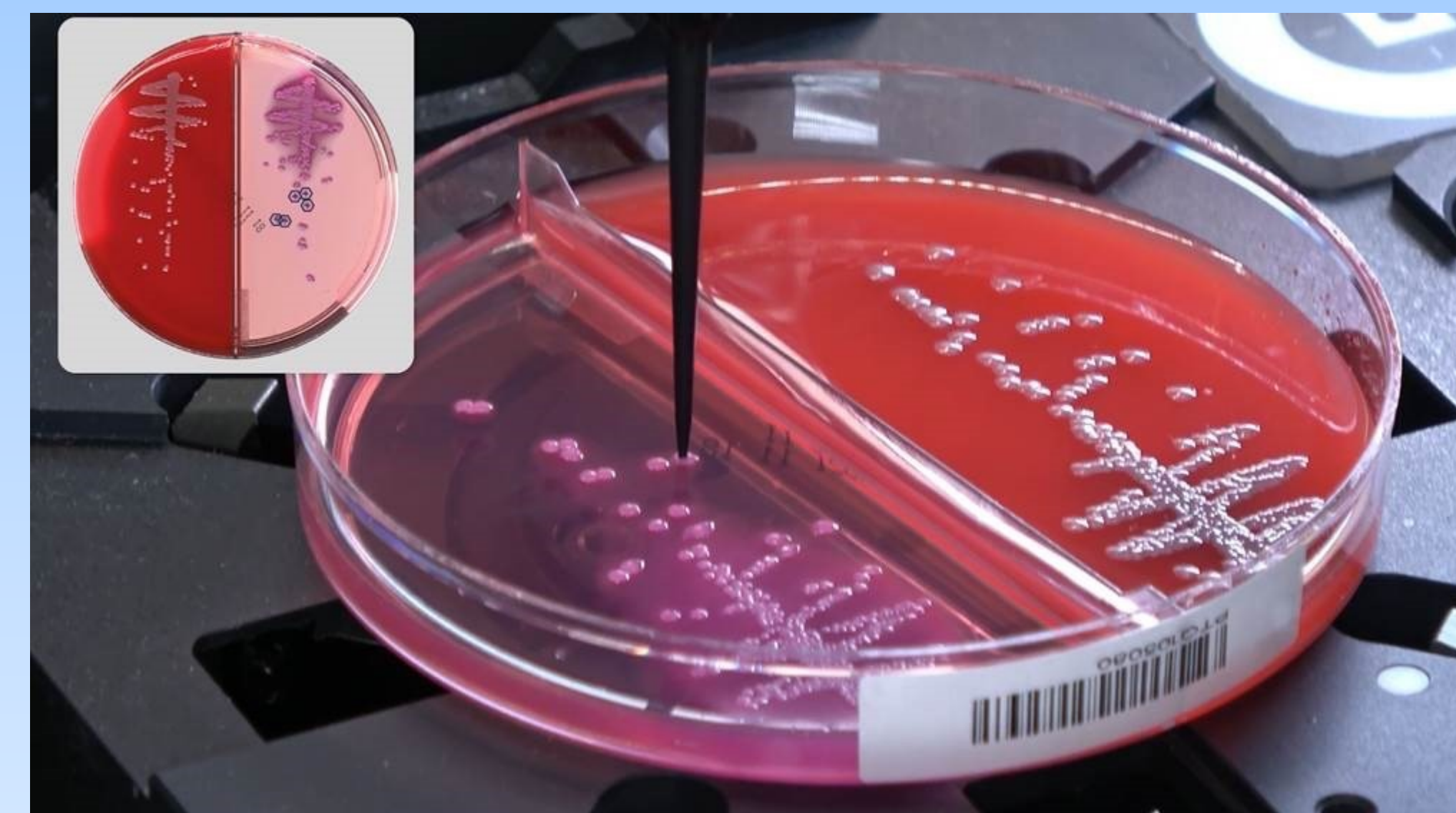

Figure 1. System plate picking

Figure 2. Automated addition of matrix

RESULTS

1. A total of 218 urine isolates and 18 QC organisms were tested
   a. Gram-negatives, 163 isolates + 12 controls (n=175)
   - E. coli n=107; Klebsiella spp n=23; Proteus spp n=16; P. aeruginosa n=5; other GNB n=11
   b. Gram-positives, 47 isolates + 6 controls (n=53)
   - E. faecalis n=15; S. aureus n=14; coag negative staph n=12; S. agalactiae n=5; other streptococci n=2

2. Total of 10 isolates (10/228, 4.4%) with no ID (would repeat manually)

3. Total of 2 errors (accuracy 216/218, 99.1%)
   a.1 S. agalactiae identified as Proteus (most likely incorrect plate tested)
   b.1 Raoultella ornithinolyticus identified as Enterobacter complex (unknown cause)

CONCLUSIONS

The Colibri instrumentation with automated spotting and addition of matrix is an accurate method for preparation of MALDI-TOF target plates. There were only 2 errors (99.1% accuracy) and only 10 isolates that yielded no identification after being spotted (these 10 would be tested manually).