P0335
Poster Session I
Rapid antimicrobial susceptibility testing

5-HOUR ANTIBIOTIC SUSCEPTIBILITY TESTING OF ENTEROCOCCUS FAECIUM AND E. FAECALIS, AND ACINETOBACTER BAUMANNII DIRECTLY FROM POSITIVE BLOOD CULTURES USING AUTOMATED MICROSCOPY

A. Shamsheyeva[1], D. Gamage[1], S. Metzger[1], B. Turng[1], D. Howson[1], **C.S. Price**[2]

[1]Assay Development, Accelerate Diagnostics, Tucson, USA ; [2]Division of Infectious Diseases Medicine, Denver Health Medical Center, Denver, USA

Objectives: Healthcare-associated *Acinetobacter baumannii* and enterococcal bloodstream infections are of increasing concern. Septic patients infected with these organisms have been shown to face elevated risk of receiving inappropriate initial therapy with broad-spectrum antibiotics. Even a few hours of delay in initiating appropriate antibiotic treatment increases the risk of severe morbidity and mortality. An innovative technology using automated microscopy was evaluated for its performance for rapid antibiotic susceptibility testing (AST) of *A. baumannii*, *E. faecium*, and *E. faecalis* directly from positive blood culture.

Methods: A total of 228 clinical isolates selected for having MICs near the CLSI breakpoints were tested; 88 *A. baumannii* isolates (45 with imipenem (IPM) and 43 with minocycline (MIN)), and 140 *E. faecium* or *E. faecalis* isolates (45 with ampicillin (AMP), 48 with vancomycin (VAN) and 47 with linezolid (LZD)). Ten to one hundred colony forming units of bacterial suspension were spiked into simulated blood culture bottles (1 part of healthy donor blood + 4 parts of BD BACTEC Standard Aerobic media), and incubated for approximate 20-24 h. One milliliter of the positive blood culture was removed and lysed. Automated gel electro-filtration (20 min) was performed to reduce sample debris. Samples were pipetted into independent flowcells of a disposable multichannel cassette, and electro-kinetic concentration immobilized cells onto the transparent lower surface of each flowcell channel (5 min). Immobilized cells were challenged with single concentration solutions of antibiotic prepared in cation-adjusted Mueller Hinton broth with 0.85% agar. Automated microscopy with image analysis software scanned and analyzed growth rates from changes in the biomass of each immobilized progenitor cell as it grew into a clone of daughter cells. A computer algorithm converted bacterial growth or inhibition in the presence of antibiotic into a minimum inhibitory concentration (MIC). A growth control was also performed. Standard CLSI frozen broth micro-dilution (BMD) was performed in parallel as a reference using overnight plated pure culture colonies. Microscopy AST results were compared to BMD results.

Results: The time from positive blood culture sample preparation to obtain MIC results for the microscopy system ranged from 3.5 to 5 h. Essential agreement (± 1 dilution) for *A. baumannii* with IPM and MIN was 98% and 95%, respectively. Essential agreement for *Enterococcus* spp. for AMP, VAN and LZD was 96%, 92%, and 94%, respectively.

Conclusion: Data in this study suggests that the new automated microscopy technology is feasible for providing rapid AST results directly from positive blood culture within 5 h. AST results were concordant with overnight pure culture using BMD. Automated microscopy offers a promising alternative for performing AST directly from positive blood culture bottles to provide rapid MIC results for tailoring treatment of *A. baumannii* and enterococcal blood stream infections.