2020. Multifaceted Approach to Optimize Blood Volume Inoculated into Culture Bottles in the Pediatric Intensive Care Unit
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Session: 234. Diagnostics – Bacterial Identification and Resistance
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Background. Blood culture is essential in diagnosing bloodstream infections (BSIs). It is well known that blood volume inoculated into the culture bottle is directly associated with blood culture positivity. However, effective interventions to encourage physicians to increase inoculum volume in pediatric patients are not known. Therefore, multifaceted interventions were performed to optimize the blood volume in our pediatric intensive care unit (PICU).

Methods. Prior to intervention, the recommended blood culture inoculum was greater than 1 ml for any child. The interventions were initiated in November, 2016. A simple guideline recommending blood volume according to body weight category (<10kg: 1ml, 10–20kg: 2ml, >20kg: 4ml for each bottle), was distributed by e-mail and placed on blood collection carts. Monthly feedback of blood volume collected and blood culture positivity was provided to all PICU medical staff who routinely perform blood sampling. Blood weight was determined by subtracting the initial bottle weight from the bottle weight on laboratory arrival. The blood volume was compared pre and post intervention.

Results. The blood volume submitted in 2,449 blood culture bottles was included in the analysis. Of these, 1,051 (42.9%) were in pre intervention and 1,398 (57.1%) were in post intervention period. The median blood volume and the percentage above recommended blood volume decreased slightly in body weight under 10 kg group (1.23 ml vs 1.13 ml, P < 0.001, 79.0% vs 73.6%, P = 0.016), but increased significantly in body weight between 10 and 20 kg group (1.42 vs. 1.79 mL, P = 0.003, 32.6% vs. 45.5%, P = 0.002) and over 20 kg group (1.60 mL vs 2.78 mL, P < 0.001, 16.4% vs. 32.4%, P = 0.001) (Figure 1). The blood culture was positive in 5.6% of the bottles that achieved recommended blood volume and 3.0% from the bottle that did not (P = 0.003).

Conclusion. The multifaceted intervention was effective in improving blood volume for culture in pediatric patients weighing over 10 kg.

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2021. The Development of an IMP Metallo-β-lactamase Detection Assay Using a Bioinformatic Approach
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Session: 234. Diagnostics – Bacterial Identification and Resistance
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Background. Carbapenem antibiotic resistance in Gram-negative bacteria is becoming increasingly common. One such resistance gene, IMP metallo-β-lactamase, has been found worldwide. With 48 different IMP alleles across many genera of bacteria, no known assay can detect all 48 IMP alleles. We developed a PCR-based assay to detect IMP genes using a novel bioinformatic approach to design primers that would recognize all gene variants.

Methods. Computer simulation of primers was accomplished by using common PCR primer design heuristics as parameters. Following primer simulation, an all-against-all gene comparison was done to calculate potential primer sets. The minimum
number of primer sets required to detect all IMP genes was subsequently found by iterative deduction. The predicted primer sets were tested against 7 IMP producing bacterial isolates (IMP-1, 4, 7, 8, 18, 27 from Serratia, Enterobacteriaceae, Pseudomonas, and Klebsiella spp.) and one synthesized gene of IMP-35. These isolates were chosen to represent the full genetic spectrum of the IMP family. The remaining 40 genes were evaluated based on gene sequences obtained from GenBank.

**Results.** The *in silico* analysis showed 6 primer sets were needed to detect all known IMP genes. PCR amplification of template DNA isolated from the 8 strains showed that primer sets 1 and 4 could detect all 8 IMP isolates while the remaining 4 sets (2, 3, 5, 6) had distinct amplification patterns that could be used together to identify a specific IMP gene group. Effectiveness of these primer sets in IMP identification was demonstrated by testing a clinical isolate containing an undetectable carbapenem resistant bacterium. The IMP-27 gene was identified by PCR amplification using the IMP-specific primers designed and confirmed by sequence analysis.

**Conclusion.** A bioinformatic approach can be used to create an assay for bacterial resistance. The assay developed with this approach can detect and classify all known IMP metallo-β-lactamase genes in carbapenem resistant Gram-negative bacteria. Such information could aid in guiding treatment and evaluating the epidemiology of IMP-producing bacteria.

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2022. **Quantiferon conversions and reversions among HIV patients on antiretroviral therapy.**

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**Background.** Tuberculosis (TB) remains a major global public health problem. Recent guidelines recommend screening of high-risk patients for latent TB infection, with the preferential use of interferon gamma release assays (IGRA) over PPD. However, there are several reports of reversions and conversion of quantiferon TB tests (QFT).

**Methods.** The primary aim of this retrospective review among HIV positive patients, >18 years of age, performed from January 2011 to April 2017, was to measure the occurrence of QFT conversions (negative to positive) and reversions (positive to negative) during routine clinical care. Continuous variables will be presented as mean, standard deviation. Categorical variables will be presented as the number and percentage of subjects in each category.

**Results.** There were a total of 381 cases, with an average age of 52.8 (±12) years of age. From those 249 patients had at least one QTF performed during the study period and 98 patients had 2 or more tests. 196 had a PPD test performed. From the patients that were initially QTF negative (N = 100), 7 converted to positive.

There were a total of 10 patients who’s QTF was positive and were retested, and from those 7 reversed to negative (see Table 1). Most cases that reversed were considered to be low positives (5/6 of those where initial values available).

The remaining 21 patients had a history of positive PPD and 16 of those had negative QTF.

**Conclusion.** Reversions were common among HIV positive patients with high CD counts. Most reversions occurred with initial low positive QTF results. Providers should consider differential testing among patients with QTF conversion and no history of TB exposure in low-risk settings. Future studies must be done to confirm these findings.

**Disclosures.** All authors: No reported disclosures.

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2022. **2023. Attenuated Total Reflectance Fourier Transform Infrared Spectroscopy for Rapid Identification of Non-Fermenting Gram-Negative Bacilli Isolated from Patients with Cystic Fibrosis**

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**Session:** 234; **Diagnosis—Bacterial Identification and Resistance**

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**Background.** Chronic respiratory infections with non-fermenting Gram-negative bacilli are a key feature of cystic fibrosis (CF). For microbiology laboratories, rapid and accurate identification of these bacteria is often challenging and labor intensive. This study was undertaken to evaluate whether attenuated total reflectance Fourier transform infrared (ATR-FTIR) spectroscopy could rapidly discriminate *Pseudomonas aeruginosa* (mucoid and non-mucoid), *Burkholderia cepacia* complex, *Burkholderia gladioli*, *Acinetobacter* spp. and *Stenotrophomonas maltophilia*.

**Methods.** A total of 263 well-characterized clinical strains isolated from respiratory samples of patients with CF attending the CHU Sainte-Justine CF clinic were included in this study. consisting of 70 *P. aeruginosa*, 83 *Burkholderia* spp., 52 *Acinetobacter* spp. and 58 *Stenotrophomonas maltophilia* isolates from the biobank. Isolates were thawed and sub-cultured twice on sheep blood (5%) agar. ATR-FTIR spectral acquisition was performed in triplicate for each isolate. Multivariate statistical analysis of the ATR-FTIR spectra was performed by hierarchical cluster analysis (HCA) and principal component analysis (PCA) in conjunction with the use of a feature selection algorithm.

**Results.** An ATR-FTIR spectral database consisting of 789 spectra of *P. aeruginosa*, *Burkholderia* spp., *Acinetobacter* spp. and *Stenotrophomonas maltophilia* was created in this study. Complete discrimination among all four genera as well as among three species within the *B. cepacia* complex and *B. gladioli* was achieved based on HCA and PCA of the spectra in the database. ATR-FTIR analysis of a validation set consisting of 30 isolates was conducted in parallel with identification by MALDI-TOF mass spectrometry and yielded >95% concordance between the two techniques.

**Conclusion.** ATR-FTIR spectroscopy is a promising tool for rapid, inexpensive and accurate identification of non-fermenting Gram-negative bacilli. Additional work is needed to further expand the spectral database, particularly with mucoid strains.

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2024. **Aminoglycoside Susceptibility Agreement between an Automated System and Broth Microdilution for Carbapenem-resistant Enterobacteriaceae**

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**Session:** 234; **Diagnosis—Bacterial Identification and Resistance**

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**Background.** CRE are a world-wide public health challenge with extremely limited treatment options. Aminoglycosides have variable activity against these organisms. At our institution, amikacin has been active against these isolates and used as part of a combination regimen for CRE treatment. In this study, we compared the susceptibility results for 3 aminoglycosides between an automated susceptibility system (Phoenix) and broth microdilution (BMD).

**Methods.** Gentamicin, tobramycin, and amikacin susceptibility were determined in our academic medical center microbiology laboratory using an automated susceptibility system (Phoenix) and broth microdilution (BMD) according to CLSI guidelines against 120 recent CRE clinical isolates. Categorical agreement was defined between methods as classification of isolates in the same susceptibility category using CLSI breakpoints. Minor, major and very major error rates were calculated for each aminoglycoside.

**Results.** The primary CRE was *K. pneumoniae* (46%), followed by *Enterobacter* spp. (32%) and *E. coli* (6%). The categorical agreement ranged 58% (gentamicin) to 68% (tobramycin). Automated susceptibility system provided significantly higher susceptibility from 14% (gentamicin) to 30% (tobramycin and amikacin).

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