Table 1. Demographics, Baseline Laboratory Values, and Clinical Outcomes for the cohort

| Variable                      | n = 185 |
|-------------------------------|---------|
| **Demographic Information**   |         |
| Age Median (IQR)              | 68 (54–78) |
| Male Gender                   | 90      | 53.3% |
| Race                          |         |
| White                         | 126     | 74.6% |
| African American              | 21      | 12.4% |
| Other                         | 18      | 10.7% |
| Unknown                       | 4       | 2.4%  |
| Ethnicity                     |         |
| Hispanic                      | 9       | 5.5%  |
| Not Hispanic                  | 141     | 76.3% |
| Unknown                       | 15      | 8.1%  |
| **Laboratory Results**        |         |
| WBC (10^9 cells/uL) Median (IQR) | 11.5 (7.4–18.6) |
| Creatinine (mg/dl) Median (IQR) | 1.1 (0.8–1.9) |
| Albumin (g/dl) Median (IQR)   | 3 (2.5–3.0) | n = 152 |
| CBT7/NAP1 B1 strain           | 17      | 10.1% |
| ICU admission                 | 24      | 14.2% |
| Collecioncy                   | 1       | 0.6%  |
| Death within 40 days          | 16      | 8.3%  |
| **Severe Clinical Outcomes - Total** |        |
| Serious Clinical Outcomes - Attributed to C. difficile |         |
| ICU admission                 | 13      | 7.7%  |
| Collecioncy                   | 1       | 0.6%  |
| Death within 40 days          | 2       | 2.4%  |
| **Severity Classifications**  |         |
| ESUSA Severe                  | 69      | 74.4% |
| ESMOD Severe                  | 69      | 76.9% |
| CAUTI Severe                  | 73      | 80.3% |
| Estimation at Severe          | 58      | 15.9% |

Figure 1. Comparison of TcdA and TcdB concentrations, as measured by Simoa, in serum and stool. Clinical cutoffs are shown: stool, 20 pg/ml for TcdA and for TcdB; serum 15.0 pg/ml for TcdA and is 26.7 pg/ml for TcdB. Signals below these cut-offs are below backgrounds and so negative.

Conclusion: In contrast to earlier published findings which reported on the presence of detectable toxin in the serum of a small number of patients with C. difficile, our work did not support this observation. Although Simoa is highly sensitive for detection of picogram quantities of TcdA or TcdB it was unable to detect either toxin in serum during C. difficile infection.

Disclosures: Alice Banz, Ph.D., BioMerieux (Employee) Kevin W. Garey, PharmD, MS, FASHP; Merck & Co. (Grant/Research Support, Scientific Research Study Investigator) Carolyn D. Alonso, MD, FIDSA, Alnylam Pharmaceuticals (Employee) Merck (Research Grant or Support) Ciyanan Kelly, MD, Artugen (Consultant)/Facile Therapeutics (Consultant)/Finch (Consultant)/First Light Biosciences (Consultant)/Matrixix (Consultant)/Merck (Consultant)/Yedanta (Consultant)

646. Adapting the modified Carbapenem Inactivation Method to assess for possible beta-lactamase mediated resistance in Piperacillin-Tazobactam resistant/ Ceftriaxone susceptible Escherichia. coli and Klebsiella pneumoniae

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Session: P-25. Diagnostics: Bacteriology/mycobacteriology

Background: A cluster of piperacillin-tazobactam resistant/ceftriaxone susceptible Escherichia coli and Klebsiella pneumoniae bacteremias were noted at our institution. A review of the literature suggested this resistance phenotype was mediated by a beta-lactamase. We sought to further corroborate this hypothesis.

Methods: We adapted the “carbapenem inactivation method” utilizing piperacillin-tazobactam and ceftriaxone discs on all E. coli and K. pneumoniae isolated from blood and demonstrating piperacillin-tazobactam resistance but with ceftriaxone susceptibility. We utilized pan-susceptible and carbapenem resistant Enterobacteriaceae reference strains as well as third generation cephalosporin resistant, piperacillin-tazobactam susceptible isolates as controls.

Results: 96% of the piperacillin-tazobactam resistant, ceftriaxone susceptible strains demonstrated the capacity to degrade the piperacillin-tazobactam discs while 100% spared the ceftriaxone discs. 75% of the piperacillin-tazobactam susceptible, ceftriaxone resistant control strains spared the piperacillin-tazobactam discs while degrading the ceftriaxone discs.

Conclusion: The resistance phenotype observed is due to beta-lactamase production and the modified carbapenem inactivation method can be adapted to probe for other beta-lactamases. Further study is required to definitively identify which beta-lactamase is responsible.

Disclosures: All Authors: No reported disclosures

647. Adoption of the updated fluoroquinolones breakpoints for Gram negative bacteria in clinical microbiology laboratories

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Session: P-25. Diagnostics: Bacteriology/mycobacteriology

Background: Despite the multiple safety warnings related to fluoroquinolones (FQs) treatment, their use remains unavoidable in several occasions due to their broad spectrum of coverage including activity against multi-drug resistant glucose non-fermenting Gram-negative bacteria such as Pseudomonas spp., and high oral bioavailability. The Clinical and Laboratory Standards Institute (CLSI) has lowered the FQs minimal inhibitory concentrations (MICs) breakpoints for Salmonella spp. in 2012 and 2013, and for the Enterobacteriaceae and P. aeruginosa in 2019. We aim to explore the number of hospitals that adopted the revised breakpoints.

Methods: We conducted a cross-sectional phone-based survey querying the 43 microbiology laboratories that serve 100% of the acute care and long-term hospitals in Connecticut to determine use of revised FQs MIC breakpoints for Gram-negative bacteria.

Results: Six laboratories refer antimicrobial susceptibility testing to another local hospital microbiology laboratory or to a national reference laboratory. Thus, we obtained information about the study question from a total of 37 microbiology laboratories. Eight laboratories (21.6%) implemented the revised breakpoints exclusively for P. aeruginosa and enterobacteria. Six laboratories (16.2%) only adopted the revised breakpoints for P. aeruginosa, while 15 (40.6%) laboratories followed by BioMérieux Vitek 2 in 13 (35.1%) laboratories. Four laboratories (10.8%) only adopted the revised CLSI FQs breakpoints for Enterobacteriaceae and P. aeruginosa, and Salmonella spp. 5 (13.5%) implemented the revised breakpoints for Enterobacteriaceae but not for salmonella spp., and 8 (21.6%) laboratories adopted the revised CLSI breakpoints for Salmonella spp. but not for Enterobacteriaceae and P. aeruginosa.

Conclusion: The use of outdated FQs breakpoints for Gram-negative bacteria remains common in the microbiology laboratories. There is an urgent need to mitigate the impact of using the outdated FQs breakpoints and reporting false susceptibility to FQs.

Disclosures: All Authors: No reported disclosures

648. BioFire FilmArray® Pneumonia plus Panel Performance Evaluation: A Multicenter, International Collaborative Study

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EME Evaluation Program Collaborative

Session: P-25. Diagnostics: Bacteriology/mycobacteriology

Background: Classical methods to identify causes of community acquired, healthcare and ventilator associated pneumonia can be insensitive and slow, leading to unnecessary or inappropriate antimicrobial therapy. The BioFire FilmArray®
Pneumonia plus Panel (PNplus) detects 15 bacteria (in semi-quantitative log bin values from $10^{-4}$ to $>10^{4}$), 7 antibiotic resistance markers (mecaA/CMRE, CTX-M, KPC, VIM, IMP, NDM, OXA-48 like), 3 atypical bacteria (ABA), and 8 viral classes directly from bronchoalveolar lavage (BAL)-like and sputum-like specimens (including endotracheal aspirations) in about 1 hr. This study compared PNplus results to standard of care (SOC).

**Methods:** 2476 samples (1234 BAL-like; 1242 sputum-like) were tested at 52 laboratories from 15 European countries and Israel by PNplus and SOC. SOC varied by site and physician prescription. Pathogen detection rates were compared. PNplus bin values and SOC descriptive or numerical quantities were evaluated for 1297 bacterial detections.

**Results:** 13 samples (0.5%) gave invalid PNplus results. 3278 bacteria in PNplus were detected by PNplus and/or SOC. SOC detected 1878 bacteria (57.1%) compared to 3128 bacteria (95.8%) for PNplus ($p<0.0001$). SOC detected 73 AB (70.9%) and 134 viruses (21.1%). PNplus detected 93 AB (90.3%) and 618 viruses (97.9%) ($p=0.0001$). Mean number of analytics/sample detected by PNplus and SOC was 1.99 and 1.44, respectively. PNplus bin values were less than SOC, equal to SOC or greater than SOC in 5.9%, 25.4% and 69.6% of results, respectively. PNplus values were on average $>1$ log than SOC values (58.5% $>1$ logs; 11.0% $=3$ logs). PNplus identified 98.2% of MRSA and SOC 55.6%. All gram-negative resistance markers were detected at least once. PNplus and SOC results were fully concordant (positive or negative) or partially concordant for 49.1% and 26.4% of samples, respectively.

**Conclusion:** PNplus detected significantly more potential pathogens than SOC. Lack of routine SOC viral testing was a missed opportunity to define the cause of pneumonia. Semi-quantification may assist in understanding the significance of the pathogens detected. Pathogen and resistance marker detection in about 1 hr could dramatically impact antimicrobial use and enhance patient outcomes.

**Disclosures:** Christine C. Ginocchio, PhD, MT(ASCP), bioMerieux (Employee) bioMerieux (Employee) Shareholder; Barbara Mauerhofer, Pharmacist, bioMerieux (Employee) Cory Rindlisbacher, n/a, BioFire Diagnostics (Employee) Carolina Garcia, BS, bioMerieux (Employee)

**649. Clinical Implementation of a Rapid Susceptibility Testing Procedure, Directly From a Positive Blood Culture Using the Vitek®2 System on Gram Negative Rods**

Charma Henry, MLS1; Dustin Evans, MT1; Daniel Navas, MLS(ASCP)3.

**Background:** The national average of detection and susceptibility for organisms isolated from positive blood culture to final susceptibility based on growth on solid media is 48 hours. The goal of this research was to prove that the Vitek®2 (bioMerieux, Inc.) system can provide an accurate and reliable susceptibility result directly from positive blood culture for Gram negative rods and reduce the turnaround time (TAT) from positive blood culture to the final susceptibility.

**Methods:** An FDA-modified validation procedure was performed on positive blood cultures directly from the bottle to the VITEK®2 System for susceptibility testing. The protocol tested and validated an aliquot of 50uL of blood directly from the positive bottle into 10mL of saline (1:200). The solution was vortexed and 3mL were placed in the VITEK®2 10mL test tubes. This protocol was intended for Gram negative bacteria using the AST-GN70, AST-GN81 & AST-GN801 cards. This protocol followed the CLSI M52 protocol.

**Results:** 351 organisms from clinical blood culture samples from July 2018 to October 2019 were evaluated. Organisms included, but were not limited to: E. coli, K. pneumoniae, Enterobacter spp., and P. aeruginosa, Proteus spp., Salmonella spp., Acinetobacter spp., and S. maltophilia. There were 5,201 drug/bug combinations. AdventHealth Orlando achieved an essential agreement of 99.32% ($k=0.53$), minor error 0.74% ($n=39$) major error 0.02% ($n=1$) and very major error 0.49% ($n=2$). A 100% agreement was achieved on detection of ESBL, CRE, and MDR organisms.

**Conclusion:** Rapid direct blood culture protocol using the VITEK®2 System and the AST-GN cards is accurate, reliable and can be performed with less than 1 minute of hands-on time. The protocol can be implemented in any laboratory at no additional costs or modification where the current VITEK®2 AST-GN panels are in use. Compared with the national average of 72 hours, the TAT obtained during this study was decreased median time from collection until arrival to the laboratory was seen was post implementation (2.0 pre- vs. 0.8 hours post-implementation, $p=0.001$). The positivity rate increased from 3967 (8.1%) pre-implementation to 6141 (11.6%) post-implementation ($p<0.001$) (Table and Figure). Staphylococcus aureus was the most frequently isolated species for both periods and had higher recovery rate with the VIRTUO system (737 (1.5%) pre- vs. 1764 (3.3%) post-implementation, $p<0.001$). Higher recovery rate was also noted for other Staphylococcus spp. in the post-implementation period (985 (2.0%) pre- vs. 1644 (3.1%) post-implementation, $p<0.001$). No difference in the organism recovery rate was noted for Streptococcus spp., Enterococcus facism, E. facialis, Pseudomonas aeruginosa, Enterobacterales, and Candida spp. The inpatient contamination rate was high post-implementation (1.5% pre- vs. 1.9% post-implementation, $p<0.001$).

**Comparison of blood culture positivity rate pre- vs. post-implementation, by culture location**

**Daily positivity rate for blood cultures processed at BJH during the study period**

**Conclusion:** The VIRTUO system showed a higher rate of positive blood cultures compared to the VersaTREK system primarily from a higher detection of Staphylococcus spp. Further studies are needed to assess whether an increased rate of positive blood cultures is associated with changes in management and clinical outcomes.

**Disclosures:** All Authors: No reported disclosures

**651. Comparative Analysis Between Bacterial And Fungal Malignant Otitis Externa**

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**Background:** Malignant otitis externa is a fatal infection of the external ear and temporal bone. Pseudomonas aeruginosa is the most common causative organism, while fungi are a rare cause of malignant otitis externa. We aimed to compare the clinical, therapeutic and evolutionary features between bacterial and fungal malignant otitis externa.

**Methods:** We conducted a retrospective study including all patients hospitalized for malignant otitis externa in the infectious diseases department between 2000 and 2018.

**Results:** Overall, we encountered 82 cases of malignant otitis externa, among which 54 cases (65.9%) of bacterial malignant otitis externa (BMO) and 28 cases (34.1%) of fungal malignant otitis externa (FMO). The males were predominating among BMO cases (57.4% vs 50%; $p=0.5$). Patients with FMO were significantly older (70.19 years vs 61.10 years; $p<0.001$) and had medical history of diabetes mellitus more frequently (96.4% vs 58%; $p=0.003$). The use of corticosteroids oral was significantly more reported among FMO cases (28.6% vs 5.6%; $p=0.006$). Otalgia (96.4% vs 81.5%), otorrhea (75% vs 66.7%), and cephalalgia (46.4% vs 42.6%) were the