640. Development of a Laboratory Verification Protocol for Concurren
tDetection of Bacterial, Fungal, and Antimicrobial Resistance Genes in a Multiplex
Syndromic Joint Infection Panel
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Session: P-29. Diagnostics: Bacteriology/mycobacteriology

\textbf{Background.} Verification is a critical component of implementing a diagnostic
test in a clinical lab and can be time consuming and costly. A verification protocol
and organism panel were developed in collaboration with ZepetoMetrics, LLC to verify all
analyte detections for the BioFire Joint Infection (JI) Panel\textsuperscript{2}. The BioFire JI Panel
detects 31 pathogens and 8 antimicrobial resistance (AMR) genes associated with joint
infections from synovial fluid specimens.

\textbf{Methods.} A protocol was developed using prototype NAT\textsuperscript{TM} controls from
ZepetoMetrics\textsuperscript{2}, synovial fluid, and the BioFire FilmArray 2.0 and the BioFire
FilmArray Torch Systems. Control materials were tested in the presence of synovial
fluid from pooled human donors. The 32 targets required for all analyte detections
were divided into 5 pools of 6-7 analytes and then tested over multiple days on sev-
eral systems.

\textbf{Results.} Preliminary outcomes were good with a cumulative positive detect-
ion rate of 100\% (310/310) and expected negative detections of 99.3\% (1182/1190) from
50 prototype BioFire JI Panel test runs. AMRs were correctly identified in 50/50
(100\%) replicates when a correlated bacterium was present. Unexpected detections of
Streptococcus spp. (7/50) and Staphylococcus lugdunensis (1/50) were likely due to
contaminants in the synovial fluid. Streptococcus spp. was confirmed by testing the
synovial fluid in isolation.

\textbf{Conclusion.} Efficient performance verification may be achieved by combining 32
organisms/8 AMR into 5 pools and can be completed with 20 test runs in 4 days. The
pooling scheme provides multiple positive/negative detections per analyte and accura-
tely detects AMR. The protocol and controls serve as a useful tool for providing reli-
able detections of targets over multiple days, operators and systems and offers a flexible
solution for supporting verification needs.

“The BioFire Joint Infection Panel is currently pending US FDA De Novo review. This
product has not been evaluated by other global regulatory agencies for in vitro diagnostic use.
Not available for sale. Panel menu subject to change.

\textbf{Disclosures.} Monica Cronin, MS, BioFire Diagnostics, LLC (Employee); Taylor K.
Fadgen, Bachelor of Science; BioFire Diagnostics, LLC (Employee); Lisa Ogden, BS,
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641. Clinical Predictors of Hospital-Acquired Bloodstream Infections
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Session: P-29. Diagnostics: Bacteriology/mycobacteriology

\textbf{Background.} Hospital-acquired bloodstream infections (HABSI) are associated
with increased mortality and decreased hospital quality metrics. This has led to an
increased focus on blood culture stewardship. Little data exists regarding predictive factors
of bacteremia in hospitalized patients. We aim to determine what clinical character-
istics are predictive of patient’s seroconversion of HABSI.

\textbf{Methods.} This is a retrospective case-control study of 540 patients with posi-
tive blood cultures admitted to our health system between September 1, 2017, to April
1, 2020. Electronic medical records of patients with positive blood cultures were
independently reviewed to determine contamination versus true bacteremia. We looked at
different clinical parameters and laboratory investigations within 24 hours of drawing blood cultures. Clinical variables were age ≥ 60 years, heart rate ≥ 90/minute, systolic blood pressure ≥ 90 mmHg or use of a vasopressor, oral tem-
perature > 38°Celsius (100.4°Fahrenheit), white blood cells (WBC) count ≥ 12,000/µL,
lymphocytes ≤ 1000/mm\textsuperscript{3}, platelets < 150,000/µL, and creatinine > 2.0 mg/ dl.
Stepwise logistic regression analysis was used for predictive statistical model development.

\textbf{Results.} In a cohort of 481 patients with hospital-acquired bacteremia, 350 cases had true bacteremia and 131 cases were contaminated blood cultures. Stepwise regression analysis showed that white blood cell (WBC) count ≥ 12,000 cells/µL, lymphocyte count ≤ 1000/mm\textsuperscript{3}, creatinine > 2.0 mg/dL, and oral tem-
perature > 38°C (100.4°F) were associated with HABSI (R\textsuperscript{2}=0.06, p value 0.002).

\textbf{Conclusion.} Our findings suggest that WBC count, lymphocyte count, creatinine,
and oral temperature together can be used to develop appropriate blood culture steward-
ship models in the inpatient setting. This may help minimize unnecessary blood culture
detections.

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642. Facility Reported vs. CLSI MIC Breakpoint Comparison of Carbapenen-
Non-susceptible (Carb-NS) Enterobacteriaceae (ENT) from 2016-2019:
A Multicenter Evaluation
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Session: P-29. Diagnostics: Bacteriology/mycobacteriology

\textbf{Background.} Carbapenem (Carb) minimum inhibitory concentration (MIC)
breakpoints are lowered by CLSI in 2010 and recognized by FDA in 2012. Adoption of
revised breakpoints is often slow, which may lead to under-reporting of Carb-
non-susceptibility (NS) by facilities. We compare facility-reported rates of Carb-NS
ENT to the CLSI MIC breakpoints for a large nationwide collection of isolates in the
United States (US) from 2016-2019.

\textbf{Methods.} All adults with a positive non-contaminant ENT culture (first isolate of a
species per 30-day period from blood, respiratory, urine, skin/wound, intra-abdomi-
 nal, or other) in ambulatory/inpatient settings from up to 300 US hospitals from 2016-
2019 were evaluated (BD Insights Research Database). Facility-reported Carb-NS was
defined as: susceptible (S), intermediate (I) or R to eteperpen (ETP), imipenem (IPM),
meropenem (MEM) and/or doripenem (DOR) per commercial panels. Where avail-
able, MICs were interpreted using CLSI 2010 MIC breakpoints (µg/mL): ≤ 0.5 (S), 1
(I), ≥ 2 (R) for ETP and ≤1 (S), 2 (I), and ≥ 4 (R) for IPM/MEM/DOR. For evaluable
ENT isolates we compared susceptibility results as reported by the facility to CLSI ENT
MIC breakpoints.

\textbf{Results.} Overall, 77.4% (937,926/1,211,845) and 90.6% (2,157,785/2,381,824)
non-duplicate ENT isolates with facility-reported susceptibility results also had inter-
pretable MIC results for ETP and IPM/MEM/DOR, respectively (Tables). ETP’s S rates
were 99.3% and 99.1% as reported by facilities and using CLSI criteria, respectively.
R rates of other Carb were 98.9% and 98.4% by facility reporting and CLSI criteria, respectively.
Systematic application of CLSI breakpoints under-reported I-R isolates by 24.2% and 16.4%,
respectively, and identification of IPM/MEM/DOR-I-R and -R isolates by 31.3% and 22.7%, respectively.

\textbf{Conclusion.} Systematic application of CLSI breakpoints in 2016-19 would have
dominated minimal impact on ENT S rates in the US. However, facility reporting failed to
identify 18.8% of ETP I-R and 26.5% of IPM/MEM/DOR I-R isolates. The clinical
relevance of these observations is unknown. Facilities should know their local
epidemiology, decide if under-reporting might be an issue, and assess if there is any
impact on their patients.

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643. Evaluation of Rapid Blood Pathogen Identification Along with Antimicrobial
Stewardship at an Academic Teaching Institution
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Session: P-29. Diagnostics: Bacteriology/mycobacteriology

\textbf{Background.} Bloodstream infections are a major cause of morbidity and
mortality in hospitalized patients. Prompt initiation of effective antimicrobials
are essential to optimize patient outcomes. New diagnostic technologies rapidly
identify bacteria, viruses, fungi, and parasites in infections of various body sites.
These are a paucity of literature determining if stewardship programs run by one
trained pharmacist with rapid diagnostics decreases time to optimal
antimicrobial therapy.

\textbf{Methods.} This was a retrospective chart review of positive bloodstream infec-
tions identified via rapid diagnostic technologies. The EHR of admitted adult patients
with positive BSI identified by BioFire FilmArray Blood Culture Identification (BCID) Panel® or Accelerate PhenoTest Blood Culture kit® between January 2018 – July 2019 were evaluated and pertinent data was collected.

**Results.** Rapid diagnostic technologies identified 108 bloodstream infections due to gram positive, 56 due to gram negative, and 6 due to Candida organisms. Mean time to optimal antimicrobial therapy was significantly lower when pharmacist recommendations were accepted versus when primary care team consulted ID for recommendations or did not accept pharmacist recommendation. Mean time to optimal therapy was 14.7, 34.3, and 217.3 hours (p < 0.0001) respectively. Median total cost of visit per patient, calculated using the average wholesale price of antibiotics multiplied by the number of doses received, was significantly lower when pharmacist recommendations were accepted ($86.40, $147.95, and $239.41, respectively).

**Baseline characteristics**

| Variable     | ASP Pharmacist recommendation accepted | ASP Pharmacist recommendation not accepted | p-value |
|--------------|----------------------------------------|-------------------------------------------|---------|
| Gram Positive | 63 (85.7%)                             | 41 (26.2%)                                | 0.0298  |
| *Staphylococcus aureus* | 38 (56.7%)                             | 35 (21.8%)                                | 0.0298  |
| *N. gonorrhoeae* | 88 (99.4%)                             | 71 (67.9%)                                | 0.0001  |
| *Rhodococcus equi* | 34 (85.0%)                             | 19 (68.2%)                                | 0.2475  |
| *Mycobacterium kansasii* | 47 (92.2%)                             | 38 (78.8%)                                | 0.0611  |

**Microbiological isolates**

| Organism Isolated | Species                                      | Genus Positive Organisms (GPOS) |
|-------------------|---------------------------------------------|---------------------------------|
| *Paenibacillus*    | *Paenibacillus*                              | *Paenibacillus*                  |
| *Staphylococcus*   | *Staphylococcus*                             | *Staphylococcus*                 |
| *Rhodococcus*      | *Rhodococcus*                                | *Rhodococcus*                    |
| *Mycobacterium*    | *Mycobacterium*                              | *Mycobacterium*                  |

**Primary outcomes: Time to optimal antimicrobial therapy**

| Variable                      | ASP Pharmacist recommendation accepted | ASP Pharmacist recommendation not accepted | p-value |
|-------------------------------|----------------------------------------|-------------------------------------------|---------|
| Time to optimal antimicrobial therapy (h) | 7.1 (5.9–12.1)                     | 25.7 (17.4–44.6)                      | <0.0001 |

**Missed cost savings**

| Variable                      | N  | Mean | Median | 25th–75th | 75th–99th |
|-------------------------------|----|------|--------|------------|-----------|
| Actual cost of end (US$)      | 42 | 862.40 | 258.41 | 156.01 | 512.18 |
| Hypothetical cost of end (US$) | 42 | 648.70 | 111.00 | 70.89 | 295.34 |
| Potential cost savings (US$)  | 42 | 213.70 | 111.00 | 85.12 | 237.72 |

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644. Phenotypic and Genomic Analysis of Novel, Fastidious, Gram-negative Bacilli Isolated from Clinical Wound Specimens

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**Session: P-29. Diagnostics: Bacteriology/mycobacteriology**

**Background.** Animal bites are considered the thirteenth leading cause of nonfatal ED visits. Epidemiology studies have shown a rise in dog bites during the COVID-19 pandemic in the U.S. In Oct. 2020, we received a facultatively anaerobic, non-hemolytic organism (OL1) from a dog bite wound for identification. 16S rRNA gene sequencing showed OL1 was 95.9% identical to *Ottowia pentelensis* in the family *Comamonadaceae*. Our historical sequence database revealed 8 additional isolates (OL2–OL9) from hand wounds/abscesses (including 3 dog bites) since 2012 that had ≥ 99.8% identity with OL1. Most other *Ottowia* sp. have been isolated from industrial and food sources, with no reports from patient samples. As these clinical isolates likely represent a novel *Ottowia* species, we aimed to characterize them using both phenotypic and genotypic approaches.

**Methods.** The OL isolates were tested in API 20 NE panels (8 conventional and 12 assimilation tests) for 4 d. Paired-end genomic DNA libraries (Nextera DNA Flex Library Prep, Illumina) were sequenced as 150 nt reads by Illumina NovaSeq. De novo assembly, annotation, functional prediction, and phylogenetic analyses were performed with Geneious, PATRIC, and web-prediction databases. Strain comparison was done with StrainTypeMer.

**Results.** All 9 OL isolates were negative for indole, urea, arginine, esculin, DNPF, glucose fermentation and carbohydrate assimilation tests. Potassium gluconate assimilation and gelatin hydrolysis were positive for 5 and 4 isolates, respectively. StrainTypeMer showed the isolates from different patients were not closely related, but 2 from the same patient were indistinguishable. The estimated genome size was ~3.1 Mbp, with 66.1% G/C, and ~3523 coding genes. Potential virulence factors (BrkB and MviM), multidrug efflux systems (MdtABC-TolC and Bcr/CIA), and 1-2 intact prophages were identified. Genomic phylogenetic analysis with RAxML showed the OL isolates clustered separately from all known *Ottowia* spp.

**Conclusion.** These OL isolates are fastidious, Gram-negative bacilli from clinical wound specimens, and are associated with dog bites. Genomic and 16S rRNA gene sequence analysis suggests these isolates constitute a novel species within the family *Comamonadaceae*.

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645. Rapid Diagnosis of Disseminated *Mycobacterium kansasii* Infection in Renal Transplant Recipients Using Plasma Microbial Cell Free DNA Next Generation Sequencing

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**Session: P-29. Diagnostics: Bacteriology/mycobacteriology**

**Background.** Disseminated *Mycobacterium kansasii* infection is rare in kidney transplant recipients. The diagnosis may not be suspected readily due to non-specific clinical presentation. The diagnosis and treatment can be further delayed due to poor sensitivity of culture (especially of extra-pulmonary sites) and slow growth in culture media. Accurate and rapid diagnosis of disseminated *M. kansasii* infections in transplant recipients is important for antimicrobial management.

**Methods.** Two cases of disseminated *M. kansasii* infections with unusual presentation in which rapid diagnosis was made using the Karius test (KT) are presented. The KT is a CLIA certified/CAP-accredited next-generation sequencing (NGS) plasma test that detects microbial cell-free DNA (mcfDNA). After mcfDNA is extracted and NGS performed, human reads are removed, and remaining sequences are aligned to a curated database of >1400 organisms. Organisms present are identified by applying a statistical threshold.

**Results.** Case 1: A 31-year female kidney transplant recipient presented with a thyroglossal duct cyst, as well as swelling of her right metacarpophalangeal joint and left 3rd finger. AFB culture of the thyroglossal cyst aspiration done on post admission day (PAD) 2 took 27 days to be identified as *M. kansasii* (on PAD 29) whereas plasma sent for KT on PAD 5 reported a positive test for *M. kansasii* at 284 molecules/μl on PAD 7. On PAD 10, the first patient presented with painless swelling of his left 3rd finger (on PAD 29) whereas plasma sent for KT on PAD 5 reported a positive test for *M. kansasii* at 1314 MPM in 3 days (on PAD 15). PET CT done simultaneously was consistent with an infection of an old AV graft in the left upper extremity. The AFB culture of the resected graft was confirmed as *M. kansasii* in 22 days on PAD 36. After the KT was available (before confirmation of *M. kansasii* on culture), the first patient underwent modification of empiric treatment and the second patient was started on specific treatment for *M. kansasii*.