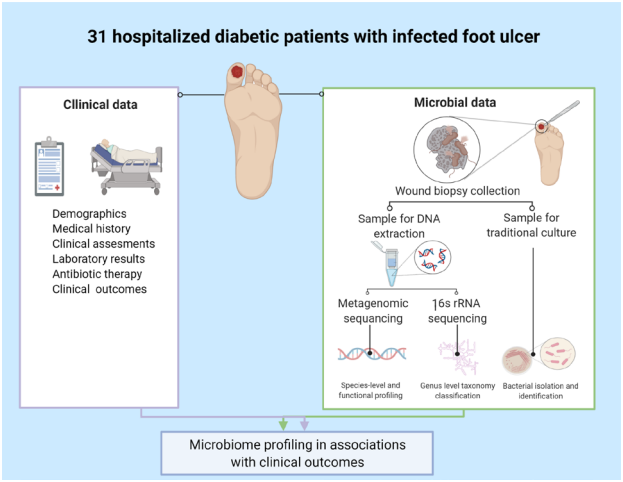
Institute of Technology, Israel, Canadian Institute for Advanced Research (CIFAR) , Canada, Haifa, Hefa, Israel

Session: P-57. Microbiome in Health and Disease

Background. Infected diabetic foot ulcers (IDFU) are a major complication of diabetes mellitus. These potentially limb-threatening ulcers are challenging to treat due to the impairment of wound healing in diabetic patients and the complex microbial environment characterizing these ulcers. Our aim was to analyze the microbiome of IDFU in association with clinical outcomes.

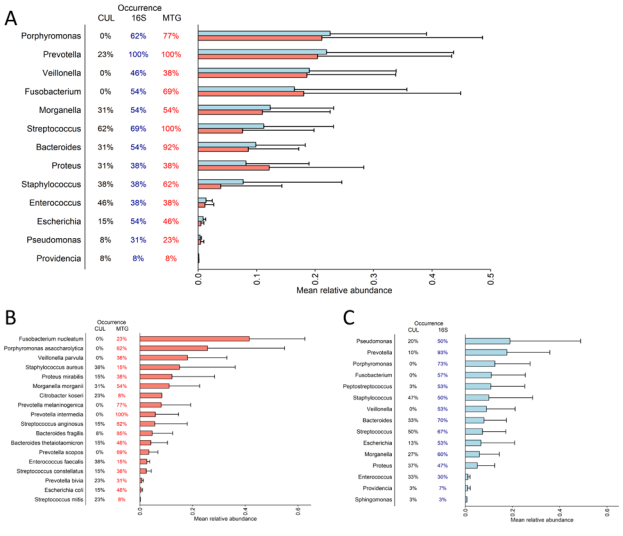
Methods. Wound biopsies from IDFU were obtained from hospitalized patients and were analyzed using traditional microbiology cultures, 16S rRNA sequencing and shotgun metagenomic sequencing. Patients' characteristics, culture-based results and sequencing data were analyzed in association with clinical outcomes.

Study Design



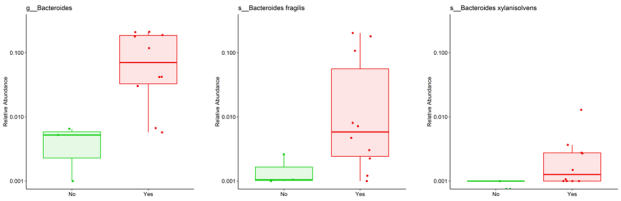
Results. 31 patients were enrolled. Significantly more anaerobic and Gram-negative bacteria were detected with sequencing methods compared to conventional cultures (59% and 76% were anaerobes according to 16SrRNA and metagenomic respectively vs. 26% in cultures, p=0.001, and 79%, 59% and 54% were Gram negative bacteria respectively, p< 0.001). Culture-based results showed that *Staphylococcus aureus* was more prevalent among patients who were conservatively treated (p=0.048). In metagenomic analysis the *Bacteroides* genus was more prevalent among patients who underwent toe amputation (p< 0.001). Analysis of metagenomic-based functional data showed that antibiotic resistance genes and genes related to biofilm production and to bacterial virulent factors were more prevalent in IDFU that resulted in toe amputation (p< 0.001).

Occurrences and mean relative abundances of the most prevalent bacteria of IDFU



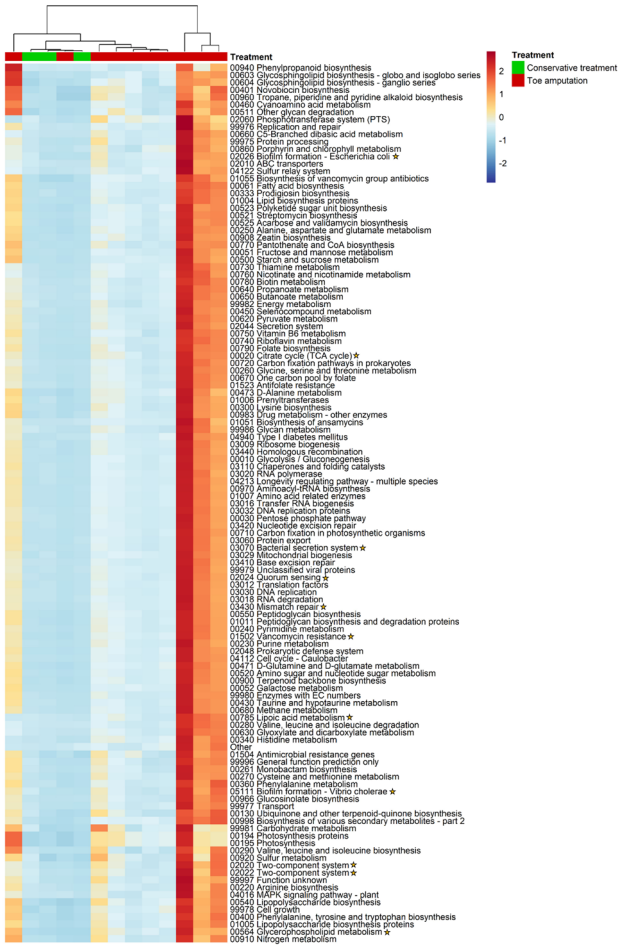
Comparison between [A] traditional cultures, 16S rRNA sequencing and metagenomic sequencing results (genera level - 12 samples) [B] traditional cultures and metagenomic sequencing results (species level – 30 samples) [C] traditional cultures and 16S rRNA sequencing results (genera level - 30 samples) CUL – cultures; 16S - 16S rRNA sequencing; MTG – metagenomic sequencing

Bacteroides genus association with toe amputation



Bacteroides genera was more common among samples of patients who underwent toe amputation compared with samples of patients who were conservatively treated (p < 0.001). Species level analysis showed that Bacteroides fragilis and Bacteroides xylanisolvens predominated IDFU of patients who underwent toe amputation (p=0.04, p=0.002 respectively). No – conservative treatment; Yes – toe amputation.

Functional genes differentiating patients who underwent toe amputation from conservatively treated



Yellow stars – indicate genes that were associated with bacterial virulent factors, biofilm formation and resistant mechanisms – all were more prevalent in patients who underwent toe amputation (with p values<>

Conclusion. Molecular sequencing tools uncover the complex biodiversity of IDFU and emphasize the high prevalence of anaerobes and Gram-negative bacteria in these ulcers. Furthermore, sequencing results highlighted the possible association between certain genera, species, and bacterial functional genes to clinical outcomes

Disclosures. **Yossi Paitan, PhD, Ilex Medical Ltd** (Employee, Other Financial or Material Support, As of 01.01.2021 I am the Laboratories Manager of Ilex Labs)

1019. Clinical Impact of a Rapid Cerebrospinal Fluid Diagnostic Stewardship Program for Suspected Central Nervous System Infections in Children

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Session: P-58. New Approaches to Diagnostics

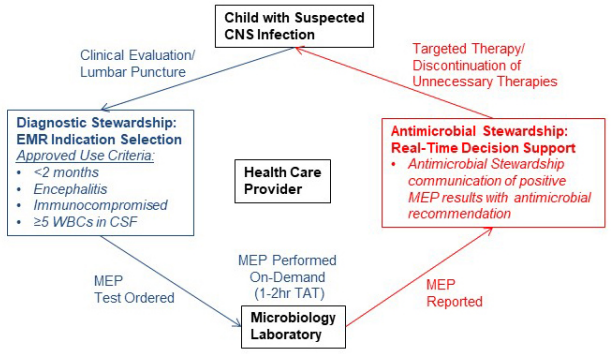
Background. Despite widespread use, the optimal implementation and clinical impact of FilmArray Meningitis Encephalitis Panel (MEP; Table 1) multiplex PCR testing of cerebrospinal fluid (CSF) in children with suspected (CNS) infections is unknown.

Table 1: FilmArray Meningitis Encephalitis Panel Test Characteristics

FilmArray® Meningitis Encephalitis Panel (MEP)
◦ FDA approved 2015 for testing CSF obtained by LP
◦ 1 hour time on machine
◦ Requires 0.2mL of specimen
◦ List price $193/test cartridge
◦ Multiplex PCR with 14 targets
(Overall: 94.2% sensitivity, 99.8% specificity)
Bacteria Targets
Escherichia coli K1
Haemophilus influenzae
Listeria monocytogenes
Neisseria meningitidis
Streptococcus agalactiae
Streptococcus pneumoniae
Virus Targets
Cytomegalovirus (CMV)
Enterovirus (EV)
Herpes simplex virus 1 (HSV-1)
Herpes simplex virus 2 (HSV-2)
Human herpesvirus 6 (HHV-6)
Human parechovirus (HPeV)
Varicella zoster virus (VZV)
Yeast Target
Cryptococcus neoformans/gattii

Methods. A pre-post quasi-experimental cohort study to investigate the impact of implementing MEP using a rapid CSF diagnostic stewardship program was conducted at Children's Hospital Colorado (CHCO). MEP was implemented with EMR indication selection to guide testing to children meeting approved use criteria: i. infants < 2mo, ii. immunocompromised, iii. encephalitis, iv. > 5 WBCs in CSF. Positive results were communicated with antimicrobial stewardship real-time decision support (Fig 1). All cases with CSF obtained by lumbar puncture (LP) sent to the CHCO microbiology laboratory meeting any of the 4 criteria above were included with pre-implementation controls (2015-2016) compared to post-implementation cases (2017-2018). Primary outcome was time-to-optimal antimicrobials (time from LP to 1st dose of antimicrobials targeted to identified pathogen, or cessation when no treatable pathogen identified).

Figure 1: Rapid Cerebrospinal Fluid Diagnostic Stewardship Program Intervention Design



Results. Post-implementation (n=1127) and pre-implementation (n=1124) group characteristics are in Table 2. Following implementation, MEP was sent in 72% of cases, largely replacing pathogen-specific singleplex CSF testing (Table 3). Time-to-optimal antimicrobials decreased by 10 hours (p< 0.0001; Fig 2). There were no differences in time-to-effective antimicrobials, hospital admissions, antimicrobial starts or length of stay. Time-to-positive CSF results was faster (4.8 vs. 9.6 hrs, p< 0.0001), IV antimicrobial duration was shorter (24 vs 36 hrs, p=0.004) with infectious neurologic diagnoses more frequently identified (15% vs. 10%, p=0.03). Overall, 3% had bacterial and 9% viral CNS infection identified. Enterovirus (n=128) was most common, then HSV (n=28) and parechovirus (n=17) with similar detection rates between groups

Table 2. Cohort Demographics, Characteristics, Inclusion Criteria Met and CSF Indices

Characteristic	Post-Implementation Cases (n=1127)	Pre-Implementation Controls (n=1124)	P Value
Demographics			
Age (median months)	43±68	42±67	0.71
Female Sex	531 (47%)	506 (45%)	0.3389
Race			0.0598
American Indian/Alaska Native/Native Hawaiian/Other Pacific Islander	17 (2%)	13 (1%)	
Asian	38 (4%)	27 (3%)	
Black or African-American	68 (7%)	77 (8%)	
White	757 (78%)	766 (81%)	
More than one race	95 (10%)	63 (7%)	
Hispanic of Latino Ethnicity	325 (31%)	315 (29%)	0.5426
Underlying medical condition	345 (31%)	343 (31%)	1
Immunocompromised	40 (12%)	31 (9%)	0.3485
Inclusion Criteria Met			
Age <2 months	552 (49%)	626 (56%)	0.0019
CSF WBC>5	474 (42%)	439 (39%)	0.1542
Immunocompromised	40 (4%)	31 (3%)	0.3404
Concern for encephalitis	602 (53%)	518 (46%)	0.0006
Cerebrospinal Fluid Indices			
CSF WBC	4 (1, 18)	3 (1, 13)	0.2263
CSF RBC	11 (1, 386)	7 (1, 280)	0.5129
CSF glucose	51 (44, 60)	51 (46, 59)	0.4959
CSF protein	49 (26, 84)	52 (26, 83)	0.8455

Figure 2: Time-to-Optimal Antimicrobial Therapy in Post-Implementation Cases (n=1127) vs. Pre-Implementation Controls (n=1124)

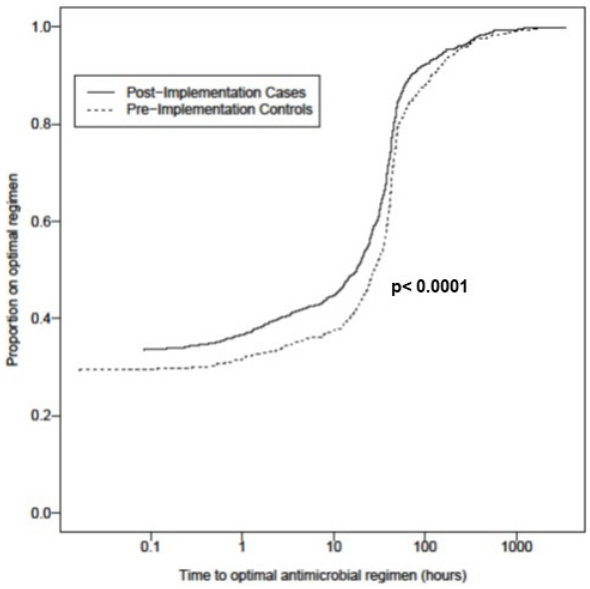


Table 3. Microbiologic Testing, Discharge Diagnoses, Outcomes			
Category	Post-Implementation Cases (n=1127)	Pre-Implementation Controls (n=1124)	P Value
Overall Testing			
Number of Singleplex PCR Tests per CSF Specimen			<0.0001
0	969 (90%)	539 (50%)	
1	97 (9%)	289 (27%)	
>2	7 (1%)	256 (24%)	
CSF Singleplex PCR Positive	4 (0%)	72 (6%)	
MEP Testing Performed	805 (72%)	N/A	
MEP Positive	102 (9%)	N/A	
Time to Positive CSF Test Result (hrs)	4.8 (2.4, 4.8)	9.6 (4.8, 16.8)	<0.0001
Bacterial Testing			
Positive Gram Stain	16 (1%)	11 (1%)	0.4243
Positive CSF culture	20 (2%)	17 (2%)	0.7109
Positive MEP for bacteria	19/805 (2%)	N/A	
Viral Testing (positive/tested)			
Enterovirus detected in CSF by singleplex PCR	1/25 (4%)	63/348 (18%)	
Enterovirus detected in CSF by MEP	54/805 (7%)	N/A	
Enterovirus neurologic cases (any site)	88 (8%)	68 (6%)	0.1189
HSV detected in CSF by singleplex PCR	3/80 (4%)	3/442 (1%)	
HSV detected in CSF by MEP	3/805 (0%)	N/A	
HSV neurologic cases (any site)	15 (1%)	13 (1%)	0.8547
Parechovirus detected in CSF by singleplex PCR	0/4 (0%)	5/72 (0%)	
Parechovirus detected in CSF by MEP	11/805 (1%)	N/A	
Parechovirus neurologic cases (any site)	12 (1%)	5 (0%)	0.1456
HHV-6 detected in CSF by singleplex PCR	0/3 (0%)	1/8 (13%)	
HHV-6 detected in CSF by MEP	13/805 (2%)	N/A	
VZV detected in CSF by singleplex PCR	0/5 (0%)	0/41 (0%)	
VZV detected in CSF by MEP	2/805 (0%)	N/A	
CMV detected in CSF by singleplex PCR	0/2 (0%)	0/16 (0%)	
CMV detected in CSF by MEP	0/805 (0%)	N/A	
Fungal Testing (positives/tested)			
Cryptococcus Ag detected in CSF	0/2 (0%)	0/2 (0%)	
Cryptococcus detected in CSF by MEP	0/805 (0%)	N/A	
Discharge Diagnosis			
Neurologic Discharge Diagnosis	499 (44%)	430 (38%)	0.0017
Infectious Neurologic Disease Discharge Diagnosis	167 (15%)	115 (10%)	0.0298
Proportion due to Viral Cause	124 (74%)	86 (75%)	
Proportion due to Bacterial Cause	35 (21%)	27 (23%)	
Non-CNS Bacterial Infection	178 (16%)	175 (16%)	0.9149
Respiratory Viral Infection	284 (25%)	253 (23%)	0.1347
Outcomes			
Hospitalized	1033 (92%)	1024 (91%)	0.6929
Length of Inpatient Stay (median days)	4 (3, 9)	4 (3, 9)	0.9635
Death During Hospitalization	13 (1%)	21 (2%)	0.2264
Death Due to CNS Infection	7 (1%)	6 (1%)	1
Started on Antibacterials	768 (68%)	799 (71%)	0.1685
Number of IV Antibacterials Received	2 (2, 3)	2 (2, 3)	0.5328
IV Antimicrobial Duration (hrs)	24 (0, 50.4)	36 (0, 60)	0.0037
IV Antimicrobial Hours (hrs)	38 (0, 98)	60 (0, 118)	0.0007
Received IV Acyclovir	284 (25%)	301 (27%)	0.42
Duration of IV Acyclovir Amongst Those Started	17 (8, 40)	24 (8, 40)	0.206
Time to Effective Antimicrobials for Cases with Treatable Etiology (hrs)	0.75 (0.43, 1.02)	0.68 (0.48, 0.93)	0.471
Time to Effective Antimicrobials for Cases with Treatable Organism in CSF	0.65 (0, 1.58)	0.43 (0, 1.7)	0.5817
Time to Optimal Antimicrobial Regimen Initiation (hrs)	18 (13, 21)	28 (25, 32)	<0.0001

Conclusion. Implementation of MEP with a rapid CNS diagnostic stewardship program improved antimicrobial use with faster results shortening empiric therapy. Routine MEP testing in high-yield cases rapidly detects common viral causes and rules out bacterial targets to enable antimicrobial optimization

Disclosures. **Samuel R. Dominguez, MD, PhD**, **BioFire Diagnostics** (Consultant, Research Grant or Support)**DiaSorin Molecular** (Consultant)**Pfizer** (Grant/Research Support) **Samuel R. Dominguez, MD, PhD**, BioFire (Individual(s) Involved: Self): Consultant, Research Grant or Support; DiaSorin Molecular (Individual(s) Involved: Self): Consultant; Pfizer (Individual(s) Involved: Self): Grant/Research Support

1020. BioFiring on all Cylinders: Validation of BioFire FilmArray Pneumonia Panel and Determination of Optimal Utility

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Session: P-58. New Approaches to Diagnostics

Background. Respiratory cultures can take up to five days to grow, time that can be crucial in treating patients with serious infections. Newer rapid microbiological identification tests are designed to shorten this delay between specimen collection and test result. The BioFire® FilmArray® Pneumonia Panel is a multiplex PCR panel that can identify 8 viral, 18 bacterial, and 7 resistance gene targets in one hour. In this study, we aimed to calculate the predictive value of this test and its utility in the clinical setting.

Methods. This retrospective study compared BioFire® FilmArray® Pneumonia Panel results to respiratory cultures run at our center from 3/1/2020 to 2/28/2021. For every BioFire sample, a respiratory culture was run concurrently. We examined correlations between these two tests using data collected from the microbiology laboratory and the electronic medical record.

Results. 190 BioFire samples from 124 patients were submitted for processing. Of these, 148 samples had a concomitant respiratory culture result that grew organisms that BioFire could detect. BioFire and culture results were compared, and sensitivity and specificity were calculated on a per-sample basis. Sensitivity was calculated at 91%, specificity at 67%, positive predictive value at 46%, and negative predictive value at 96%.

BioFire detected 30 resistance genes total, including *mec*A/C and MREJ, CTX-M, and KPC. The sensitivity and negative predictive value for BioFire resistance gene detection was 100%. However, specificity was 94-98%, and the positive predictive value ranged between 25-41% when compared to culture.

Conclusion. Despite the promise of faster results and better screening, our data suggests that further study is needed to determine the utility of the BioFire pneumonia panel. The strength of the panel appears to lie in its negative predictive value and sensitivity, but as a positive predictive tool, it is suboptimal.

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

1021. Utility of Cell-Free DNA Sequencing in Diagnosing *Murine typhus* in Children

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Session: P-58. New Approaches to Diagnostics

Background. Murine typhus is a zoonotic infection caused by Rickettsia typhi and transmitted through infected fleas. Geographic distribution within the United States is limited primarily to South Texas and Southern California. Infection is typically associated with a triad of fever, headache, and rash, although is only present in one-third of cases. Immunofluorescence assay (IFA) is currently the gold standard for diagnosis, but it has its limitations as it is dependent on the time to seroconversion and has low specificity due to cross-reactivity among other rickettsial species. Cell-free DNA (cfDNA) sequencing for broad-range pathogen detection may offer higher sensitivity at the early stages of the disease.

Methods. We performed a retrospective electronic medical record search of children with cfDNA sequencing detection of Murine typhus hospitalized at Driscoll Children's Hospital, Corpus Christi, Texas, between June 2020 and May 2021.

Results. We found 4 children (range 9-15 year-old) positive for R. typhi by cfDNA sequencing. All patients presented with fever of unknown origin and rash. Also, 2 patients were diagnosed with pneumonia. One patient exhibited severe illness with acute kidney injury, elevation of transaminases and encephalitis that warranted admission to the pediatric intensive care unit. All patients defervesced and improved within 48 hours of doxycycline initiation; average length of stay 6 days (range 3-12 days). In one patient, M. typhus was detected by Karius® test only, in the other three was concordant with serology.

Conclusion. We highlight next-generation cfDNA sequencing as a useful tool in identifying the etiologic agent of patients with fever of known origin, where murine typhus is one of the possible etiologies. Preventing extensive laboratory workup and subsequent delay of assessment and management. The rapid turnaround time of cfDNA test allows for de-escalation of therapy and initiation of appropriate treatment.

Disclosures. **Jaime Fergie, MD**, **AstraZeneca** (Scientific Research Study Investigator)**Explify** (Speaker's Bureau)**Karius** (Speaker's Bureau)**Pfizer, Merck, AstraZeneca, and Sanofi** (Speaker's Bureau)**Pfizer, Merck, Sanofi, and Moderna** (Consultant, Advisor or Review Panel member)

1022. Evaluating the Impact of GenMark Dx ePlex® Blood Culture Identification (BCID) on Gram-negative Bloodstream Infections

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Session: P-58. New Approaches to Diagnostics

Background. The GenMark Dx ePlex BCID Gram-Negative (GN) panel utilizes electrowetting technology to detect the most common causes of GN bacteremia (21 targets) and 6 antimicrobial resistance (AMR) genes from positive blood culture (BC) bottles. Rapid detection of extended spectrum β-lactamases (ESBL: CTX-M & carbapenemases: KPC, NDM, IMP, VIM, OXA 23/48), and highly resistant bacteria such as *S. maltophilia* should enable early optimization of antimicrobial therapy.

Methods. In this prospective study, aliquots of positive BC bottles with GN bacteria detected on Gram stain (GS) (n=108) received standard of care (SOC) culture and antimicrobial susceptibility testing (AST). Additionally, samples were evaluated with the BCID-GN panel but only SOC results were reported in the EMR and available to inform clinical decisions. Chart reviews were performed to evaluate the impact of the BCID-GN panel on the time to organism identification, AST results, and optimization of antimicrobial therapy.

Results. A total of 108 patients are included in the analysis (Table 1). *Escherichia coli* was the most common bacteria identified followed by *Klebsiella pneumoniae, Pseudomonas aeruginosa,* and *Enterobacter* species (Table 2). There were 11 (10.2%) polymicrobial bacteremias. Repeat BCs were obtained in 68 (63%) patients of which 13 (19%) were persistently positive. Eight (7%) patients had evidence of additional gram-positive (GP) pathogens. Organism identification occurred 26.7 hours faster than culture. In conjunction with GS, negative pan-GP marker data could have helped providers make the decision to remove GP antibiotic coverage in 63 (58%) patients. Narrowing from empiric meropenem could have occurred in 5 patients. Of 10 individuals infected with resistant isolates (1 *S. maltophilia*, 1 OXA 23/48, and 8 CTX-M) empiric therapy was ineffective in 4 (40%) cases. Optimization of antimicrobial therapy for 9 (8.3%) patients could have occurred an average of 52.4 hours earlier than standard methods.