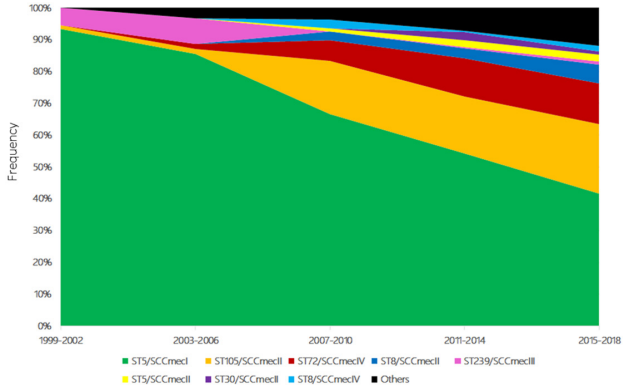
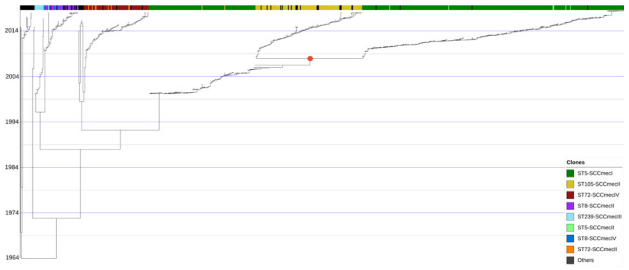
Figure 2. Relative frequency of MRSA clones from 1999 to 2018.



The genomes were grouped according to their isolation dates. Most frequent MRSA clones are represented by colored sections.

Figure 3. Maximum clade credibility tree from the molecular clock analysis of the 798 MRSA genomes.



A Bayesian molecular clock analysis was performed with BEAST using the isolation date of each genome as a calibrator. The colored strip showed the most frequent clones. The red dot shows a major event of divergence in 2008.

Conclusion. The ChC clone remains the most prevalent MRSA in Chile. However, our data is consistent with the evolution of this clone and a progressive replacement of with ST105 and ST72 genetic lineages.

Disclosures. **Lorena Diaz, PhD** , Nothing to disclose **Cesar A. Arias, M.D., MSc, Ph.D., FIDSA**, **Entasis Therapeutics** (Grant/Research Support)**MeMed Diagnostics** (Grant/Research Support)**Merk** (Grant/Research Support)

667. Next Generation Sequencing of Microbial Cell Free DNA in the Diagnosis and Treatment of Infectious Disease in Children: When Does the Result Justify the Cost?

Rachel Downey Quick, MSN, RN, CNS[1]; Kelli A. Martinez, MD[2]; Susan M. Russo, MSN, APRN, CPNP-PC[1]; Sarah E. McGwier, MSN, APRN, CPNP-PC[3]; Rachel A. Quirt, MSN, APRN, CPNP-PC[3]; Sarmistha Bhaduri. Hauger, MD[4]; Mariosl Fernandez, MD[4]; Donald Murphey, MD[5]; [1]Dell Children's Medical Group, Austin, TX; [2]Dell Medical School at the University of Texas at Austin, Austin, TX; [3]Dell Children's Medical Group, Autsin, TX; [4]Dell Children's Medical Center, University of Texas at Austin Dell Medical School, Austin, TX; [5]Dell Children's Medical Center; University of Texas at Austin Dell Medical School, Austin, TX

Session: P-30. Diagnostics: Typing/sequencing

Background. Pathogen testing using next-generation sequencing of microbial cell-free DNA (NGS cfDNA) is a promising diagnostic tool to identify pathogens that might not be detected using conventional lab evaluation. Considering the cost of this test, it is important to determine when it is most useful to the plan of care (POC).

Figure 1. Unit of admission among cases

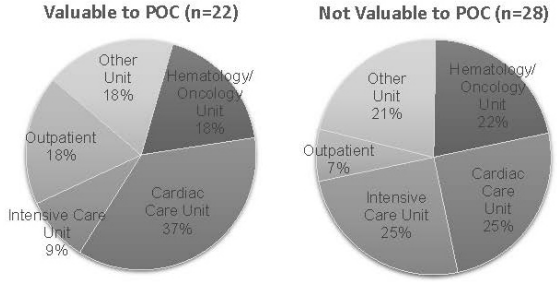
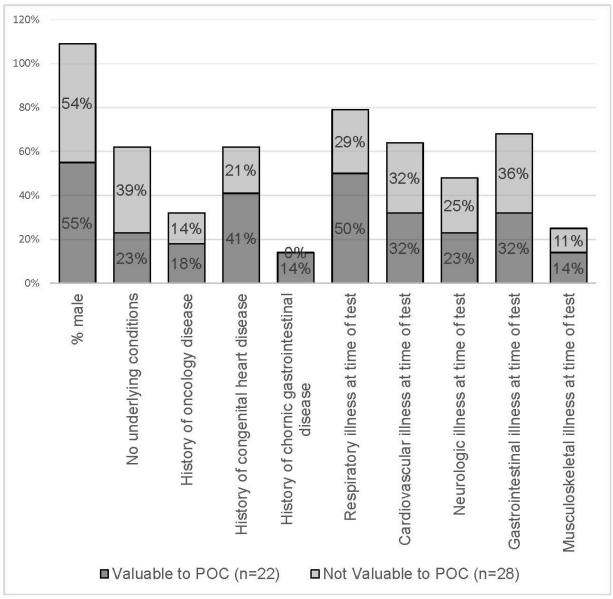


Figure 2. Patient characteristics in cases determined to be valuable and not valuable to the plan of care (POC)



Methods. In this retrospective study, we collected data from the medical charts of 50 consecutive NGS cfDNA tests in a free-standing children's hospital. We evaluated patients for demographics, underlying conditions, diagnosis at time of testing, conventional laboratory testing and timing, medical treatment, and NGS cfDNA test results for clinical relevance or false negative results compared to conventional testing. The primary goal was to identify patients for whom the NGS cfDNA testing affected the POC. Charts were reviewed, and determinations regarding whether the result influenced the POC were confirmed by a provider.

Results. We were unable to differentiate patients with clinically valuable NGS cfDNA results (Fig 1 & 2). Among those with NGS cfDNA results valuable to the POC (n=22), both negative and positive testing guided POC (13 valuable negative vs. 9 diagnostic cases). In the total sample, 5 cases (10%) had a clinically relevant pathogen identified through conventional testing, but not through NGS cfDNA and 2 cases had antimicrobial resistance on culture, which is not detected by NGS cfDNA.

Conclusion. While we did not find a specific clinical profile for NGS cfDNA use, positive results were essential to the diagnosis in 18% of cases with otherwise negative laboratory evaluation for the pathogen identified in NGS cfDNA. Negative tests affected the POC in 26% of cases by avoiding unnecessary antimicrobials in high risk immunocompromised patients and patients that presented with low-risk of infection, but unclear disease process.

Caution must be exercised with reliance on this test with respect to antimicrobial resistance and risk of false negative results.

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

668. Restricting Ordering of Multiplex Gastrointestinal Panel Improves Test Utilization

Don Bambino Geno Tai, MD, MBA[1]; Anisha Misra, PhD[1]; Priya Sampathkumar, MD[1]; John C. O'Horo, Sr., MD, MPH[1]; [1]Mayo Clinic, Rochester, Minnesota

Session: P-30. Diagnostics: Typing/sequencing

Background. The multiplex gastrointestinal pathogen panel (GIP) is a convenient and quick diagnostic test for determining the infectious etiology of diarrhea. It identifies several of the most common pathogens associated with gastroenteritis. However, it is expensive, and test results may not impact care, given that several of the pathogens in the panel are managed expectantly. We describe our experience with a diagnostic stewardship initiative to resolve the overuse of this testing method.

Methods. We performed a pre/post study of GIPs ordered for inpatients 18 years old and older from December 19, 2018, to December 18, 2020, at Mayo Clinic hospital in Rochester, Minnesota. GIP orders for inpatients were limited to the first 72 hours of hospitalization starting December 19, 2019. Orders after 72 hours were encouraged to be changed to *Clostridioides difficile* NAAT testing or sent to an infectious disease provider to override on a case-by-case basis. Our hospitals used BioFire® FilmArray® Gastrointestinal Panel (BioFire Diagnostics, Salt Lake City, Utah).

Results. A total of 2,641 GIPs were performed during the study period. There were 1,568 GIPs (3.3/100 hospitalizations) in the pre-intervention period compared to 1,073 (2.6/100 hospitalizations) post-intervention, representing a drop of 21.2%. The most common pathogen detected was *C. difficile* (toxin A/B) (48.8%, n=402), followed by norovirus (17.5%, n=144). The overall test positivity rate was 27.9% (n=736). The test positivity rate decreased 1.8% from 28.6% (n=448) to 26.8% (n=288) after the restriction (p=0.33). The proportion of *C. difficile* among all pathogens detected increased from 48.5% to 49.7% (*p*=0.67).

Table 1. Pre- and Post-Intervention Test Positivity Rate of Specific Pathogens in GIP

Pathogen	Pre-intervention n (%)	Post-intervention n (%)	% Change	*p*-value
Bacteria				
Campylobacter (jejuni, coli, and upsaliensis)	13 (0.83)	13 (1.21)	0.38	0.32
Clostridioides difficile (toxin A/B)	238 (15.18)	164 (15.28)	0.11	0.94
Enteroaggregative *Escherichia coli*	16 (1.02)	5 (0.47)	-0.55	0.12
Enteropathogenic *E. coli*	59 (3.76)	32 (2.98)	-0.78	0.28
Enterotoxigenic *E. coli*	8 (0.51)	3 (0.28)	-0.23	0.37
Shiga-like toxin-producing *E. coli*	10 (0.64)	6 (0.56)	-0.08	0.8
Shigella/Enteroinvasive *E. coli*	6 (0.38)	-	-0.38	-
Plesiomonas shigelloides	-	2 (0.19)	0.12	-
Salmonella	10 (0.64)	5 (0.47)	-0.17	0.56
Yersinia enterocolitica	4 (0.26)	5 (0.47)	0.21	0.36
Vibrio species	1 (0.06)	-	-0.06	-
Viruses				
Adenovirus F40/41	2 (0.13)	1 (0.09)	-0.03	0.8
Astrovirus	4 (0.26)	3 (0.28)	0.02	0.9
Norovirus GI/GII	74 (4.72)	70 (6.52)	1.8	**0.04**
Rotavirus A	21 (1.34)	-	-1.34	-
Sapovirus (I, II, IV, and V)	10 (0.64)	9 (0.84)	0.2	0.55
Parasites				
Cryptosporidium	6 (0.38)	7 (0.65)	0.27	0.33
Cyclospora cayetanensis	4 (0.26)	2 (0.19)	-0.07	0.72
Entamoeba histolytica	-	-		
Giardia lamblia	4 (0.26)	3 (0.28)	0.02	0.9

Conclusion. Our study showed that restricting the ordering of GIP to the first 72 hours of hospitalization and directing providers to standalone *C. difficile* NAAT testing resulted in a reduction of GIPs performed. There were marginal changes in the test positivity rate of GIP. A limitation of our study is that the timing of post-intervention coincided with the COVID-19 pandemic, which had unpredictable effects on hospital practice and patient admissions. Ideally, future quality improvement projects should increase the test positivity of pathogens other than *C. difficile* while lowering the GIP use in diagnosing *C. difficile* colitis.

Disclosures. **John C. O'Horo, Sr., MD, MPH**, **Bates College and Elsevier Inc** (Consultant)

669. Metagenomic Plasma Microbial Cell Free DNA-Sequencing Assists in Diagnosis of Infections and Critical Antimicrobial Changes in Immunocompromised Hosts

Nicole C. Vissichelli, MD[1]; Megan M. Morales, MD[2]; Bharadhwaj Kolipakkam, MBBS[1]; Alexandra L. Bryson, Ph.D., D(ABMM)[1]; Daniel Nixon, DO, PhD[1]; Roy T. Sabo, PHD[3]; Amir A. Toor, MD[1]; [1]Virginia Commonwealth University Health System, Midlothian, Virginia; [2]Virginia Commonwealth University Health, Richmond, Virginia; [3]Virginia Commonwealth University, Richmond, Virginia

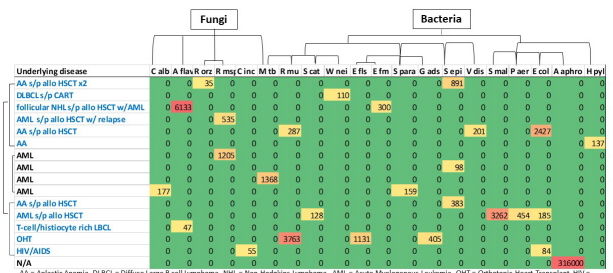
Session: P-30. Diagnostics: Typing/sequencing

Background. Metagenomic next-generation sequencing of plasma cell-free DNA (Karius®) (plasma mcf-DNA-seq) is a noninvasive approach that may have a unique role for the diagnosis of infectious complications in immunocompromised patients. The rapid turnaround time and noninvasive nature makes this a promising supplement to standard of care.

Methods. The aim of this study is to investigate the utility of plasma-mcf-DNA-seq in clinical practice; how it changes management, correlations between organism abundance over time from symptom onset and the value of negative tests. Retrospective review of plasma-mcf-DNA-seq performed, January 2020 -March 2021. Organism abundance was displayed as a heat map and graphed over time from initiation of antimicrobials. Management changes and concordance with standard of care results were compared for positive and negative tests. This study was approved by the Virginia Commonwealth University Institutional Review Board.

Results. Thirty-six adult patients included: 92% immunosuppressed (11 with T cell deficits (solid organ transplant, malignancy, human immunodeficiency virus), 8 with B-cell deficits (hematologic malignancy, diabetes mellitus), and 14 with both (hematopoietic stem cell transplant, aplastic anemia)). Most tests evaluated fever (67%) and/or pneumonia (72%). Patients received a median 7 days of antimicrobials prior to testing. Twenty-one (58%) tests detected 1-5 organisms (14/21 bacteria, 8/21 fungi, and 6/21 viruses). A positive test prompted therapy changes in 14/21 patients. Of the bacterial species identified, 8/20 were considered clinically pathogenic, 3 prompted targeted treatment; 7/8 fungi identified were clinically pathogenic and resulted in antifungal therapy changes to target the species identified. Antimicrobials were de-escalated in 3 patients with negative tests. There was an exponential relationship between the abundance of pathogenic fungi over time from symptom onset, but no such relationship was seen with bacteria.

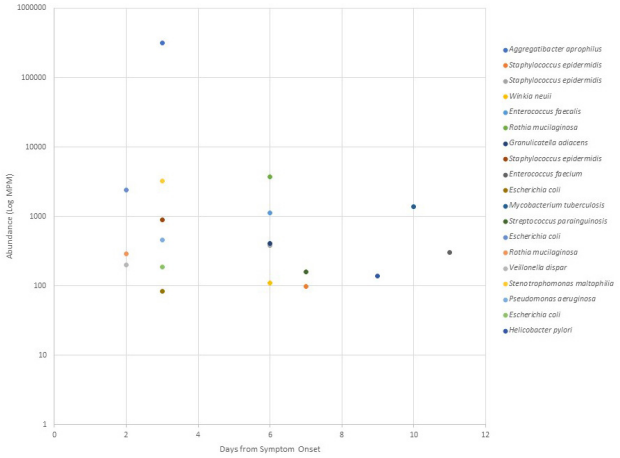
Abundance of fungi and bacteria detected on plasma mcf-DNA-seq test



AA = Aplastic Anemia, DLBCL = Diffuse Large B cell lymphoma, NHL = Non-Hodgkins Lymphoma, AML = Acute Myelogenous Leukemia, OHT = Orthotopic Heart Transplant, HIV = Human Immunodeficiency Virus, AIDS = Acquired Immunodeficiency Syndrome, LBCL = Large B Cell Lymphoma, C alb = *Candida albicans*, A flav = *Aspergillus flavus*, R orz = *Rhizopus oryzae*, R msp = *Rhizopus microspores*, C inc = *Candidabolus incongruous*, M tb = *Mycobacterium tuberculosis*, R mu = *Rothia mucilaginosa*, S cat = *Streptomyces cattleya*, W nei = *Winkia neuii*, E fs= *Enterococcus faecalis*, E fm = *Enterococcus faecium*, S par = *Streptococcus parasanguinosis*, G ads = *Granulicatella adiacens*, S epi = *Staphylococcus epidermidis*, V dis = *Veillonella dispar*, S mal = *Stenotrophomonas maltophilia*, P aer = *Pseudomonas aeruginosa*, E col = *Escherichia coli*, A aphro = *Aggregatibacter aprophilus*, H pyl = *Helicobacter pylori*

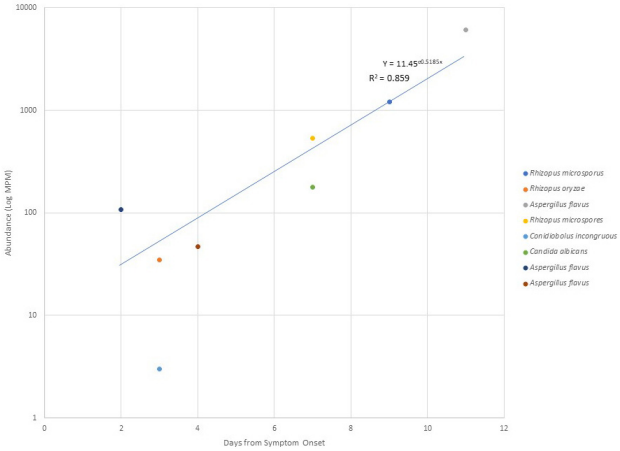
Abundance of bacteria and fungi detected on plasma mcf-DNA-seq test. Data classified by organism and level of immunosuppression. Abundance is expressed in microbial cell free DNA per microliter. Warmer colors towards red represent higher abundance.

Figure 1. Bacteria abundance from date of symptom onset.



There was no clear trend in bacterial abundance over time from symptom onset. Most bacteria detected were not considered clinically pathogenic.

Figure 2. Fungi abundance from date of symptom onset



There was an increasing trend in the abundance of fungi detected from time of symptom onset. Seven of the 8 fungi detected were considered clinically pathogenic.

Conclusion. Plasma-mcf-DNA assisted in making critical management changes including initiation of treatment for identified organisms and de-escalation of antimicrobials. Plasma-mcf-DNA is a promising approach for a non-invasive rapid diagnosis.

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

670. Precision Metagenomic (PM) Sequencing Outperforms Conventional Urine Culture in Detecting Clinically Relevant Microorganisms

Rita C. Stinnett, PhD, MHS[1]; Bethany Kent, PhD[2]; Marta Mangifesta, PhD[1]; Anagha Kadam, PhD[1]; Heng Xie, PhD[1]; Stacie Stauffer, BS[1]; Jamie Lemon, PhD, D(ABMM)[1]; Benjamin Briggs, MD, PhD[1]; Lauge Farnaes, MD, PhD[1]; Robert Schlaberg, MD, MPH[1]; [1]IDbyDNA, Salt Lake City, Utah; [2]PathGroup Labs, Nashville, Tennessee

Session: P-30. Diagnostics: Typing/sequencing

Background. Morbidity from urinary tract infection (UTI) is high. Urine culture is the reference method for UTI diagnosis. Its diagnostic yield is limited as prior antibiotic use prevents growth of established uropathogens, many emerging uropathogens do not grow under routine culture conditions, and results interpretation can be subjective. Faster, more comprehensive diagnostics could help manage recurrent and/or drug-resistant infections. We evaluated the diagnostic yield of a precision metagenomic (PM) workflow for pathogen detection & antimicrobial resistance (AMR) characterization directly from urine.

Methods. Residual urine samples from symptomatic adults evaluated by culture & susceptibility were identified by a combination of consecutive & stratified random sampling (n=480; 79% culture positive). DNA was extracted with modifications to the Quick-DNA Urine Kit (Zymo). Libraries were generated with Illumina DNA Prep