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

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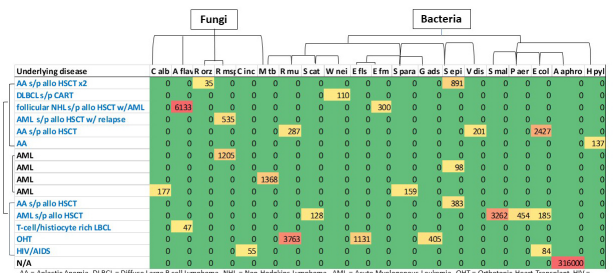
Session: P-30. Diagnostics: Typing/sequencing

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.

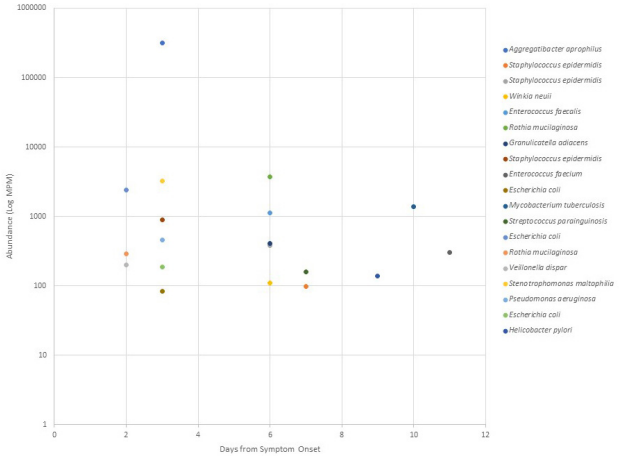
Abundance of fungi and bacteria detected on plasma mcf-DNA-seq test



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*

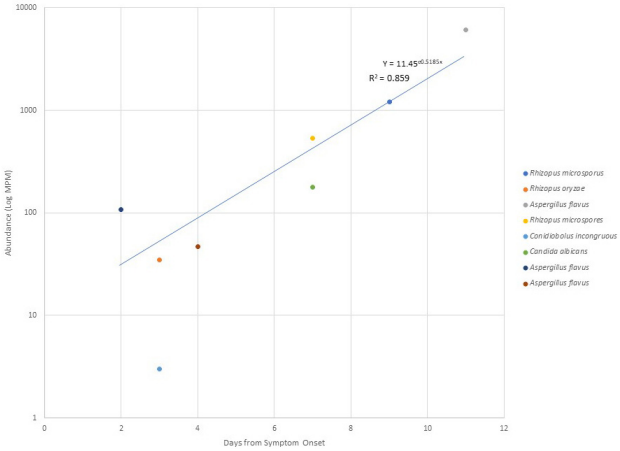
Abundance of bacteria and fungi detected on plasma mcf-DNA-seq test. Data classified by organism and level of immunosuppression. Abundance is expressed in microbial cell free DNA per microliter. Warmer colors towards red represent higher abundance.

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

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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

with Enrichment for clinically relevant targets (191 pathogens, 1976 AMR markers) with the Explify Urinary ID/AMR Panel (UPIP, IDbyDNA). Enriched libraries were sequenced on the NextSeq550 (Illumina) and data analyzed with the Explify UPIP Data Analysis Solution (IDbyDNA).

Results. For bacterial uropathogens, 94% positive agreement was observed between this PM workflow and culture. PM detected fastidious and/or anaerobic potential uropathogens in 30% and 7% of samples reported as culture-negative or positive for other bacteria, respectively. Total agreement between AMR marker detection and phenotypic resistance was 78%. Notably, PM predicted phenotypes of ESBL *E. coli* and *K. pneumoniae* (10/10), MRSA (9/9), and vancomycin-resistant *E. faecium* (4/5). PM also detected pathogens associated with sexually-transmitted infection (*C. trachomatis*, HSV) and bacterial vaginosis (*G. vaginalis*). PM produced complete results within 24-36 hours of sample receipt (vs culture & susceptibility: 42-72 hrs).

Conclusion. The sensitivity of PM for uropathogen detection was noninferior to culture (Δ = 0.05; Nam RMLE; $p < 0.0005$). PM predicted antimicrobial resistance phenotypes for common uropathogens and identified potential pathogens not detected by conventional culture. Future studies should assess the impact of PM-guided management on clinical outcomes.

Disclosures. **Rita C. Stinnett, PhD, MHS**, **IDbyDNA** (Employee) **Marta Mangifesta, PhD**, **IDbyDNA** (Employee) **Anagha Kadam, PhD**, **IDbyDNA** (Employee) **Heng Xie, PhD**, **IDbyDNA** (Employee) **Stacie Stauffer, BS**, **IDbyDNA** (Employee) **Jamie Lemon, PhD, D(ABMM)**, **IDbyDNA** (Employee) **Benjamin Briggs, MD, PhD**, **IDbyDNA** (Employee) **Lauge Farnaes, MD, PhD**, **Cardea Bio** (Advisor or Review Panel member)**IDbyDNA** (Employee) **Robert Schlaberg, MD, MPH**, **IDbyDNA** (Consultant, Shareholder, Co-founder)

671. Initial Impression of Explify Respiratory Testing: Is It an Effective Diagnostic Tool?

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Session: P-30. Diagnostics: Typing/sequencing

Background. Respiratory infections are a common cause of hospital admissions resulting in significant morbidity and mortality. Isolating specific pathogens from the respiratory tract is a diagnostic challenge. Traditional testing modalities are prone to contamination, time consuming, and have low sensitivity. Next generation genetic sequencing technology has made possible the development of a number of hypothesis free, fast, and highly accurate genome-based identification tests. In this study, we aim at assessing the initial use and performance of one of these tests, the Explify Respiratory panel, at a large quaternary hospital in west Michigan.

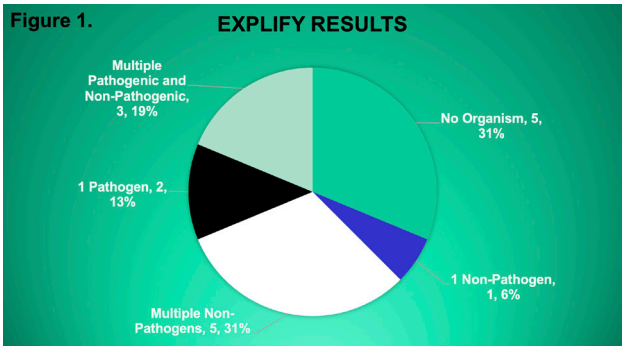
Table 1. Explify Respiratory Testing
Next Generation Sequence Testing
Capable of Identifying > 900 pathogens (bacteria, fungi, virus, parasites)
Sample type: Sputum, NP swab, BAL (PREFERRED)
Turn around time: 5 days (including shipping time)
Cost: $470 (compared to $1284.18 for Universal PCR Testing)
Concerns
False positives due to contaminants or host DNA
False positives due to identification of nonpathogenic organisms

Methods. We performed retrospective analysis on 16 patients with suspected lower respiratory infections. Subjects were chosen for inclusion in the analysis based on the suspicion of pulmonary infection without an identified pathogen. The patient population included 5 immunocompromised patients, 3 with hematologic malignancy, 4 with solid tumor malignancy, and 2 transplant recipients.

Table 2. Patient Characteristics
9 male, 7 female
Ages 40 - 77
5 immunocompromised patients
3 with known hematologic malignancy
4 with history of solid organ malignancy
1 solid organ transplant recipient (heart)
1 hematopoietic stem cell transplant recipient

Results. The test resulted in: lack of identified organism (5 patients), identification of non-pathogenic organisms (6 patients), and identification of organisms that were either identified by other traditional testing or did not impact provider's therapeutic plan (5 patients). The results of Explify testing in all 16 patients did not have a clinical impact on patient care or treatment plan.

Figure 1.



Conclusion. Explify testing seemed to be an appealing cost-effective tool that could replace other available testing modalities such as culture, other sequencing tests, and serological testing with faster turn-around time and less cost. However, it failed to demonstrate any benefit to clinicians in identifying respiratory pathogens while resulting in added cost burden to the patient. Moreover, it resulted in clinical delays of further investigation while awaiting the results. It remains unclear if the lack of clinical impact results from the extensive interventions and treatments that patients receive prior to Explify testing or from the poor sensitivity and performance of the test.This study emphasizes the importance of continuous evaluation of new diagnostic testing before widespread implementation to improve patient care and minimize cost burden.

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

672. Diagnosis of Rocky Mountain Spotted Fever Using Plasma Metagenomic Next-Generation Sequencing

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Session: P-30. Diagnostics: Typing/sequencing

Background. Rocky mountain spotted fever (RMSF), caused by *Rickettsia rickettsii*, incurs significant morbidity and mortality, especially in children. Early in the course of illness, standard diagnostic tests are of limited sensitivity, and diagnosis is often based on clinical symptoms and local epidemiology. The diagnosis can be missed in areas where RMSF is not endemic, and a delay in initiation of therapy may lead to poor clinical outcomes. Plasma metagenomic next-generation sequencing (mNGS), with turnaround times approaching 48 hours, may be a useful adjunctive tool in the diagnosis of RMSF.

Methods. We describe four children hospitalized with RMSF between January 1, 2017 to May 15, 2021 at a tertiary children's hospital in southern California. All had plasma mNGS and rickettsial serologic testing as part of clinical care.

Results. mNGS detected *Rickettsia rickettsii* in all 4 patients. Only 2 subjects had positive serologic testing initially and required repeat testing in the convalescent stage to confirm RMSF. The mean turnaround time for mNGS was 2.75 days, which was comparable to serologic testing. Antibiotic therapy was changed in three subjects as a result of the plasma mNGS result.

Conclusion. Plasma mNGS may be a useful diagnostic modality early in the disease course of RMSF.

Disclosures. **Lauge Farnaes, MD, PhD**, **Cardea Bio** (Advisor or Review Panel member)**IDbyDNA** (Employee)

673. Unbiased Microbial Cell-free DNA Next-Generation Sequencing Resolves HHV6 Variant Specificity and Demonstrates a Predominance of HHV6B over HHV6A in Real-World Clinical Experience

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Session: P-30. Diagnostics: Typing/sequencing

Background. *Human herpesvirus 6* (HHV6) has been classified in two distinct variants, HHV6A and HHV6B. Although distinct epidemiology, disease association, biological and immunological properties, their genomes are 90% homologous. Less is known about HHV6A which is typically asymptomatic but can cause severe infection in patients with neurological disorders or HIV. HHV6B infection occurs during childhood, but can reactivate after solid organ and stem cell transplantation. Antibody assays may indicate previous, recent or current infection. Quantitative PCR cannot differentiate active from latent infections and some available qualitative PCR assays are unable to differentiate the two variants. A single open-ended test through plasma-based microbial cell-free DNA (mcfDNA) metagenomic next-generation sequencing (NGS) may overcome these limitations.