

No cross reactivity was observed with any of the malarial species tested. *Babesia* MO1, *Babesia duncani* and all bacterial isolates tested were negative by the BMPCR. Intra-run, inter-run and day to day reproducibility of the assay was 100%.

Conclusion. The *B. microti* real time PCR assay developed by Northwell Health Laboratories is rapid, sensitive, specific and reproducible. With the sample to result turnaround time of 2.5 hours and hands on time of only 5 minutes per sample, BMPCR can be used as screening assay for *B. microti* in clinical laboratories.

Disclosures. All authors: No reported disclosures.

2088. A Novel Diagnostic Method for Malaria Using Loop Mediated Isothermal Amplification (LAMP) and MinION Nanopore Sequencer

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Background. Simply and accurately diagnostic tool for Malaria is required for clinical diagnosis and epidemiological survey. We have developed a novel diagnostic tool for Malaria using loop mediated isothermal amplification (LAMP) with MinION nanopore sequencer.

Methods. In this study, we have designed human *Plasmodium* parasites-specific LAMP primers targeting for the lesion of 18S rDNA gene, which were locating on the conserved sequences across all five *Plasmodium* species; *Plasmodium falciparum*, *P. vivax*, *P. ovale* (*P. wallikeri* and *P. crutisi*), *P. knowlesi* and *P. malariae*, containing each species-specific sequence within F1-B1 primer pairs. The sensitivities were evaluated using 10-fold serially diluted plasmids harboring the sequences of 18S rDNA. We also applied our protocol to human blood samples collected and stored with FTA elute cards derived from 30 Malaria patients, who are clinically diagnosed as Malaria in Indonesia. Its analytical sensitivities and specificities were also evaluated while comparing the results of previously described nested PCR methods. Finally, we performed amplicon sequencing of our LAMP methods using MinION nanopore sequencer to identify each *Plasmodium* species.

Results. Our LAMP method could amplify all targeting 18S rDNA gene on constructed plasmids and its detection limits were 10 - 100 copies/reaction respectively. In clinical samples, obtained LAMP results were completely consistent with the results of nested PCR. Additionally, identifications of *Plasmodium* species based on the sequence analysis with MinION were also consistent with the sequence of each constructed plasmid and could consistently confirmed its *Plasmodium* species with the highest homology of reference *Plasmodium* parasite sequence.

Conclusion. Our innovative diagnostic technology with LAMP and MinION could become a powerful tool for identification of *Plasmodium* parasites even in resource-limited situation.

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2089. Accelerating Time to Pathogen-adapted Antibiotic Treatment through Culture-independent Antimicrobial Susceptibility Testing in Patients Suffering from Sepsis

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Background. Accurate and fast pathogen identification and consecutive antimicrobial susceptibility testing (AST) is of vital importance for patient outcome in patients suffering from sepsis.

Methods. The Accelerate Pheno™ system is a new, fully automated, culture-independent diagnostic method for both pathogen identification (ID) and antimicrobial susceptibility testing (AST). We analyzed positive blood cultures from critically ill patients with new onset of sepsis according to the new sepsis guidelines, using both conventional standard methods (VITEK, MALDI-TOF) and Accelerate Pheno™ system. ID/AST results of the Accelerate Pheno™ system were not reported to treating physicians as part of our internal evaluation process.

Results. Accelerate Pheno™ system correctly detected 74 pathogens [Gram-negative (GN) (*n* = 27), Gram-positive (GP) (*n* = 47)] straight out of 84 positive blood culture bottles. Gram-negative (GN) pathogens were identified as *E. coli* (*n* = 15; concordance rate 100%), *K. pneumoniae* (*n* = 7; 71,4%), *S. marcescens* (*n* = 3; 100%), *E. cloacae* (*n* = 2; 50%), *P. mirabilis* (*n* = 1; 100%) and *P. aeruginosa* (*n* = 1; 33%). Gram-positive pathogens were identified as CNS (*n* = 24; 82,6%), *S. aureus* (*n* = 15; 88,2%), *E. faecium* (*n* = 6; 100%) and *E. faecalis* (*n* = 2; 100%). The Accelerate Pheno™ system generated a GN-AST result in 70,4% (19 of 27 samples) and a GP-AST result in 61,7% (29 of 47 samples) when compared with routine AST. Growth control, analysis

and mechanical failure led to reduced results in comparison to conventional ID/AST. Accelerate Pheno™ delivered correct MIC-results for most of the panel antibiotics [e.g., meropenem: 83,3%, gentamicin: 88,9%, ertapenem: 100%].

Conclusion. The use of the Accelerate Pheno™ system significantly improved time-to-ID/AST and would have led to a reduced time-to-treatment in patients suffering from sepsis if results would have been reported. The system currently has some weakness in the detection of polymicrobial and streptococcal infections but due to the short hands-on-time, culture-independence and fast generation of results, it represents a promising new diagnostic method for the consecutive antibiotic treatment of septic patients.

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2090. T-Cell Immunity Panel Measures CMV-Specific CD4 and CD8 T-Cell Responses

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Background. Infection and disease from human cytomegalovirus (CMV) is a major complicating factor for both solid organ and hematopoietic stem cell transplant recipients. Antiviral therapy is often used to control CMV infections, but presents problems of toxicity, antiviral resistance and excessive costs. Currently, treating physicians are limited in the information and data available to assess a patient's ability to control a potential CMV infection post-transplant. Recent studies have shown that measuring a patient's CMV specific T cell mediated immunity may provide valuable information to physicians for monitoring CMV infection/disease in transplant patients and may aid in determining which patients need antiviral therapy.

Methods. For this purpose, a flow cytometry assay was developed to determine the percentages of CD4+ and CD8+ T cells that respond to stimulation with CMV antigen. Assessment of CMV specific response is based upon the cellular activation surface marker CD69 in conjunction with IFN γ , TNF α , and IL-2 cytokine production. Three CMV antigens were used to assess patient immunity; a whole viral lysate, a peptide pool of pp65, and a peptide pool of IE-1.

Results. Our data indicate that CD8 T cells respond primarily to the pp65 and/or IE-1 peptide pools while the CD4 T cells respond primarily to the viral lysate. Detection of both CD4 and CD8 responding populations at levels above background, $\geq 0.2\%$ of the parent population, indicates that a patient's immune system has previously been exposed to CMV. Validation of 23 CMV seropositive samples demonstrated immune responses for all 23 samples above 0.2% for at least one of the three intra-cellular cytokines and at least one of the three CMV antigens. Validation of five CMV seronegative samples demonstrated immune responses below 0.2% (when excluding underlying, unrelated immune responses). Included for each sample is a positive (Staphylococcal Enterotoxin type B) control to assess patient's overall ability to mount an immune response and negative (media) control to capture the presence of an underlying immune response.

Conclusion. This assay evaluates a patient's pre-existing CMV specific T cell immunity and their global T cell function.

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2091. Application of Laser Light Scattering Technology in Rapid Diagnosis of Urinary Tract Infections and Antimicrobial Susceptibility Testing in a Tertiary Children's Hospital

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Background. Timely and accurate microbiology testing is crucial in the diagnosis and management of urinary tract infections (UTIs). The ability to rapidly screen for potential UTIs can lead to early rule out and judicious use of antimicrobial therapy. This study examines the application of laser scattering for bacterial detection and antimicrobial susceptibility testing (AST) directly from urine.

Methods. Residual urine samples collected for routine culture were tested using the BacterioScan™ 216Dx™ UTI System and 216R AST System. Continuous collection of light refraction patterns generated growth curve that was used to determine whether the sample was likely positive or negative for bacteria. Further curve analysis ruled out mixed flora at lower concentrations, and "qualified" samples were identified directly on MALDI-TOF MS. AST for ampicillin, cefazolin, ceftriaxone and ciprofloxacin was performed concurrently on the instrument. Samples were incubated for up to 16 hours with results available as early as 2 hours.