248. Thirty-Day Mortality Among Patients with Candidemia Diagnosed by T2Candida Assay Alone: Influence of Risk Factors and Candida Species
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Background. Candidemia is a common cause of healthcare-associated bloodstream infection with high mortality rates despite antifungal therapy. Risk factors include prolonged ICU stay, immunosuppression, and exposure to broad-spectrum antibiotics. Blood cultures (BC) remain the gold standard for diagnosis, but lack sensitivity and can take days to result. T2Candida (T2C) is a rapid diagnostic test utilizing PCR and magnetic resonance technology to detect five Candida species in whole blood in less than 6 hours. In this study we examined characteristics of patients with positive T2C assays in the absence of positive BC including risk factors and 30-day mortality rates.

Methods. We conducted a retrospective analysis of positive T2C cases at UAB Medical Center from 2016 to 2018 with either negative or no BC. For each patient we determined if clinical signs (e.g., hypotension, leukocytosis) and risk factors for candidemia were present at the time of collection. Our primary outcome of interest was 30-day mortality. Data were compared by multivariate analysis.

Results. A total of 173 patients with T2C positivity alone were included in the analysis. The most common risk factor was the use of broad-spectrum antibiotics followed by CVC (Table 1). The mean number of risk factors per patient was 3.6 (Figure 1). Overall 30-day mortality was 41%. Patients with a T2C result of C. albicans/C. tropicalis were almost 2.5 times more likely to die at 30 days (aOR 2.401, CI 1.159–4.974) compared with those with other positive results. Increasing number of risk factors (aOR 1.457, CI 1.126–1.886) and increasing age (aOR 1.052, CI 1.026–1.079) were significantly associated with increased odds of death at 30 days (Table 2).

Conclusion. In this study we demonstrate a significant association between increasing number of risk factors, older age, and A/T result with higher odds of 30-day mortality among patients with T2C positivity alone. While concern for false-positives exists when using T2C, our data suggest that this is an acutely ill population which warrants early and aggressive antifungal therapy. The lower limit of detection of T2C (1 cfu/mL) as compared with BC may explain lack of paired positive cultures in these patients despite clinical signs of and risk factors for candidemia.

Table 1. Demographics and risk factors

| Age (years) | Mean (SD) | Median (QR) |
|-------------|-----------|-------------|
| Male | 45.1 (15.8) | 50 (45–60) |
| Female | 54.3 (15.0) | 58 (45–68) |

| Race or ethnic group | n (%) |
|---------------------|-------|
| White | 110 (61.6) |
| Black | 66 (35.8) |
| Hispanic | 4 (0.5) |
| Other | 3 (1.6) |

| Location type | n (%) |
|---------------|-------|
| ICU | 112 (65) |
| Floor | 61 (35) |
| T2C result | n (%) |
| C. albicans/C. tropicalis | 74 (42.8) |
| C. parapsilosis | 47 (26.7) |
| C. krusei/C. glabrata | 17 (9.8) |
| Other | 35 (19.7) |

| Clinical signs | n (%) |
|---------------|-------|
| Fever/Sepsis | 112 (65) |
| Leukocytosis | 35 (19.7) |
| Hypotension | 79 (43) |
| Risk factors | n (%) |
| Broad-spectrum antibiotics | 109 (60.9) |
| Central venous catheter | 39 (21.8) |
| ICU stay | 73 (40.6) |
| Mechanical ventilation | 48 (27) |
| Steroids/immunosuppression | 43 (23.7) |
| Diabetes | 47 (27) |
| Intra-abdominal surgery | 90 (50) |
| Total parenteral nutrition | 90 (50) |
| Necrotizing pancreatitis | 20 (11) |

249. Limited Diagnostic Utility of Extended Aerobic Blood Culture Incubation for Fungal Pathogen Detection
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Background. Blood cultures are an important diagnostic tool for the detection of fungemia. At our institution, fungal blood cultures consist of aerobic blood culture with incubation extended from the standard 5 days to 14 days. Orders for fungal blood cultures exist in multiple electronic order sets for selected populations, including oncology and bone marrow transplant services.

Methods. To determine the yield of fungal blood cultures at our institution, a 570-bed tertiary-care referral center, we extracted all fungal blood culture results over a 4.5-year period (January 1, 2014-May 15, 2018) from a Laboratory Information System.

Results. Of the 21,657 fungal blood cultures performed, only 202 (0.9%) demonstrated growth and 189 (0.9%) grew fungal organisms. The majority (90%, n = 182/202) of positive fungal blood cultures grew a Candida or other yeast species. 96% (n = 174/182) of the fungal cultures that grew yeast would have been detected with standard bacterial blood culture. Eight of these cultures became positive during the extended hold period and grew a Candida species. All 8 cultures were collected from patients who had previous positive cultures for the same Candida species detected by standard incubation. Five fungal blood cultures from 4 patients turned positive after 5 days of incubation. Among these, two additional fungal pathogens were identified including 2 cases of Lomentospora prolificans and 2 cases of Fusarium. In both cases of L. prolificans and one case of Fusarium, the patients had previous positive blood cultures that detected the same organism with standard incubation. One patient with Fusarium had no previous positive blood cultures, but had multiple tissue cultures positive for Fusarium. The remaining cultures that turned positive after 5 days of incubation contained bacterial organisms, a number of which were considered clinically insignificant (e.g., Cutibacterium species).

Conclusion. These data suggest that extended incubation of aerobic blood culture bottles has limited diagnostic utility beyond standard bacterial blood culture for detection of fungemia. Fungal blood cultures represent an opportunity for improved diagnostic test stewardship, and use should be restricted to selected situations in consultation with Infectious Diseases or Laboratory Medicine.

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Table 2. Multivariate analysis of factors associated with 30-day mortality

| Variable | OR (95% CI) | P-value |
|----------|-------------|---------|
| Sum of risk factors | 1.457 (1.126-1.886) | 0.004 |
| Age | 1.052 (1.026-1.079) | <0.001 |
| A/T positive | 2.401 (1.159-4.974) | 0.018 |
| Gender | 0.920 (0.447-1.895) | 0.822 |

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250. Comparison of T2Candida Assay with Blood Culture, Candida Sepsis Score and Serum β-D-glucan in Diagnosis of Candidemia
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Background. Although blood cultures are the clinical diagnostic standard for candidemia, their delay in results and low sensitivity has led to increasing the use of alternate tests and diagnostic algorithms. The T2Candida magnetic resonance assay
251. Implementation of the Sōna Coccidioides Antibody Lateral Flow Assay in the Clinical Laboratory Proves to Reduce Cost and Decrease Turnaround Time When Compared with Send out Immunodiffusion and Complement Fixation Testing

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Background. Coccidioidomycosis (Valley fever) is an airborne, invasive fungal infection endemic to Arizona, California, Mexico, and Central and South America. The dominant method of diagnosis is serology, which includes complement fixation (CF), immunodiffusion (ID), and complement fixation assay (EIA). These serological assays require highly trained personnel and are time consuming, with turnaround times (TAT) that range anywhere from 5 days to 2 weeks. Costs of send outs and long TAT, Valley fever presents a diagnostic challenge to physicians and laboratories. IMMY developed the sōna Coccidioides Antibody Lateral Flow Assay (LFA), a rapid and simple diagnostic assay that detects anti-Coccidioides antibodies in patient serum within 30 minutes.

Methods. We tested the sōna Coccidioides antibody LFA using 315 patient specimens and compared cost-analysis and TAT to a send-out reference labs ID and CF assays.

Results. In this study, we found that after implementing the sōna Coccidioides Antibody LFA, the diagnostic test, the cost of send outs reduced by 84%, and the cost of all testing reduced by 68%. The TAT for sending out testing averaged 5–10 days, whereas the sōna Coccidioides Antibody LFA averaged a total TAT of <24 hours.

Conclusion. The sōna Coccidioides Antibody LFA offers a rapid, simple, and inexpensive method for accurately detecting antibodies against Coccidioides spp. in patient serum.

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252. Development and Evaluation of a Novel MultiCode Real-Time PCR Assay for the Detection of Pneumocystis jirovecii in Bronchoalveolar Lavage Fluid and Induced Sputum

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Background. Pneumocystis jirovecii is a medically important fungal pathogen responsible for opportunistic infections in immunocompromised hosts with high morbidity and mortality. Compared with standard microscopy based assays, home-brew nucleic acid amplification tests (NAAT) have emerged as sensitive tools for the diagnosis of P. jirovecii pneumonia, but their sensitivities vary depending upon selected genetic targets. Recent studies suggest that the mitochondrial small subunit (mtSSU) is a better NAAT target given its higher copy number and stable expression in the disease process. We aimed to develop and evaluate a mtSSU-targeted MultiCode real-time PCR assay that incorporates a sample processing control (SPC) and enables detection of P. jirovecii in bronchoalveolar lavage fluid (BALF) and induced sputum.

Methods. Firstly, we compared manual DNA extraction using Zymo Quick DNA kit with automated extraction using the NucliSENS easyMAG system after sample pre-treatment with either FastPrep mechanical grinding or vortex-based bead beating. We then determined the mouse hepatitis virus SPC (Luminex) spike-in amount, and optimized the PCR conditions on the ABI 7500 PCR system. A new Pneumocystis mtSSU run control was generated and using mtSSU gene as a generally engineered E. coli strain, and quantified with a home-brew quantitative TaqMan PCR. Lastly, the performance characteristics of the MultiCode PCR assay were determined.

Results. Mechanical grinding of BALF or sputum before the easyMAG based extraction was better than the other extraction protocols as evidenced by lower CT of mtSSU or SPC. Diluted SPC added to samples before DNA extraction made its CT within 34–31. With 32 mtSSU run control, the limit of detection of the new assay was 80 copies/mL. No cross-reactivity was found with 9 respiratory viruses, 8 bacteria and 11 fungi. The assay has high reproducibility for three-day detection of the same sample aliquots for mtSSU (CT: 30.0–30.3; CV%: 0.5–1.6) and SPC (CT: 32.1–32.2; CV%: 0.8–2.4).

Conclusion. We developed a novel MultiCode real-time PCR assay for detection of P. jirovecii in BALF and sputum, which demonstrated high analytical sensitivity, specificity and reproducibility and warrants further clinical validation.

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