

**Title: Rapid, ultrasensitive isolation of pathogenic *Candida* species directly from whole human blood**

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**Background:** There is a compelling need for a product which rapidly isolates the causative agents of candidemia from whole blood, permitting accelerated identification and characterization. Candidemia is a bloodstream infection associated with a high mortality rate. This delay in accurate diagnosis and administration of appropriate therapy in patients with candidemia is directly associated with poor clinical outcomes. The current gold standard diagnostic for candidemia, blood culture, requires extended incubation periods up to 96 hours before cultures turn positive with additional time required for speciation and characterization of antifungal sensitivity. nanoMR has developed a culture-free pathogen capture system (PCS) based on immunomagnetic capture, of fungal pathogens directly from whole patient blood in under 60 minutes. The PCS was tested against samples of whole human blood spiked with clinically relevant concentrations of pathogenic *Candida* species. Results from these spike and recovery studies demonstrate that the PCS can recover *Candida* at clinically relevant levels directly from human blood.

**Methods:** Individual samples of whole human blood at 10 ml volumes were collected from healthy volunteers in green-top sodium heparin Vacutainers™. The Vacutainers™ were spiked with a nominal 100 CFU of the following species; *C. albicans* (Type A and B), *C. tropicalis*, *C. parapsilosis*, *C. glabrata* and *C. krusei* for a level of 10 CFU pathogen/ml blood. Ten replicates per species were tested, one unique blood donor per replicate. Samples were processed using the nanoMR PCS. The sample matrix consisted of the full blood sample volume plus *Candida* Capture Reagent (paramagnetic beads conjugated with custom polyclonal antibodies raised against the target species) and Capture Assay Buffer. After incubation, samples were magnetically separated and washed. Total process time for samples was 45 minutes. Samples and controls were plated on yeast-mold agar and enumerated. Percent recovery for each species was determined by comparing recovered sample CFU to recovered control CFU.

**Results:** Percent recoveries for experimental samples ranged from 55% (*C. tropicalis*) to 110% (*C. albicans*, Type A) with minimal variability between replicates. PCS isolation performance for each species is shown in Figure 1.

**Conclusion:** The nanoMR PCS is a rapid, ultrasensitive culture-free system designed to isolate clinically relevant levels of pathogenic *Candida* spp. directly from whole human blood in less than 60 minutes. Whole human blood spiked with clinically relevant concentrations of *Candida* spp. and processed with the nanoMR PCS showed robust recovery of all species. The PCS is capable of either delivering whole cells for growth-based assays (as described here) or purified DNA from captured targets for gene-based analysis. The ability of the nanoMR PCS to isolate *Candida* spp. directly from patient blood in less than 60 minutes has the potential to positively impact patient outcomes by significantly decreasing time to diagnosis for suspected cases of candidemia.