

## **Real-time PCR of Whole Blood Specimens Transported in PrimeStore MTM® to Detect and Monitor MTB Bacteremia**

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**Background:** *Mycobacterium tuberculosis* (MTB) is an important cause of bacteremia and sepsis in HIV patients residing in sub-Saharan Africa. Many patients with MTB sepsis go undiagnosed and die within 18 days of presentation, making culture inadequate for detecting MTB in the blood.

**Objective:** To determine the feasibility of ambient temperature transport of blood in PrimeStore MTM® to a distant lab for real-time PCR detection of MTB bacteremia and to monitor clearance of MTB from the blood after therapy.

**Methods:** BALB/c female mice were injected intravenously with 0.2 mls of ethanol killed MTB (approximately 10<sup>5</sup> CFU/mL). Two anti-MTB opsonophagocytic bactericidal MABs were used to simulate treatment of MTB sepsis. Mouse monoclonal antibodies (MAB LHN-AB9 or LHN-GG9) or control were given IP using 0.3 mls of sterile PBS 24 hours before MTB challenge. To monitor MTB in the blood, mice were bled at 3 time points: within 15 minutes of injection with MTB and again at 4 and 24 hours after MTB challenge. Collected blood was placed into citrate tubes and 0.1ml was transferred into PrimeStore MTM®. Samples were transported at ambient temperature from Gaithersburg, MD to San Antonio, TX. DNA was extracted from blood in PrimeStore MTM® and replicate real-time polymerase chain reactions (PCR) were performed using PrimeMix® MTB Complex on an ABI 7500 Instrument.

**Results:** Blood PCRs on specimens obtained 15 minutes after MTB challenge were positive with an average CT value of 29.8 (range 29.2-30.6). Mice treated with PBS control had MTB detected in the blood by PCR at all time points (at 15 min, 4 and 24 hours post challenge). Mice given anti-TB opsonic MABs cleared the MTB from the blood either by 4 or 24 hours (CT=40).

**Discussion:** Blood specimens were efficiently transported to a central lab to detect MTB bacteremia by PCR. In addition, PCR may be useful to monitor patient treatment, similar to viral load testing for HIV. Using PrimeStore MTM® to ship specimens safely and rapidly at ambient temperature to a regional facility for PCR analysis may provide rural hospitals in sub-Saharan Africa the opportunity to diagnose MTB sepsis and monitor treatment.