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Passive Concentration Of Burkholderia Pseudomallei Capsular Polysaccharide From Melioidosis Urine Samples For The Enhanced Detection On The Inbios Active Melioidosis Detecttm Lateral Flow Immunoassay

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Burkholderia pseudomallei is a Gram-negative bacterium that is the causative agent of melioidosis, an infectious disease with an estimated mortality rate of 89,000. It is also a Tier-1 biothreat because it can be aerolsolized and dispersed. The saprophytic bacterium is endemic to Southeast Asia and northern Australia, though a more global prevalence is suspected from reports of incidence in South Asia and Africa. Disease manifestations can vary depending on the route of infection ranging from non-specific fever and myalgia to pneumonia and sepsis. Successful treatment involves an expensive and protracted regimen of antibiotics due to resistance to most frontline antibiotics. Early detection provides the best odds for successful treatment and cure and has been instrumental in lowering mortality. The gold standard for diagnosing melioidosis is culturing bacteria from a sample, which can take up to seven days to produce a result. Therefore, a more rapid diagnostic testing method is urgently needed. Towards that end, the InBios Active Melioidosis DetectTM Plus (AMD Plus) was developed in a user- friendly lateral flow format. It detects the capsular polysaccharide (CPS) of B. pseudomallei, which has been identified to be a reliable biomarker of melioidosis. To be effective in diagnosing the diverse manifestations of infection, it was intended for use in matrices including urine, serum, pus and respiratory secretions. Urine has emerged as a highly desirable matrix for near- patient use due to the non- invasive nature of sampling and evidence that circulating CPS is excreted in urine. The AMD Plus performs with high specificity, but its sensitivity in urine has been variable, because CPS levels in the small volumes of urine used for testing can be undetectable. To overcome this, we identified a passive urine concentration device to enrich for CPS. Here we examine the utility of the device in improving the sensitivity of AMD Plus on urine samples from melioidosis patients in 2 endemic regions, Australia and Thailand. In the studies, AMD Plus performance was assessed on urine samples from the same patient pre and post concentration. We also assessed the specificity of the AMD Plus on pre and post concentrated urine samples from healthy individuals and from patients with other urinary tract infections. Study results show that passive concentration can improve the sensitivity of AMD Plus on urine samples, as well as produce stronger test line intensities compared to unconcentrated urine samples, while not affecting the specificity of AMD Plus. Our findings demonstrate that passive concentration of urine samples can enrich for CPS and support the inclusion of the concentration device in the AMD Plus workflow, thereby increasing the confidence of the clinician to incorporate the AMD Plus in a melioidosis diagnosis algorithm.
The InBios Active Melioidosis DetectTM Plus can be an effective early, near patient diagnostic tool and can fill an existing gap in the case management workflow, enabling early confirmation and treatment. Given the high cost of melioidosis management, this is a significant product that is in line with DTRA's policies of creating stockpiles for effective management of biothreats.

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