

## REVOLUTIONARY DIAGNOSTICS – NONTRADITIONAL APPROACHES FOR DEVELOPING BREAKTHROUGH CAPABILITIES AGAINST EMERGING THREATS

### Immediate Results: An Fpa For Detection Of Glanders And Melioidosis Exposure.

**Michael Norris** University of Florida Emerging Pathogens Institute    **Andrew P. Bluhm** Department of Geography and Emerging Pathogens Institute, University of Florida    **Jason K. Blackburn** Department of Geography and Emerging Pathogens Institute, University of Florida    **Miladin Kostovic** EllieLab LLC

**Background:** Melioidosis and glanders are two diseases caused by *Burkholderia pseudomallei* and *Burkholderia mallei*, respectively. Melioidosis is an environmentally mediated disease associated with moist-soil environments throughout the tropics, causing infections in humans and animals. Glanders is restricted to ungulate hoof-stock such as horses and donkeys. Rapid detection of exposures by measuring serological responses is key to identifying exposed animals and chronically infected animals. Current technologies such as ELISA have been employed to screen serological samples for exposure to either pathogen but can take hours to run and require host-species optimization.

**Purpose:** Enable rapid detection of exposures to *Burkholderia pseudomallei* and *Burkholderia mallei*.

**Objective:** Develop a fluorescence polarization assay (FPA)-based diagnostic for melioidosis and glanders in all animal and human serum samples.

**Rationale:** ELISA assays are sensitive but are host species specific, requiring optimization to each species tested. Moreover, ELISAs use whole LPS of the bacteria as the antigen, thus lacking test specificity. Fluorescence polarization assays are O-chain polysaccharide-based assays, with high sensitivity and specificity and can identify samples containing bound antibodies regardless of the host-species. FPA is faster than ELISA and provides a homogenous platform for cross-species measurements. Samples can be read using a few steps procedure in seconds to minutes compared to hours that are common to ELISAs.

**Relationship to other areas of study:** one health, public health, veterinary science

**Methods:** The University of Florida has partnered with Ellie LLC, the maker of the brucellosis FPA and Sentry 300 portable FPA instrument, to develop a glanders/melioidosis FPA assay compatible with Sentry FPA instruments.

**Preliminary results:** An acapsular purine auxotroph of *B. pseudomallei* was engineered to allow biosafe purification of the *B. pseudomallei* OPS antigen. Another mutation in the *oacA* gene allows biosafe purification of the *B. mallei* OPS antigen structure. The OPS was isolated, purified and conjugated to the tracer. The tracer samples were used to measure antibody levels by ELISA and by testing on the Sentry 300.

**Preliminary conclusions:** FPA on the Sentry 300 is a highly portable, battery powered, rapid detection method for glanders and melioidosis that can be used with serum from any host-species and completed in minutes.