COMBATTING FUTURE BIOLOGICAL THREATS – HOST-DIRECTED INTERVENTIONS TO EMERGING THREATS FOR RAPID RESPONSE

Validation Of Omv Vaccine Composition Through Immune Reactivity And Mass Spectrometry To Generate Assays For Manufacturing And Clinical Trials

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Burkholderia pseudomallei is the causative agent of melioidosis, which can have a mortality of 10-50% in humans. B. pseudomallei is a gram-negative soil bacterium found primarily in Southeast Asia and Northern Australia. However, the worldwide distribution is not completely understood and was recently discovered in Mississippi. To date, no FDA licensed vaccine exists to prevent melioidosis. A novel outer membrane vesicle (OMV) vaccine is being developed to prevent melioidosis. The vesicles for this vaccine are naturally produced from an attenuated B. pseudomallei isolate generating a complex multi-component vaccine that could mimic bacterial infection and provide broad protection. Initial preclinical evaluation using research grade material yielded promising protection after aerosol challenge in a rhesus macaque model and GMP production of the vaccine is currently underway for additional protection and immunogenicity studies. The goal of our study was to comprehensively determine the proteomic content and immune reactivity of the OMV vaccine for manufacturing and clinical trial assay development. To that end, we performed shotgun proteomics using liquid chromatography mass spectrometry (LC MS/MS) using a Thermo Orbitrap Ascend followed by serology assays. With this highly sensitive method, we have identified greater than 1,300 different proteins in OMV produced lots. The OMV protein identification information was used to design a comprehensive OMV-specific nucleic acid protein programable array (NAPPA) to detect antibody responses generated during vaccination. To this end, protein coding regions were synthesized to produce proteins for ~1600 B. pseudomallei NAPPA proteins with an emphasis on OMV proteins that are predicted to localize to the outer membrane or the extracellular space. This OMV specific NAPPA was synthesized and antibody responses specific to the OMV proteins were determined. The array was probed using samples collected from OMV vaccinated rhesus macaques at Tulane NPRC. Key antigens include OmpA, Lipoprotein, and FlgK, though there were many others. Antibody reactivity for OmpA was also observed using melioidosis patient samples suggesting human reactivity for this vaccine antigen. The identified antigens will be used to monitor potential OMV vaccine induced antibody correlates of protection by adding the purified antigens to a Luminex serology assay to track antigen reactivity in animal studies, melioidosis patients, and future OMV clinical trials. In addition, monoclonal antibodies to these antigens are being developed to aid in vaccine production and quality control evaluation. In culmination, the OMV vaccine is positioned for GMP manufacturing and follow-up animal studies and clinical trials with the hope of licensing a vaccine to prevent melioidosis.

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