RESEARCH REPORT
SWINE HEALTH

Title: Multi-institutional development and validation of a multiplex fluorescent microsphere immunoassay for the diagnosis of multiple agents in serum and oral fluid – NPB #11-143

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Date Submitted: December 26, 2013

Scientific Abstract:
Serologic detection of antibodies remains the primary tool for the diagnosis of infection and for the purpose of conducting disease surveillance in negative populations. Fluorescent microsphere immunoassay (FMIA), also known as "Luminex" technology, represents a major advancement in serology by allowing the detection of antibodies against multiple antigens in a single, small volume of sample. This multiplexing capability has the potential to replace traditional serology-based technologies, such as ELISA. Previous funding was used to establish the utility of the technology in the detection of antibodies against PRRSV, PCV2 and SIV antigens. Based on detection of antibodies against nsp7, PRRS antibody detection was further divided into the detection of type 1 and type 2 genotypes. A recent advancement in technology is the incorporation of magnetic beads and the introduction of the MAGPIX instrument, which is lower in cost, possess a smaller footprint, and improved ease of operation. The goal of this proposal was to implement this technology for everyday use in diagnostic labs. The overall approach was to assemble a multi-institutional effort involving researchers at KSU, SDSU, and ISU. The advantages of the collaborative approach included access to an extensive number of serum and oral fluid samples from field and experimental studies, and the unique expertise of each lab with respect to the test development and standardization. The first objective was to establish a panel of serum and oral fluid samples for the standardization of PRRSV, SIV and PCV2 Luminex assays. SDSU prepared a panel of serum and oral fluid samples from pigs infected with a European-like type 1 PRRSV. The second objective was to determine reproducibility across the three laboratories, which included the development of a standard set of samples and protocols delivered to a commercial company. The third objective was to conduct a large scale inter-laboratory field validation incorporating field samples. Objective 3 identified the presence of false positive results in some pigs. After the engagement of a company, the second and third objectives were modified to include the transfer of all materials for the development of a commercial kit.