RESEARCH ABSTRACT
SWINE HEALTH

Title: PEDV antibody-based diagnostic test improvement for evaluation of immunity in milk, feces, and serum **- NPB 14-175** revised

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Scientific Abstract: Porcine epidemic diarrhea virus (PEDV), an enteric coronavirus related to transmissible gastroenteritis virus (TGEV), appeared suddenly in the United States in April 2013. Epidemic sow herd outbreaks, characterized by severe diarrhea, vomiting, and high mortality in nursing pigs for several weeks, continue to spread the disease. Virus can be detected in feces using a real-time PCR assay. Antibodies to the virus can be detected in serum samples using an ELISA assay based on the nucleocapsid (N) protein, but the sensitivity and specificity need to be improved. Our objective for this proposal was to create an improved ELISA for serum, optimize an ELISA for use with milk, colostrum, and feces, and examine these assays to determine predictors of protection for piglets and protection against re-exposure for sows. We were able to detect anti-PEDV antibodies in milk and colostrum for IgG and IgA isotypes. In milk, PEDV-specific IgA antibodies to the spike proteins are the most prevalent. In colostrum, both IgG and IgA antibodies are prevalent, again IgA antibodies are most reactive to the spike proteins and IgG antibodies are mainly to N and S2. Fecal IgA antibodies were observed in some samples, but were not consistently detected. Refolding of the N protein was able to slightly increase the antibody reactivity in serum, colostrum, and milk samples. Refolding of the other protein antigens was unsuccessful, but a spike protein fragment containing just the predicted antigenic region is being produced for further testing. An alternative sandwich ELISA coupling N antigen to HRP was produced but lacked specific reactivity. Other proteins may be promising if they can be produced in a soluble form under physiological conditions. Lactogenic immunity was examined as a predictor of piglet protection and consistently high levels of IgG antibodies were observed in colostrum to N and S2, while IgA antibodies were mainly observed in colostrum to S1 and S2. Milk contained IgG antibodies, but the levels decreased quickly suggesting that colostral spike antigens might be the best indicators of lactogenic immunity. Analysis of predictors of protection after re-exposure suggested that IFA is a better predictor of protection than ELISA, since ELISA values did not suggest the animals would be protected, when in fact, they were protected against disease. Overall, the PEDV N and S2 ELISAs seem to be the most useful predictors of protection in both IgG and IgA isotypes, refolding of the N protein is able to increase sensitivity of the assay, and serum and colostrum reactivity are mainly IgG antibodies against the N and S2 proteins, while milk reactivity is IgA antibodies against the spike proteins. This study has increased our knowledge of the antibody presence following infection or re-infection as well as the best ways to detect predictors of protection.