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Development of a high-throughput format assay for viral quantification

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Emergence of the porcine epidemic diarrhea virus (PEDV) in the U.S in 2013 resulted in severe economic damage, and loss of over one quarter of the swine population. The enteric disease primarily causes high mortality of up to 90% in neonatal pigs. Conventional methods for quantiufication of the virus include TCID50 and plaque assays. Virus quatification is important for vaccine and diagnostic test development. However, both methods are tedious to perform and subjective in analysis. Therefore, they are unsuitable for high-volume testing in diagnostic laboratories. In this study, a colorimettric assay using a tetrazolium dye called MTT, which measures cell viability, was developed to quantify PEDV cultures. A comparison of the optical density (OD) output with the visual read outs was assessed by Receiver Operator Characteristics (ROC) analysis. The specificity (Sp) and sensitivity (Se) of the MTT_{50} assay, in comparison to the conventional TCID50 assay were 98% and 100% respectively, at an OD cutoff value of 0.52. The agreement between the tests as determined by a kappa statistic value was 0.83 with a 95% confidence interval of 0.780 to 0.874. In the future the assay will be adapted to virus neutralization assays to enable the measurement of protective antibody titers. The assay will also simplify high throughput testing of PEDV in diagnostic laboratories.

Biography

Sheela Ramamoorthy is a Veterinarian and Virologist, who obtained her Bachelors in Veterinary Medicine (BVSc) from the Madras Veterinary College in India, MS in Microbiology and Molecular Genetics from the Oklahoma State University, followed by a PhD in Biomedical Sciences from Virginia Tech. She served as the Section Head of Diagnostic Virology and Serology at the University of Georgia Tifton Veterinary Diagnostic Laboratory, before moving to N Dakota State University, where her research is focused on studying vaccine-mediated immunity against viral infections. Her well-funded research group works on translating basic findings into novel vaccines and diagnostics, for agents involved in the porcine respiratory disease syndrome. She had authored over 40 peer-reviewed publications, is a Board Member of the American Association of Veterinary Immunology, she serves as a Reviewer for reputed journals and as a Grant Reviewer for several agencies including the United States Department of Agriculture and National Institutes of Health.

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