Development and Optimization of a Reverse Genetics System for Porcine Reproductive and Respiratory Syndrome Virus

Changhee Lee[1], Jay G. Calvert[2], Siao-Kun Welch[2], and Dongwan Yoo[1]*
Department of Pathobiology, Ontario Veterinary College, University of Guelph[1]
and Pfizer Animal Health[2]
519-824-4120 ext 54729, dyoo@uoguelph.ca

Background

Porcine reproductive and respiratory syndrome virus (PRRSV) is an emerged virus in swine and causes significant economic losses in the pork industry world-wide. PRRS is represented by reproductive failures in sows and gilts and severe respiratory distress in grower/finisher pigs. Pathogenic mechanisms of PRRS virus are largely uncharacterized, and biological function of individual viral proteins are poorly understood. An infectious full-length cDNA clone is a useful tool for genetic analysis of virus allowing us to introduce a mutation to a specific region of the viral genome and accordingly a mutant virus can be generated (reverse genetics). Development of such molecular tool is useful for studying virus replication, pathogenesis, and function of viral proteins, as well as to develop a genetically modified vaccine candidate.

Objective

To develop and optimize a functional reverse genetics system for PRRSV so that the PRRSV genome can be freely engineered.

Results

A full-length cDNA clone for PRRSV was assembled. When transfected cells with the assembled genomic clone, infectious PRRSV was recovered from transfected cells, indicating that the full-length cDNA clone was infectious. A shuttle plasmid was constructed to facilitate the introduction of a mutation to the structural region of the virus, and a genetic marker was incorporated into the shuttle plasmid. Subsequently, the genetic marker was rescued to the genomic clone and an infectious recombinant PRRSV was successfully generated.

Take Home Message

A functional reverse genetics system was developed for PRRSV. The reverse genetic system was further optimized to facilitate genetic modifications of the viral genome and to distinguish the engineered virus from the parental virus. This is a useful genetic tool for molecular studies of PRRSV. The developed system will allow us to device a safe and effective vaccine for PRRS in pigs.

This study was partly supported by funding from Ontario Pork and the Ontario Ministry of Agriculture and Food (OMAF) Animal Program.