Development of an Alphavirus Replicon Classical Swine Fever Virus Vaccine Candidate

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Summary and Implications

Classical swine fever virus (CSFV) E2 glycoprotein was expressed in an alphavirus based replicon expression system. Vaccinated pigs developed CSFV-specific antibodies. This is the first known use of this technology against CSFV.

Introduction

Classical swine fever virus is the cause of hog cholera. CSFV was eradicated from the US in 1978, but remains a serious threat to the swine industry. Alphavirus-derived replicon particles (RP) are safe and effective vectors that have been developed for use in veterinary vaccines.

Materials and Methods

The CSFV E2 gene was codon-optimized and *de novo* synthesized prior to cloning into the replicon DNA plasmid. A modified signal sequence was added to the 5' end of the gene, and a histidine tag sequence was added to the 3' end of the gene to facilitate detection in immunoassays. Replicon RNA was generated by *in vitro* transcription using T7 RNA polymerase. The CSFV-E2 RP were generated by co-electroporation of replicon RNA and helper RNAs into Vero cells. The RP were harvested by affinity chromatography and quantified by immunofluorescence. Vero cells were infected with CSFV-E2 RP, and cell lysates were used for Western blot expression analysis. Vero cells were also infected with CSFV-E2 RP and fixed with acetone:methanol for indirect immunofluoresence assay (IFA). Young pigs (n=3) were vaccinated twice with CSFV-E2 RP, and serum samples were tested for reactivity against fixed, RP-infected Vero cells.

Results and Discussion

CSFV E2 expression was confirmed by Western blot (Figure 1). Serum from vaccinated pigs reacted with CSFV-E2-positive cells by IFA, while control pig serum did not. Future work will determine the level of neutralizing antibody in vaccinated pigs, and CSFV challenge studies will be conducted to evaluate the level of protection afforded by RP vaccination.

Figure 1. Western blot of CSFV E2 expressed in Vero cells.