Hydrogels as Adjuvants for PRRS Modified Live Virus Vaccine

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ABSTRACT SUMMARY

We recently demonstrated that a novel peptide nanofiber hydrogel (H9e) could act as a potent adjuvant for a killed H1N1 influenza vaccine. The objective of this study was to evaluate H9e as an adjuvant for porcine reproductive and respiratory syndrome virus (PRRSV) modified live virus (MLV) vaccines. Pigs were vaccinated with Ingelvac PRRS-MLV with or without H9e adjuvant before being challenged with the VR-2332 (parental vaccine strain) or MN184A (genetically diverse strain) PRRSV. Pigs vaccinated with MLV+H9e had more persistent and higher levels of circulating vaccine virus, earlier onset of PRRSV-specific antibodies, and higher titers of neutralizing antibodies than that in pigs vaccinated with MLV alone. More importantly, pigs vaccinated with MLV+H9e had improved protection against challenge by both PRRSV strains, as demonstrated by reduced challenge-induced viremia compared with pigs vaccinated with MLV alone. Taken together, our studies suggest that the peptide nanofiber hydrogel H9e, when combined with the PRRSV MLV vaccine, can enhance vaccine efficacy against two different PRRSV strains by modulating host immune responses.

INTRODUCTION

Adjuvants including oil-in-water emulsions and nanoparticles have been tested in combination with modified live vaccines in an effort to reduce the antigenic load and improve vaccine efficacy. Peptide hydrogels also might be a promising delivery system for vaccines due to their high water content, polymer network and reversible sol-gel (solution to gel) formation. Peptide hydrogels have been well studied as drug delivery systems, for tissue engineering applications, and in 3-D cell culture and show promising results. We recently developed a novel peptide that can form a flexible nanofiber hydrogel (H9e) and functions as a potent adjuvant for killed H1N1 influenza vaccines [1]. To further characterize the capabilities of the H9e hydrogel, we evaluated H9e as an adjuvant for PRRSV MLV vaccines.

EXPERIMENTAL METHODS

Thirty five female/unvaccinated (3 weeks old) Large White-Duroc crossbred PRRSV-free pigs were divided into 7 groups (n = 5) and housed in separate pens within the Large Animal Research Center (LARC) at Kansas State University. These piglets were confirmed sera-negative for antibodies to PRRSV by ELISA and PRRSV-free in serum by RT-PCR. Pigs were immunized intramuscularly on day 0 with placebo, PRRS-MLV (1x10^6 TCID50/pig), or PRRS-MLV+H9e (1x10^6 TCID50 + 6mg H9e/pig). Twenty-eight days post vaccination (DPV), the pigs were challenged with either homologous PRRSV VR-2332 (1x10^6 TCID50) or heterologous MN184A (5x10^5 TCID50). All pigs were humanely euthanized 15 days post challenge (DPC). All animal experiments were approved by the Institutional Animal Care and Use Committee at Kansas State University.

PRRSV-specific and virus neutralizing antibody titration PRRSV-specific ELISA antibody titers were measured using the Herdcheck Porcine Reproductive and Respiratory Syndrome Antibody test Kit (IDEXX Laboratories, Westbrook, ME) as described by the manufacturer. Virus neutralizing antibody titer in the serum was analyzed as previously described [2].

RESULTS AND DISCUSSION

To determine if vaccinated pigs were protected from homologous or heterologous virus challenge, titers of circulating virus were measured 7 days post challenge (DPC). The pigs vaccinated with MLV+H9e were able to significantly clear both the VR-2332 and MN184A strains circulating in the blood, whereas pigs vaccinated with MLV alone were able to significantly clear only the homologous VR-2332 virus strain (Fig.1A, B). These results suggest that the addition of H9e adjuvant to PRRSV MLV vaccines can enhance protection against genetically distinct stains of PRRSV.
Figure 1. H9e adjuvant enhances protection efficacy of MLV to homologous and heterologous PRRSV infection in pigs. Pigs (3-week-old) were vaccinated with MLV or MVL+H9e and challenged with the VR-2332 (A) or MN184A (B) strain of PRRSV 28 days post vaccination. Viral RNA in the serum (TCID₅₀ equivalent/mL) was measured on 7 days post challenge (DPC) by RT-PCR. Viremia data are shown as means ± SEM (n=5). * p <0.05), ** p <0.01).

To determine if the H9e affects the production of neutralizing antibodies, the PRRSV neutralizing antibody titers (VN titers) were analyzed. On 28dpv, MLV+H9e vaccinated pigs had equivalent VN titers to both VR-2332 and MN184A as MLV-alone vaccinated pigs (data not shown). After homologous VR-2332 viral challenge, pigs vaccinated with MLV+H9e had significantly higher VN titer to VR-2332 compared with pigs vaccinated with MLV alone (Fig. 2).

Figure 2. Pigs vaccinated with MLV+H9e produce more PRRSV-specific antibodies. Serum samples were titrated individually in MARC-145 cells for the levels of anti-PRRSV neutralizing antibodies 14 days post challenge (DPC) determined as the highest dilution that inhibited CPE. Data are shown as mean ± SEM (n=5). ** p <0.01.

CONCLUSION
Results show that the addition of H9e to MLV enhanced protection of pigs to both homologous and heterologous strains of PRRSV. Compared with pigs vaccinated with MLV alone, animals vaccinated with MLV+H9e developed more robust PRRSV-specific neutralizing antibodies. PRRS MLV vaccine formulated with H9e may have higher efficacy against various types of PRRS viruses.

REFERENCES

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