Evaluation of the response to PRRSGard® administration in weaned pigs

C. Smith[1], DVM; F. Chamba[1], DVM, MS, PhD, SHM-EVP; J. Pittman[2], DVM, MS, DABVP; G. Rawal[3], DVM, MS; J. Zhang[3], MD, MS, PhD; C. Francisco[1], DVM, MS, SHM-EVP

[1]Pharmgate Animal Health, Wilmington, North Carolina; [2] Smithfield Hog Production-North Region, Waverly, Virginia; [3]Iowa State University, Ames, Iowa

Introduction

Porcine reproductive and respiratory syndrome virus (PRRSV) continues to cause significant economic and performance losses in swine production in the United States.[1] Multiple practices have been employed in attempts to control the disease, including modified-live virus (MLV) vaccines to reduce clinical signs, viremia and lung lesions, which improve health and performance in pigs. There are several different PRRSV MLV products commercially available to producers. The choice between products may be influenced by cost, efficacy, decreased performance or setback directly following vaccination (drag), and the ability of the vaccine to spread to non-target or non-vaccinated pig populations.

PRRSGard® is a unique PRRSV MLV vaccine with a chimeric virus composed from a proprietary, highly attenuated backbone and structural proteins from a highly virulent isolate (MN184) within ORF5 lineage 1.[2] Additionally, a 23-nucleotide insert has been positioned between ORF1b and ORF2 to be used as a genetic marker for differential rRT-PCR diagnostic testing. The objective of this report is to summarize the immunological response, shedding profile, and production performance drag of weaned pigs vaccinated with PRRSGard®.

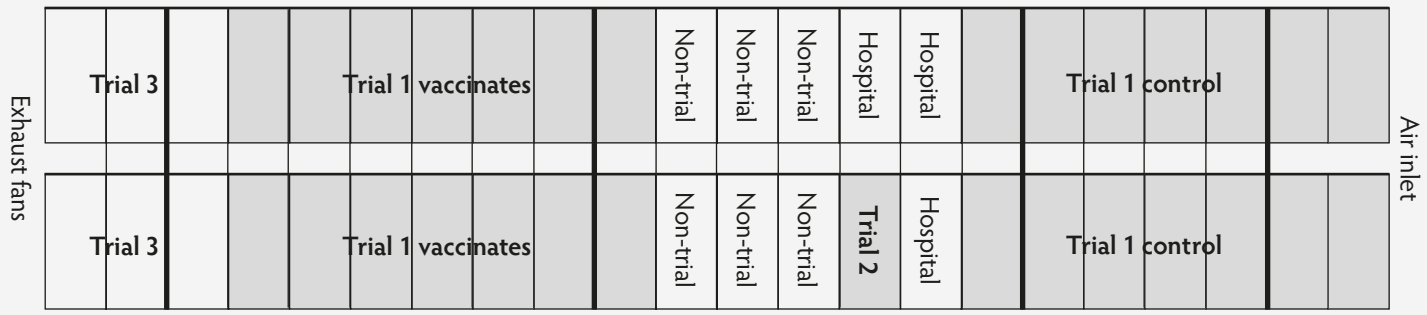
Materials and methods

Weaned pigs approximately 3 weeks old and known to be naïve for PRRSV were sourced from multiple sow farms. Eight hundred fifty-three mixed breed pigs were sorted by sow farm source and randomly allocated into 35 of 44 total pens in a commercial, tunnel ventilated, wean-to-finish barn following the farm's standard operating procedures. The day after placement, pens were assigned to various trials based upon location within the barn and air flow in order to limit potential vaccine virus spread to other trial pigs. The assignments are illustrated in Figure 1. Pigs in pens not associated with a trial were not vaccinated with PRRSGard® or sham vaccine. The farm's standard operating procedures included a "pull program" which involved leaving several hospital pens empty at placement with multiple fixed time identification and removal points of the slowest growing pigs throughout the barn. These pigs were used to populate the hospital pens. Any pigs removed from their original pen and placed in hospital pens were immediately removed from their respective trials and no further information was collected.

Trial 1: Performance drag

Twenty-eight total pens were selected and assigned to either vaccinated or sham-vaccinated control groups. All pigs within each pen were individually identified by ear-tag, weighed, and vaccinated with the appropriate product. The vaccinated group received 1ml of PRRSGard® intramuscularly and the sham-vaccinated control group received 1ml of vaccine diluent intramuscularly. Six oral fluid samples were taken on day 0 and 48 post-vaccination in the control pens, and tested by a commercial PRRSV rRT-PCR to ensure no virus circulation within the controls. Pigs were individually weighed again 48 days post-vaccination. Average daily gain (ADG) and survivability were estimated utilizing generalized linear and logistic regression models in the R statistical software[3] packages lme4[4] and lsmeans.[5] The analysis was adjusted by start weight and block (barn location) effects. A *P*-value of <0.05 was used to indicate statistical significance.

Figure 1: Barn diagram with trial and treatment assignments.



Trial 2: Immune response characterization

Twenty-five pigs in one pen were vaccinated with 1ml of PRRSGard® intramuscularly. Serum samples were collected from each pig on days 0, 7, 14, 21, 28, 35, and 41 post-vaccination. Serum samples were tested by a commercial PRRSV rRT-PCR at day 0. All samples were tested by the PRRSGard® specific rRT-PCR utilizing the integrated genetic marker, a commercial PRRSV ELISA and a PRRSV174 serum neutralization (SN) assay. The PRRSGard® specific rRT-PCR was developed and performed by Dr. Jianqiang Zhang and Dr. Gaurav Rawal at the Iowa State University Veterinary Diagnostic Laboratory. The PRRSGard® specific rRT-PCR did not cross react with many PRRSV field and vaccine strains as well as other common swine pathogens. The analytical sensitivity based on ct values was similar to a current commercial PRRSV rRT-PCR. Plotting of those results was done using the R package ggpubr.[6]

Trial 3: Shed and spread

Six pens containing a total of 144 pigs were selected and pigs within each pen were individually identified by ear tag and randomly assigned to the vaccinated or sham-vaccinated groups. All six individual pens were composed of 50% vaccinated and 50% control pigs. The vaccinated group received 1ml of PRRSGard® intramuscularly and the sham-vaccinated control group received 1ml of vaccine diluent intramuscularly. Serum samples were collected from all pigs at vaccination on day 0 and again on day 41. Day 0 samples were tested by a commercial rRT-PCR. All samples were tested by a commercial PRRSV ELISA and the PRRSGard® specific rRT-PCR. A sample with an S/P ratio of ≥0.4 was considered ELISA positive in all trials. A sample with a ct value of < 40 was considered rRT-PCR positive in all trials. The basic reproduction number (R_0) of PRRSGard® spread during those 6 weeks was estimated using the attack rate method in the R package R0.[7]

Results

Trial 1: Performance drag

Oral fluid samples tested PRRSV negative by the commercial rRT-PCR at days 0 and 48 post-vaccination. There was not enough evidence of differences in ADG and survivability between the PRRSGard® and control groups. The descriptive and statistical analysis of performance parameters is summarized in Table 1.

Trial 2: Immune response characterization

All weaned pigs were confirmed to be PRRSV negative by rRT-PCR and ELISA prior to vaccination. Two pigs were removed from the trial between days 14 and 21 per the farm's standard operating procedure to identify the slowest growing pigs and placed them in a hospital pen. Viremia was detected in sera of 19/25, 22/25, 19/23, 21/23, 20/23 and 19/23 at 7, 14, 21, 28, 35, and 41 days post-vaccination, respectively. Viremia begin dropping off 35 days post-vaccination. The mean rRT-PCR cycle threshold (ct) values over time are illustrated in Figure 2. As expected, ELISA results were negative until the 2nd sampling event (14 days post-vaccination) with a markedly increase in the percentage of positives in subsequent sample events. The ELISA results are summarized in Figure 3. Serum neutralization assay results were unavailable at the time of paper submission but they will be summarized in the accompanying presentation.

Trial 3: Shed and spread

All weaned pigs were confirmed to be PRRSV negative by rRT-PCR and ELISA prior to vaccination. At 41 days post-vaccination, 57/71 (80%) of the negative control pigs tested PRRSGard® rRT-PCR negative. Only one of the negative control pigs tested positive in the ELISA with an S/P ratio of 0.454. The basic reproduction number (R_0) for viremic pigs and serological positive pigs was 1.1 and 1.0, respectively. Our results are indicative of the limited transmission of PRRSGard® to contact-naïve pigs during those 6 weeks of evaluation.

Discussion

PRRSV continues to be a problem for many swine producers. Modified-live vaccines remain one of the best tools to control the clinical signs of disease associated with PRRSV. Traditional attenuation methods rely on altering field viruses, so they are hopefully less virulent, but similar enough to stimulate a protective immune response to subsequent natural infection. This balance between altering a virus, while also keeping it similar, may often be difficult to achieve. PRRSGard® takes a unique approach by the application of a chimera. The replication machinery in ORF1a and ORF1b are from a proprietary strain that has been well attenuated. However, the structural proteins (ORF3-7) are from a highly virulent isolate (MN184) that was not modified using traditional attenuation methods. The purpose of the chimera pro-

Table 1: PRRSGard® performance drag summary

Group	No. pigs	ADG (lb/day)	95% CI	Survivability %	95% CI
PRRSGard®	345	1.17	1.15 – 1.19	96.7	94.1 – 98.1
Control	339	1.19	1.17 – 1.21	96.5	93.8 – 98.0
Difference	-	0.02	-	0.02	-
P-value	-	0.14	-	0.87	-

Figure 2: PRRSGard® viremia over time.

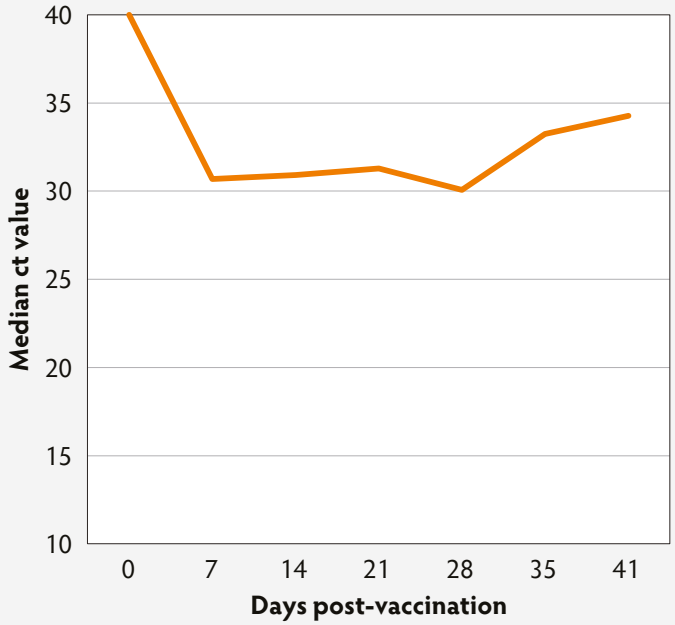
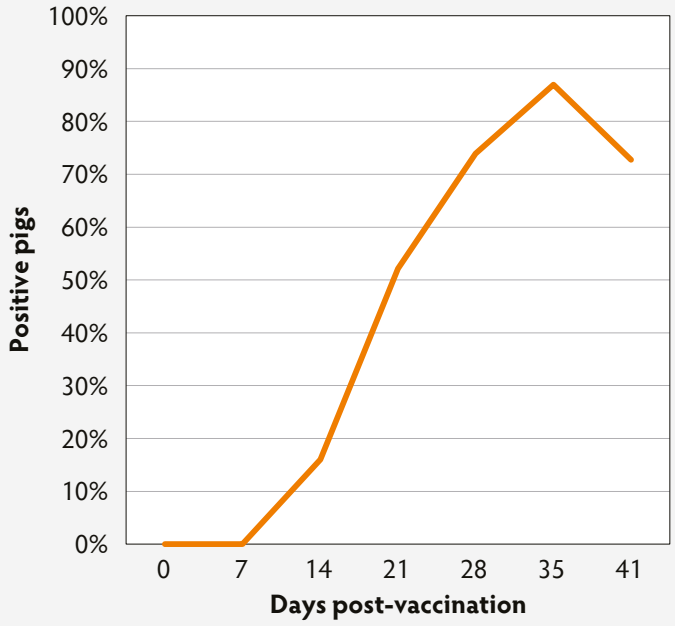


Figure 3: ELISA positive pigs over time.



duction is a vaccine virus that is highly infectious and stimulates a strong, relevant immune response but is lowly transmissible and does not negatively impact performance through signs similar to disease expression.

Vaccination of weaning age pigs with PRRSGard® did not result in reduction of performance as measured by average daily weight gain and survivability when compared to sham-vaccinated pigs. This limited impact of PRRSGard® on performance is important in high health flows where pigs need to be vaccinated given the high risk of lateral introductions. Additionally, PRRSGard® induced high levels of replication seven days post-vaccination and a subsequent immune response two weeks later. Finally, PRRSGard® spreads slowly within naïve populations with direct contact and following vaccination of 50% of the animals. This last point is important because it suggests that there was limited exposure of non-target animals to vaccine virus under field conditions. Even more, a PRRSV vaccine with limited transmission is important when considering the risk of area spread, especially transmission to naïve sow farms. A PRRSV vaccine with limited transmission and performance drag can be applied in other production situations that warrant further exploration. In summary, these trials begin characterizing the benefits of PRRSGard® as a new tool to aid in the control of PRRSV in the US and worldwide.

References

1. Holtkamp D. et al. Assessment of the economic impact of porcine reproductive and respiratory syndrome virus on the United States pork producers. *Swine Health and Production.* 2013. 21:72-84.

2. Chamba F. et al. Experimental safety and efficacy of a unique MLV PRRSV vaccine: PRRSGard®. *AASV proceedings.* 2018. 170-174.

3. R: A language and environment for statistical computing v. 3.4.2 (R Foundation for Statistical Computing, Vienna, Austria, 2017).

4. Bates, D., Mächler, M., Bolker, B. & Walker, S. Fitting linear mixed-effects models using lme4. arXiv preprint arXiv:1406.5823 (2014).

5. Lenth, R. V. Least-Squares Means: The R Package lsmeans. Journal of Statistical Software 69, doi:10.18637/jss.v069.i01 (2016).

6. ggpubr: "ggplot2" based publication ready plots. R package version 0.1.7 (2018).

7. R0: Estimation of R0 and Real-Time Reproduction Number from Epidemics v. R package version 1.2-6 (2015).