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CHARACTERIZATION OF THE PRRSV STRAIN CIRCULATING IN A PRRSV TYPE 1 POSITIVE HERD

BEFORE, DURING, AND AFTER VACCINATION WITH A PRRSV TYPE 1 VACCINE

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Introduction

PRRSV is endemic in Denmark with approx. 50 % of the swine herds being infected (<u>www.spf-sus.dk</u>). The economic losses due to PRRSV are ~15 mill€/year. Both PRRSV Type 1 and Type 2 are circulating in the Danish pig herds. Vaccination with live-attenuated vaccines against both genotypes is widely used to control the disease.

The aim of the project was to investigate if it was possible to eradicate PRRSV from the herd by the use of vaccination. The PRRSV dynamic in the herd was observed during a period of 2 years (Aug. 2012 -July 2014) before, during, and after vaccination with at Type 1 PRRSV MLV.

Materials and methods

The case farm was positive for PRRSV Type 1 and Mycoplasmas and contained four sections of 7-30 kg pigs and eight sections of 30 kg- slaughter pigs. All sections were located at the same site. The herd receives 470 7 kg PRRSV-negative pigs every other week. The new arrivals are housed in clean and emptied pens. At 30 kg the pigs were moved to an empty and clean section where they stayed until slaughter. Pigs were not moved between pens and sections. The farm has been positive for PRRSV Type 1 for several years based on serology.

Blood samples were collected from the herd approx. every other month, before, during, and after vaccination with a Type 1 PRRSV MLV (figure 1). Following blitz vaccination in February 2013, all new arrivals were vaccinated prior to housing until ultimo August 2013. In January 2014, the last vaccinated pigs were slaughtered. To screen for PRRSV, RNA was extracted from serum pools (10 samples/pool) using QIAsymphony RNA Kit on QIAsymphony SP extraction robot. RNA was initially tested by an in-house PRRSV real-time RT-PCR¹. To investigate the genetic diversity over time, RNA was extracted from selected samples from each collection day and ORF5 was sequenced using a previously described method². Furthermore, partial NSP2 was sequenced². The sequences were analyzed using CLC Main Workbench v7.5.1. All samples were tested for antibodies (Ab) by ELISA and IPMA.

Results

At least one serum pool from each collection day was found PRRSV Type 1 positive by real-time RT-PCR, except samples from Oct. 2012 and Nov. 2013, which were negative (Figure 1.). Samples collected Nov. 2013 were also negative for Ab by ELISA. The real-time RT-PCR results obtained in Dec. 2012 confirmed that the case farm was infected with PRRSV Type 1. Nucleotide comparisons of ORF5 sequences from samples obtained in Aug. and Dec. 2012 showed resemblance to other Danish Type 1 PRRSV² but only 85 % identity to the Type 1 vaccine strain. During the vaccination period, the ORF5 sequences obtained were close to 100 % identical to the Type 1 vaccine strain. After a period with PRRSV negative pigs, PRRSV Type 1 was again detected. The ORF5 sequences from these samples showed 98.35-99.67% homology to the vaccine strain. To further investigate the origin of these viruses partial NSP2 was sequenced. The results showed deletions in the same region as the vaccine strain.

Figure 1. Timetable for collection of blood and vaccination. Asterisk: negative real-time RT-PCR.

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Aug. '12	Oct '12	Dec. '12	Feb. '13	Apr. '13	June '13	aug. '13	Nov. '13	Jan. '14	Mar. '14	May '14	July '14

Discussion

Following vaccination with a PRRSV Type 1 vaccine, the circulating viruses in the herd shifted from a heterologous to homologous PRRSV Type 1 vaccine strain. In November 2013, PRRSV was not detected in the herd which indicated that the vaccination program had indeed eradicated PRRSV from the herd. However, in January 2014 the herd tested positive for PRRSV again and remained PRRSV positive until the end of the study. Comparison of the ORF5 of the viruses isolated during the vaccination period with the re-emerged viruses showed high level of homology indicating that it was the vaccine strain that now circulated in the herd. This was supported by the analysis of the partial-NSP2 sequences of the re-emerged viruses showing that the isolates harbored a deletion that was also present in the vaccine strain. In conclusion, vaccination pushed out the existing PRRSV strain but the vaccine strain seemed to persist in the herd also after vaccination was terminated.

References

1. Wernike et al. (2012). J. Vet. Diagn. Invest 24, 855-66.

2. Kvisgaard et al. (2013). Virus Res. 178, 197-205.