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Porcine Reproductive and Respiratory Syndrome Virus Serotype-distinguishing performance of the Immunoperoxidase Monolayer Assay and the double-Blocking ELISA

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Introduction

Porcine Reproductive and Respiratory Syndrome virus (PRRSV) can lead to profound economic losses in pig production. The European and the American PRRSV strains may give rise to distinct serological profiles (Murtaugh, 1995). In Europe, since the introduction of a US-type-based vaccine (Ingelvac PRRS- MLV®, Boehringer Ingelheim, Ingelheim, Germany), there is an increasing desire for reliable serological tests to monitor vaccination strategies in EU-wild type endemically infected herds. These tests should allow for the distinguishing of pigs infected with an EU or a US strain of PRRSV or pigs infected with both serotypes at the same time. Among these serological tests, the differentiating-immunoperoxidase monolayer assay (d-IPMA) and the double-blocking ELISA (Sorensen , 1998) are readily performed.

The aim of this study was to establish the EU and US-antibody-distinguishing capabilities of these serological tests. The trial was performed as a randomised blind and controlled laboratory study.

Materials and Methods

Fifty, 5-6 week old pigs derived from a PRRSV-negative farm, were randomly assigned to the following 5 groups. Group I: negative control; intranasally inoculated (i.n.) with 2 ml of sterile water on day 0 and day 84; Group II: Vac; vaccinated with 2 ml of Ingelvac®PRRS MLV intramuscularly (i.m.) on day 0 and on day 84, 2 ml of sterile water i.m. Group III: DK; 2 ml of a virulent Danish Field strain i.n. on day 0 and 84 days later 2 ml of sterile water i.m.;

Group IV: DK+Vac; 2 ml of a virulent Danish field strain i.n. On day 0 and 84 days later 2 ml of Ingelvac®PRRS MLV i.m.; Group V: Vac+DK; 2 ml of Ingelvac®PRRS MLV i.m. on day 0 and 84 days later 2 ml of a virulent Danish Field strain i.n..

All animals were bled on days –14, 0, 7, 14, 28, 42, 56, 70, 84, 91, 98, 112, 126, 140, 154, and 168 after first inoculation or vaccination, for determination of antibody titers by $IPMA_{DK}$, $IPMA_{VAC}$ and Blocking ELISA ratio. The IPMA results are expressed as reciprocal titers. A titer exceeding 1:50 is considered positive. The ELISA results are expressed as the ratio of the optical densities as determined in the DK and the US setup of the ELISA. A ratio exceeding 2 is considered positive for US-type antibodies, whereas a ratio below 1 is considered positive for DK-type of antibodies.

All serological tests were performed by SVIV, Lindhom, Denmark, according to established and published laboratory techniques.

Results

No clinical signs could be detected after inoculation.

No PRRSV-directed antibodies could be detected in any of the pigs before being challenged. The control group remained negative throughout the study. In Group II both the $IPMA_{DK}$ and $IPMA_{VAC}$ showed a positive titer from 14 days after inoculation. However, the $IPMA_{VAC}$ titer was higher than the $IPMA_{DK}$ titer throughout the study The ratio of the ELISA always exceeded 2. In Group III, the $IPMA_{DK}$ showed a higher titer than the $IPMA_{VAC}$ at all times. The ratio always remained below 1. In Group IV, the $IPMA_{DK}$ titer was higher than the $IPMA_{VAC}$ titer throughout the study in spite of vaccination at day 84. The ratio changed from below 1 prior to day 84 to above 2 from day 119 until the end of the study.

In group V, the $IPMA_{VAC}$ had a higher titer than the $IPMA_{DK}$ throughout the study in spite of DK-inoculation at day 84. The ratio remained above 2 during the entire study.

Conclusion

The d-IPMA and the ELISA could accurately show infections with the Danish field strain or the vaccine strain. When both strains are consecutively being inoculated, the d- IPMA is not able to show exposure of a heterologous strain in previously PRRSV-infected pigs. The ELISA performed only accurate if vaccine virus is introduced in previously DK-field virus infected pigs, but not vise versa.

Reference

Sørensen, K.J., Strandbygaard, B., Bøtner, A., Madsen, E.S., Nielsen, J., and Have, P: Blocking ELISA's for the destinction between antibodies against European and American strains of porcine reproductive and respiratory syndrome (PRRS) virus. Vet. Microbiol., 1998, 60, 169-177.