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Impact of a novel inactivated PRRS virus vaccine on virus replication and virus-induced pathology in fetal implantation sites and fetuses upon challenge

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Abstract

Preventing congenital infection is important for the control of porcine reproductive and respiratory syndrome (PRRS). Recently, in our laboratory, an inactivated porcine reproductive and respiratory syndrome virus (PRRSV) vaccine has been developed. Promising results in young pigs encouraged us to test the vaccine potency to prevent congenital infection. In the present study, the performance of this experimental inactivated vaccine was investigated in pregnant gilts. An advanced protocol was used to test the PRRSV vaccine efficacy. This protocol is based on recent insights in the pathogenesis of congenital PRRSV infections. Three gilts were vaccinated with an experimental PRRSV 07V63 inactivated vaccine at 27, 55, and 83 days of gestation. Three unvaccinated gilts were included as controls. At 90 days of gestation, all animals were intranasally inoculated with 10^5 tissue culture infectious dose 50 ($TCID_{50}$) of PRRSV 07V63. Twenty days postchallenge animals were euthanized and sampled. The vaccinated gilts quickly developed virus neutralizing (VN) antibodies starting from 3 to 7 days postchallenge (1.0 to 5.0 $\log_2$). In contrast, the unvaccinated gilts remained negative for VN antibodies after challenge. The vaccinated gilts had shorter viremia than the control gilts. Gross pathology (mummification) was observed in 8% of the fetuses from vaccinated gilts and in 15% of the fetuses from unvaccinated gilts. The number of fetuses with severe microscopic lesions in the fetal implantation sites (a focal detachment of the trophoblast from the uterine epithelium; a focal, multifocal, or full degeneration of the fetal placenta) was lower in the vaccinated (19%) versus unvaccinated (45%) gilts ($P < 0.05$). The number of PRRS-positive cells in the fetal placentae was higher in unvaccinated versus vaccinated gilts ($P < 0.05$). In contrast, the number of PRRS-positive cells in the myometrium/endometrium was higher in vaccinated versus unvaccinated gilts ($P < 0.05$). Fifty-seven percent of the fetuses from the vaccinated gilts and 75% of the fetuses from the unvaccinated gilts were PRRSV-positive. In conclusion, implementation of the novel experimental inactivated PRRSV vaccine primed the VN antibody response and slightly reduced the duration of viremia in gilts. It also reduced the number of virus-positive fetuses and improved the fetal survival, but was not able to fully prevent congenital PRRSV infection. The reduction of fetal infection and pathology is most probably attributable to the vaccine-mediated decrease of PRRSV transfer from the endometrium to the fetal placenta.

Keywords: PRRSV; Inactivated vaccine; Transplacental infection; Placenta; Monograph

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1. Introduction

Porcine reproductive and respiratory syndrome virus (PRRSV) is a major disease affecting swine livestock worldwide. The etiologic agent is an RNA virus which causes reproductive disorders in pregnant sows and respiratory dysfunctions in young pigs. Prevention of porcine reproductive and respiratory syndrome (PRRS) and eradication of the virus is challenging. At present, there is a gap in understanding the PRRSV biology, pathogenesis and immunity. This lack of knowledge hampers the research-based design of vaccines that are able to protect animals against a wide range of PRRSV strains. Today's difficulties in the field of PRRSV vaccinology are explicitly described in a number of recent reviews [1–3]. For the control of PRRSV-induced reproductive problems, attenuated and inactivated vaccines are commercially available. However, there are concerns on both safety and efficacy. Attenuated vaccines are able to induce virus neutralizing (VN) antibodies, prevent the virus replication in target cells, viremia, and clinical symptoms of the disorder [4–6], but a full protection is only achieved against homologous strains. Moreover, it has been shown that an attenuated vaccine virus can return to virulence and cause disease [7,8]. Transplacental spread of the attenuated vaccine virus from mother to fetuses has also been reported [9]. In contrast, the existing commercial inactivated vaccines are safe, but do not protect animals against infection, even after challenge with homologous strains [10–15]. Our laboratory is doing a lot of efforts to design new vaccines based on new insights in the PRRSV infection pathogenesis and immune response [3]. As a result, an inactivated PRRSV vaccine has recently been developed [15]. During the development of this vaccine, the following aspects were fulfilled: (1) a quality-controlled viral inactivation procedure was designed and applied assuring complete killing of PRRSV, while conserving epitopes of the virus which are important for the induction of a protective immunity; (2) a high dose of inactivated virus was used; and (3) a suitable adjuvant was used. Vaccination of young pigs with the experimental vaccine resulted in a strong VN antibody response by vaccination on itself with a clear reduction of viremia after homologous PRRSV challenge [15]. If this vaccine would also be able to prevent congenital infection, it might become an interesting candidate vaccine for use in pregnant animals.

Preventing the virus spread from dam to fetus is an important step toward control of PRRSV-related reproductive problems characterized by late-term abortions, early farrowing, and an increase in the number of dead and mummified fetuses and weak-born piglets. Congenital PRRSV infection has a unique pathogenesis because fetuses are susceptible to PRRSV at any stage of gestation after direct inoculation [16,17]; however, PRRSV crosses from dam to fetus and induces reproductive failures mainly in late gestation [18,19]. The exact mechanism of the virus-induced reproductive failure remains unknown. The absence of severe microscopic lesions in the internal organs of aborted fetuses or stillborn piglets suggests that fetal death is not a direct result of PRRSV replication in the internal organs [20–22]. Recent findings have shown that PRRS efficiently replicates in the fetal implantation sites (endometrium/fetal placenta) during late gestation and causes apoptosis in infected and surrounding cells [23]. Hence, PRRSV infection in the fetal implantation sites might be a reason for fetal death.

At present, there is no standardized protocol to test PRRSV vaccines in pregnant animals. In previous studies, the postvaccination maternal immune response and viremia upon viral challenge were commonly examined, as well as the challenge virus in the fetal internal organs. Although this approach delivers valuable information concerning a vaccine performance, examination of the endometrium and fetal placenta [23,24] may improve the evaluation of the vaccine efficacy. Therefore, in the present study, an advanced protocol (based on the recent insights into pathogenesis of congenital PRRS infection) to test the performance of the experimental inactivated vaccine in pregnant gilts was used.

2. Materials and methods

2.1. Vaccine preparation

The PRRSV strain 07V063 (GenBank No: GU737264) was used for vaccine production and challenge [25]. Propagation, purification, binary ethylenimine-inactivation, and analysis of the complete virus inactivation were performed as previously described [15].

2.2. Experimental set-up and sample collection

Six gilts from a PRRSV-free herd were randomly assigned to two groups and were kept in isolation rooms. Three gilts (V1, V2, and V3) were intramuscularly vaccinated three times at 27, 55, and 83 days of gestation with 2 mL of the experimental vaccine (1 mL of the inactivated virus in a 1 mL oil-in-water diluent that is used in the commercial pseudorabies virus vaccine Suvaxyn Aujeszky, Fort Dodge Animal Health, Kelmis, Belgium), containing 10^8 tissue culture infectious dose 50 ($TCID_{50}$) of the binary ethylenimine-