



## Virological and serological characterization of vaccinated and non-vaccinated piglet subpopulations coming from vaccinated and non-vaccinated sows



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### ABSTRACT

The present study describes the virological and serological profiles of PCV2 vaccinated (V) and non-vaccinated (NV) piglet subpopulations coming from V and NV sows in a PCV2 subclinically infected farm. Four hundred seventy-six piglets born from V or NV sows were further subdivided in a total of four groups: NV sows–NV pigs (NV–NV), NV sows–V pigs (NV–V); V sows–NV pigs (V–NV) and V sows–V pigs (V–V). Seventy-five pigs were randomly selected at the beginning of the trial from each group and they were bled at 4, 8, 12, 16, 21 and 25 weeks of age. All animals included in the trial were weighed at 4 and 25 weeks of age and their average daily weight gain (ADWG) was calculated. Serum samples obtained at different time points were used to assess PCV2 infection (viremia) and the level of antibodies by means of immunoperoxidase monolayer assay (IPMA) against this pathogen. IPMA titers (classified in high, medium or low) and PCR results (positive or negative) were analyzed using a multiple correspondence and K-means cluster analysis. According to these tests, animals included in the study were classified into the following four clusters: (1) 93 piglets that were viremic mainly from 12 to 25 weeks of age and with PCV2 antibody titers increasing over time; (2) 75 piglets with late PCV2 infection and seroconversion (later than 16 weeks of age); (3) 26 piglets with high but decreasing PCV2 antibody titers and low percentages of PCV2 PCR positive serum samples; and (4) 105 piglets with medium and high IPMA titers throughout the trial and sporadic PCR positive samples. The defined subpopulations of piglets were observed in all experimental groups (NV–NV, NV–V, V–NV and V–V) although in variable percentages. Thus, animals from clusters 1 and 2 belonged mainly to the NV–NV and V–NV groups and animals from clusters 3 and 4 were distributed mainly into the NV–V and V–V groups. Finally, the ADWG of pigs belonging to clusters 3 and 4 was significantly higher ( $p=0.02$ ) than that of pigs belonging to clusters 1 and 2. Within each cluster, no statistically significant differences were found in ADWG between treatment groups.

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## 1. Introduction

Porcine circovirus type 2 (PCV2)-systemic disease (SD) is considered one of the major swine diseases worldwide. Infection with PCV2 is a necessary but not sufficient condition for pigs to develop the disease. Thus, management factors are important for disease development (Woodbine et al., 2007; Alarcon et al., 2010) and secondary pathogens, such as *Mycoplasma hyopneumoniae*, porcine reproductive and respiratory syndrome virus (PRRSV) and porcine parvovirus (PPV), are also considered infectious risk factors for disease triggering (Opriessnig et al., 2004). Traditionally, PCV2-SD control was based on counteracting infectious and non-infectious triggering factors, such as management improvement, control of co-infections and changes of the boar genetic background (Grau-Roma et al., 2011). However, PCV2 vaccines have demonstrated to be very efficient to control PCV2-SD and PCV2 infections under experimental and natural conditions (Beach and Meng, 2012).

PCV2 vaccines can be applied in sow and/or in piglets (Grau-Roma et al., 2011). PCV2 sow (and/or gilt) vaccination leads to an increase of PCV2 antibody titers in sow serum, a reduction in viremia as well as viral shedding in milk and colostrum and an improvement of piglet production records (mortality rates and average daily weight gain [ADWG]) (Gerber et al., 2011; Kurmann et al., 2011). Piglet PCV2 vaccination results in a significant reduction of viral-induced microscopic PCV2-SD lymphoid lesions, decrease mortality and cull rates, decrease the frequency of co-infections and improve ADWG (Segalés et al., 2009; Martelli et al., 2011). Indeed, a meta-analysis study of 66 published field trials found a positive effect of all PCV2 commercially available vaccines on productivity, with no statistically significant differences among them (Kristensen et al., 2011). Alternatively, a not yet extensively studied strategy is the double (sow and piglet) PCV2 vaccination (Opriessnig et al., 2010; Pejsak et al., 2010; Fraile et al., 2012a). This strategy achieved the best productive results although no statistically significant differences were observed in comparison with a protocol based only in piglet vaccination (Fraile et al., 2012a). On the other hand, a recent publication (Oh et al., 2014) confirmed that the combination of sow and pig (at 49 days of age) vaccination significantly reduced PCV2 viremia, induced higher neutralizing antibody titers, and resulted in higher proportion of CD4(+)CD8(+)IFN- $\gamma$ (+) lymphocyte subsets in piglets compared to the other groups of animals tested (non-vaccinated piglets coming from vaccinated sows, only vaccinated piglets at 21 or 49 days of age, and piglets vaccinated at 21 days of age coming from vaccinated sows). Thus, the combination of sow and pig (49 days of age) vaccination could be more effective for controlling PCV2 infection if PCV2 the infection occurs during the growing–finishing period in herds. From an economical perspective, PCV2 vaccination is considered a worldwide great success. However, these vaccines are imperfect in the sense that they prevent clinical signs but not infection (Kekarainen et al., 2010). Moreover, the seroconversion elicited by the vaccine can be affected by the antibody titers (Fraile et al., 2012a,b) and the vaccine efficacy may depend on the timing of natural infection (Beach and Meng, 2012). As a

consequence, in a PCV2 vaccinated community, subpopulations of animals not equally protected in front of a natural PCV2 challenge may exist. Optimization of PCV2 vaccination protocols would probably require a better characterization of piglet subpopulations coming from piglet, sow or both vaccination programs. To the authors' knowledge, this kind of information is not yet available in the literature. Thus, the main goal to this research work was to characterize, by means of PCV2 antibody and virological profiles, the different piglet subpopulations generated by piglet and/or sow vaccination. A secondary objective was to describe the ADWG observed in the different piglet subpopulations.

## 2. Material and methods

### 2.1. Study design

Data analyzed in this study was taken from a previously published field study (Fraile et al., 2012a). Briefly, the study was conducted in a 1500-sow Spanish farm with clinical history of PCV2-SD in which no PCV2 vaccination had been ever used. Pigs included in the study were born from Landrace (50%)  $\times$  Large white (50%) sows mated with Pietrain (100%) boars. One month before the beginning of the trial, PCV2-SD was diagnosed (fulfilling the internationally accepted disease case definition) (Segalés et al., 2005) in 5 out of 10 animals showing a wasting condition. One week before mating, a population of 57 sows was distributed into two groups: vaccinated (V,  $n=26$ ) and non-vaccinated sows (NV,  $n=31$ ). V sows received 2 ml of Porcilis® PCV (MSD Animal Health, The Netherlands) and NV received 2 ml of phosphate buffer saline (PBS). The parity average was not significantly different between V (3.1) and NV (3.2) sows; animals with different treatments were mingled in the same farrowing and gestation units. Blood samples were taken from the sows 2 weeks pre-mating and at 3, 10 and 17 weeks post-mating. All healthy piglets ( $n=476$ ) born from these 57 sows were included (at 4 weeks of age) in a sow and/or piglet vaccination strategy as follows: NV sows–NV pigs (NV–NV,  $n=134$ ), NV sows–V pigs (NV–V,  $n=135$ ); V sows–NV pigs (V–NV,  $n=104$ ) and V sows–V pigs (V–V,  $n=103$ ). The housing and husbandry conditions were standard for pigs reared under intensive conditions in Spain. Briefly, animals were housed in confinement with controlled environmental conditions (temperature and ventilation). In particular, cross-fostering was not allowed for the litters under study and the weaning age was close to 4 weeks of age ( $27 \pm 2$  days). At weaning, V piglets received 2 ml of Porcilis® PCV by intramuscular route and NV ones received 2 ml of PBS by the same route. Piglets with different treatment were mingled in the same growing and finishing units. Mortality during the study was recorded. No evidence of PCV2-SD was observed in the studied batch. The sampling size of the study was carried out to be able to detect differences in ADWG close to 20 g/day between vaccinated and non-vaccinated piglets (Fraile et al., 2012a). Nevertheless, the final number of piglets included in this study allowed having statistical power to detect differences between the proportion of the piglets present in the different groups equal or higher than 15%.

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