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Salmonella transmission from the gilt to her offspring

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ABSTRACT

The identification of gilts as a key factor in the salmonellosis dynamics is an important issue to the implementation of specific control programs in herds. This paper aims to assess the transmission of *Salmonella enterica* from the gilt to her offspring. The study was carried out in a multiple sites farrow-to-finish farm, built before the study to house 4500 sows, populated gradually with gilts weaned with less than 9 days of age. To determine the *Salmonella* infection prevalence in gilts, 1000 blood samples, 719 fecal samples and 236 mesenteric lymph nodes were collected from ten groups of gilts at an average age of 150 days. After that, a longitudinal study of the newborn piglets from the breeding herd was carried out for 3 consecutive weeks, which were followed from 10 to 150 days of age by serology (ELISA) and bacteriology (ISO 6579/02). The relatedness among the *Salmonella* isolates recovered was determined by *XbaI*-PFGE. A significant variability in the average of seropositive gilts among groups (from 0.00 to 31.52%) and low *Salmonella* shedding (1.4%) were found in the breeding herd at 150 days of age, but a wide range of *Salmonella* serovars ($n=11$) were isolated from slaughtered gilts. In the serological profile of the offspring, none of the pigs were found seropositive between 35 and 90 days of age, and bacteriology allowed to recover *S.* Derby from pigs only after 90 days of age. This suggests that offspring infection may not be taking place in the farrowing unit. The *S.* Schwarzengrund isolates recovered from gilts showed mainly the same *XbaI*-PFGE pattern, whereas *S.* Derby patterns of the strains obtained from gilts were different and also differed from the single *XbaI*-PFGE pattern isolated from the offspring. All these results suggest that serotype specific passive immunity would protect pigs from infection by *S. enterica* strains present in sows during their stay in the farrowing facilities, but fattening pigs can be infected by *Salmonella* from different sources of infection.

1. Introduction

Salmonella infections in swine is an important issue because of the impact on the production performance caused by the clinical presentation (salmonellosis), and also because of the implications for public health, of human disease attributed to consumption of contaminated pork and pork products (Griffith et al., 2006).

Specifically in swine production, salmonellosis is related to an increase in mortality rates and septicemic episodes, in addition to decrease in the production performance by affecting the daily weight gain and feed conversion, which economic cost has been estimated in £16.200/year/100 sow in United Kingdom (Muirhead and Alexander, 2001).

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The in-herd prevalence shows important differences between farms. In a meta-analysis based on 46 studies in 15 European and North American countries, Fosse et al. (2009) calculated a herd prevalence estimate of 21.8%, whlie a prevalence estimate of 6.7% was calculated for *Salmonella* shedding.

Most *Salmonella* control programs are based on monitoring systems used to assess the infection prevalence and the possible dynamics of infection in swine herds. These programs include serological testing, as well as, bacteriology testing of both fecal samples (FS) and mesenteric lymph nodes (MLN) (Arnold and Cook, 2009; Funk et al., 2001; Funk, 2003; Nowak et al., 2007; Rostagno et al., 2012).

The detection of specific antibodies against *Salmonella* spp. by ELISA in serum from pigs of different ages allows for the identification of time and level of the herd infection (Baum et al., 2005; Nielsen et al., 1995; Nollet et al., 2005), as well as the passive transfer of immunity from the sow to the newborn piglet (Kranker et al., 2003; Proux et al., 2000). Moreover, the bacteriology of FS allows for detection of pigs shedding *Salmonella enterica*, and the isolates recovered can be serotyped to determines serovars present in the herd. In addition, molecular analysis tools, such as pulsed-field gel electrophoresis (PFGE), can be applied to establish the genetic diversity, to infer or confirm transmission sources and pathways (Magistrali et al., 2008; Mannion et al., 2012; Rao et al., 2010; Weigel et al., 2007).

Through the study of risk factors and the analysis of key components, several aspects related to *Salmonella* transmission in swine herds have been identified, such as the farm biosecurity, hygienic measures, farm staff, animal health, the season and *Salmonella* serovar (Cardinale et al., 2010; Lurette et al., 2009; Lo Fo Wong et al., 2002; Rostagno and Callaway, 2012).

However, at present the role of *Salmonella* infection in sows, and more especially in gilts, related to on –farm transmission and dissemination still remains unclear (Funk and Gebreyes, 2004). Lurette et al. (2009) suggested that early infection, occurring between birth and weaning, is a critical point of *Salmonella* spread within a herd, whereas, to Kranker et al. (2003) the transmission at this stage may not be relevant, due in part to the presence of passive immunity in piglets. More specifically, acquired replacement gilts are considered by several authors to be an important source for introduction and dissemination of new *Salmonella* serovars in a herd (Davies et al., 2000; Silva et al., 2006), although, others consider it unlikely (Penmetchsa et al., 2009).

The identification of gilts as a key factor in the salmonellosis dynamics is a very important issue for the implementation of specific control programs in herds and swine production. Therefore, this paper aims to assess the transmission of *S. enterica* from the gilt to her offspring, by molecular subtyping and serology.

2. Materials and methods

The study was carried out in a multiple sites farrow-to-finish farm that had strict biosecurity measures. The farm had been recently built before the study to house 4500 sows and had a site used only for boars. The farm was populated gradually with groups of 760 gilts per week, with a total of 10 groups included in the study. All the gilts were brought from the same farm, they shared the same genetics and had been weaned with less than 9 days of age. Previous to the weaning, each piglet was medicated with tulatromicine (Draxxin, Pfizer Animal Health), following to the manufacturer´s recommendations. Each weekly group was taken to an individual room equipped to house the gilts from 9 to 150 days of age, at this time, the gilts that would constitute the breeding herd were selected. The gilts that were not selected were sent to slaughter.

2.1. Determination of Salmonella infection prevalence in the gilts

The software *WIN Episcope* 2.0 (www.clive.ed.ac.uk/winepiscope) was used to determine the number of gilts to be sampled, necessary to estimate the prevalence of the gilts shedding *Salmonella* spp. in every group. It is known that *Salmonella* shedding in infected animals is intermittent (Nielsen et al., 1995), which could lead to the shedding prevalence at certain time to be lower than the number of infected animals (Fosse et al., 2009). Because of that, we considered an expected prevalence of 8%, with 95% confidence level and 5% error, resulting in $n=100$ animals per group. The 100 gilts were selected at random in every group, at an average age of 150 days. Rectal fecal samples and blood samples (5 ml) were collected from each pig. The blood was placed in tubes, where the serum was separated and then kept at -20 °C. The fecal samples were refrigerated until they were processed. A total number of 1000 blood samples from the 10 groups and 719 fecal samples from 8 gilt groups were collected during a 10-week sampling period.

Mesenteric lymph nodes (MLN) were taken from 236 gilts that were sent to the slaughterhouse, following the procedures proposed by the European Union (Anonymous, 2006).

2.2. Determination of Salmonella infection prevalence in the offspring

A longitudinal study of the newborn piglets from the selected breeding herd was carried out for three consecutive weeks (weeks 1, 2 and 3). Blood samples from 10 pigs were taken at 10, 35, 56, 90, 120 and 150 days of age in each week. The number of samples necessary to detect pigs infected with *Salmonella* spp., was estimated at an expected prevalence of 25%, with 95% confidence level. At least 4–6 pigs were randomly selected and necropsied on each sampling day. MLN and contents of cecum (CC) samples were collected from each pig.

2.3. Sample processing

2.3.1. Serology

All serum samples were analyzed for the presence of *Salmonella* antibodies using a commercially available indirect mix-ELISA (Herd-Check Swine *Salmonella*, IDEXX Laboratories, Inc., Maine, USA) according to the manufacturer´s instructions. This kit combines different LPS