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Serological examination of fattening pigs reveals associations between *Ascaris suum*, lung pathogens and technical performance parameters

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ABSTRACT

Diagnosing the presence of the highly prevalent and economically important pig parasite *Ascaris suum* on fattening farms has so far been challenging. Currently, only the number of livers affected at slaughter is routinely used to measure parasite exposure. However, recently, a new serological test was developed based on the detection of antibodies to the *A. suum* haemoglobin molecule. The test showed to be highly sensitive for the detection of exposure to *A. suum* in fattening pigs. In this study we first compared the performance of *A. suum* serology versus the percentage of affected livers at slaughter, subsequently we investigated potential associations between *A. suum* infection levels and exposure to important lung pathogens and finally we identified correlations between serological data and technical performance parameters (TPIs) from 20 Belgian and 20 German pig fattening farms. In both Belgian and German farms, a significant relationship was detected between elevated average *Ascaris* serology and percentages of affected livers ($\rho = 0.63$ and $\rho = 0.75$, respectively). On the Belgian farms, both *Ascaris* serology and the percentage of affected livers were negatively correlated with average daily gain (ADG) ($\rho = -0.69$ and $\rho = -0.56$, respectively). Using the German dataset, only a borderline negative association was detected between the percentage of affected livers and the ADG ($\rho = -0.44$, $P = 0.053$). In contrast, only in the German farms, correlations between the percentage of affected lungs at slaughter and elevated presence of *A. suum* and several other airway pathogens were detected. To conclude, this study indicates that serological screening for *A. suum* on fattening farms is an attractive new diagnostic tool that can be used to indicate the presence of roundworm infection by measuring infection intensity. Furthermore the results of this study also add weight to the evidence that both roundworm infections as well as herd exposure to airway pathogens have a significant impact on farm productivity and hence, that all these factors should to be taken into account when assessing pig health and farm productivity.

1. Introduction

Although several studies have previously reported that infections with the intestinal roundworm *Ascaris suum* result in significant economic losses to the pig industry (Bernardo et al., 1990; Hale et al., 1985; Kipper et al., 2011; Knecht et al., 2012; Miskimins et al., 1994; Nilsson, 1982; Stewart and Hale, 1988; Van Meensel et al., 2010), the prevalence of this parasite in commercial pig farms remains high (Eijck and Borgsteede, 2005; Haugegaard, 2010; Joachim et al., 2001; Vlaminck et al., 2012). This is mainly because an *Ascaris* infection is in most cases a

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subclinical disease and, second, exposure of animals to this parasite cannot unambiguously be diagnosed (reviewed by Vlaminck et al., 2014).

Recently, we reported on the development and evaluation of a serodiagnostic test that can be used to measure parasite exposure of fattening pigs more accurately (Vlaminck et al., 2012). The test is based on the recognition of a haemoglobin protein (AsHb) produced by the parasite by systemic IgG antibodies of exposed animals and has shown to have a diagnostic sensitivity and specificity in experimentally infected pigs of almost 100% (Vlaminck et al., 2012). Further evaluation of the test under field conditions showed that it had a superior sensitivity for the detection of *A. suum* infections in comparison to faecal egg counts (Vlaminck et al., 2012). Faecal egg counts are however hardly ever used to diagnose *A. suum* infection in practise, as adult worm populations are extremely over-dispersed within a group of exposed pigs. On the other hand, the percentage of livers showing white spots at slaughter is widely used as a parameter to estimate *A. suum* infection levels in fattening pigs. Therefore, the first aim of this study was to compare the performance of this new serological test with the percentage affected livers at slaughter. As other infectious diseases causing lung pathology have also shown to reduce growth and carcass quality of fattening pigs (Er et al., 2014; Kristensen et al., 2011; Nieuwenhuis et al., 2012; Ostanello et al., 2007; Schlepers et al., 2013) and the fact that general inadequate hygiene management may negatively affect the infection pressure of both lung pathogens as well as parasitic infections, the second objective of this study was to investigate potential associations between *A. suum* infection levels and exposure to the most important lung pathogens including Porcine Reproductive and Respiratory Syndrome Virus (PRRSV), *Mycoplasma hyopneumoniae* (*M. hyo*), Swine Influenza Virus (SIV), *Actinobacillus pleuropneumonia* (APP) and Porcine Circovirus Type 2 (PCV2), as measured by serology. Finally, since parasitic- and lung infections have both been associated with reduced farm productivity, the final aim of this study was to investigate potential associations between serology for all tested pathogens and technical performance parameters (TPIs) including average daily growth (ADG) and feed conversion efficiency (FCE) in fattening pigs.

2. Materials and methods

2.1. Selection of the farms

In Belgium, 20 pig farms were selected that were part of the integration of the cooperative Covavee cvba. In Germany, 20 pig farms were selected from the Southeast of Bavaria and were all a member of the Bavarian grower association. All selected farms were conventional indoor fattening farms. In the Belgian farms, pigs were fattened from approximately 10 weeks old till slaughter weight was reached. Pigs were kept on a fully- or semi-slatted concrete floor and an all-in/all-out management system was employed. Of the 20 German farms, 5 farms were farrow-finish farms; the other 15 farms fattened their pigs starting from approximately 10 weeks old. Additional information on the housing or other management factors of the German farms was unavailable.

2.2. Collection of blood samples

A total of 20 (Belgium) or 15 (Germany) blood samples were randomly collected at the slaughter line from pigs from one fattening period of each selected farm. The blood was collected in a blood tube and after transportation to the lab, blood samples were allowed to clot for 1 additional hour at 37 °C and subsequently refrigerated for another hour. Samples were spun at 4,000 × *g* during 5 min at 4 °C and serum was collected and frozen at −20 °C until used.

2.3. Slaughter line data and technical performance indicators (TPIs)

Liver and lung lesions were assessed by official meat inspection personnel during routine post-mortem meat inspection at the slaughterhouse. The percentage of affected livers was calculated as the percentage of pig livers from one slaughtered batch of pigs from one farm on which the meat inspection personnel reported the presence of at least 1 white spot lesion. The percentage of affected lungs was calculated as the percentage of lungs from the same batch of pigs on which meat inspection personnel reported visible pneumonic lesions. Average daily growth and average feed conversion efficiency of that particular round of pigs was provided by Covavee cvba for 14 out of the 20 Belgian farms, and by the Federal Bavarian Consultancy (LKV Bayern) for the 20 German farms.

2.4. Analysis of the serum samples

The *Ascaris* ELISA test was essentially performed as described in Vlaminck et al. (2012). ELISA results are reported both as average sample to positive (S/P) value per farm (S/P sample = $(OD_{sample} - OD_{NegativeControl})/(OD_{PositiveControl} - OD_{Negativ\ \ Control}))$ and percentage of the animals that tested positive. The serology for mycoplasma, PRRSV, SIV, APP2 and PCV2 was performed with commercially available test kits (summarized in Table 1) according to manufacturers' instructions.

2.5. Statistical analysis

Associations between the different variables were investigated using the Spearman's rank correlation test and linear regression. Probability (*P*) values < 0.05 were considered to indicate significant correlations. Multivariate linear regression analyses were conducted to assess the associations between the dependent variable (% affected livers and lungs, ADG and FCE) and the independent variables (average farm S/P for *Ascaris*, APP2, PRRSV, PCV2, SIV, *M. hyo*). No herd management factors were included in these analyses. The herd was the unit of analysis. A forward stepwise selection procedure was used to select the variables that were significantly (*P* < 0.10) associated with the different independent variables tested. The variable that resulted