The effect of a porcine reproductive and respiratory syndrome outbreak on genetic parameters and reaction norms for reproductive performance in pigs[1]

Austin M. Putz,[†,2] Clint R. Schwab,[‡] Alysta D. Sewell,[‖] Derald J. Holtkamp,[§] Jeffery J. Zimmerman,[§] Kimberlee Baker,[§] Nick V. L. Serão,[†] Jack C. M. Dekkers[†]

[†]Department of Animal Science, Iowa State University, Ames, IA 50011; [‡]The Maschhoffs, 7475 State Route 127, Carlyle, IL 62231; [‖]Purina Animal Nutrition LLC, 100 Danforth Drive, Gray Summit, MO 63039; and [§]Department of Veterinary Diagnostics and Production Animal Medicine, College of Veterinary Medicine, Iowa State University, Ames, IA 50011

ABSTRACT: The objective of this study was to estimate genetic parameters of antibody response and reproductive traits after exposure to porcine reproductive and respiratory syndrome virus. Blood samples were taken approximately 60 d after the outbreak. Antibody levels were quantified as the sample-to-positive ratio (S/P ratio) using a fluorescent microsphere assay. Reproductive traits included total number born (**TNB**), number born alive (**NBA**), number stillborn (**NSB**), number mummified (**NBM**), and number born dead (**NBD**). Mortality traits were log transformed for genetic analyses. Data were split into prior, during, and after the disease outbreak phases using visual appraisal of the estimates of farm-year-week effects for each reproductive trait. For NBA, data from all phases were combined into a reaction norm analysis with regression on estimates of farm-year-week effects for NBA. Heritability for S/P ratio was estimated at 0.17 ± 0.05. Heritability estimates for reproduction traits were all low and were lower during the outbreak for NBA but greater for mortality traits. TNB was not greatly affected during the outbreak, as many sows that farrowed during the outbreak were mated prior to the outbreak. Heritability for TNB decreased from 0.13 (prior) to 0.08 (after). Genetic correlation estimates between prior to and during the outbreak were high for TNB (0.86 ± 0.23) and NBA (0.98 ± 0.38) but lower for mortality traits: 0.65 ± 0.43, −0.42 ± 0.55, and 0.29 ± 1.39 for LNSB, LNBM, and LNBD, respectively. TNB prior to and after the outbreak had a lower genetic correlation (0.32 ± 0.33). In general, genetic correlation estimates of S/P ratio with reproductive performance during the outbreak were below 0.20 in absolute value, except for LNSB (−0.73 ± 0.29). Based on the reaction norm model, estimates of genetic correlations between the intercept and slope terms ranged from 0.24 ± 0.50 to 0.54 ± 0.35 depending on the parameterization used, indicating that selection for the intercept may result in indirect selection for steeper slopes, and thus, less resilient animals. In general, estimates of genetic correlations between farm-year-week effect classes based on the reaction norm model resembled estimates of genetic correlations from the multivariate analysis. Overall, compared to previous studies, antibody S/P ratios showed a lower heritability (0.17 ± 0.05) and low genetic correlations with reproductive performance during a porcine reproductive and respiratory syndrome outbreak, except for the LNSB.

Key words: antibody, litter size, PRRS, S/P ratio, reaction norm, genetic parameters

J. Anim. Sci. 2019.97:1101–1116
doi: 10.1093/jas/sky485

[1]This project was funded by the Iowa Pork Producers Association.

[2]Corresponding author: putz.austin@gmail.com

Received September 23, 2018.
Accepted December 21, 2018.

INTRODUCTION

Porcine reproductive and respiratory syndrome virus (**PRRSV**) severely affects both the breeding and growing sectors of the swine industry. In the breeding sector, PRRS causes abortions, stillborns, mummified piglets, pre-weaning mortality, and embryonic death (Lunney et al., 2016). It was estimated that $302 million (~45%) out of the annual $663 million in costs associated with problems caused by PRRS are due to losses in the breeding sector (Holtkamp et al., 2013). This was very different than estimates from a previous study by Neumann et al. (2005), where only ~12% was due to breeding herd losses. A relatively large amount of work has been done on the growing pig sector to reduce the economic impact of PRRS (Lunney et al., 2011) and less attention has been focused on reducing the effects of the disease in breeding herds (Lunney et al., 2016). One reason is that, prior to the availability of high-density genotyping, genetic analyses required a pedigree, which is typically not available for sows at the commercial level due to pooling of semen. Because nucleus and multiplier herds are managed to maximize biosecurity and minimize the risk of exposure to major pathogens such as PRRS, studies on outbreaks in herds with pedigreed sows are rare. Although pedigrees are not required with genomics, genotyping is still relatively expensive, and it still requires high-quality data, which is typically collected in nucleus herds. It is also much more expensive to set up experimental infection trials for reproductive traits in sows, as done in the PRRS Host Genomics Consortium nursery pig trials (Lunney et al., 2011). Breeding for increased resistance to PRRS is difficult in growing pigs and the problem becomes even more difficult for reproductive performance.

Antibody level in sows following an outbreak with a PRRSV could be used as an indicator trait for selection. Serão et al. (2014) demonstrated that antibody level measured as sample-to-positive (S/P) ratio from a commercial IDEXX ELISA analysis of blood samples taken after a PRRS infection was highly heritable (0.45) and had moderate-to-strong genetic correlations with many reproduction traits during the outbreak (~0.7 in absolute value for several traits). Since antibody levels under a real challenge may be impractical for commercial breeding programs, Serão et al. (2016) suggested that antibody following vaccination with a modified live virus (**MLV**) could be used as an indicator trait to select for reproductive performance under PRRS (Madapong et al., 2017). To the best of our knowledge, Serão et al. (2014) is the only study that has investigated genetic relationships between antibody level and reproductive performance under a PRRSV challenge. Therefore, it is necessary to validate these findings in a larger, independent PRRSV outbreak study.

To date, multivariate and reaction norm models are the two main methods that have been used for analysis of disease outbreak data. Lewis et al. (2009) first split reproductive data from a commercial herd that experienced a PRRS outbreak into healthy and PRRS phases and found that splitting the data based on trait rolling averages was better than using diagnostic lab confirmation dates. Estimated genetic correlations of reproductive performance between healthy and PRRS phases ranged from −0.13 to 0.98, although many genetic correlations were moderate or low between phases (Lewis et al., 2009). Lewis et al. (2009) only separated traits into two phases, while it is known that PRRS is a persistent infection (Wills et al., 2003; Lunney et al., 2016), suggesting that the post-outbreak phase may need to be considered as a separate phase, creating three phases (prior, during, and after the outbreak). In addition, estimates of genetic correlations between reproductive traits within phase may also give some insight into how disease changes the relationship among traits during the different phases of an outbreak.

Reaction norm models are a common way to model genotype-by-environment interactions (G × E) but they have only more recently been utilized for litter size in pigs. Reaction norm models are an application of random regression (longitudinal) models that regress the response variable on a continuous environmental variable. These reaction norm models yield estimates of breeding values for an intercept term that is highly correlated to estimates of breeding values from routine genetic evaluations (Knap and Su, 2008) and estimated breeding values for a slope term that describes the additive genetic sensitivity to changes in the environment (when using an additive genetic relationship matrix). Reaction norm models have been used to analyze disease outbreak data by regressing phenotypes on estimates of continuous farm-year-week effects or on an index of challenge load from multiple traits (Mathur et al., 2014; Rashidi et al., 2014; Herrero-Medrano et al., 2015).

The objectives of the current study were to (i) estimate the genetic parameters of reproduction traits prior, during, and after a PRRSV outbreak, and among traits within each phase, (ii)

estimate heritability and genetic correlations of sample-to-positive (S/P) ratio from blood during the PRRS outbreak with reproductive performance during the outbreak to validate findings from Serão et al. (2014), and (iii) evaluate a reaction norm model to model the effect of PRRS on NBA.

MATERIALS AND METHODS

Blood sampling of sows was approved by the Institutional Animal Care and Use Committee (IACUC, 4-15-8006-S). Reproductive data was retrieved from an existing database that included data collected as part of a routine breeding program and, therefore, did not require approval from an animal care and use committee.

Outbreak, Inoculation, and Antibody test

Three breeding farms from The Maschhoffs (Carlyle, IL), located in close proximity to each other in Illinois, USA, broke with a PRRSV strain in the spring of 2015. These farms included pedigreed purebred Yorkshire (**YORK**) and Landrace (**LR**) sows. Farms 1 and 3 contained both breeds, while farm 2 contained only the LR breed. After suspect abortions, samples were sent for diagnostics and the 1-7-4 restriction fragment length polymorphism (**RFLP**) pattern (strain) of PRRS was confirmed by PCR analysis. This strain is a highly virulent strain. A nearby farm first broke with this PRRSV strain. To protect some high indexing sows in farm 1, they were preemptively sent to a quarantine facility to be tested for PRRS. After clearing these tests, these sows were then transferred to one of the other farms to farrow. Farm 1 was confirmed positive on March 5 and was then depopulated to try to protect other nearby farms 2 and 3. Subsequently, farms 2 and 3 broke with PRRS and were then closed to new animals. Farm 2 was confirmed positive on April 16 (42 d after farm 1) and farm 3 on April 9 (35 d after farm 1). After the initial confirmed outbreak with several positive samples, all sows on farms 2 and 3 were inoculated with live virus of a 1-7-4 RFLP pattern (strain) that was isolated at each farm (same strain) approximately 3 wk later, on May 5 for farm 2 and on April 30 for farm 3. Inoculation was intranasal at farm 2 and intramuscular at farm 3. All sows were injected with an MLV vaccine ~30 d after the inoculation. Blood samples for antibody levels were taken from the anterior vena cava with vacutainer serum tubes from sows on farm 2 on June 18 and from farm 3 on June 16, 17, and 19. This was ~60 d after the initial outbreak. Antibody levels taken at this time point should have plateaued for most animals (Lunney et al., 2016). Serum tubes were centrifuged at the farm and these serum samples were sent to Kansas State University for analysis. Antibody level against the PRRSV N-protein was measured using a fluorescent microsphere immunoassay (Luminex) and converted into a standardized sample-to-positive (S/P) ratio using positive and negative controls. This assay is conceptually similar to an indirect ELISA.

Reproductive Data and Phases

Reproductive data obtained from routine data collection in these breeding herds included total number born (**TNB**), number born alive (**NBA**), number stillborn (**NSB**), number mummified (**NBM**), and number born dead (**NBD**; the sum of NSB + NBM). Raw weekly means for each farm are presented in Figure 1. For genetic analyses, mortality traits NSB, NBM, and NBD were all log transformed as ln(phenotype + 1) (Lewis et al., 2009) and will be referred to as LNSB, LNBM, and LNBD, respectively. Records on reproductive traits were separated into three phases (prior, during, and after the outbreak) based on estimates of farm-year-week (**FYW**) effects (extracted from the farrowing date) for each reproductive trait that were obtained from the following linear mixed model for each trait separately

$$y_{ijklm} = \mathrm{PAR}_i + \mathrm{FARM}_j + \mathrm{BREED}_k + \mathrm{fyw}_l + \mathrm{sow}_m + e_{ijklm},$$

where PAR is the parity effect (i=1,…,8), FARM is the farm effect (j=1,2,3), and BREED is the breed effect (k=1,2), which were fitted as fixed effects, while fyw is the random FYW effect, assumed to follow ~$N(0, \boldsymbol{I}\sigma^2_{\mathrm{fyw}})$ in which σ^2_{fyw} is the FYW variance and $\boldsymbol{I}$ is an identity matrix, and sow is a random sow effect, assumed to follow ~$N(0, \boldsymbol{I}\sigma^2_{\mathrm{sow}})$ in which σ^2_{sow} is the sow variance (following Rashidi et al., 2014). To make all traits comparable, estimates of FYW effects for each trait were standardized by their respective overall SDs (based on the variance estimated for FYW using lme4 in R; Bates et al., 2015) and plotted over time (Figure 2). Visual appraisal was used to split the data for each trait into three phases because it was known when the outbreak occurred (dates given above, similar to Serão et al., 2014). Preliminary analysis showed only minor changes in variance component estimates (<0.1 for genetic correlations) when slightly different dates (by 1 or 2 wk) were used to split

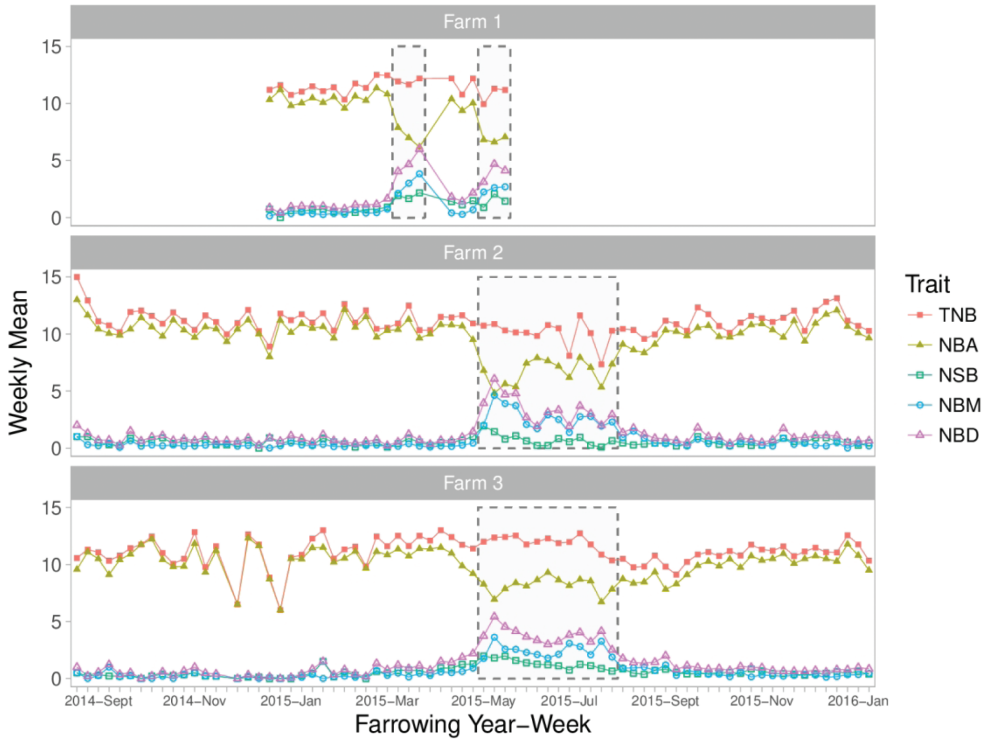

Figure 1. Weekly means for litter size traits from each farm. Shaded boxes represent the PRRS outbreak phase.

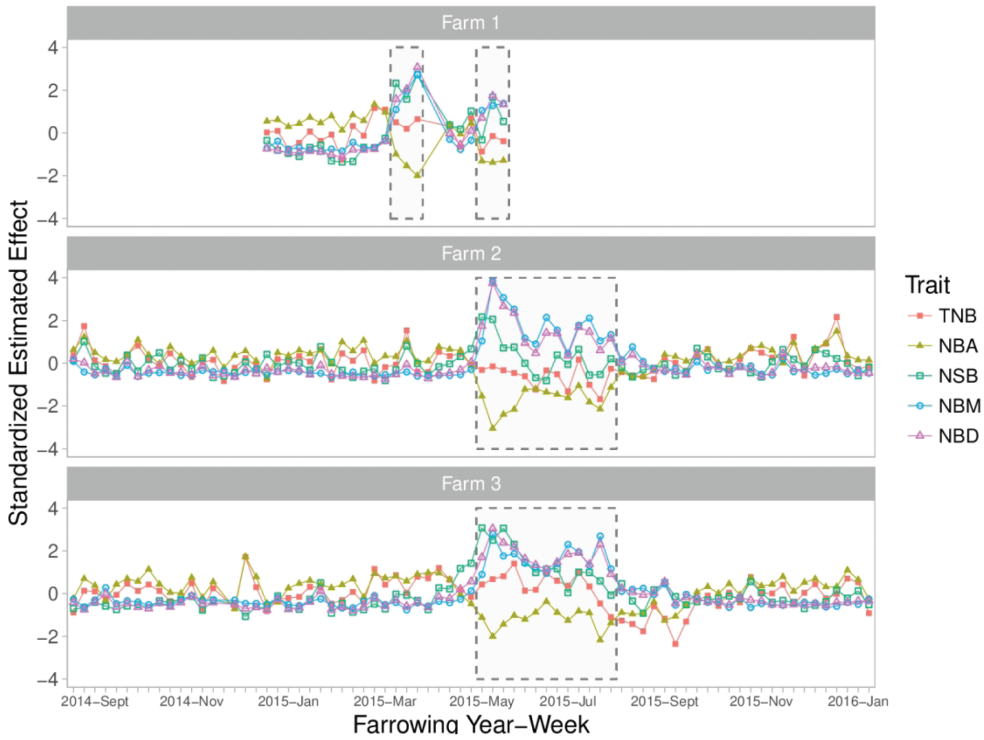

Figure 2. Standardized estimates of farm-year-week random effects for litter size traits from a linear mixed model. Shaded boxes represent the PRRS outbreak phase.

the data into phases because these different dates changed the data sets very little. The PRRS outbreak phase for farm 1 was identified to be from March 12 to April 1 (20 d) and also from May 7 to May 27 (20 d), after sows were moved. The PRRS outbreak phase for farms 2 and 3 was from May 7 to August 5 (90 d). Based on these dates, data from each reproductive performance trait were separated into three traits (prior, during, and after), which were designated with subscripts p, d, and a, respectively, on the trait acronym (e.g., TNB_p for TNB prior to the outbreak). Transition

periods were masked for this analysis by removing data just prior to and after the outbreak phase (following Herrero-Medrano et al., 2015) because these records represented a "grey area" for classification. For farm 1, data from 1 wk prior to the first outbreak and from the 3 wk between the two outbreaks were removed. For farm 2, data from 1 wk prior to and 2 wk after the outbreak were removed. For farm 3, data from 1 wk prior to the outbreak phase and 7 wk after the outbreak were removed. The latter were removed because rolling averages for TNB fluctuated continuously during these weeks, possibly from a rebound after the outbreak, which made it unclear how these data should be classified (see Figure 2).

Multivariate Variance Components

Variance components among traits were estimated both between phases (e.g., TNB_p with TNB_d) and within phase (e.g., TNB_d with NBA_d) by basic bivariate animal models, using ASReml4 (Gilmour et al., 2015). Heritability estimates for a trait were averaged over the bivariate analyses. The model used for reproductive traits was as follows:

$$\boldsymbol{y} = \boldsymbol{X\beta} + \boldsymbol{Zu} + \boldsymbol{e},$$

where $\boldsymbol{\beta}$ included the fixed effects of parity (1 through 8), farms (1, 2, or 3), breed (YORK, LR), and farm-year-month (FYM), and the 30-d rolling herd average of the trait analyzed as a fixed covariate (following Lewis et al., 2009). The vector $\boldsymbol{u}$ represents the random additive genetic effect of the sow [$\sim N(0, \boldsymbol{A}\sigma^2_{\text{sow}})$] where σ^2_{sow} is the sow variance and $\boldsymbol{A}$ is a matrix of additive genetic relationships among pigs, and the vector $\boldsymbol{e}$ represents the random residual term [$\sim N(0, \boldsymbol{I}\sigma^2_e)$]. Very few sows had repeated records for traits prior to and after the outbreak and in these cases, the second record in the dataset was removed such that a repeatability model was not needed. The final multivariate dataset included 2,014, 1,428, and 1,626 records for the prior, during, and after phases, respectively. The model used for S/P ratio (only recorded during the outbreak) also was a simple animal model, with parity, breed, date of sample collection (June 16 to 19), and the plate of the assay (96-well plates used) as fixed effects. Collection date was confounded with farm (see above) and, therefore, farm was not fit in the model. Random effects were the same as for the reproduction traits. The pedigree included at least three generations to calculate the numerator relationship matrix (**A**), for a total of 6,202 animals. Animal models for variance components were analyzed with ASReml 4 (Gilmour et al., 2015).

Reaction Norm Analysis

Reaction norms were used to analyze NBA by regressing on estimates of farm-year-week (FYW) effects for NBA (estimates ranging from −4.11 to 2.33) that were obtained using the animal models described above for each phase. For this analysis, the entire dataset was kept intact for each trait, without splitting it into phases. This dataset included 6,328 records from 3,378 sows. These sows recorded between one (1,397), two (1,209), three (575), and four (197) records (farrowings). The healthy phase started at approximately −1 on the FYW scale, which would include data from both prior to and after the PRRS outbreak phase. The model used was as follows:

$$\boldsymbol{y} = \boldsymbol{Xb} + \boldsymbol{Qa} + \boldsymbol{Zpe} + \boldsymbol{e},$$

where the fixed effects vector $\boldsymbol{b}$ included breed, farm, parity, status (prior, during, after), and the fixed covariate of FYW effect estimates for NBA, with corresponding design matrix $\boldsymbol{X}$. Matrix $\boldsymbol{Q}$ contains coefficients for the random additive genetic effects ($\boldsymbol{a}$), which included correlated random intercepts ($\boldsymbol{a_i}$) and random slopes ($\boldsymbol{a_s}$) on FYW effect estimates for each individual in the pedigree, connected through the pedigree relationship matrix. The variance–covariance structure of $\boldsymbol{a}$ was as follows:

$$\text{Var}[\boldsymbol{a}] = \text{Var}\begin{bmatrix}\boldsymbol{a_i}\\ \boldsymbol{a_s}\end{bmatrix} = \boldsymbol{G}_{RN} \otimes \boldsymbol{A} = \begin{bmatrix}\boldsymbol{A}\sigma^2_{a_i} & \boldsymbol{A}\sigma_{a_i,a_s}\\ \boldsymbol{A}\sigma_{a_i,a_s}\boldsymbol{A}\sigma^2_{a_s}\end{bmatrix},$$

where $\boldsymbol{G}_{RN}$ is the genetic (co)variance matrix, with σ_{a_i,a_s}, $\sigma^2_{a_i}$, and $\sigma^2_{a_s}$ denoting the covariance and additive genetic variances for intercept and slope, respectively. Three different scales were used for the random regression coefficients for the 170 unique FYW classes ($\boldsymbol{\Phi}$ matrix): (i) the raw scale of estimated FYW effects, (ii) Legendre polynomial terms from the FYW effects (leg function in ASReml), and (iii) orthogonal polynomial terms based on the pol function in ASReml (Gilmour et al., 2015). No information is reported on model fit, as these were equivalent models. Vector **pe** contains random permanent environmental effects for animals with records, with matrix **Z** being diagonal with only an

intercept term (1/0), as in a normal repeatability model. More complex models for the permanent environmental effects did not converge, possibly due to the low number of records per sow (Meyer, 2005). Finally, vector **e** contains residuals which, after a preliminary analysis (fitting many different group sizes), were fitted using heterogeneous residual variances with five discrete classes based on the NBA FYW effect estimates {−Inf, −2, −1, 0, 1, Inf}. Thus, the residual variance was structured as follows:

$$\text{Var}\begin{bmatrix}\boldsymbol{e}\end{bmatrix} = \text{Var}\begin{bmatrix}\boldsymbol{e}_1\\ \boldsymbol{e}_2\\ \boldsymbol{e}_3\\ \boldsymbol{e}_4\\ \boldsymbol{e}_5\end{bmatrix} = \begin{bmatrix}\boldsymbol{I}\sigma^2_{e_1} & 0 & 0 & 0 & 0\\ 0 & \boldsymbol{I}\sigma^2_{e_2} & 0 & 0 & 0\\ 0 & 0 & \boldsymbol{I}\sigma^2_{e_3} & 0 & 0\\ 0 & 0 & 0 & \boldsymbol{I}\sigma^2_{e_4} & 0\\ 0 & 0 & 0 & 0 & \boldsymbol{I}\sigma^2_{e_5}\end{bmatrix}.$$

Estimates of genetic correlations of NBA between FYW classes were obtained from the estimated genetic covariance matrix $\hat{\boldsymbol{G}}_{RN}$ using $\boldsymbol{\Phi}\hat{\boldsymbol{G}}_{RN}\boldsymbol{\Phi}'$, which results in a square, symmetric matrix with dimensions equal to the number of FYW effects that is used to calculate $\boldsymbol{\Phi}$. Estimated breeding values for each animal for each FYW level were calculated as $\boldsymbol{\Phi}_{(q\times 2)}\hat{\boldsymbol{U}}'_{(2\times n)}$, where $\hat{\boldsymbol{U}}$ is a matrix of estimates of the random intercept and slope effects from the reaction norm model, where q is equal to the number of FYW levels (170), and n is the number of animals in the pedigree (n = 6,451). For the current data, this resulted in a 170 × 6,451 matrix of EBVs.

RESULTS

Summary Statistics Across Phases

Table 1 shows means and SD for the five reproductive traits during the three phases (prior, during, and after). The average TNB was similar across phases, although slightly lower after the outbreak. All other traits were greatly affected by the PRRS infection. The average NBA dropped from 10.6 prior to the outbreak to 7.7 during the PRRS phase. The average NSB rose from 0.5 to 1.2 per litter during the outbreak. The average NBM went from 0.3 prior to the outbreak to 2.6 per litter during the outbreak. Finally, average NBD went from 0.8 to 3.8 during the PRRS outbreak. All four traits (removing TNB) returned to their pre-challenge average after the outbreak.

Identification of Outbreak Phases

Figure 1 displays the raw averages by FYW for each trait. The severity of the disease during the PRRS outbreak in the spring of 2015 is evident. Weekly means for TNB did not show large changes over time but did trend downward starting midway through the PRRS phase. All other traits were affected much more severely by the PRRS outbreak as expected. NBA dropped below 7 pigs in all three farms. For farm 2, the mean for NBD was higher than the mean NBA for 1 wk. Farm 2 had a spike in NSB immediately after the outbreak and then returned to a normal level after approximately 5 wk. Farm 3 was slightly less affected by the outbreak in terms of NBA and NBD. An important note is that the majority of the in utero mortality during the PRRS phase was due to mummies (68%) and not stillborns. Prior to and after the outbreak, the percent of deaths due to mummies was 36% and 42%, respectively. Standardized estimates of FYW effects followed the same trends as the raw means (Figure 2). The PRSS outbreak corresponded to spikes in mortality traits and drops in the estimates of FYW effects for TNB and NBA. The most extreme standardized effects were from farm 2, where some estimates were as high as four. Trends in estimates over time were similar for all traits, except for TNB. For farm 2, the NSB returned to baseline quicker than farm 3.

Heritability of Reproduction Traits Across Phases

Heritability estimates were ≤0.13 for all reproductive traits for all phases (Table 2). The estimate

Table 1. Mean reproductive performance prior to, during, and after the PRRS outbreak (with SD in parentheses)

Phase[1]	Count	TNB[2]	NBA[2]	NSB[2]	NBM[2]	NBD[2]
Prior	2,478	11.4 (3.4)	10.6 (2.3)	0.5 (0.9)	0.3 (0.7)	0.8 (1.3)
During	1,455	11.5 (3.5)	7.7 (4.0)	1.2 (1.6)	2.6 (3.3)	3.8 (3.6)
After	1,632	11.2 (3.4)	10.4 (3.3)	0.5 (0.9)	0.3 (0.9)	0.8 (1.3)

[1] Phases were split using a mixed linear model, fitting farm year week (FYW) as a random effect and extracting the predicted values. Visual appraisal was used to split phases into prior, during, and after the PRRS outbreak

[2] TNB = total number born, NBA = number born alive, NSB = number stillborn, NBM = number born mummified, NBD = number born dead.

of heritability for S/P ratio was also relatively low at 0.17 ± 0.05. Heritability estimates for mortality traits (LNSB, LNBM, and LNBD) ranged from 0.01 to 0.06 prior to the outbreak, increased during the outbreak (0.06 to 0.13), likely because of the higher incidence of mortalities during the outbreak, and then reduced again after the outbreak. However, only the estimate of heritability of LNBM returned to its estimate prior to the outbreak, while estimates for both LNSB and LNBD remained slightly elevated after the outbreak (0.09 ± 0.04 and 0.06 ± 0.04, respectively). The estimate of heritability of TNB (0.13 ± 0.05) did not change during the outbreak but reduced to 0.08 ± 0.04 after the outbreak.

Genetic Correlations of Reproduction Traits Between Phases

Estimates of genetic correlations of traits between the three phases are displayed in Table 3. Estimates of the genetic correlation between prior to and during the outbreak for TNB and NBA were >0.85, indicating similar genetic backgrounds. Genetic correlations for TNB and NBA were much lower between prior to and after the outbreak (0.32 ± 0.33 and 0.27 ± 0.42, respectively). Again, this may be expected when sows are being bred during the outbreak. The genetic correlation between TNB during and after the outbreak was higher (0.72 ± 0.55) than for NBA (0.21 ± 0.54). Estimates of genetic correlations for mortality traits prior to and during the outbreak were inconsistent; they were positive for LNSB and LNBD but negative for LNBM (−0.42 ± 0.55). Trends in estimates of genetic correlations for reproductive performance between prior to and after the outbreak were similar to those between during and after the outbreak.

Genetic Correlations Between Reproductive Traits and S/P Ratio

Estimates of genetic correlations of S/P ratio with reproductive traits are presented in Table 4. Prior to the outbreak, estimates of the genetic correlations ranged from 0.05 to 0.85, but with very large SE for LNBM and LNBD. Most estimates of the genetic correlation of S/P ratio with reproduction traits during the outbreak, which was of main interest, were close to zero, with the exception of LNSB, which had an estimate of −0.73 ± 0.29 with

Table 2. Heritability estimates (with SE in parentheses) for S/P ratio and reproductive traits prior to, during, and after the outbreak

Trait[1]	Prior[2]	During[2]	After[2]
TNB	0.13 (0.05)	0.12 (0.05)	0.08 (0.04)
NBA	0.11 (0.04)	0.05 (0.03)	0.05 (0.04)
LNSB	0.06 (0.03)	0.13 (0.06)	0.09 (0.04)
LNBM	0.03 (0.03)	0.12 (0.06)	0.03 (0.04)
LNBD	0.01 (0.02)	0.06 (0.04)	0.06 (0.04)
S/P Ratio[3]	NA[3]	0.17 (0.05)	NA[3]

[1] TNB = total number born, NBA = number born alive, LNSB = log number stillborn, LNBM = log number born mummified, LNBD = log number born dead, S/P = sample-to-positive ratio of the PRRS antibody levels.

[2]Phases were split using a mixed linear model, fitting farm year week (FYW) as a random effect and extracting the predicted values. Visual appraisal was used to split phases into prior, during, and after the PRRS outbreak.

[3] S/P ratio was only collected during the PRRS outbreak.

Table 3. Estimates of genetic correlations (with SE in parentheses) for reproductive traits between the three phases relative to the outbreak (prior to, during, and after)

Trait[1]	Prior–during[2]	Prior–after[2]	During–after[2]
TNB	0.86 (0.23)	0.32 (0.33)	0.72 (0.28)
NBA	0.98 (0.38)	0.27 (0.42)	0.21 (0.54)
LNSB	0.65 (0.43)	0.40 (0.41)	0.81 (0.28)
LNBM	−0.42 (0.55)	−0.40 (0.88)	−0.36[3]
LNBD	0.29 (1.39)	0.69 (1.52)	0.07 (0.48)

[1]TNB = total number born, NBA = number born alive, LNSB = log number stillborn, LNBM = log number born mummified, LNBD = log number born dead.

[2]Phases were split using a mixed linear model, fitting farm year week (FYW) as a random effect and extracting the predicted values. Visual appraisal was used to split phases into prior, during, and after the PRRS outbreak.

[3]Completed with remlf90 from BLUPF90 programs in place of ASReml due to convergence issues, no SE available.

Table 4. Estimates of genetic correlations (with SE in parentheses) of S/P ratio with reproductive traits prior to, during, and after the outbreak

Trait[1]	Prior[2]	During[2]	After[2]
TNB	0.27 (0.25)	−0.10 (0.26)	−0.17 (0.29)
NBA	0.19 (0.25)	0.05 (0.35)	−0.12 (0.32)
LNSB	0.05 (0.35)	−0.73 (0.29)	−0.06 (0.31)
LNBM	0.85 (0.90)	0.02 (0.42)	0.05[3]
LNBD	0.69 (0.79)	−0.18 (0.28)	−0.20 (0.32)

[1]TNB = total number born, NBA = number born alive, LNSB = log number stillborn, LNBM = log number born mummified, LNBD = log number born dead.

[2]Phases were split using a mixed linear model, fitting farm year week (FYW) as a random effect and extracting the predicted values. Visual appraisal was used to split phases into prior, during, and after the PRRS outbreak.

[3]Completed with remlf90 from BLUPF90 programs in place of ASReml due to convergence issues, no SE available.

S/P ratio. The negative genetic correlation estimates of S/P ratio with LNSB and LNBD during the outbreak were in the favorable direction (i.e., sows with higher antibody level are expected to have fewer stillborn pigs phenotypically/genetically). After the outbreak, estimates of genetic correlations of S/P ratio with reproductive traits were low (−0.20 to 0.05). Negative genetic correlations may be as expected because producing more antibody during the infection may have diverted resources away from reproduction while the sow was cycling during the outbreak, leaving fewer embryos/fetuses to develop and be born after the outbreak.

Genetic Correlations Among Reproduction Traits Within Phases

Genetic correlations among reproductive traits within phase are displayed in Table 5. TNB and NBA had high genetic correlations prior to and after the outbreak (>0.90) but the correlation dropped to 0.71 ± 0.16 during the outbreak, likely due to greater prenatal mortality during the outbreak. TNB was positively correlated, genetically, with all mortality traits during all three phases (0.23 to 0.56), as expected, but correlations were slightly stronger during and after the outbreak (0.56 ± 0.23 with LNBD during the outbreak). NBA had close to zero genetic correlation estimates with mortality traits prior to and after the outbreak but slightly negative estimates during the outbreak (−0.14 to −0.22). Estimates of genetic correlations among mortality traits within phase were all positive (0.23 to 0.98) for all phases. Prior to and after the outbreak, LNSB and LNBD were genetically highly correlated, at 0.94 ± 0.07, likely because most mortalities at those times are due to stillborns rather than mummies. The estimate of the genetic correlation between LNSB and LNBD dropped to 0.73 ± 0.23 during the outbreak but the estimate of the genetic correlation between LNBM and LNBD increased from 0.68 ± 0.28 prior to the outbreak to 0.80 ± 0.15 during the outbreak, as a greater proportion of mortalities was due to mummies during the outbreak. The estimate of the genetic correlation between LNSB and LNBM was moderate prior to the outbreak (0.40 ± 0.47) and low during the outbreak (0.23 ± 0.48). All estimates of genetic correlations among mortality traits after the outbreak were >0.83.

Reaction Norm Model

The three parameterizations of the reaction norm model only differed in estimates of genetic variances for the intercept and slope, and in estimates of the genetic covariance or correlation between intercept and slope (Table 6). However, estimates of genetic variances and covariances for NBA at given FYW levels were unaffected, as expected. Estimates of the genetic variance of the intercept and slope ranged from 0.52 to 0.81 and from 0.07 to 1.21, respectively. All estimates of the genetic covariance between intercept and slope were positive (0.11 to 0.51). Estimates of the genetic correlation between the intercept and slope were 0.54 ± 0.35, 0.24 ± 0.50, and 0.52 ± 0.36 for the raw, Legendre, and the polynomial (pol) function of ASReml, respectively. Estimates of residual variance increased slightly from the first FYW level to the second (estimated from 12.21 to 12.73) and then reduced as the FYW effect increased (10.54,

Table 5. Estimates of genetic correlations (with SE in parentheses) between reproductive traits within each of the three phases relative to the outbreak (prior to, during, and after)

Traits[1]	Prior[2]	During[2]	After[2]
TNB-NBA	0.96 (0.02)	0.71 (0.16)	0.92 (0.04)
TNB-LNSB	0.23 (0.26)	0.32 (0.28)	0.33 (0.28)
TNB-LNBM	0.23 (0.42)	0.34 (0.36)	0.33 (0.38)
TNB-LNBD	0.28 (0.26)	0.56 (0.23)	0.33 (0.27)
NBA-LNSB	0.00 (0.27)	−0.20 (0.35)	−0.05 (0.31)
NBA-LNBM	0.05 (0.44)	−0.22 (0.41)	0.01 (0.42)
NBA-LNBD	0.03 (0.28)	−0.14 (0.33)	−0.05 (0.31)
LNSB-LNBM	0.40 (0.47)	0.23 (0.48)	0.84 (0.36)
LNSB-LNBD	0.94 (0.07)	0.73 (0.23)	0.98 (0.06)
LNBM-LNBD	0.68 (0.28)	0.80 (0.15)	0.90 (0.16)

[1] TNB = total number born, NBA = number born alive, LNSB = log number stillborn, LNBM = log number born mummified, LNBD = log number born dead.

[2]Phases were split using a mixed linear model, fitting farm year week (FYW) as a random effect and extracting the predicted values. Visual appraisal was used to split phases into prior, during, and after the PRRS outbreak.

Table 6. Estimates of variance components (with SE in parentheses) for three parameterizations of the reaction norm model, regressing on the predicted farm-year-week (FYW) effects for number born alive (**NBA**)

	Reaction norm parameterization		
Variance component	Raw scale[1]	Legendre[2]	Polynomial[3]
Var(intercept) ($\sigma^2_{a_i}$)	0.54 (0.17)	0.81 (0.38)	0.52 (0.17)
Var(slope) ($\sigma^2_{a_s}$)	0.07 (0.06)	0.15 (0.28)	1.21 (0.90)
Cov(intercept,slope) (σ_{a_i,a_s})	0.11 (0.07)	0.51 (0.38)	0.41 (0.27)
Cor(intercept,slope) (r_{a_i,a_s})	0.54 (0.35)	0.24 (0.50)	0.52 (0.36)
Var(permanent envir.) (σ^2_{pe})	1.30 (0.24)	1.30 (0.24)	1.30 (0.24)
Residual Variance 1 ($\sigma^2_{e_1}$)	12.21 (1.06)	12.21 (1.06)	12.21 (1.06)
Residual Variance 2 ($\sigma^2_{e_2}$)	12.73 (0.62)	12.73 (0.62)	12.73 (0.62)
Residual Variance 3 ($\sigma^2_{e_3}$)	10.54 (0.58)	10.54 (0.58)	10.54 (0.58)
Residual Variance 4 ($\sigma^2_{e_4}$)	8.70 (0.31)	8.70 (0.31)	8.70 (0.31)
Residual Variance 5 ($\sigma^2_{e_5}$)	6.84 (0.45)	6.84 (0.45)	6.84 (0.45)

[1]Predicted values of farm-year-week (FYW) effects used as covariate.

[2]Predicted FYW effects scaled using leg function in ASReml 4.

[3]Predicted FYW effects scaled using the pol function in ASReml 4.

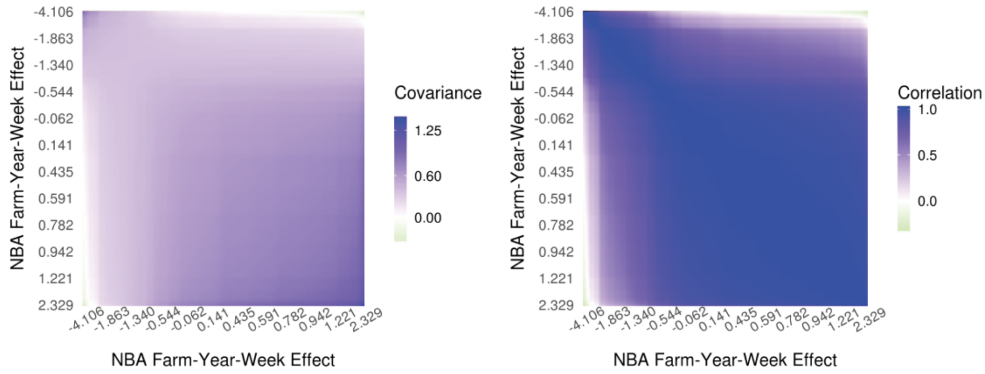

Figure 3. Matrix of genetic covariances (left) and correlations (right) for number born alive (**NBA**) based on the reaction norm model as a function of estimates of NBA farm-year-week effects. The PRRS outbreak phase begins at approximately −1 for the NBA farm-year-week effect. Same for all parameterizations of the reaction norm intercept and slopes.

8.70, and 6.84). Figure 3 shows estimates of genetic covariances (left) and correlations (right) from the reaction norm model for NBA between FYW levels which, as indicated, were the same for all three parameterizations. Estimates of genetic covariances (diagonals) showed the expected quadratic trend (given the first-order model) and were negative only between the most extreme FYW classes. Estimates of genetic correlations for NBA between FYW levels showed two fairly distinct blocks during the healthy and diseased phases (diseased in the top left, healthy in the bottom right). The transition from diseased to healthy started for NBA FYW effects around −1 (with a very small overlap of the two phases, see Figure 2). Genetic correlations between the two blocks were moderate, except for the very extreme FYW levels, which was consistent with the multi-trait analysis of traits defined by phase.

Table 7 shows estimates of correlations of EBV from the multivariate analysis of NBA by phase with EBV obtained from the reaction norm model using the raw FYW scale. Correlations of EBV for the additive genetic intercept and slope terms with EBV for FYW levels equal to −4, −2, 0, and 2 are also shown, with the first two (−4 and −2) being during the outbreak and the other two (0 and 2) from the two healthy phases (representing a combination of prior to and after the PRRSV outbreak). The EBV for the intercept terms from the reaction norm model had the highest correlation with EBV for NBA_p (0.82), while EBV for NBA_p and NBA_d were highly correlated with EBV from the reaction norm model at FYW equal to −2 and 0 (0.78 to 0.80). The EBV for the intercept was almost perfectly correlated with EBV for the reaction norm at FYW equal to 0 (as expected) and were also highly correlated with EBV for the reaction

Table 7. Correlations among EBV from the multivariate analysis of number born alive (**NBA**) prior to, during, and after the outbreak (NBA_p, NBA_d, and NBA_a) and with EBV obtained from the reaction norm (RN) model using the raw scale for farm-year-week (FYW) estimates, including EBV for intercept (RN_{int}) and slope (RN_{slope}), along with the EBV for NBA for FYW estimates equal to −4, −2, 0, and 2 (RN_{-4}, RN_{-2}, RN_0, and RN_2)

	NBA_d	NBA_a	RN_{int}	RN_{slope}	RN_{-4}	RN_{-2}	RN_0	RN_2
NBA_p	0.99	0.48	0.82	0.42	0.36	0.80	0.78	0.68
NBA_d		0.45	0.81	0.39	0.38	0.80	0.76	0.66
NBA_a			0.75	0.59	0.09	0.64	0.77	0.74
RN_{int}				0.59	0.34	0.93	0.97	0.88
RN_{slope}					−0.55	0.26	0.77	0.91
RN_{-4}						0.66	0.11	−0.15
RN_{-2}							0.82	0.64
RN_0	Sym.							0.97

norm at FYW equal to −2. Correlations between EBV at different FYW levels were very similar to the estimates of genetic correlations from the reaction norm model, showing decreasing correlations with increasing distance between FYW levels.

DISCUSSION

Genetic Parameters for S/P Ratio

Estimates of heritability and genetic correlations for S/P ratio with reproductive performance during a PRRS outbreak were mostly inconsistent with previous studies. Serão et al. (2014) reported a heritability of 0.45 ± 0.13 for S/P ratio after a PRRS outbreak in a multiplier herd in Canada, which was validated in a more complex independent study (Serão et al., 2016). The estimate of heritability of S/P ratio from the current study was, however, substantially lower at 0.17 ± 0.05. Estimates of genetic correlations of S/P ratio with reproductive traits also did not completely agree with previous results, except for the genetic correlation of S/P ratio with LNSB (Serão et al., 2014). Although this is favorable, most of the prenatal mortality (68%) during the PRRS outbreak in this study was due to mummified piglets, as mentioned above. This is important because although S/P ratio was more correlated with LNSB, it would not change overall mortality as much because more piglet mortality stems from mummified piglets. Serão et al. (2014) found that S/P ratio tended to have moderate/strong genetic correlations with reproduction traits, ranging from −0.72 (NSB) to 0.73 (NBA); the lowest estimate in absolute value was 0.27 (NBD). The only genetic correlation that was similar in the current study was for LNSB (−0.73). Both TNB and NBA were not strongly associated with S/P ratio in the current study. Note, however, that these estimates come with large standard errors when dealing with small sample sizes and lowly heritable reproductive traits, therefore strong conclusions cannot be drawn until further studies are conducted.

One notable difference between the current study and the studies of Serão et al. (2014, 2016) is the use of different antibody assays for semi-quantification of antibody levels. The IDEXX PRRS X3 ELISA (at the same lab) was used in both Serão et al. (2014) and Serao et al. (2016), while the Luminex (Luminex Corp., Austin, TX) microsphere assay was utilized in the present study. Although the IDEXX is considered an industry/research gold standard for measuring PRRS antibody (Sattler et al., 2014), the microsphere (or microbead) assay is rising in popularity because the Luminex multiplex system allows for the detection of numerous analytes within a single biological sample, saving cost, time, and labor (Lin et al., 2011). The Luminex assay was also used by the same lab in the study of Hess et al. (2018) on nursery pigs following experimental PRRSV infection, resulting in a moderate-to-high heritability estimate. The microbead assay is not a traditional ELISA but is conceptually similar to an indirect ELISA, as both measure antibodies against the nucleocapsid (N) protein (inside the complete PRRS virus). Lin et al. (2011) compared an earlier version of the standard single plex ELISA (IDEXX Herdchek PRRSV 2XR kit) and the microsphere-based immunoassay and found the spearman rank correlation to be 0.72 for PRRS antibody. The sensitivity and specificity between the assays were 91% and 93% for PRRS in young pigs, respectively (κ coefficient of 0.67). Commonly, young pigs are used for testing and validating assay results for several reasons (cost, ease of sampling, availability, etc.). In adult pigs, however, 64% (16/25) of samples were found to be positive by the Luminex assay but negative by the IDEXX

HerdChek PRRS X3 assay (Giménez-Lirola et al., 2014). Giménez-Lirola et al. (2014) used the newest IDEXX (HerdChek PRRS X3), the same test used in Serão et al., (2014, 2016). Adults pigs (sows and boars) may have higher background reactivity than young pigs (Giménez-Lirola et al., 2014), possibly due to a more mature immune system and antigens seen later in life and will need to be investigated further. Although we do not have direct evidence that the differences in variance components estimated between the present study and Serão et al., (2014, 2016) can be attributed to the differences between the IDEXX and Luminex platform, it should be a major consideration in future research, along with the age of the animal being tested.

There are several other possible reasons for the difference in estimates of genetic parameters for S/P ratio between the current and previous PRRS outbreak studies. These could include other aspects of the assay such as in-house diagnostic target variations, time of year, the strain of the virus, sample processing, and other unknown environmental effects. In contrast to Serão et al. (2014), sows in the current study were inoculated 3 wk after the confirmed outbreak, followed by MLV vaccination. Vaccination is not expected to impact antibody levels at 40 d after inoculation, as a secondary type of response in a relatively short time after infection is not expected due to the persistency of infection of the PRRS virus (Lunney et al., 2016; discussed below). However, it still could contribute to differences. Typically, antibody response studies in pigs are conducted in designed experiments with one injection given simultaneously to all animals. In a natural disease challenge, this consistency is lost and sows in a large farm are consistently re-exposed to antigens, some possibly due to "rebound" animals (Boddicker et al., 2012). In Serão et al. (2016), antibody levels were measured on gilts following acclimation across many commercial farms, which represented a range of times following exposure, either through infection or MLV vaccination, again strengthening the idea that some of these other factors may not play a large role.

Multiple factors make determining the cause of differences between estimates of genetic parameters for S/P between studies hard to understand. These will continue to be an issue as what is best for measuring antibody response in research (i.e., this study) may not be optimal for production and clearing the virus from a commercial farm, such as inoculation and vaccination observed in the current study. This may provide some insight into the difficulty of conducting this type of research in field conditions and therefore alternatives will be needed (e.g., separate, carefully designed challenge studies in sows). Perhaps other measures such as interferon-γ (IFN-γ) response after inoculation might be useful, along with antibody response. Collecting antibody response at multiple time points may also be helpful to determine the approximate date of infection, but this would be expensive and not feasible on a commercial farm.

Genetic Parameters for Reproduction Traits

Ranges of heritability estimates for litter size and mortality traits were consistent with previous estimates (0.01 to 0.13; Bidanel, 2011). Trends in heritabilities between phases generally followed results by Lewis et al. (2009). Heritability for NBA was lower during the outbreak, while estimates of heritability for mortality traits were higher during the outbreak, most likely due to the increased incidence of mortality under PRRS challenge. Heritability of TNB was not affected by the PRRS phase like the other traits but was after the outbreak. Biologically, this makes sense, as sows that farrowed during the outbreak were bred prior to the outbreak and all fetuses would be counted in the total born. However, sows bred during the outbreak farrowed later during or after the outbreak, which affects the total born observed due to possibly fewer oocytes being fertilized or fetuses being absorbed. There is no verification of this because pregnancies were not evaluated by ultrasound. Low to moderate genetic correlations for reproduction traits between prior to and during the PRRS phase likely indicate the influence of disease resistance QTL during the outbreak phase, making them different traits. Serão et al. (2014) found similar trends as observed in the present study for estimates of heritability prior to and during the outbreak for NBM, NBD and NSB but their estimates of heritability for NBA was higher during the outbreak.

Estimates of genetic correlations for reproduction traits between phases were fairly consistent with previous estimates (Lewis et al., 2009; Rashidi et al., 2014; Herrero-Medrano et al., 2015), although these studies combined data from prior and after the outbreak into one trait. For instance, the genetic correlation from Lewis et al., (2009) for NBA was 0.56 between healthy and diseased phases, which would be a combination of the current estimates for NBA between prior and during (0.98) and between during and after (0.21). Rashidi et al. (2014) estimated genetic correlations for NBA and NBD at 0.87 and 0.57 between healthy and diseased

phases, respectively. Herrero-Medrano et al. (2015) estimated these same correlations at 0.75 and 0.74, respectively.

Differences between studies in estimates of genetic correlations for reproduction traits between diseased and healthy phases in PRRS outbreak herds can also be due to other factors. The strain that caused the outbreak in the current study was a very severe strain of the PRRS virus. More studies are needed to determine whether results from Serão et al. (2014) also hold for other virus strains, such as those used in the PRRS host genetics consortium and associated trials (Hess et al., 2016; Waide et al., 2018). New viral strains develop and results from previous antibody studies may not apply. For instance, a new PRRS strain that developed in China in the last decade shows very different clinical signs than normal strains (see Figure 2 from Tian et al., 2007). Diseases such as PRRS can change and antibody measures as indicator traits need to continually be re-evaluated for effectiveness in a breeding program.

Reproductive Performance After the PRRS Outbreak

To date, all studies have divided the reproduction data from PRRS outbreak herds into only healthy and diseased phases. A finding from this research was that estimated genetic correlations may support keeping the time period after the PRRS phase as a separate trait from prior to the PRRS outbreak, although standard errors were large. PRRS can be a persistent infection (Lunney et al., 2016) and, thus, it is possible that PRRS still affects reproductive performance after the outbreak has cleared, perhaps subclinically. Lunney et al. (2016) discussed the three stages of a PRRS infection: acute, persistent, and extinct. The virus can persist in tonsils and lymph nodes and has been identified in animals as long as 175 to 251 d postinfection (Wills et al., 2003; Molina, 2008), although most cleared within three to four months (Wills et al., 2003). In the present study, the after phase included ~4 mo of data. It is possible that the large farm sizes contributed to the persistent nature of the infection. It is also possible that the less than one genetic correlation between traits prior and after the outbreak was caused by genotype-by-environment interactions due to reasons such as seasonality, which will need to be investigated further in another study. The outbreak phase for the current study was during the late spring/summer months.

Another reason why the after-period may need to be analyzed separately is that some sows that farrowed after the outbreak were bred during the outbreak, which could result in some residual effects. Any sow bred during the PRRS phase could suffer reduced TNB from reduced fertilization, embryos not surviving, or fetuses being absorbed. In contrast, most sows that farrowed during the outbreak phase were bred during the healthy phase prior to the outbreak and, thus, TNB should not be severely affected due to the piglets already being fully-formed, as observed in the present study, especially in farm 3. This was also reinforced by the low estimate of the genetic correlation for TNB between before and after the outbreak (0.32 ± 0.03). Thus, it may be better to consider leaving the after phase a separate trait or to remove this data for routine genetic evaluation. Further research will be needed to determine how long this period extends.

Reproduction Traits During the Outbreak

Previous reproductive disease outbreak studies have separated reproduction data into two phases (healthy and diseased) but, to our knowledge, this is the first study to report estimates of genetic correlations within phase (e.g. NBA_p with $LNSB_p$ or TNB_d with NBA_d). The phase prior to infection represents typical variance components for litter size without major disease (Su et al., 2007; Putz et al., 2015). Not separating data from herds that experience disease outbreaks into three phases could affect estimates of genetic correlations between traits (e.g., TNB and NBA or their genetic correlation with mortality traits). For example, the genetic correlation between TNB and LNBD was 0.28 ± 0.26 prior to infection and 0.56 ± 0.23 during the PRRS outbreak. This should be as expected, as more total born during the outbreak would allow more pigs to be affected by disease and die prior to farrowing.

Reaction Norm Model

One possible downside of the use of reaction norms for analysis of disease outbreak data is that they do not differentiate records obtained prior to and after the outbreak. Separate stressors may cause dips in performance. This may be a disadvantage of the reaction norm models, especially for other situations such as outbreaks from different pathogens or different strains of the same virus (observed in Herrero-Medrano et al., 2015). Multivariate analyses may also not be able to disentangle causes if multiple pathogens are involved

in the infection. In Herrero-Medrano et al. (2015), both PRRS and a coronavirus caused outbreaks that led to a high challenge load (described by Mathur et al., 2014) of over 15 index units (the challenge load). The reaction norm model treats both of these outbreaks being very similar traits (in terms of regressor values, here the FYW effects), when in fact they most likely have different genetic backgrounds in terms of genetic resistance. For example, two FYW effects around −1 NBA may be the result of separate environmental changes; one could be the result of porcine epidemic diarrhea and the other from heat stress for example. Any FYW effect would be a combination of any management environments and challenges/stressors. Regardless of this, given enough data, these models should still result in sows that are more resilient/robust to environmental challenges. The advantage of the reaction norm is that it would average over all of these effects in one parsimonious model without regard to specific causes. Knap and Su (2008) stressed the need to have large datasets for reaction norm models to be effective. The optimal breeding objective should include general resilience/robustness to any number of stressors, including different diseases, not to single diseases or stressors. Therefore, for the reaction norm models to be effective, it would be advantageous to have a large number of environments classified from many different farms with as many different management practices as possible, such that the values used to regress on capture as many stressors and different environments as possible (Knap, 2005). When thinking about testing a sire, it would be best to have as many daughters in as many different farms/environments as possible. One issue is that there can be high leverage on the slope of the reaction norm slope for extreme FYW observations (Pool et al., 2000).

It is known that different parameterizations of the regressor value (FYW estimates) lead to different variances and covariances for the intercept and slope terms. Therefore, it can be dangerous to interpret these estimates, as was done by Knap and Su (2008). Another factor that can influence the (co)variances is using different contemporary group sizes (week, month, or season). Knap and Su (2008) utilized estimates for herd-year-season contemporary groups instead of herd-year-week contemporary groups used in the current analysis. The estimate of the genetic correlation between the intercepts and slopes from the three parameterizations used in the current study was different (as expected), although all were positive. This indicates that selection for improved NBA of animals with the standard animal model (related to the reaction norm intercept) would result in animals with greater reaction to changes in the environment. Again, as expected, estimates of genetic covariances and correlations between EBVs from each FYW from the reaction norm model were not affected by the parameterization of the model (Figure 3).

The correlation between EBV for the intercept term from the reaction norm model and EBV from a typical animal model was high, which agrees with previous research (Knap and Su, 2008). The current analysis expanded this by calculating the correlations of EBV from the multivariate phases (prior, during, and after) model with the reaction norm estimates of intercepts and slopes and EBV at discrete FYW levels. Knap and Su (2008) found EBV from the multivariate animal model and EBV for the intercept terms from the reaction norm to be correlated 0.78 to 0.85, similar to the current analysis, which found correlations between 0.75 and 0.82 (for prior, during, and after for NBA). EBV from the reaction norm model at different levels of FYW was also correlated with EBV from the multivariate model. EBV from the reaction norm between −-2 and +2 were moderate to highly correlated with EBV from the multivariate analysis (between 0.64 and 0.80). The highest correlation between the multivariate EBV and the EBV at −4 from the reaction norm was for NBA during the outbreak, as expected (0.38), however, this was at the very extreme of the outbreak phase. The EBV for the slope from the reaction norm was negatively correlated with the EBV at −4 (−0.55) and strongly positively correlated with EBV at 2 (0.91), which is as expected. Animals with EBV for the slope that deviate from zero are considered sensitive to environmental changes. Therefore, the optimum selection would be for animals with a high EBV for the intercept and an EBV for the slope close to zero, indicating high producing animals that produce uniformly (in ranking) across environmental gradients.

The reaction norm can capture more than just health, which may contribute to the difference in genetic correlations observed between the multivariate and reaction norm models. Guy et al. (2012) discussed resilience to not only health challenges but also other environmental challenges. In commercial data, challenges for pigs can encompass social, environmental, metabolic, immunological, and human interactions (Martínez-Miró et al., 2016). Seasonality encompasses effects of heat stress and disease and both affect FYW estimates. For instance, there is a positive seasonality effect

during the summer months for pigs weaned/sow/year (Stalder, 2017). Sevillano et al. (2016) showed that seasonal infertility can be impacted by photoperiod and not just by ambient temperatures. So, as long as data are captured over long periods of time and plenty of heterogeneous environments, the reaction norm should also be thought of as general resilience, instead of only disease resilience. Of course, the multivariate model could also pick up effects from other stressors.

Bishop and Woolliams (2014) stated that the requirement to measure resistance phenotypes is a rate-limiting step in breeding for disease resistance. One problem for both the multivariate and reaction norm models is that it is difficult to get enough records in the diseased phase to obtain accurate EBV for disease resilience; most of the information for either model will come from correlated data, i.e., from the "healthy" phase in the multivariate case. For the reaction norm model, most data are from healthy weeks when an outbreak has not occurred for an extended period of time. For instance, in farm 1 of Herrero-Medrano et al. (2015), only one outbreak occurred over a 6-yr span of the data and only five total outbreaks occurred in the three farms. The reaction norm is only observed on part of the FYW estimates for many animals, especially because many sows are culled early (especially in nucleus environments), although this is partially overcome by the use of the pedigree relationships. The use of random regression models for a reaction norm is different from many other situations in which random regression models are used for genetic analyses, such as milk yield in dairy cattle, growth or feed intake in pigs, and egg production in poultry. In those situations, animals have repeated records that span most of the lactation, growth period, or egg-laying cycle, leading to more accurate estimates of breeding values than obtained for the sparser reaction norm model (Knap and Su, 2008). In the current study, sows had between one and four records for the reaction norm model. Meyer (2005) stated that using higher order polynomials when a substantial proportion of animals have fewer records than the order of polynomials fitted can lead to erratic and implausible estimates. One should be careful before applying complex models to this type of data. The total range in estimates of NBA FYW effects was 6.43 on the original scale. A total of 47%, 70%, and 89% of sows had phenotypes in contemporary groups that ranged <0.5, 2.0, and 3.0, respectively, in NBA FYW effects (i.e., the *x*-axis). This may contribute to the poor accuracy referred to by Knap and Su (2008).

Future Work

There is some work needed prior to the swine industry adopting antibody response to PRRS outbreaks or MLV vaccination. Novel strains of PRRSV are continuing to show up because of the high mutation rate of the PRRS virus and predictive ability in terms of genetic correlations should be regularly checked. Antibody tests continue to change over time and differences among labs exist, although the HerdChek PRRS X3 antibody test seems to be very repeatable within and across labs (Kittawornrat et al., 2012). This possible instability over time in other antibody assays such as the Luminex (or future IDEXX assays) is risky for implementation into the swine breeding industry. One important validation needed is to send samples to multiple veterinary diagnostic labs and with multiple tests (e.g., IDEXX vs. Luminex) to verify results for each test and each lab to make sure genetic analyses agree. Perhaps even lab replicates will need to be performed to determine the repeatability. Thus, at this point, it is unclear whether selection on antibody response (possibly to PRRS vaccines) will be highly useful to the swine breeding industry.

CONCLUSIONS

Antibody level in sows to PRRS following a PRRS outbreak, measured as S/P ratio, was low to moderately heritable (0.17 ± 0.05) and had low genetic correlations with reproductive traits except for LNSB (-0.73 ± 0.29). Standard errors for variance component estimates were large because of a relatively small dataset and lowly heritable traits, so no strong conclusions can be drawn. More research will be needed to understand why these results did not completely validate previous findings on S/P ratio heritability and genetic correlations with reproductive performance. It is possible that the differences in the antibody assay were the cause, but this is still unknown. The genetic correlation between reproductive performance prior to and during the PRRS outbreak was high for both TNB and NBA. The only negative genetic correlation between performance prior to and during the PRRS outbreak was for LNBM. TNB had a genetic correlation of 0.32 between prior to and after the outbreak. It may be useful to consider reproductive performance several months after the outbreak as a separate trait from performance prior to the outbreak, as sows farrowing after the outbreak were bred during the outbreak. The reaction norm model for NBA

showed similar trends in genetic correlations as the multivariate model that considered reproductive performance prior, during, and after the outbreak as separate traits, although it considered data from prior and after the outbreak as having overlapping environments. Overall, future work will need to address some of the differences from previous research observed in the current study.

ACKNOWLEDGMENTS

We would also like to thank The Maschhoffs for allowing us to take samples in their barns following the outbreaks and their farm staff for help with sample collection. Several Iowa State University undergraduate students also helped with data collection, including Allysa Koethe, Jake Baker, Zach Sauer, Dylan Lathrum, and Kaylyn Hodge.

Conflict of interest statement. None declared.

LITERATURE CITED

Bates, D., M. Maechler, B. Bolker, S. Walker. 2015. Fitting linear mixed-effects models using lme4. J. Stat. Softw. 67:1–48. doi:10.18637/jss.v067.i01

Bidanel, J. 2011. Biology and genetics of reproduction in genetics of the pig. Rothschild, M. F. and A. Ruvinsky, editors. The Genetics of the PIg. 2nd ed. Oxfordshire (UK): CAB International; p. 218–241.

Bishop, S. C., and J. A. Woolliams. 2014. Genomics and disease resistance studies in livestock. Livest. Sci. 166:190–198. doi:10.1016/j.livsci.2014.04.034

Boddicker, N., E. H. Waide, R. R. Rowland, J. K. Lunney, D. J. Garrick, J. M. Reecy, and J. C. Dekkers. 2012. Evidence for a major QTL associated with host response to porcine reproductive and respiratory syndrome virus challenge. J. Anim. Sci. 90:1733–1746. doi:10.2527/jas.2011-4464

Gilmour, A. R., B. J. Gogel, B. R. Cullis, and R. Thompson. 2015. ASReml user guide release 4.1 structural specification. Hemel Hempstead, UK: VSN International Ltd.

Giménez-Lirola, L. G., P. F. Gerber, R. R. Rowland, P. G. Halbur, Y. W. Huang, X. J. Meng, and T. Opriessnig. 2014. Development and validation of a 4-plex antibody assay for simultaneous detection of igg antibodies against torque teno sus virus 1 (TTSuV1), TTSuV2, and porcine reproductive and respiratory syndrome virus types 1 and 2. Res. Vet. Sci. 96:543–550. doi:10.1016/j.rvsc.2014.02.014

Guy, S. Z., P. C. Thomson, and S. Hermesch. 2012. Selection of pigs for improved coping with health and environmental challenges: breeding for resistance or tolerance? Front. Genet. 3:281. doi:10.3389/fgene.2012.00281

Herrero-Medrano, J. M., P. K. Mathur, J. ten Napel, H. Rashidi, P. Alexandri, E. F. Knol, and H. A. Mulder. 2015. Estimation of genetic parameters and breeding values across challenged environments to select for robust pigs. J. Anim. Sci. 93:1494–1502. doi:10.2527/jas.2014-8583

Hess, A. S., Z. Islam, M. K. Hess, R. R. Rowland, J. K. Lunney, A. Doeschl-Wilson, G. S. Plastow, and J. C. Dekkers. 2016. Comparison of host genetic factors influencing pig response to infection with two North American isolates of porcine reproductive and respiratory syndrome virus. Genet. Sel. Evol. 48:43. doi:10.1186/s12711-016-0222-0

Hess, A. S., B. R. Trible, M. K. Hess, R. R. Rowland, J. K. Lunney, G. S. Plastow, and J. C. M. Dekkers. 2018. Genetic relationships of antibody response, viremia level, and weight gain in pigs experimentally infected with porcine reproductive and respiratory syndrome virus. J. Anim. Sci. 93:3565–3581. doi:10.1093/jas/sky229

Holtkamp, D. J., J. B. Kliebenstein, E. J. Neumann, J. J. Zimmerman, H. F. Rotto, T. K. Yoder, C. Wang, P. E. Yeske, C. L. Mowrer, C. A. Haley. 2013. Assessment of the economic impact of porcine reproductive and respiratory syndrome virus on United States pork producers. J. Swine Health Prod. 21:72–84.

Kittawornrat, A., C. Wang, G. Anderson, A. Ballagi, A. Broes, S. Carman, K. Doolittle, J. Galeota, J. Johnson, S. Lizano, et al. 2012. Ring test evaluation of the repeatability and reproducibility of a porcine reproductive and respiratory syndrome virus oral fluid antibody enzyme-linked immunosorbent assay. J. Vet. Diagn. Invest. 24:1057–1063. doi:10.1177/1040638712457929

Knap, P. W. 2005. Breeding robust pigs. Austral. J. Exp. Agric.. 45:763–773. doi:10.1071/EA05041

Knap, P. W., and G. Su. 2008. Genotype by environment interaction for litter size in pigs as quantified by reaction norms analysis. Animal 2:1742–1747. doi:10.1017/S1751731108003145

Lewis, C. R., M. Torremorell, L. Galina-Pantoja, and S. C. Bishop. 2009. Genetic parameters for performance traits in commercial sows estimated before and after an outbreak of porcine reproductive and respiratory syndrome. J. Anim. Sci. 87:876–884. doi:10.2527/jas.2008-0892

Lin, K., C. Wang, M. P. Murtaugh, and S. Ramamoorthy. 2011. Multiplex method for simultaneous serological detection of porcine reproductive and respiratory syndrome virus and porcine circovirus type 2. J. Clin. Microbiol. 49:3184–3190. doi:10.1128/JCM.00557-11

Lunney, J. K., Y. Fang, A. Ladinig, N. Chen, Y. Li, B. Rowland, and G. J. Renukaradhya. 2016. Porcine reproductive and respiratory syndrome virus (PRRSV): pathogenesis and interaction with the immune system. Annu. Rev. Anim. Biosci. 4:129–154. doi:10.1146/annurev-animal-022114-111025

Lunney, J. K., J. P. Steibel, J. M. Reecy, E. Fritz, M. F. Rothschild, M. Kerrigan, B. Trible, and R. R. R. Rowland. 2011. Probing genetic control of swine responses to PRRSV infection: current progress of the PRRS host genetics consortium. BMC Proc.. 5(Suppl. 4):S30. doi:http://www.biomedcentral.com/1753-6561/5/S4/S30

Madapong, A., G. Temeeyasen, K. Saeng-Chuto, T. Tripipat, W. Navasakuljinda, A. Boonsoongnern, A. Tantituvanont, and D. Nilubol. 2017. Humoral immune responses and viral shedding following vaccination with modified live porcine reproductive and respiratory syndrome virus vaccines. Arch. Virol. 162:139–146. doi:10.1007/s00705-016-3084-4

Martínez-Miró, S., F. Tecles, M. Ramón, D. Escribano, F. Hernández, J. Madrid, J. Orengo, S. Martínez-Subiela, X. Manteca, and J. J. Cerón. 2016. Causes, consequences and biomarkers of stress in swine: an update. BMC Vet. Res. 12:171. doi:10.1186/s12917-016-0791-8

Mathur, P. K., J. M. Herrero-Medrano, P. Alexandri, E. F. Knol, J. ten Napel, H. Rashidi, and H. A. Mulder. 2014. Estimating challenge load due to disease outbreaks and other challenges using reproduction records of sows. J. Anim. Sci. 92:5374–5381. doi:10.2527/jas.2014-8059

Meyer, K. 2005. Random regression analyses using B-splines to model growth of Australian Angus cattle. Genet. Sel. Evol. 37:473–500. doi:10.1051/gse:2005012

Molina, R. M. 2008. Porcine reproductive and respiratory syndrome virus: understanding and managing persistent infection. Theses and Dissertation. https://lib.dr.iastate.edu/rtd/15733/

Neumann, E. J., J. B. Kliebenstein, C. D. Johnson, J. W. Mabry, E. J. Bush, A. H. Seitzinger, A. L. Green, and J. J. Zimmerman. 2005. Assessment of the economic impact of porcine reproductive and respiratory syndrome on swine production in the United States. J. Am. Vet. Med. Assoc. 227:385–392.

Pool, M. H., L. L. Janss, and T. H. Meuwissen. 2000. Genetic parameters of legendre polynomials for first parity lactation curves. J. Dairy Sci. 83:2640–2649. doi:10.3168/jds.S0022-0302(00)75157-5

Putz, A. M., F. Tiezzi, C. Maltecca, K. A. Gray, and M. T. Knauer. 2015. Variance component estimates for alternative litter size traits in swine. J. Anim. Sci. 93:5153–5163. doi:10.2527/jas.2015-9416

Rashidi, H., H. A. Mulder, P. Mathur, J. A. van Arendonk, and E. F. Knol. 2014. Variation among sows in response to porcine reproductive and respiratory syndrome. J. Anim. Sci. 92:95–105. doi:10.2527/jas.2013-6889

Sattler, T., E. Wodak, S. Revilla-Fernndez, and F. Schmoll. 2014. Comparison of different commercial ELISAs for detection of antibodies against porcine respiratory and reproductive syndrome virus in serum. BMC Vet. Res. 10:300. doi:10.1186/s12917-014-0300-x

Serão, N. V., R. A. Kemp, B. E. Mote, P. Willson, J. C. Harding, S. C. Bishop, G. S. Plastow, and J. C. Dekkers. 2016. Genetic and genomic basis of antibody response to porcine reproductive and respiratory syndrome (PRRS) in gilts and sows. Genet. Sel. Evol. 48:51. doi:10.1186/s12711-016-0230-0

Serão, N. V., O. Matika, R. A. Kemp, J. C. Harding, S. C. Bishop, G. S. Plastow, and J. C. Dekkers. 2014. Genetic analysis of reproductive traits and antibody response in a PRRS outbreak herd. J. Anim. Sci. 92:2905–2921. doi:10.2527/jas.2014-7821

Sevillano, C. A., H. A. Mulder, H. Rashidi, P. K. Mathur, and E. F. Knol. 2016. Genetic variation for farrowing rate in pigs in response to change in photoperiod and ambient temperature. J. Anim. Sci. 94:3185–3197. doi:10.2527/jas.2015-9915

Stalder, K. J. 2017. 2016 Pork Industry Productivity Analysis. National Pork Board. https://www.pork.org/wp-content/uploads/2017/11/2017-pork-industry-productivity-analysis.pdf

Su, G., M. S. Lund, and D. Sorensen. 2007. Selection for litter size at day five to improve litter size at weaning and piglet survival rate. J. Anim. Sci. 85:1385–1392. doi:10.2527/jas.2006-631

Tian, K., X. Yu, T. Zhao, Y. Feng, Z. Cao, C. Wang, Y. Hu, X. Chen, D. Hu, X. Tian, et al. 2007. Emergence of fatal PRRSV variants: unparalleled outbreaks of atypical PRRS in china and molecular dissection of the unique hallmark. PLoS One 2:e526. doi:10.1371/journal.pone.0000526

Waide, E. H., C. K. Tuggle, N. V. L. Serão, M. Schroyen, A. Hess, R. R. R. Rowland, J. K. Lunney, G. Plastow, and J. C. M. Dekkers. 2018. Genomic prediction of piglet response to infection with one of two porcine reproductive and respiratory syndrome virus isolates. Genet. Sel. Evol. 50:3. doi:10.1186/s12711-018-0371-4

Wills, R. W., A. R. Doster, J. A. Galeota, J. H. Sur, and F. A. Osorio. 2003. Duration of infection and proportion of pigs persistently infected with porcine reproductive and respiratory syndrome virus. J. Clin. Microbiol. 41:58–62. doi:10.1128/JCM.41.1.58-62.2003