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> Biometeorology and animal welfare Full-length research article

Behavioral and physiological performance of different gilt breeds during lactation

Runze Liu¹ D, Wenbo Ji¹ D, Chao Wang¹ D, Ran Yi¹ D, Peng Zhao¹ D, Lei Pan¹ D, Honggui Liu^{1,2} D, Jun Bao^{1*} D

¹ Northeast Agricultural University, College of Animal Science and Technology, Harbin, China.

² Key Laboratory of Swine Facilities Engineering, Ministry of Agriculture, Harbin, China.

ABSTRACT - We evaluated the maternal behavior, physiology, and reproductive performance of both Damin (Min-pig × Large White) and Large White gilts to identify the advantages hybrid sows offer with regard to stress relieve and improvement of the welfare level of sows during late lactation. First-parity Damin gilts (n = 40) and firstparity Large White gilts (n = 40) were farrowed in individual pens. Video surveillance was used to monitor the occurrence of lateral recumbency and compare it to other postures, such as ventral recumbency, defecation, urination, tail posture, sham-chewing, and bar-biting behaviors. Monitoring was conducted from 07:00 to 09:00 h and from 13:00 to 15:00 h on days 3 and 6 of each week from the third to the fifth week postparturition. In addition, the concentrations of tumor necrosis factor- α , interleukin-6, and salivary α -amylase were assessed. During the fourth week postpartum, Damin gilts showed a higher frequency of postural changes from lateral recumbency to other postures and less ventral recumbency, sham-chewing, and bar-biting behavior compared with Large White gilts. However, no significant differences were found between Damin and Large White gilts with regard to urination, defecation, tail wagging, and "tail low" behaviors. The concentrations of serum interleukin-6, salivary α -amylase, and serum tumor necrosis factor- α were higher in Damin gilts than in Large White gilts during the fifth week postpartum. Damin gilts partly achieve lower stress levels during late lactation and better animal welfare than purebred Large White gilts.

Keywords: breed, gilt, lactation, physiology, welfare

1. Introduction

In recent years, welfare considerations have increased the interest in loose-housing systems (large individual house for breeding) for parturient and lactating sows (Damm et al., 2005). Growing evidence indicates that a barren environment during lactation negatively impacts sows, and environmental enrichment might be a useful procedure to improve their welfare (Jarvis et al., 2006; Van de Weerd and Day, 2009). Stress reduction is an important aspect to improve welfare of gilts during late lactation.

This study compares the stress level of two gilt breeds during lactation. The Min pig, a local breed that can be commonly found in north-eastern China, displays good maternal instincts (Cui et al., 2011; Liu et al., 2013). The majority of relevant research has focused on common commercial pig breeds (such as the Large White), and far less is known about how traditional breeds may cope in these intensive production systems. Thus, utilizing maternal advantages, Damin gilts (Large White × Min gilts) were used to evaluate the behavioral and physiological animal welfare of gilts during late lactation.

Interest for the animal welfare of farm animals is increasing to achieve an improvement of our understanding of both animal welfare and health (Mendl and Paul, 2004; Boissy et al., 2007). So far, researchers in this field have focused on sham-chewing (Mason, 1993), bar-biting (Lawrence and Terlouw, 1993), and excessive excretion (Mendl et al., 1997; Mendl and Paul, 2004; Zupan et al., 2016). The results indicated that these behaviors are closely associated with negative emotions (Mendl et al., 1997). Numerous studies have shown that tail postures are indicators of behavioral and psychological responses (Forkman et al., 2007; Reefmann et al., 2009; Boissy et al., 2011; Jones and Boissy, 2011; Marcet-Rius et al., 2019).

It is generally assumed that long-term stress exerts an inhibitory effect on the immune system (Groot et al., 2000). Therefore, IFN- γ (interferon gamma), TNF- α (tumor necrosis factor alpha; which mediates cellular immunity), as well as IL-10 (interleukin) and IgG (immunoglobulin G), both of which mediate humoral immunity, are immune factors that can be used for the evaluation whether animals are in a state of infection or chronic stress. To evaluate the advantages of hybrid sows, this study assessed the behavior and physiology of both Damin and Large White sows during the late lactation period.

2. Material and Methods

All procedures used for the present study were approved by the local Institutional Animal Care and Use Committee (case no. IACECNEAU20121013). This study was performed in Harbin, Heilongjiang Province, China (45°31'12" E longitude, 126°57'11" N latitude, and 126 m altitude).

Eighty first-parity gilts (40 Damin and 40 Large White gilts) that had been mated with Duroc (Canadian) males, were selected. No significant differences were found between both breeds (Damin vs. Large White) with regard to backfat and weight when gilts were mated at an age of 7-8 months (backfat: 18.99 ± 1.22 vs 19.06 ± 0.91 ; weight: 113.39 ± 6.10 vs 115.99 ± 5.01 kg, respectively). Both groups were housed in identical pens (5.5 m width, 5.6 m length), each containing 10 pregnant gilts, a sow feeder, a water nipple drinker, and concrete flooring with straw. A heating pad was set up in the lying area to automatically heat; the gas environment was controlled by an automatic detection system to prevent harmful gases from exceeding standards; and average temperature and humidity were maintained at 24 °C and 65%, respectively. Sows were transferred to parturition pens seven days before the expected due date. Each gilt was randomly assigned to a parturition house. The litter sizes of Damin and Large White gilts were 14.0 ± 1.4 and 12.3 ± 1.5 , respectively (P = 0.018), and their survival rates were 91.80 and 89.69%, respectively (P = 0.08).

The structure of an enriched farrowing pen was used in this study (Figure 1). A wall partitioned the parturition area and piglet activity area in each parturition pen; however, a space between the wall and the floor allowed piglets free access. Each parturition pen held one sow and was made of galvanized tubes with an inset straw board, which was divided into lying and excreting areas. Two drinkers were provided at different heights so that both sows and piglets had convenient access to water. The concrete floor had a slope of 18°. All parturition pens contained straw as litter material (with a thickness of 0.05 m), which was supplemented after cleaning every morning.

Gilts were fed twice daily (at 07:30 and 16:00 h) in their parturition pens. The health of piglets was assessed, and sick piglets were removed from parturition pens at 06:30 h each morning. Approximately 2 cm of bedding was added to parturition pen floors at 07:30 h each weekend. All gilts were restrictively fed 3 kg of complete feed per day, which contained the following constituents per kg: 12.9 MJ ME, 185.0 g crude protein, 50.0 g crude fat, 80.0 g crude ash, and 12.0 g lysine. During the study period, both the temperature and relative humidity of the interior and exterior of the parturition pens were measured daily using a hygrothermograph (Kestrel 4000 Pocket Weather Tracker; Kestrel, Santa Cruz, CA, USA). Daily temperature and humidity inside the parturition pens in August were 21.6 °C and 58.7%, respectively, while in September, the values were 14.7 °C and 51.3%, respectively. Piglets were weaned on day 35 after birth, and neither tails nor teeth were docked.

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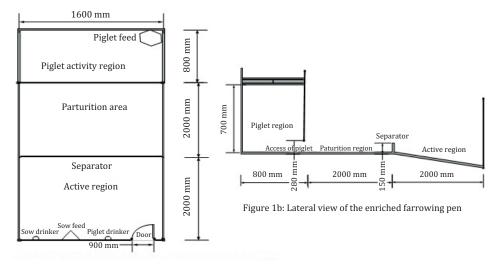


Figure 1a: Vertical view of the enriched farrowing pen

Figure 1 - Structure of the enriched farrowing pen.

Behaviors of gilts were recorded with a video surveillance system (DS-IT5, Hangzhou Hikvision Digital Technology Co., Ltd., Hangzhou, China) to prevent artificially delimited observation times from impacting the test results. From the third to the fifth post-parturition weeks, all behaviors were recorded on video via focal sampling continuous recording from 07:00 to 09:00 h and 13:00 to 15:00 h on days 3 and 6 of each week; this could avoid stockman feeding time and sow sleeping time, and the observation interval twice a week was to increase the behavior of sows at each observation and then prevent the observer's subjective judgment from affecting sow behavior statistics, in addition to avoiding man-made disturbance of sow behavior caused by the noise from cleaning the outside walls and corridors of the farm every Sunday. The behaviors were observed via instantaneous sampling (20-s interval) for the statistics of status behavior in a more accurate and less demanding manner. Data of continuous behaviors were recorded as the number of their occurrence. The parameters and their definitions are presented in Table 1.

Behavior	Definition		
Ventral recumbency ²	Sow's chest and abdomen touching the floor and front legs stretched or folded under the body $^{\rm I}$		
Lateral recumbency to other postures ³	Posture changing from lateral recumbency to other positions including ventral recumbency, sitting, and standing $^{\rm 1}$		
Defecation ³	Elimination of feces from the body ¹		
Urination ³	Discharge of urine from the body ¹		
Tail in curl ²	Tail coiled up in a curl on top of the body ⁴		
Tail wagging ²	Tail swinging in any direction, but mostly from side to side ⁴		
Tail low ²	Tail hanging down against the body ⁴		
Sham-chewing ³	Chewing actions performed without the presence of food in the oral cavity ¹		
Bar-biting ³	Stereotyped biting, gnawing, or sliding of the mouth on accessible part (usually metal bars) of an enclosure ¹		

Table 1 - Behavioral parameters and their definitions¹

¹ Parameters of behaviors and their definitions quoted from Hurnik et al. (1995).

² Presented as percentages of time.

³ Presented as number of occurrences per hour.

⁴ Parameters of behaviors and their definitions quoted from Reimert et al. (2013).

Blood samples of each sow were collected at the 7th day of weeks 3, 4, and 5 of lactation. Immediately after the collection of blood from the ear vein, serum was prepared. The concentrations of IL-6 and TNF- α in serum were measured with a commercial porcine ELISA kit (Nanjing Jiancheng Bioengineering Institute, Nanjing, China). Concentrations of salivary α -amylase in the saliva were assayed using a commercial porcine ELISA kit (Nanjing Jiancheng Bioengineering Institute) (Cook et al., 1996). The sponge block was wrapped in sterile gauze before both sides of the sponge were fastened with sterile ribbon gauze. Then, both sides of the ribbon gauze were connected with a 0.1 m elastic rubber hose. All equipment was disinfected after use.

All dependent variables were examined for normal distribution, using PROC UNIVARIATE of SAS (Statistical Analysis System, version 9.0), considering skewness, kurtosis, the Shapiro-Wilks test (for normality), and a normal probability plot. A GLM procedure of SAS was used to test behavioral differences between both breeds and time-dependent observations. The model for behavioral analysis after parturition included the following effects:

 $Y_{iik} = \mu + Breed_i + Time_i + Breed_i \times Time_i + e_{ii}$

in which μ represents the overall average, *Breed*_i represents the effect of breed on behavior data, *Time*_j represents the effect of the number of weeks after parturition (W₃, W₄, and W₅) on behavior data, and *Breed*_i × *Time*_j indicates the interaction between breeds and observation time. All data are presented as mean ± standard errors.

3. Results

There were no significant differences in behaviors between Damin and Large White gilts during weeks 3 and 4, with the exception of tail curling in the fourth week (P>0.05); however, significant differences were found between Damin and Large White gilts with regard to movement from lateral recumbency to other postures, such as ventral recumbency, sham-chewing, and bar-biting behavior in week 4 (P = 0.030, P = 0.007, P = 0.004). During the study period, no differences were found between Damin and Large White gilts with regard to urination, defecation, tail wagging, and "tail low" behavior (P>0.05). The frequency of movements from lateral recumbency to other postures gradually increased in Damin and Large White gilts over extended lactation (Table 2).

No significant differences were found between both breeds of gilts in serum concentrations of TNF- α and IL-6 during the third and fourth weeks after parturition (P>0.05). The concentration of salivary α -amylase in the third and fourth weeks after parturition also did not significantly differ between both breeds (P>0.05); however, significant differences were found between both breeds in the fifth week in concentrations of TNF- α , IL-6, and salivary α -amylase (P = 0.028, P = 0.035, and P = 0.022, respectively). During extended lactation period, concentrations of TNF- α and salivary α -amylase decreased progressively. Serum concentration of IL-6 in Damin gilts followed an increasing tendency. The concentration of IL-6 of Large White gilts initially followed an increasing trend and then decreased (Table 3).

4. Discussion

Ventral recumbency and movement from lateral recumbency to other postures represent two important indicators of a gilt's refusal to lactate (Gonyou et al., 1998; Valros et al., 2003). In the present study, observations of ventral recumbency at weeks 3-5 postpartum showed that both breeds gradually refused to lactate. This trend toward this type of behavior may indicate that the use of more maternal gilt breeds (i.e., Damin pigs) might still not eliminate the resulting effects on piglets. However, at the fifth week after parturition, Damin gilts spent markedly less time in ventral recumbency than Large White gilts. This might indicate that Damin gilts had particular advantages when dealing with late lactation under stress conditions, and may have a genetically favorable maternal ability and inherited tolerance traits of the Min pig breed (Liu et al., 2013).

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Behavior parameter	Breed	Time			TT:
		3w	4w	5w	Time effect
Ventral recumbency	Damin	10.1a±2.70	12.9ab±4.04	15.2bx±4.62	0.122
	Large White	12.3a±3.11	13.7ab±5.43	17.6by±5.33	NS
	Breed effect	NS	0.030	NS	NS
Lateral recumbency to other postures	Damin	1.4a±0.41	1.7ab±0.42	1.9bx±0.52	0.230
	Large White	1.2a±0.52	1.5ab±0.43	1.6by±0.54	NS
	Breed effect	NS	0.002	NS	NS
Defecation	Damin	0.7±0.20	0.7±0.31	0.8±0.10	0.067
	Large White	0.8±0.10	0.9±0.28	0.9±0.10	NS
	Breed effect	NS	0.071	NS	NS
Urination	Damin	0.8±0.10	0.9±0.10	0.9±0.20	0.210
	Large White	0.7±0.10	0.9±0.20	1.0±0.30	NS
	Breed effect	NS	0.081	NS	NS
Tail in curl	Damin	5.7a±0.20	2.0bx±0.60	3.2c±0.40	0.025
	Large White	4.8a±0.34	5.4by±1.20	2.8c±0.80	NS
	Breed effect	NS	0.027	NS	NS
Tail wagging	Damin	17.4a±0.48	12.5b±0.23	8.6c±0.30	0.011
	Large White	15.4a±0.56	11.3b±0.17	4.1c±0.20	NS
	Breed effect	NS	0.243	NS	NS
Tail low	Damin	76.8a±1.40	85.4b±0.98	91.1c±1.50	0.035
	Large White	79.7a±1.09	84.2b±1.30	93.0c±2.01	NS
	Breed effect	NS	0.601	NS	NS
Sham-chewing	Damin	0.6±0.13	0.8±0.12	0.8x±0.10	0.025
	Large White	0.9a±0.13	1.0ab±0.12	1.1by±0.10	NS
	Breed effect	NS	0.007	NS	NS
Bar-biting	Damin	0.8±0.10	0.9±0.11	1.1y±0.12	0.010
	Large White	0.7a±0.10	1.1ab±0.15	2.0by±0.12	NS
	Breed effect	NS	0.004	NS	NS

Table 2 - Observations of behavioral differences and	changes in two different breeds of gilts kept in the same
environment for 3 to 5 weeks after farrowing	5

NS - not significant.

a,b - Different letters in a row indicate significant differences between the observed weeks (P<0.05).

 x_y - Different letters in a column indicate significant differences between breeds (P<0.05). Data are shown as means and standard errors of the mean.

Physiological index	Time	Damin	Large White
TNF-α (pg mL ⁻¹)	W ₃	195.21a±17.58	186.52a±11.83
	W ₄	165.87b±21.56	162.23b±15.67
	W ₅	159.83bx±14.21	143.34cy±15.68
IL-6 (pg mL ⁻¹)	W ₃	4.74a±0.89	4.83a±0.82
	W_4	5.13ab±0.23	5.42b±1.00
	W ₅	6.35bx±0.17	4.04by±0.45
$\alpha\mbox{-Salivary}$ amylase (pg $\mu\mbox{L}^{\mbox{-1}}$)	W ₃	152.1a±5.56	153.4a±5.23
	W_4	148.14ab±4.65	130.12ab±4.26
	W ₅	142.14bx±6.45	124.23by±5.80

Table 3 - Effect of breed on the physiological indices of gilts at different stages

 $TNF-\alpha$ - tumor necrosis factor alpha; IL-6 - interleukin 6. x,y - Different letters in a row indicate significant differences between observation weeks (P<0.05). a,b,c - Different letters in a column indicate significant differences between breeds (P<0.05).

Data are shown as means and standard deviations.

Excessive excretion has been reported to be closely related to negative animal welfare and stress being experienced by the animals (Jones and Boissy, 2011); however, in this study, both defecation and urination behaviors of both gilt breeds were neither directly affected by late lactation environment nor by piglets. This may be related to the enriched farrowing environments that were used for both gilt breeds in the present study. Results of both gilt breeds were derived from conditions in which the gilts were not sufficiently stressed during the late lactation phase. Nevertheless, other authors have found that excessive excretion might be caused by severe environmental constraints. For example, excessive excretion has been reported for sows that were kept in restricted spaces (Chapinal et al., 2010).

In the present study, the frequency of tail wagging decreased gradually under conditions of extended lactation. This indicates that tail wagging may be a very useful index for animal emotional behavior; however, the association between tail wagging and positive animal welfare still requires a more formal verification (Newberry et al., 1988; Kleinbeck and McGlone, 1993; Terlouw and Porcher, 2005). In the present study, the tail low position occurred more often during the fourth and fifth weeks than during the third week after parturition. This indicates that the tail low position may represent a potentially novel indicator of negative emotional expression in pigs. The expression of tail curling did not appear to follow any particular rule in both breeds during the experimental period. Therefore, it cannot be used as an indicator of animal welfare (with an acceptable standard of accuracy) and, therefore, does not seem to be useful when determining late-lactation behavior and welfare of sows.

Large White gilts exhibited more sham-chewing and bar-biting behaviors during the fourth and fifth weeks postpartum than Damin gilts. This indicates that Large White gilts had a more negative emotional state during late lactation than Damin gilts, which is consistent with previously published observations (Chapinal et al., 2010). Furthermore, we propose abnormal behaviors, such as sham-chewing and bar-biting, to be valuable indicators of negative animal welfare in late-lactating sows.

The concentration of salivary α -amylase tended to gradually decrease during extended lactation, which is consistent with psychological observations in humans. Thus, it is possible that the decrease in salivary α -amylase concentration is regulated by chronic stress (Grigoriev et al., 2003; Wolf et al., 2008). In the present study, the decrease in salivary α -amylase concentration might indicate sustained stress in both gilt breeds during the late lactation period. Therefore, salivary α -amylase can be considered as a valuable indicator for assessment of psychological state of gilts.

Serum concentration of IL-6 was significantly higher in Damin gilts than in Large White gilts during the fifth week after parturition. The major reason for this observed difference might be the dual function of IL-6 as a cytokine that displays both pro- and anti-inflammatory properties (Zaldivar et al., 2006). Thus, the insensitivity of serum IL-6 levels to stress, which was observed in the present study, may be associated with the anti-inflammatory action of IL-6. The exposure of Damin gilts to continuous stress during the fifth week might have caused them to adapt to the environment during the late lactation period via a compensatory increase of the levels of beneficial inflammatory cytokines (Chrousos, 2000; Marshall Jr. and Agarwal, 2000). However, during the fifth week of lactation, the ability of Large White gilts to adapt to environmental stress decreased, which might have led to a decline in the production of immunological factors. Combined with observations of the abovementioned behaviors, this outcome might also indicate that Damin gilts had a higher capacity to adjust to late lactation stress and a superior level of psychological health compared with Large White gilts. With extended lactation period, the serum concentration of IL-6 in Damin gilts increased significantly, while that in Large White gilts increased briefly before following a decreasing trend. These observations were consistent with previously reported results (DeRijk, et al., 1997; Tuchscherer et al., 2009). Such changes might also reflect the psychological component of the stressor, which has been characterized by an uncontrolled and unpredictable appearance of anxiety in animals (Mormede et al., 1988; Brosschot et al., 1998; Peters et al., 1999).

In the present study, serum concentration of TNF- α in Damin gilts was higher than that in Large White gilts during the fifth week of lactation. Since it is conventionally accepted that glucocorticoids specifically suppress the release of pro-inflammatory cytokines (Besedovsky and del Rey, 2000; Sapolsky et al., 2000), the reasons for the differences observed in this study might be due to the relatively

lower degree of stress that was experienced by Damin gilts under the influence of glucocorticoids compared with Large White gilts under similar states of stress. Extending the lactation period placed both gilt breeds under increased stress, and consequently, the serum concentration of TNF- α gradually decreased, which is similar to previously reported results (DeRijk et al., 1997; Besedovsky and Rey, 2000; Sapolsky et al., 2000). A possible explanation for this psychological stress might be beyond the scope of modulated and cell-derived TNF- α . These present findings emphasize the particular importance of TNF- α for assessing animal welfare, health, and psychosocial stress in gilts.

Tumor necrosis factor- α and IL-6 are also important immune-regulatory factors; TNF- α induces Th1-type effector cell differentiation and is a major mediator of cellular immunity (Salem, 2004). Similarly, the cytokine IL-6 is predominantly secreted by Th2-differentiated T-cells and plays a major role in humoral-mediated immunity (Jarvis et al., 1997). In general, Th1-type effector T-cells and Th2 effector T-cells both secrete factors that restrict each other's functions with regard to inflammation and host immunity. In doing so, immunological homeostasis is maintained at the levels of normal cellular and humoral immune competence and functional dynamic balance (Tsai et al., 1997). In the present study, extending the lactation period caused significant increases in concentrations of TNF- α and IL-6 in Damin gilts following parturient readjustment and the balance of Th1/Th2 cells in the blood. However, the immune response of Damin gilts was not only restrained, but also showed an increasing tendency. This result might be related to a compensatory mechanism of increasing selected cytokines and establishing a new set-point for immunological homeostasis in an attempt to adapt to the stress the lactation environment induces. The Th1/Th2 cells in the blood of Large White gilts were in a state of disequilibrium at the fifth week postpartum. This also supports the fact that Large White gilts showed potential for immunological injury during extended lactation.

5. Conclusions

Hybrid Damin gilts show improved maternal behavior and physiological immune performance compared with purebred Large White gilts during late lactation. This indicates that hybrid sows inherited good maternal characteristics and immune capacity. However, both production performance and welfare of sows cannot be improved based on this experiment.

Conflict of Interest

The authors declare no conflict of interest.

Author Contributions

Conceptualization: R. Liu, C. Wang and J. Bao. Data curation: R. Liu, C. Wang, P. Zhao and L. Pan. Formal analysis: R. Liu, C. Wang, R. Yi and P. Zhao. Funding acquisition: R. Yi, H. Liu and J. Bao. Investigation: W. Ji and P. Zhao. Methodology: C. Wang and H. Liu. Project administration: J. Bao. Resources: R. Liu, W. Ji and P. Zhao. Software: R. Liu, W. Ji, C. Wang and L. Pan. Validation: C. Wang. Visualization: C. Wang. Writing-original draft: R. Liu, C. Wang and P. Zhao. Writing-review & editing: R. Liu, W. Ji, C. Wang, P. Zhao. L. Pan, H. Liu and J. Bao.

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