ORIGINAL RESEARCH ARTICLE

Effect of pig birth weight and castration age on growth performance, hematological and antioxidant parameters in suckling period

Efecto del peso al nacimiento de los cerdos y la edad de castración sobre el crecimiento, parámetros hematológicos y antioxidantes durante el periodo de lactancia

Efeito do peso ao nascimento de suinos e da idade de castração sobre o crecimento, os parámetros hematológicos e antioxidantes durante o periodo de aleitamento

Eva S. Safaie[1]; Chan-Ho Kwon[1]; Jannell A. Torres[1]; Young-Dal Jang[1]*.

[1]*Department of Animal and Dairy Science, University of Georgia, Athens, GA 30602, USA*

Received: January 21, 2025. Accepted: May 22, 2025.

***Corresponding author:** *Mailing address: 425 River Road, Athens, Georgia 30602, USA. Tel.: +1-706-542-4098 Fax: +1-706-542-0399, Email: youngdal.jang@uga.edu*

eISSN: 2256-2958

Rev Colomb Cienc Pecu
https://doi.org/10.17533/udea.rccp.e359581

To cite this article:

Safaie ES, CH Kwon, Torres JA, Jang YD. Effect of pig birth weight and castration age on growth performance, hematological and antioxidant parameters in suckling period. Rev Colomb Cienc Pecu. *Year, Vol, number, and pages pending.* DOI : https://doi.org/10.17533/udea.rccp.e359581

Abstract

Background: Castration is one of the most stressful events for suckling pigs. Increasing castration age may help alleviate the stress associated with castration, with the effect depending on the pig's birth weight. **Objective:** This study was conducted to investigate the effect of birth weight and castration age on growth performance, hematological and antioxidant parameters of suckling pigs. **Methods:** At birth, a total of 32 male pigs from 8 litters were assigned to castration age and birth weight. Treatments were: 1) d4-HBW: high-birth weight (HBW; ≥1.5 kg) pigs castrated on d 4 of age, 2) d9-HBW: HBW pigs castrated on d 9 of age, 3) d4-LBW: LBW (≤1.3 kg) pigs castrated on d 4 of age, and 4) d9-LBW: LBW pigs castrated on d 9 of age. Growth performance, hematological and antioxidant parameters were measured at weaning. **Results:** The HBW pigs had a greater ADG ($p<0.05$) than LBW pigs in the suckling period, while there was no difference between pigs castrated at d 4 and d 9 of age. With an interaction between birth weight and castration age ($p<0.05$), the d4-HBW pigs had a lower ADG in d 4 to 9 of age than d9-HBW pigs, while no difference was observed between d4-LBW and d9-LBW pigs. At weaning, d9-castration pigs had greater hemoglobin levels ($p<0.05$), but lower basophil percentage ($p=0.07$, tendency) and plasma malondialdehyde ($p<0.05$) levels than d4-castration pigs. The HBW pigs showed lower hemoglobin ($p<0.05$) and hematocrit ($p=0.06$, tendency) levels than the LBW pigs. **Conclusion:** The castration age did not affect the growth performance of pigs until weaning although early castration for the HBW pigs may temporarily slow their growth rate. Late castration resulted in greater hemoglobin levels and reduced oxidative stress compared to early castration. Despite growing faster than the LBW pigs, the HBW pigs had lower hemoglobin and hematocrit levels.

Keywords: *antioxidant capacity; blood cells; birth weight; castration age; growth; hematology; hemoglobin; immune; oxidative stress; suckling pigs.*

Resumen

Antecedentes: La castración es uno de los eventos más estresantes para los lechones. Aumentar la edad de castración podría contribuir a reducir el estrés asociado con la castración, siendo este efecto dependiente del peso al nacimiento del lechón. **Objetivo**: Este estudio fue realizado para investigar el efecto del peso al nacimiento y la edad de castración en el crecimiento, los parámetros hematológicos y antioxidantes de los lechones. **Métodos:** Al nacer, un total de 32 lechones machos de 8 camadas fueron asignados a diferentes edades de castración y pesos al nacer. Los tratamientos fueron: 1) d4-HBW: cerdos de peso alto al nacer (HBW; ≥ 1.5 kg) castrados a los 4 días de edad, 2) d9-HBW: cerdos HBW castrados a los 9 días de edad, 3) d4-LBW: cerdos de peso bajo al nacer (LBW; ≤ 1.3 kg) castrados a los 4 días de edad, y 4) d9-LBW: cerdos LBW castrados a los 9 días de edad. Las variables evaluadas fueron el rendimiento de crecimiento, los parámetros hematológicos y antioxidantes en el momento del destete. **Resultados**: Los cerdos HBW tuvieron una mayor ganancia diaria de peso (ADG) ($p<0.05$) que los cerdos LBW durante el periodo de lactancia, mientras que no hubo diferencia entre los cerdos castrados a los 4 y 9 días de edad. Con una interacción entre el peso al nacer y la edad de castración ($p<0.05$), los cerdos d4-HBW mostraron una menor ADG entre los 4 y 9 días de edad que los cerdos d9-HBW, mientras que no se observó diferencia entre los cerdos d4-LBW y d9-LBW. Al destete, los cerdos castrados a los 9 días tuvieron mayores niveles de hemoglobina ($p<0.05$), pero menores porcentajes de basófilos ($p=0.07$, tendencia) y menores niveles de malondialdehído plasmático ($p<0.05$) que los cerdos castrados a los 4 días. Los cerdos HBW mostraron niveles más bajos de hemoglobina ($p<0.05$) y hematocrito ($p=0.06$, tendencia) que los cerdos LBW. **Conclusión**: La edad de castración no afectó el crecimiento de los cerdos hasta el destete, aunque la castración temprana en los cerdos HBW podría retrasar temporalmente su tasa de crecimiento. La castración tardía resultó en mayores niveles de hemoglobina y menores niveles de estrés oxidativo en comparación con la castración temprana. A pesar de crecer más rápido que los cerdos LBW, los cerdos HBW presentaron menores niveles de hemoglobina y hematocrito.

Palabras clave: *capacidad antioxidante; células sanguíneas; crecimiento; edad de castración; estrés oxidativo; hematología; hemoglobina; inmunidad; lechones; peso al nacimiento.*

Resumo

Antecedentes: A castração é um dos eventos mais estressantes para os leitões. Aumentar a idade de castração pode ajudar a reduzir o estresse associado à castração, sendo esse efeito dependente do peso ao nascimento do leitão. **Objetivo**: Este estudo foi realizado para investigar o efeito do peso ao nascimento e da idade de castração no crescimento, nos parâmetros hematológicos e antioxidantes dos leitões. **Métodos**: Ao nascer, um total de 32 leitões machos de 8 ninhadas foram designados para diferentes idades de castração e pesos ao nascimento. Os tratamentos foram: 1) d4-HBW: leitões de peso alto ao nascimento (HBW; ≥1.5 kg) castrados aos 4 dias de idade, 2) d9-HBW: leitões HBW castrados aos 9 dias de idade, 3) d4-LBW: leitões de peso baixo ao nascimento (LBW; ≤1.3 kg) castrados aos 4 dias de idade e 4) d9-LBW: leitões LBW castrados aos 9 dias de idade. As variáveis avaliadas foram o desempenho de crescimento, os parâmetros hematológicos e antioxidantes no momento do desmame. **Resultados**: Os leitões HBW apresentaram maior ganho diário de peso (ADG) ($p<0.05$) do que os leitões LBW durante o período de amamentação, enquanto não houve diferença entre os leitões castrados aos 4 e 9 dias de idade. Com uma interação entre o peso ao nascimento e a idade de castração ($p<0.05$), os leitões d4-HBW mostraram um menor ADG entre os 4 e 9 dias de idade do que os leitões d9-HBW, enquanto não foi observada diferença entre os leitões d4-LBW e d9-LBW. No desmame, os leitões castrados aos 9 dias apresentaram maiores níveis de hemoglobina ($p<0.05$), além de menores percentagens de basófilos ($p=0.07$, tendência) e menores níveis plasmáticos de malondialdeído ($p<0.05$) do que os leitões castrados aos 4 dias. Os leitões HBW apresentaram níveis mais baixos de hemoglobina ($p<0.05$) e hematócrito ($p=0.06$, tendência) do que os leitões LBW. **Conclusão**: A idade de castração não afetou o crescimento dos leitões até o desmame, embora a castração precoce em leitões HBW possa retardar temporariamente sua taxa de crescimento. A castração tardia resultou em maiores níveis de hemoglobina e menores níveis de estresse oxidativo em comparação com a castração precoce. Apesar de crescerem mais rápido do que os leitões LBW, os leitões HBW apresentaram menores níveis de hemoglobina e hematócrito.

Palavras-chave: *capacidade antioxidante; células sanguíneas; crescimento; estresse oxidativo; hematologia; hemoglobina; idade de castração; imunidade; leitões; peso ao nascimento.*

Introduction

Surgical castration of male pigs is typically performed within first 10 days of life in commercial swine production to prevent boar taint and reduce aggressiveness. However, the procedure can induce stress and behavioral problems including elevated plasma cortisol concentrations and increased stress vocalizations, changes in behavior such as lying without contact, and other pain-like behaviors (Carroll *et al.*, 2006; Sutherland *et al.*, 2012) and potentially affect preweaning growth and weaning weight of pigs (Kielly *et al.*, 1999; Morales *et al.*, 2017). In previous studies, Kielly *et al.* (1999) reported that castration of pigs at d 3 of age resulted in temporarily slow growth but this impact disappeared by weaning. McGlone *et al.* (1993) reported that pigs castrated at d 1 of age were lighter at weaning than those castrated at d 14 of age. Therefore, the level of pain and distress caused by castration may depend on castration age (Kielly *et al.*, 1999; Rault *et al.*, 2011) and increasing castration age may potentially alleviate negative impacts of castration.

With increasing litter size in hyperprolific sows, pig birth weight decreases while the number of low-birth weight (LBW) pigs and pre-weaning mortality increase (Charneca *et al.*, 2023). The LBW pigs have lower colostrum and milk intake, less-developed digestive system including intestinal barrier functions and absorption capacity, and decreased immunity at weaning compared normal and high-birth weight (HBW) pigs (Verso *et al.*, 2020; Ayuso *et al.*, 2021). Thus, the impact of castration age may be influenced by pig birth weight (Morales *et al.*, 2017). Morales *et al.* (2017) reported that reduced weaning weight by castration was only observed in HBW pigs but not in mid- and light-birth weight pigs potentially due to stress by handling heavyweight pigs, while Kielly *et al.* (1999) reported that lightweight pigs may be more prone to negative impact of early castration than heavyweight pigs. In addition, Fjelkner *et al.* (2024) reported that castration may result in lower hemoglobin levels than female pigs due to blood loss, indicating the impact of castration in hematological parameters. Therefore, it is still unknown if late castration could be beneficial or more stressful for pigs due to the increased body weight at later ages and how castration age and birth weight affect piglet growth, blood hematology and antioxidant status. As surgical castration induces tissue damage and physiological stress, leading to increased cortisol levels that can stimulate the production of reactive oxygen species (Prunier *et al.,* 2005; Giergiel *et al.,* 2021), it is important to evaluate how both procedure and the age at castration affect the antioxidant status of pigs. Therefore, this study was conducted to investigate the effect of castration age and birth weight on preweaning growth rate, hematological and antioxidant parameters at weaning.

Materials and Methods

Ethical considerations

The experiment was conducted under protocols (A2024 01-023-Y1-A0) approved by the Institutional Animal Care and Use Committee of the University of Georgia (GA, USA).

Location

This experiment and sample collections were carried out in an environmentally controlled room at the Swine Research Unit of University of Georgia.

Animals, Experimental Design, and Housing

A total of 32 newborn male pigs (Camborough × PIC337) from 8 sow litters with 10-14 pigs were used in a 2 × 2 factorial arrangement as main factors of birth weight category and castration age. At birth (d 0 of age), 2 sow litters with similar litter size were paired, resulting in 4 pairs. Within each pair, the litters were randomly assigned to 2 castration age treatments: castration at d 4 and 9 of age (average litter size: 11.75 ± 0.75 for castration at d 4 of age and 11.25 ± 0.75 pigs for castration at d 9 of age). Then, male pigs from each litter within the castration age groups (4 litters per age group) were weighed and categorized into 2 birth weight groups: HBW (≥1.5 kg; 1.5-2.2 kg; initial body weight: 1.69 ± 0.18 kg) and LBW (≤1.3 kg; 0.8-1.3 kg; initial body weight: 1.20 ± 0.13 kg). The piglets were then kept within their respective littermates for the duration of the study. This approach follows the method used by Carroll *et al.* (2006) as it is difficult to have male pigs that meet both the birth weight criteria and the castration age requirements within the same litter. Therefore, treatments were: 1) d4-HBW: HBW pigs castrated on d 4 of age (n=6), 2) d9-HBW: HBW pigs castrated on d 9 of age (n=12), 3) d4-LBW: LBW pigs castrated on d 4 of age (n=9), and 4) d9-LBW: LBW pigs castrated on d 9 of age (n=5).

Animal Housing, Processing, and Castration

At 3 d after birth, the pig processing including 200 mg of iron dextran injection (UNIFERON® 200, Pharmacosmos, Inc., Watchung, NJ, USA) into the neck muscle, ear notching, and tail docking was performed under standard operational protocol of the University of Georgia swine farm by the trained personnel. At the assigned age (d 4 and 9 of age), male pigs were carefully castrated by trained personnel using the open method of castration as described by Carroll *et al.* (2006). A sterile scalpel blade was used to make two vertical cuts posterior to each testicle and each testicle was completely removed by cutting the spermatic cord. After the castration process, pigs were immediately returned to their pen. Cross-fostering was not performed, and creep feed was not provided to pigs during the whole experimental period to avoid potential influence from variation in creep feed intake and pigs from different litters. All pigs were housed with their littermates in individual farrowing crates (2.00 m × 2.00 m) at the environmentally controlled farrowing facility. All sows were *ad libitum* access to a common corn-soybean meal-based lactation diet and water by individual feeders and nipples during the whole lactation period. The lactation diet contained 3,300 kcal/kg of metabolizable energy, 22.4% crude protein, and 1.10% total lysine that met or exceeded NRC (2012) nutrient requirement estimates.

Data and Sample Collection and Blood Analysis

All pigs were weighed at d 0, 4, 9 and weaning (20.7 ± 0.9 d of age) and average daily gain (ADG) was calculated by using body weight obtained at each day.

At weaning, blood samples (10 mL) from 4 pigs per treatment selected based on average body weight within each treatment from each litter were collected via jugular venipuncture in disposable vacutainer tubes containing K_3 EDTA (Becton Dickinson, Franklin, NJ, USA) as an anticoagulant. Whole blood samples collected were analyzed for complete blood count including hemoglobin, hematocrit, red blood cell (RBC), red cell distribution width (RDW), mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), platelet count, mean platelet volume (MPV), white blood cell, segmented neutrophils, lymphocytes, monocytes, eosinophils, and basophils performed by the ADVIA 120 Hematology System (Siemens Healthcare Diagnostics, Tarrytown, NY, USA) at the University of Georgia Veterinary Diagnostic Laboratories (Athens, GA, USA). Plasma samples were obtained by centrifugation at 2,500 × g for 30 min at 4°C and stored at –80°C until analysis. Plasma samples were analyzed for superoxide dismutase (SOD) activity and malondialdehyde (MDA) level using colorimetric kits (Cayman Chemical Company, Ann Arbor, MI, USA) and a spectrophotometer (Thermo Fisher Scientific, Waltham, MA, USA).

Statistical Analysis

All data were tested for normality and homogeneity of variances using PROC UNIVARIATE and analyzed using PROC MIXED of SAS (ver. 9.4. SAS Inst. Inc., Cary, NC, USA) in a 2 × 2 factorial arrangement as main factors of birth weight category and castration age. An individual pig was used as an experimental unit for data analysis. The models included birth weight and castration age treatments and birth weight × castration age interaction as fixed effects and the litter pair as a random effect for growth performance and blood parameters. The unequal number of pigs per treatment for growth performance measurement was due to variations in litter size, the number of males within each litter and the distribution of males across birth weight categories, which could not be controlled and may have affected the statistical power of the analysis. To account for this imbalance, the least square means were reported. The least square means were separated using the PDIFF option of SAS. Statistical differences were established at $p<0.05$ and tendencies were established at $p<0.10$. However, the small sample size may have limited statistical power to interpret the data, particularly interaction effects, so non-significant results should be interpreted with caution.

Results

There were no significant differences in weaning weight and overall growth rate during suckling period among the castration age groups (Table 1) except for the body weight at d 4 of age tended to be lower in pigs in the d4 castration treatment than the d9 castration treatment ($p=0.08$), although this difference was not attributable to the treatment effect. However, the HBW pigs had greater ADG ($p<0.05$) than the LBW pigs in the entire suckling period except for d 0 to 4 of age resulting in 2.04 kg heavier weaning weight ($p<0.05$). There was no interaction between birth weight and castration age in body weight and ADG except for an interaction ($p<0.05$) in ADG from d 4 to 9 of age in which the HBW pigs castrated at d 4 of age had lower ADG than the HBW pigs castrated at d 9 of age, while no difference was observed between d 4 and d 9 castration age in the LBW pigs.

Table 1. Growth performance in high and low birth weight pigs with different castration age[1].

	Birth weight			Castration age			p-value		
	High	Low	SEM	d 4	d 9	SEM	Birth weight	Castration age	Interaction
Body weight, kg									
d 0[2](at birth)	1.69	1.20	0.04	1.40	1.49	0.04	<.01	0.20	0.20
d 4	2.18	1.55	0.08	1.78	1.95	0.08	<.01	0.08	0.66
d 9	3.48	2.27	0.21	2.76	3.00	0.21	<.01	0.18	0.14
Wean[3]	6.39	4.35	0.27	5.28	5.47	0.27	<.01	0.62	0.25
Average daily gain, kg/d									
d 0-4	0.121	0.092	0.01	0.095	0.118	0.01	0.11	0.19	0.44
d 4-9	0.261	0.146	0.03	0.196	0.210	0.03	<.01	0.52	0.04[4]
d 9-wean	0.252	0.167	0.02	0.215	0.204	0.02	<.01	0.59	0.23
d 0-9	0.198	0.121	0.02	0.151	0.169	0.02	<.01	0.31	0.28
d 0-wean	0.229	0.151	0.01	0.189	0.191	0.01	<.01	0.90	0.24

[1]Treatments were: 1) d4-HBW: high-birth weight pigs castrated on d 4 of age (n=6), 2) d9-HBW: high-birth weight pigs castrated on d 9 of age (n=12), 3) d4-LBW: low-birth weight pigs castrated on d 4 of age (n=9), and 4) d9-LBW: low-birth weight pigs castrated on d 9 of age (n=5).

[2]day of experiment (age).

[3]Weaning age (average 20.7 ± 0.9 d) was not significantly different among treatments.

[4]Interaction ($p<0.05$) between birth weight and castration age: 0.231, 0.290, 0.161, and 0.130 kg/d (SEM 0.03) for pigs in d4-HBW, d9-HBW, d4-LBW, and d9-LBW treatments, respectively.

Pigs castrated at d 9 of age had greater hemoglobin levels than pigs castrated at d 4 of age ($p<0.05$; Table 2) at weaning. Pigs castrated at d 9 of age tended to have a lower basophil percentage ($p=0.07$). The HBW pigs had lower hemoglobin ($p<0.05$; Table 2) levels and tended to have lower hematocrit ($p=0.06$), and MCH ($p=0.06$) levels than the LBW pigs at weaning. There was an interaction with a tendency in eosinophil percentage ($p=0.07$) in which the LBW pigs castrated at d 4 of age tended to have greater values than the HBW pigs castrated at d 4 of age, while no difference was observed between HBW and LBW pigs castrated at d 9 of age.

Table 2. Complete blood count in high and low birth weight pigs with different castration age at weaning[1,2,3].

	Birth weight			Castration age			p-value		
	High	Low	SEM	d 4	d 9	SEM	Birth weight	Castration age	Interaction
Hemoglobin, g/dL	119.84	128.13	2.44	120.63	127.34	2.44	0.02	0.04	0.19
Hematocrit, %	37.98	41.81	1.25	38.31	41.48	1.25	0.06	0.11	0.80
RBC, 10^6/μl	6.54	6.43	0.17	6.27	6.70	0.17	0.66	0.11	0.28
MCV, fl	58.98	61.94	1.92	61.24	59.68	1.92	0.22	0.50	0.85
MCH, pg	19.51	20.78	0.40	20.28	20.01	0.40	0.06	0.66	0.78
MCHC, g/dL	33.01	33.64	0.57	33.13	33.52	0.57	0.30	0.50	0.76
RDW, %	21.73	20.60	1.00	20.73	21.61	1.00	0.22	0.33	0.44
WBC, 10^3/μl	13.57	10.71	1.92	11.64	12.64	1.92	0.28	0.70	0.46
Neutrophil, 10^3/μl	4.52	4.78	1.18	3.95	5.36	1.18	0.86	0.35	0.55
Lymphocyte, 10^3/μl	8.33	5.35	1.17	6.98	6.69	1.17	0.11	0.86	0.61
Monocyte, 10^3/μl	0.58	0.41	0.09	0.52	0.47	0.09	0.19	0.72	0.50
Eosinophil, 10^3/μl	0.12	0.11	0.03	0.10	0.13	0.03	0.70	0.49	0.66
Basophil, 10^3/μl	0.08	0.07	0.01	0.09	0.07	0.01	0.75	0.33	0.75
Neutrophil, %	33.15	41.60	4.19	36.01	38.73	4.19	0.19	0.66	0.76
Lymphocyte, %	61.10	52.78	4.16	57.74	56.14	4.16	0.20	0.79	0.82
Monocyte, %	4.26	3.90	0.60	4.54	3.62	0.60	0.52	0.13	0.77
Eosinophil, %	0.83	0.94	0.10	0.86	0.90	0.10	0.39	0.77	0.07[4]
Basophil, %	0.56	0.79	0.13	0.85	0.50	0.13	0.22	0.07	0.14
Platelet, 10^3/μl	527.49	560.25	56.70	517.75	569.99	56.70	0.67	0.51	0.97
MPV, fl	9.86	10.13	0.30	10.08	9.91	0.30	0.42	0.61	0.96

[1]n = 4 per treatment.

[2]Treatments were: 1) d4-HBW: high-birth weight pigs castrated on d 4 of age, 2) d9-HBW: high-birth weight pigs castrated on d 9 of age, 3) d4-LBW: low-birth weight pigs castrated on d 4 of age, and 4) d9-LBW: low-birth weight pigs castrated on d 9 of age.

[3]RBC: red blood cell, MCV: mean corpuscular volume, MCH: mean corpuscular hemoglobin, MCHC: mean corpuscular hemoglobin concentration, RDW: red cell distribution width, WBC: white blood cell, and MPV: mean platelet volume.

[4]Interaction (p = 0.07) between birth weight and castration age: 0.675, 0.976, 1.050, and 0.825% (SEM: 0.132) for pigs in d4-HBW, d9-HBW, d4-LBW, and d9-LBW treatments, respectively.

The pigs castrated at d 9 of age had lower plasma MDA levels at weaning than the pigs castrated at d 4 of age ($p<0.05$; Table 3) while there was no significant difference in plasma SOD activity (Table 3) by birth weight or castration age. There was no significant interaction between body weight and castration age in both plasma MDA and SOD levels.

Table 3. Plasma malondialdehyde (MDA) level and superoxide dismutase (SOD) activity in high and low birth weight pigs with different castration age at weaning[1,2].

	Birth weight			Castration age			p-value		
	High	**Low**	**SEM**	**d 4**	**d 9**	**SEM**	**Birth weight**	**Castration age**	**Interaction**
MDA, µM	11.67	11.32	0.95	13.39	9.60	0.95	0.79	0.02	0.23
SOD, U/mL	3.05	4.04	0.41	3.74	3.34	0.41	0.13	0.51	0.57

[1]n = 4 per treatment.

[2]Treatments were: 1) d4-HBW: high-birth weight pigs castrated on d 4 of age, 2) d9-HBW: high-birth weight pigs castrated on d 9 of age, 3) d4-LBW: low-birth weight pigs castrated on d 4 of age, and 4) d9-LBW: low-birth weight pigs castrated on d 9 of age.

Discussion

Castration is a common practice in swine production that is performed using a surgical procedure within 10 d of age to prevent boar taint and reduce aggressiveness. However, it could negatively affect growth performance and behavior. Due to severe pain and stress associated with castration, there have been alternatives to the surgical castration method introduced such as immuno-castration and using anesthesia to minimize pain and stress and improve animal welfare. Although these alternatives have been considered humane methods of castration, concerns remain regarding their effectiveness, the potential stress with multiple vaccinations, meat quality, and consumer acceptance (Weiler *et al.*, 2021). As a result, surgical castration of male piglets remains common in most countries (Bonneau *et al.*, 2019). In surgical castration, the impact of castration may vary depending on castration age (Kielly *et al.*, 1999) and birth weight of pigs (Morales *et al.*, 2017). Therefore, this study evaluated the effect of birth weight and castration age and their interaction on preweaning growth performance, hematological and antioxidant parameters of pigs at weaning.

In the current study, there was no effect of castration age in overall growth performance during the suckling period. This result agrees with Kielly *et al.* (1999) who reported that pigs castrated at d 3 of age had similar weaning weight compared with those castrated at d 10 of age. In contrast, McGlone *et al.* (1993) reported that pigs castrated at d 1 of age had lower preweaning growth rate and weaning weight compared with those castrated at d 14 of age. Castration causes pain and discomfort resulting in behavioral changes and stress response as evidenced by elevated blood cortisol levels (Carroll *et al.*, 2006; Sutherland *et al.*, 2012). However, this cortisol level decreased to the baseline within 3-24 hours (Carroll *et al.*, 2006; Sutherland *et al.*, 2012), which indicates that major stress causing immediate response of pigs may disappear within a day or two. Therefore, this result indicates that castration after d 4 of age may not negatively affect weaning weight of piglets although there could be potential impact remaining until weaning such as hematological and antioxidant status observed in the current study. However, Kielly *et al.* (1999) also reported that there was temporarily slower growth when pigs were castrated at d 3 of age although this was not observed when pigs were castrated at d 10 of age. In the current study, there was an interaction observed between birth weight and castration age in growth rate during d 4 to 9 of age when the castration was already performed for d 4 castration pigs, while the pigs in d 9 castration group remained intact yet. In this period, the HBW pigs castrated at d 4 of age had slightly lower growth rate than the HBW pigs in the d 9 castration group that were not castrated yet whereas there was no impact on growth rate between castration ages in the LBW pigs. This result is consistent with Kielly *et al.* (1999) and indicates that the temporary slow growth due to early castration is more pronounced in pigs with heavier weight compared to those that were not yet castrated. Morales *et al.* (2017) reported that castration reduced growth and weaning weight only in HBW pigs but not in mid- and light-birth weight pigs. Therefore, this result may be attributed to increased stress and discomfort caused by handling of pigs during castration in larger pigs, which could affect milk consumption (Morales *et al.*, 2017). In addition, as there was no interaction between birth weight and castration age in weaning weight and overall growth rate in the suckling period, later castration for the LBW pigs did not have additional benefit in pre-weaning growth performance compared with early castration. It is possible that the elevated stress levels in LBW pigs caused by their lighter weight may hinder the effect of castration age on growth performance as Novais *et al.* (2020) reported an increased liver oxidative stress in LBW pigs at weaning compared with normal-birth weight pigs. However, further study is needed to investigate the relationship between castration age and pig weight at castration, and how LBW affects castration-related stress.

As expected, growth rate was greater in HBW pigs than LBW pigs throughout the suckling period resulting in greater weaning weight difference between HBW and LBW pigs (2.04 kg) compared with the birth weight difference (0.49 kg). This result agrees with previous studies reporting that the HBW pigs grow faster resulting in heavier weaning weight as they could occupy teats having higher milk production compared with the LBW pigs (Škorjanc *et al.*, 2007; Fix *et al.*, 2009; Surek *et al.*, 2019). In addition, lower gene expressions for nutrient digestion, intestinal barrier function, absorption capacity and immune defense in the LBW pigs than the HBW pigs (Verso *et al.*, 2020; Ayuso *et al.*, 2021) may result in increased difference in weaning weight between birth weight categories compared with that in birth weight. Therefore, it can be noted that increasing birth weight can be one of effective ways to enhance weaning weight and survival rate of pigs (Muns *et al.*, 2016) although the other factors (e.g., colostrum intake, litter weight uniformity, etc.) are also important as they can affect weaning weight of pigs (Charneca *et al.*, 2023).

In complete blood count analysis, pigs castrated at earlier age (d 4 of age) had lower hemoglobin levels than those castrated at later age (d 9 of age). There is limited information available regarding the effect of castration age on hematological parameters. Castration could cause substantial blood loss due to cutting spermatic cord (Abendschön *et al.*, 2020; Schmid *et al.*, 2021). Fjelkner *et al.* (2024) reported that pigs castrated at d 3 of age had significantly lower hemoglobin levels than female pigs at weaning and that hemoglobin levels could be affected by the blood loss by castration. Although the current study did not measure blood loss by castration, this blood loss may have a greater impact on iron needs and hemoglobin levels in pigs castrated at an earlier age than later since the proportion of blood loss by castration relative to total blood volume may be greater when pigs are younger than older due to greater total blood volume in pigs at older age. Also, as there is a significant correlation between hemoglobin levels at weaning and postweaning growth rate (Bhattarai and Nielsen, 2015), this difference in hemoglobin levels between castration ages might affect postweaning growth performance, which warrants further investigations.

Interestingly, basophil percentage tended to be greater in pigs castrated at d 4 of age compared with those castrated at d 9 of age. As a greater basophil percentage indicates a greater immune or inflammatory response, this result indicates that pigs castrated at earlier age may be more sensitive to stress compared with older pigs that may have more developed immune system and less pronounced immune or inflammatory response (Yang *et al.,* 2022).

At weaning, the HBW pigs had lower hemoglobin levels and tended to have lower hematocrit and MCH levels than the LBW pigs. It is attributed to growth rate and body weight differences between HBW and LBW pigs. Heavier and faster-growing pigs have a greater blood volume and the iron provided by a single iron injection at birth may not be enough to support the iron needed for hemoglobin synthesis due to the expansion of blood volume, resulting in lower hemoglobin levels in pigs with higher weaning weight (Joliff and Mahan, 2011).

Although there was no significant statistical difference, the LBW pigs had slightly lower lymphocyte counts than the HBW pigs. Although there is limited information available, this result agrees with Amdi *et al.* (2020) who reported that LBW pigs with intrauterine growth retardation had lower percentage of lymphocytes than normal pigs at d 24 of age due to less-developed immune system. With an interaction between birth weight and castration age, the LBW pigs castrated at d 4 of age tended to have greater eosinophil percentage compared to the HBW pigs castrated at d 4 of age, while birth weight did not affect the eosinophil percentage when pigs were castrated at d 9 of age. Eosinophil count and percentage increase when there is an immune or inflammatory response (Cooper *et al.*, 2014). Therefore, this result suggests that early castration may trigger a stronger immune or inflammatory response in LBW pigs than the HBW pigs as they have less-developed immune system (Yang *et al.,* 2022).

Regarding antioxidant parameters, there were no significant differences by birth weight and castration age, and the interaction between birth weight and castration age in plasma SOD activity. However, plasma MDA levels were lower in the pigs castrated at later age than earlier age. Surgical castration increased plasma adrenocorticotropin hormone, cortisol, and lactate indicating stress and tissue damage due to castration (Prunier *et al.,* 2005; Sutherland *et al.,* 2012). Cortisol as a stress hormone can stimulate metabolic processes such as gluconeogenesis and lipolysis that can produce the source of reactive oxygen species (Giergiel *et al.,* 2021). Therefore, stress from castration could induce oxidative stress in pigs. In this study, pigs castrated early resulted in greater oxidative stress as shown by greater plasma MDA levels than those castrated at later age, which indicates that early castration may be more prone to oxidative stress due to castration compared with late castration as castration at older age can be less stressful for pigs than castration early due to more mature immune system (Kielly *et al.,* 1999; Yang *et al.,* 2022). Sutherland *et al.* (2012)

reported that plasma cortisol levels of pigs increased after castration up to 2 hours and then decreased to have no difference from the sham-castrated pigs at 3 hours after castration, which indicates that immediate response to the castration may disappear in 24 hours. Although there was no difference in weaning weight and overall preweaning growth rate by castration age, increased plasma MDA levels in pigs castrated at earlier age than older age indicates that there could be potential prolonged stress (i.e., oxidative stress) during the suckling period due to early castration. Therefore, late castration could provide better welfare for young piglets by reducing oxidative stress and maintaining higher hemoglobin levels in pigs at weaning, supporting their overall health and well-being.

It should be noted that the small sample size in the current study limits generalizability of the findings. Therefore, the results should be interpreted with caution, and further research in larger-scale setting is needed to validate these findings and confirm the potential effects of castration age and birth weight on pig health and performance with larger scale setting.

In the current study, birth weight had no impact on oxidative stress of pigs at weaning. Novais *et al.* (2020) reported that there were no significant differences in antioxidant and oxidative stress parameters in plasma, intestine and kidney between normal birth weight (average 1.73 kg) and LBW (average 1.01 kg) pigs at weaning, which agrees with the current study. However, this study also reported that LBW pigs had greater oxidative stress in the liver at weaning compared with the normal birth weight pigs, which can be attributed to the ATP concentrations that are slightly lower in the LBW pigs than normal birth weight pigs. This result indicates that the response of pigs in antioxidant and oxidative stress parameters could vary depending on specific body tissues and blood analyzed. Further studies with larger populations are necessary to clarify the correlation between birth weight and antioxidant and oxidative stress status in various organs during the suckling period.

Conclusions

The castration age did not affect the growth performance of pigs until weaning although early castration for the HBW pigs may temporarily slow their growth rate. Late castration resulted in greater hemoglobin levels and reduced oxidative stress at weaning compared to early castration. The HBW pigs grew faster than LBW pigs, resulting in a heavier weaning weight, but they had lower hemoglobin and hematocrit levels at weaning. Although late castration did not provide additional benefits in growth rate or weaning weight for LBW pigs, it could be beneficial by reducing oxidative stress and maintaining higher hemoglobin levels compared to early castration. The birth weight and castration age might have slight effects on the production of white blood cells in pigs. Further studies are needed to investigate the potential benefits of late castration on growth rate and health status after weaning, particularly in terms of reducing oxidative stress and increasing hemoglobin levels at weaning compared to early castration, especially in large-scale or commercial settings due to the limitations associated with the small sample size used in the current study, which may reduce its applicability.

Declarations

Acknowledgments

This work is supported by the Hatch Project (#7007665) from the U.S. Department of Agriculture's National Institute of Food and Agriculture and the Georgia Research Foundation. Appreciation is also expressed to Russell Utley for the care of pigs.

Funding

This research was funded by UGA College of Agricultural & Environmental Sciences Undergraduate Research Initiative program.

Conflict of interest

We certify that there is no conflict of interest with any financial organization regarding the material discussed in the manuscript.

Author Contributions

ESS, CHK, JAT, and YDJ were responsible for the design and conception of the study, collecting data, writing, reviewing, and critical reading of the paper. YDJ was responsible for administering the project and editing the paper.

Use of artificial intelligence (AI)

No AI or AI-assisted technologies were used during the preparation of this work.

References

Abendschön N, Senf S, Deffner P, Miller R, Grott A, Werner J, Saller AM, Reiser J, Weiß C, Zablotski Y, Fischer J, Bergmann S, Erhard MH, Baumgartner C, Ritzmann M, Zöls S. Local anesthesia in piglets undergoing castration-a comparative study to investigate the analgesic effects of four local anesthetics based on defensive behavior and side effects. Animals. 2020; 10(10): 1752. https://doi.org/10.3390/ani10101752

Amdi C, Lynegaard JC, Thymann T, Williams AR. Intrauterine growth restriction in piglets alters blood cell counts and impairs cytokine responses in peripheral mononuclear cells 24 days post-partum. Sci Rep. 2020; 10: 4683. https://doi.org/10.1038/s41598-020-61623-w

Ayuso M, Irwin R, Walsh C, Van Cruchten S, Van Ginneken C. Low birth weight female piglets show altered intestinal development, gene expression, and epigenetic changes at key developmental loci. FASEB J. 2021; 35(4): e21522. https://doi.org/10.1096/fj.202002587R

Bhattarai S, Nielsen JP. Association between hematological status at weaning and weight gain post-weaning in piglets. Livest Sci. 2015; 182: 64-68. https://doi.org/10.1016/j.livsci.2015.10.017

Bonneau M, Weiler U. Pros and cons of alternatives to piglet castration: welfare, boar taint, and other meat quality traits. Animals. 2019; 9(11): 884. https://doi.org/10.3390/ani9110884

Carroll JA, Berg EL, Strauch TA, Roberts MP, Kattesh HG. Hormonal profiles, behavioral responses, and short-term growth performance after castration of pigs at three, six, nine, or twelve days of age. J Anim Sci. 2006; 84(5): 1271-1278. https://doi.org/10.2527/2006.8451271x

Charneca R, Freitas A, Nunes J, Le Dividich J. Effects of the mean weight of uniform litters on sows and offspring performance. Animals. 2023; 13(19): 3100. https://doi.org/10.3390/ani13193100

Cooper C, Nonnecke E, Lönnerdal B, Murray J. The lactoferrin receptor may mediate the reduction of eosinophils in the duodenum of pigs consuming milk containing recombinant human lactoferrin. Biometals. 2014; 27:1031-1038. https://doi.org/10.1007/s10534-014-9778-8

Fix J, Cassady J, Herring W, Holl J, Culbertson M, See M. Effect of piglet birth weight on body weight, growth, backfat, and longissimus muscle area of commercial market swine. Livest Sci. 2009; 127(1): 51-59. https://doi.org/10.1016/j.livsci.2009.08.007

Fjelkner J, Sannö A, Emanuelson U. Iron status in piglets at three days of age and at weaning and possible seasonal effects on the blood haemoglobin levels in a Swedish outdoor pig-producing farm. Acta Vet Scand. 2024; 66(13). https://doi.org/10.1186/s13028-024-00735-z

Giergiel M, Olejnik M, Jabłoński A, Posyniak A. The markers of stress in swine oral fluid. J Vet Res. 2021; 65(4): 487-495. https://doi.org/10.2478/jvetres-2021-0065

Joliff JS, Mahan DC. Effect of injected and dietary Fe in young pigs on blood hematology and postnatal pig growth performance. J Anim Sci. 2011; 89(12): 4068–4080. https://doi.org/10.2527/jas.2010-3736

Kielly J, Dewey CE, Cochran M. Castration at 3 days of age temporarily slows growth of pigs. J Swine Health Prod. 1999; 7(4): 151-153. https://www.aasv.org/shap/issues/v7n4/v7n4p151.pdf

McGlone JJ, Nicholson RI, Hellman JM, Herzog DN. The development of pain in young pigs associated with castration and attempts to prevent castration-induced behavioral changes. J Anim Sci. 1993; 71(6): 1441-1446. https://doi.org/10.2527/1993.7161441x

Morales J, Dereu A, Manso A, de Frutos L, Piñeiro C, Manzanilla EG, Wuyts N. Surgical castration with pain relief affects the health and productive performance of pigs in the suckling period. Porc Health Manag. 2017; 3:18. https://doi.org/10.1186/s40813-017-0066-1

Muns R, Nuntapaitoon M, Tummaruk P. Non-infectious causes of pre-weaning mortality in piglets. Livest Sci. 2016; 184: 46-57. https://doi.org/10.1016/j.livsci.2015.11.025

National Research Council. Nutrient requirements of swine. Washington DC, USA: National Academies Press. 2012.

Novais AK, Martel-Kennes Y, Roy C, Deschêne K, Beaulieu S, Bergeron N, Laforest JP, Lessard M, Matte JJ, Lapointe J. Tissue-specific profiling reveals modulation of cellular and mitochondrial oxidative stress in normal- and low-birthweight piglets throughout per peri-weaning period. Animal. 2020; 14(5): 1014-1024. https://doi.org/10.1017/S1751731119002829

Prunier A, Mounier AM, Hay M. Effects of castration, tooth resection, or tail docking on plasma metabolites and stress hormones in young pigs. J Anim Sci. 2005; 83(1): 216-222. https://doi.org/10.2527/2005.831216x

Rault JL, Lay Jr DC, Marchant-Forde JN. Castration induced pain in pigs and other livestock. Appl Anim Behav Sci. 2011; 135(3): 214-225. https://doi.org/10.1016/j.applanim.2011.10.017

Schmid SM, Genter CI, Heinemann C, Steinhoff-Wagner J. Impact of tearing spermatic cords during castration in live and dead piglets and consequences on welfare. Porc Health Manag. 2021; 7(17). https://doi.org/10.1186/s40813-021-00200-7

Škorjanc D, Brus M, Čandek Potokar M. Effect of birth weight and sex on pre-weaning growth rate of piglets. Arch Anim Breed. 2007; 50(5): 476–486. https://doi.org/10.5194/aab-50-476-2007

Surek D, Almeida LM, Panisson JC, Krabbe EL, Oliveira SG, Alberton GC, Maiorka A. Impact of birth weight and daily weight gain during suckling on the weight gain of weaning piglets. Arq Bras Med Vet Zootec. 2019; 71(6): 2034-2040. https://doi.org/10.1590/1678-4162-10786

Sutherland MA, Davis BL, Brooks TA, Coetzee JF. The physiological and behavioral responses of pigs castrated with and without anesthesia or analgesia. J Anim Sci. 2012; 90(7): 2211-2221. https://doi.org/10.2527/jas.2011-4260

Verso LL, Matte JJ, Lapointe J, Talbot G, Bissonnette N, Blais M, Guay F, Lessard M. Impact of birth weight and neonatal nutritional interventions with micronutrients and bovine colostrum on the development of piglet immune response during the peri-weaning period. Vet Immunol Immunopathol. 2020; 226: 110072. https://doi.org/10.1016/j.vetimm.2020.110072

Weiler U, Font-I-Furnols M, Tomasevič I, Bonneau M. Alternatives to piglet castration: from issues to solutions. Animals. 2021; 11(14): 1041. https://doi.org/10.3390/ani11041041

Yang S, Yang N, Huang X, Li Y, Liu G, Jansen CA, Savelkoul HFJ, Liu G. Pigs' intestinal barrier function is more refined with aging. Dev Comp Immunol. 2022; 136: 104512. https://doi.org/10.1016/j.dci.2022.104512