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Effects of iron injection in suckling piglets on growth performance, fecal score, and hematological criteria

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Abstract

A total of 16 sows (average parity = 3.5 ± 0.10, standard deviation = 2.05) and their newborn pigs (Duroc × [Landrace × Yorkshire], initial body weight 1.53 ± 0.07 kg) were used in a 21-day study. On day 3 of age, piglets along with their dam were divided into two groups: CON and TRT. CON group piglets did not receive any iron injection while, TRT group piglets received 200 mg of injectable iron (GleptoForte) in a single dose. The administration of iron at day-3 of birth improved weaning body weight and overall average daily gain in sucking piglets compared with their control counterparts. For blood criteria, injection of 200 mg of iron improved serum iron values and hematocrit and decreased total iron binding capacity (TIBC) during the day 21 period. In summary, 200 mg iron injection at birth resulted in greater growth performance and blood iron status. These results indicate that providing 200 mg of injectable iron is sufficient to optimize the growth performance and blood iron status of suckling piglets.

Key words: gleptoferron, growth performance, hematocrit, iron, weaning

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Introduction

Iron is an indispensable micromineral owing to its involvement in many biological functions; for instance, oxygen metabolism, binding and transport of oxygen, and regulation of cell growth to maintain homeostasis within the body (Pantopoulos et al., 2012). Compared with other microminerals that are regulated through excretion, the conservation of whole-body iron homeostasis is through regulating iron absorption (Williams et al., 2020). In newborn piglets, inefficient absorption of iron decreases the number of circulating red blood cells resulting in anemia with negative influence in growth performance (Kim et al., 2017). Iron deficiency and anemia develop prior to weaning due to iron low stores at birth, rapid growth rate, and low intake of sow colostrum which has more iron content (Hurley, 2015; Mazgaj et al., 2020; Sureshkumar and Kim, 2021). Because of this, an iron injection within 3 days of birth is commonly used in the pig industry to prevent iron deficiency.

The administration of a single 200 mg iron dose is commonly practiced within the first week post-farrowing. However, limited research exists on the specific day of age when the iron injected is administered to optimize performance and iron status. Egeli and Framstad (1999) reported that the supplementation of iron dextran 60 mg injected to newborn piglets from days 1, 3 or 4 after birth showed no evidence in hemoglobin values at 14 or 21 d after birth. However, Peters and Mahan (2008) found that 200 mg injection of iron from iron dextran in a single dose could increase bodyweight and serum iron status of weaning pigs.

GleptoForte (Dr. K, Woogene B&G Co., Ltd., Seoul, Korea) is an injectable iron source that contains gleptoferron. Gleptoferron is a macro-molecule complex that has the potential for increased bioavailability which could allow for improved iron status at weaning and improve growth performance. The optimal dose of iron from gleptoferron in pigs were previously determined to be 200 mg to optimize growth and blood iron status (Williams et al., 2020; Williams et al., 2021). In this study, all pigs were administered iron injection on d 3 after farrowing. However, we are unaware of any studies evaluating the timing of gleptoferron administration after birth. Therefore, the objective of this study was to determine the effects of injectable iron in newborn pigs performance, fecal score and hematological criteria.

Materials and Methods

All procedures involving animals were conducted in line with the protocol approved by the Animal Ethics Committee of Dankook University (Protocol number: DK-2-2114).

Expreimntal design, animals and diets

A total of 16 sows (average parity = 3.5 ± 0.10, standard deviation = 2.05) and their newborn pigs (Duroc × [Landrace × Yorkshire], initial body weight1.53 ± 0.07 kg were used in a 21 study. On day 3 of age, piglets along with their dam were divided into two groups: CON and TRT. CON group piglets did not receive any iron injection while, TRT group piglets receive 200 mg of injectable iron GleptoForte (Dr. K, Woogene B&G Co., Ltd., Seoul, Korea) in a single dose. All diets were formulated to meet the National Research Council (NRC, 2012) requirements of nutrient standards for gestation and lactating sows. The ingredients and compositions of the basal diet are shown in Table 1.

Sows were housed in individual stalls (5.20 m × 4.30 m) and fed twice (07:00 and 15:00) a day with a constant amount of 3 kg during late gestation. Five days before farrowing, the sows were moved into individual farrowing crates (2.10 m × 1.80 m), with parturitions being observed frequently in all groups. The farrowing room was strictly controlled, with disturbances avoided as much as possible, and the inner temperature was kept approximately at 20 to 25℃ by air conditioning system. All the farrowing crates were equipped with a feeder and a nipple drinker for sows; and a heating lamp were provided for suckling piglets. Sows were not provided with feed on their farrowing day but were fed a lactation diet twice a day (06:30 and 17:00) from the next day until weaning. The initial amount of feed was 1.5 kg on the first day postpartum which was increased daily by 0.5 kg until 7 d postpartum, and then the sows were fed ad libitum until weaning. They refused feed was weighed and removed every morning, and feed intake was recorded by subtracting the rejected feed from the feed offered. No creep feed was offered to the piglets throughout the lactation period, and piglets did not have access to the sows' feed. Piglets were treated according to routine management practices that included teeth clipping, tail docking, and ear notching.

Table 1. Ingredient composition and chemical analysis of the basal diet (g·kg^{-1} as-fed basis unless stated otherwise)[x].

Item	Gestation diet	Lactation diet
Ingredients		
Maize (ground)	577.5	510.0
Soybean meal (480 g crude protein·kg^{-1})	100.0	267.3
Wheat bran	110.0	10.0
Rice bran	60.0	50.0
Rapeseed meal	47.0	35.0
Tallow	35.9	60.5
Molasses	36.0	35.0
Dicalcium phosphate	15.2	16.4
Limestone	9.9	7.6
Sodium chloride	6.0	5.0
L-lysine-HCl (780 g·kg^{-1})	0.5	1.2
Vitamin premix[y]	1.0	1.0
Trace mineral premix[z]	1.0	1.0
Analysis compositions (g·kg^{-1})		
Metabolizable energy (MJ·kg^{-1})	13.05	14.47
Crude protein	132.2	183.4
Crude fat	70.1	91.6
Lysine	6.7	10.8
Calcium	8.5	10.6
Total phosphorus	7.6	7.3
Iron (mg·kg^{-1})	20.0	25.0

[x] Maize was replaced by bacterial-iron with the concentration of 2 and 1 g·kg^{-1} for gestation and lactation period, respectively.
[y] Provided per kilogram of complete diet: vitamin A, 12,100 IU; vitamin D3, 2,000 IU; vitamin E, 48 IU; vitamin K3, 1.5 mg; riboflavin, 6 mg; niacin, 40 mg; d-pantothenic, 17 mg; biotin, 0.2 mg; folic acid, 2 mg; choline, 166 mg; vitamin B6, 2 mg; and vitamin B12, 28 μg.
[z] Provided per kilogram of complete diet: copper (as blue vitriol), 15 mg; Zinc (as zinc sulfate), 50 mg; manganese (as manganous oxide), 54 mg; iodine (as potassium iodide), 0.99 mg; and selenium (as sodium selenite), 0.25 mg.

Sample collection and measurements

To determine the Body weight difference individual sow's body weight (BW) was measured at initial, before and after farrowing and at weaning (21 days). The backfat thickness (6 - 8 cm from the midline of the 10th rib) of each sow was measured using piglet 105, SFK Tech real-time ultrasonic instrument (SFK Technology A/S, Herlev, Denmark) at initial, pre- and post-pregnancy and at weaning to determine the backfat- thickness difference. At farrowing, the number and weights of the piglets born, born alive, stillbirths and mummies were recorded. The feed intake and the leftovers were calculated during gestation and lactation to determine the sows' average daily feed intake (ADFI).

The initial (INO) and the final number (FNO) of piglets were also recorded to calculate the survival ratio of piglets during the lactation period. Individual piglet's BW was measured at initial and at weaning. The average daily gain (ADG) of the piglets was calculated by the birth weight (kg) - weaning weight (kg)/length of lactation (day) × 1,000. The weight of the piglets was measured after cross-fostering and at day 7 and day 21 after farrowing, and the numbers of piglets were also recorded to calculate the litter weight gain and average daily gain (ADG) during lactation. The stool consistency of individual pig was recorded (at 08:00 and 20:00) on a pen basis. The fecal score was calculated according to the method described by Zhao et al. (2015) in which scoring system which ranged from 1 to 5; 1, dry pellet; 2, formed feces; 3, moist feces that retains shape; 4, unformed feces that assumes the shape of container; 5, watery liquid that can be poured.

Blood sample were collected via jugular venipuncture in 5-mL vacuum tube with menadione ethylenediaminetetraacetic acid and whole blood (K3EDTA: Becton Dickinson Vacutainer Systems, Franklin Lakes, NJ, USA) tube using 22-gauge, 3 cm needles from 2 piglets from each pen (one barrow and one gilt) on days 3 and 21. Hematological criteria measured included hemoglobin, and hematocrit value using an ADVIA 2021i Hematology System (Siemens Healthcare Diagnostics, Tarrytown, NY, USA) and serum iron concentrations and total iron-binding capacity using a COBAS C501 Chemistry Analyzer (Roche Diagnostics, Indianapolis, IN, USA). The whole blood samples were analyzed for red blood cells (RBC) using an automatic blood analyzer (ADVIA 120, Bayer, Tarrytown, NY, USA).

Statistical analysis

All data were subjected to the GLM procedures of SAS (2014) as a randomized complete block design (SAS Inst. Inc., Cary, NC, USA) according to their BW, with pen as the experimental unit. Differences between the two treatments were separated by student T-test. The variability in the data was expressed as standard error of mean (SEM). Probability values less than 0.05 ($p < 0.05$) were considered as significant.

Results

As shown in Table 2, there was no significant difference in reproduction performance among the treatments. The intramuscular administration of 200 mg Fe into piglets led to a greater overall ADG and BW at day 21 compared to CON group (Table 3). No effects were detected on fecal score of piglets between treated and CON group (Table 4). Piglets receive iron injection, had only increased ($p < 0.05$) serum iron and hematocrit and decreased total iron binding capacity while no difference was observed on red blood cell and hemoglobin at day 21 of lactation (Table 5).

Discussion

Iron is an essential micronutrient that is vital for maintaining homeostasis in mammals as it is involved in numerous cellular processes such as electron transport, energy production, DNA synthesis, and mitochondria function (Aisen et al., 2001). Owing to low iron storages at birth and milk iron being insufficient to meet growth and sustenance needs, newborn pigs are at a serious risk for developing iron deficiency anemia (Mazgaj et al., 2020). Furthermore, low amounts of iron are provided from sow's colostrum with each containing approximately 2.84 and 1.96 $\mu g \cdot mL^{-1}$, respectively (Hurley, 2015). Increasing iron content and/or supplying different sources in the sow's diet in an effort to increase iron given from colostrum to increase iron status or growth have shown to be inconclusive (Novais et al., 2016). Because environmental sources of iron that are available to suckling pigs during lactation are inadequate to meet the iron growth requirement and prevent anemia, an exogenous source of iron is needed. A single intramuscular injection of 200 mg of iron is commonly used in the swine industry to prevent iron deficiency. Knight and Dilger (2018) reported that piglets not provided with injectable iron at birth had greater hematocrit, and hemoglobin and growth rate was accomplished by offering liquid milk-replacer diet with 21.3 $mg \cdot L^{-1}$ iron. Thus, supplemental iron is very vital for young pigs and under commercial rearing conditions and is commonly provided as an iron injection after birth to support piglets and subsequent nursery growth performance.

Table 2. The effect of reproduction performance in lactating sows[w].

Item	Iron (mg)		SEM	p-value
	0	200		
Parity	3.5	3.5	0.4	
Litter size				
Total birth (head)	13.3	12.9	0.7	0.67
Total alive (head)	13	12.8	0.6	0.74
Stillbirth (head)	0.1	0.1	0.1	0.34
Mummification (head)	0.1	0	0.1	0.73
SUR1[x] (%)	98.21	99.17	1.12	0.38
Body weight (kg)				
Initial	251.9	242.4	6.6	1
After farrowing	236.5	226.9	6.6	0.92
Weaning	221.3	211.4	6.5	0.97
Body weight difference 1[y]	15.4	15.5	0.3	0.25
Body weight difference 2[y]	15.2	15.5	0.3	0.18
Backfat thickness (mm)				
Initial	21.3	21.1	0.2	0.67
After farrowing	19.8	19.9	0.2	0.58
Weaning	17.5	17.5	0.3	0.61
Backfat thickness difference 1[z]	-1.5	-1.3	0.2	0.71
Backfat thickness difference 2[z]	-2.3	-2.4	0.2	0.43
Body condition score				
Initial	3.3	3.4	0.1	0.34
After farrowing	3.2	3.2	0.1	0.64
Weaning	2.4	2.5	0.1	0.67
Average daily feed intake (kg)				
Before farrowing	2.75	2.78	0.02	0.43
Lactation	6.21	3.22	0.01	0.12

SEM, standard error of the mean.

[w] A total of 192 pigs (Duroc × [Landrace × Yorkshire]) from 16 litters were used in a 21-day experiment with 12 pigs per sow of each treatment within sow.

[x] Survival rate of number of alive pigs per number of totals born pigs.

[y] Body weight difference: 1, before farrowing to after farrowing; 2, after farrowing to weaning.

[z] BFT, backfat thickness difference: 1, before farrowing to after farrowing; 2, after farrowing to weaning.

Table 3. The effect of Iron injection on growth performance in suckling piglets[x].

Item	Iron (mg)[y]		SEM	p-value
	0	200[z]		
INO	13	12.75	0.12	0.43
FNO	12.63	12.25	0.23	0.40
SUR2 (%)	97.12	95.99	1.95	0.41
Body weight (kg)				
Birth weight	1.46	1.47	0.03	0.52
Weaning	5.33b	6.97a	0.14	0.05
Overall				
ADG (g)	231b	261a	7.00	0.03

INO, initial number of piglets; FNO, final number of piglets; SUR2, survival rate during lactation; SEM, standard error of the mean; ADG, average daily gain.

[x] A total of 192 pigs (Duroc × [Landrace × Yorkshire]) from 16 litters were used in a 21-day experiment with 12 pigs per sow of each treatment within sow.

[y] Iron (Dr. K, Woogene B&G Co., Ltd., Seoul, Korea) dosage administered 3 day after farrowing.

[z] Pigs were administered 200 mg at 3 d after farrowing.

a, b: Values in a row with no common superscripts differ significantly ($p < 0.05$).

Table 4. The effect of Iron injection on Fecal score in suckling piglets[w].

Item	Iron[x] (mg)		SEM	p-value
	0	200[y]		
Fecal score[z]				
Week 1	3.79	3.77	0.02	0.14
Week 2	3.70	3.73	0.04	0.57
Weaning	3.35	3.30	0.02	0.97

SEM, standard error of the mean.

[w] A total of 192 pigs (Duroc × [Landrace × Yorkshire]) from 16 litters were used in a 21-day experiment with 12 pigs per sow of each treatment within sow.

[x] Iron (Dr. K, Woogene B&G Co., Ltd., Seoul, Korea) dosage administered 3 day after farrowing.

[y] Pigs were administered 200 mg at 3 d after farrowing.

[z] Fecal scores were determined using the following fecal scoring system: 1, dry pellet; 2, formed feces; 3, moist feces that retains shape; 4, unformed feces that assumes the shape of container; 5, watery liquid that can be poured.

Table 5. The effect of Iron injection on homatological criteria in suckling piglets[x].

Item	Iron[y] (mg)		SEM	p-value
	0	200[z]		
Day 3				
Serum iron ($\mu mol \cdot dL^{-1}$)	64	51	8	0.44
Total iron binding capacity ($g \cdot dL^{-1}$)	385	358	28	0.77
Red blood cell ($10^6 \cdot \mu L^{-1}$)	4.6	4.53	0.13	0.10
Hemoglobin ($g \cdot dL^{-1}$)	7.2	7.2	0.1	0.45
Hematocrit (%)	34.3	33.9	0.4	0.59
Weaning (Day 21)				
Serum iron ($\mu mol \cdot dL^{-1}$)	167	200	22	0.01
Total iron binding capacity ($g \cdot dL^{-1}$)	427	344	29	0.04
Red blood cell ($10^6 \cdot \mu L^{-1}$)	6.19	6.36	0.15	0.14
Hemoglobin ($g \cdot dL^{-1}$)	11.1	11.6	0.3	0.73
Hematocrit (%)	50.2	64	1.5	0.02

SEM, standard error of the mean.

[x] A total of 192 pigs (Duroc × [Landrace × Yorkshire]) from 16 litters were used in a 21-day experiment with 12 pigs per sow of each treatment within sow.

[y] Iron (Dr. K, Woogene B&G Co., Ltd., Seoul, Korea) dosage administered 3-day after farrowing.

[z] Pigs were administered 200 mg at 3 d after farrowing.

Previous research has determined that effectiveness of a single intramuscular injection of 200 mg of iron to support growth requirements (Morales et al., 2018; Williams et al., 2020; Williams et al., 2021). But, few studies assume that a single intramuscular injection of 200 mg of iron is inadequate to support growth requirements. This is owing to faster growing or larger pigs having serious risk of exhibiting low iron status at weaning and could reduce growth performance (Almond et al., 2017; Gillespie, 2019). Perri et al. (2016) suggest that piglets showing lower iron status at weaning were 0.82 kg lighter 3 weeks post-weaning than pigs with normal iron status. The administration of a single intramuscular injection 200 mg of iron would only support 4 kg of growth before weaning and assessed that 390 mg of iron is needed to prevent the development of iron deficiency before weaning (Van Gorp et al., 2012). Bruininx et al. (2000) reported that the administration of 200 mg of iron 3 days after birth plus a supplemental 200 mg of injected iron 7 day before weaning did not increase in growth compared with a single injection of 200 mg of iron 3 day after birth. Joliff and Mahan (2011) reported that growth performance has not improved when providing 300 mg of iron against 200 mg. Chevalier (2019) observed that piglets receiving an injection of 150 mg iron first day after birth and a supplemental 150 mg injection of iron four day before weaning exhibited higher nursery average daily gain and final body weight compared to pigs receiving a single injection of 150 mg of iron one day after birth. On the other hand, Almond et al. (2017) observed that administration of a second injection with 150 mg of iron approximately 5 to 7 days before weaning provided inconsistent responses in subsequent post-weaning growth compared with a single injection of 150 or 200 mg of iron at 3 to 5 days of age. Williams et al. (2020) found no difference in weaning weight or subsequent weaning growth performance when piglets were provided a single 200 mg iron injection on day 3 after compared with pigs receiving a 200 mg of iron on day 3 after birth and a supplemental 100 mg 10 day before birth.

Hemoglobin concentration is one of the most widely used blood criteria to measures and to estimate iron deficiency and anemia in pigs. Defined normal iron as a Hemoglobin concentration > 11 $g \cdot dL^{-1}$, iron deficiency as a Hemoglobin concentration > 9 $g \cdot dL^{-1}$ but ≤ 11 $g \cdot dL^{-1}$, and anemia as a Hemoglobin concentration ≤ 9 $g \cdot dL^{-1}$ (Bhattarai and Nielsen, 2015b). The negative impact of no iron injection after farrowing on hemoglobin concentrations through weaning and subsequent nursery performance has been established (Peters and Mahan, 2008; Muniyappan et al., 2022). Bhattarai and Nielsen (2015a) reported a positive association between hemoglobin and ADG with an increase in 10 g $Hemoglobin \cdot L^{-1}$ blood corresponding to a weight gain improvement of 17 g daily weight gain 3 weeks post-weaning. In the present study, a significant increase was observed on piglet weaning body weight and overall average daily gain and no difference in fecal scores between iron 200 mg injected treatment compared with piglets that not treated with iron. Also results agree with Wang et al. (2014) who reported that piglets injected with iron had increased individual body weights compared to piglets not injected with iron at day 21 of lactation. Wang and Kim (2012) found a significant increase in body weight and average daily gain of piglets with the administration of 200 mg iron injection. Williams et al. (2021) reported that pig receiving 100 mg of iron 3 day after birth have similar performance to that of pigs receiving 200 mg of iron up to weaning, but Hemoglobin values at weaning were decreased in the pigs receiving 100 mg of iron compared to the pigs receiving 200 mg of iron.

Together with hemoglobin, the hematocrit is also widely used as blood criteria to monitor the iron status of pigs. Other researchers showed that range for hematocrit indicating normal blood iron status as hematocrit value > 30 to 40% and iron deficiency hematocrit value > 30 to 40% (Perri et al., 2017; Chen et al., 2019). Similar to that of Kegley et al. (2002) and Dong et al. (2020), an administration of a single 200 mg injection iron from gleptoferron resulted in improved hematocrit values at weaning and initially post-weaning compared to piglets that were not treated with iron. As with hemoglobin, the performance of pigs receiving 200 mg of iron after birth have improved growth performance of pigs than pigs not treated with iron after birth, but hematocrit values at weaning are improved in pigs receiving 200 mg of iron after birth compared to pigs that are not treated with iron after birth. Similar to that of hemoglobin, a positive relationship was observed between hematocrit values at weaning and growth performance initially post-weaning in pigs (Bhattarai and Nielsen, 2015a). Our study would agree with these results and shows the higher blood iron status with a single injection of 200 mg of iron. Nevertheless, hemoglobin and hematocrit are commonly used as indicators to estimate iron deficiency and anemia in weaning piglets, some scholars reported that these blood criteria may undervalue the iron requirement of piglets because the sensitivity and specifically of these criteria for diagnosis of iron deficiency and anemia are low (Svoboda et al., 2005; Chen et al., 2019).This is because hemoglobin and hematocrit may not accurately indicate early iron deficiency because erythrocytes have a slow turnover rate and are measures of erythropoietic activity in mature erythrocytes (Cook, 2005). Bhattarai and Nielsen (2015b) reported a serum iron and Total iron binding capacity may be more suitable indicators to determine iron deficiency as they are earlier indicators of erythropoietic activity in piglets. Total iron binding capacity is the evaluation of total serum transferrin and revels the quantity available for binding and transfer of iron in the body. Limited reference values for serum iron and total iron binding capacity are available in swine to determine pigs that are iron deficient. Perri et al. (2017) reported that pigs with a serum iron values < 43.0 to 47.0 $\mu mol \cdot L^{-1}$ and total iron binding capacity values < 121.0 - 125.0 $\mu mol \cdot L^{-1}$ would be considered iron deficient.

In the current study, piglets provided a single 200 mg iron injection from gleptoferron from day 3 to after birth had improved serum iron values at day 21. This result agrees with Williams et al. (2021) who reported that a single injection of 200 mg of iron after birth is sufficient to maintain serum iron values above iron deficiency levels at weaning. Furthermore, research has consistently shown that a single injection of 200 mg of iron after birth will improve serum iron values at weaning (Zhao et al., 2015; Li et al., 2018; Morales et al., 2018). Joliff and Mahan (2011) found that a single injection with 200 mg of iron had improved serum iron values above iron deficiency levels at weaning. The current study also observed that a single injection of 200 mg of iron after birth decreased total iron binding capacity values at weaning, indicating more serum transferrin was transporting iron throughout the body and an improved blood iron status. Research has shown that pigs receiving a single injection of 200 mg of iron exhibit lower total iron binding capacity (TIBC) values at weaning and would agree with the results from the study herein (Sperling et al., 2018; Williams et al., 2020; Williams et al., 2021). Morales et al. (2018) also observed that serum iron decreased from days 14 to 21 in pigs injected with 200 mg of iron after birth from either gleptoferron or iron dextran, similar to the study herein.

In summary, our studies have provided evidence that administering 200 mg iron from GleptoFortefrom day 3 of age would optimizes weaning piglets overall average daily gain. Administering iron injection after birth improved body weight, overall average daily gain and blood serum status at weaning.

Conflict of Interests

No potential conflict of interest relevant to this article was reported.

Ethical Approval

All procedures involving animals were conducted in line with the protocol approved by the Animal Ethics Committee of Dankook University (Protocol number: DK-2-2114).

Data Availability Statement

The data presented in this study are available on request from the corresponding author.

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Author Contributions

MM and IHK: Conceptualization and designed the trials. MM: writing–original draft preparation, performed the animal trials, MM and KH: Software, Methodology, Formal analysis, Writing–review and editing, IHK: Supervision. All authors contributed to the article and approved the submitted version.

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