# Effects of iron dosage administered to newborn piglets on hematological measures, preweaning and postweaning growth performance, and postweaning tissue mineral content

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## Summary

**Objective:** To evaluate the effect of iron dosage given at birth on pig growth performance, the course of the preweaning and postweaning blood profile, and postweaning tissue mineral concentration.

**Materials and methods:** Crossbred pigs (n = 70) were assigned to 1 of 5 iron dosages (0, 50, 100, 200, and 300 mg iron) administered by injection on day 0. Body weight and blood samples were collected at day 0, 1, 2, 3, 4, 6, 8, 11, 17, 22, 23, 24, 25, 29, 38, and 52. All blood samples were analyzed for complete blood count (CBC) profile. On day 22, 38, and 52, tissues from 3 pigs per treatment were obtained for analysis of trace minerals (Fe, Zn, Cu, and Mn).

**Results:** Pigs receiving no iron at birth had the slowest growth and lowest hematological profile demonstrating that iron deficiency anemia (IDA) was induced. Hemoglobin concentrations were increased as early as day 6 and continued to increase until day 17 for the 200 and 300 mg iron treatments. Body weight, other hematological measures, and tissue iron content were greater for pigs that received an iron injection at birth.

**Implications:** Pigs that did not receive an iron injection shortly after birth developed IDA resulting in poor growth, low blood hematological measures, and depleted tissue iron reserves. Supplying an iron injection at birth improved preweaning and postweaning growth performance and CBC profile. The magnitude and timing of peak hematological responses was dose dependent.

**Keywords:** swine, iron dextran, iron injection, iron deficiency, dosage

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**Resumen - Efectos de la dosis de hierro administrada a lechones recién nacidos en medidas hematológicas, el rendimiento del crecimiento antes y después del destete y el contenido de minerales tisulares después del destete**

**Objetivo:** Evaluar el desempeño de la dosis de hierro administrada al nacimiento sobre el rendimiento del crecimiento de los cerdos, la trayectoria del perfil sanguíneo antes y después del destete, y la concentración de minerales tisulares después del destete.

**Materiales y métodos:** A los cerdos híbridos (n = 70) se les asignó 1 de 5 dosis de hierro (0, 50, 100, 200, y 300 mg de hierro) administradas por inyección el día 0. El peso corporal y las muestras de sangre se recogieron el día 0, 1, 2, 3, 4, 6, 8, 11, 17, 22, 23, 24, 25, 29, 38, y 52. Todas las muestras de sangre se analizaron para determinar el perfil del conteo de células completo (CBC). Los días 22, 38, y 52, se obtuvieron tejidos de 3 cerdos por tratamiento para el análisis de minerales traza (Fe, Zn, Cu, y Mn).

**Resultados:** Los cerdos que no recibieron hierro al nacer tuvieron el crecimiento más lento y el perfil hematológico más bajo, lo que demuestra que se indujo anemia por deficiencia de hierro (IDA). Las concentraciones de hemoglobina aumentaron desde el día 6 y continuaron aumentando hasta el día 17 para los tratamientos de hierro de 200 y 300 mg. El peso corporal, otras medidas hematológicas y el contenido de hierro en los tejidos fueron mayores en los cerdos que recibieron una inyección de hierro al nacer.

**Implicaciones:** Los cerdos que no recibieron una inyección de hierro poco después del nacimiento desarrollaron IDA que resultó en un crecimiento deficiente, medidas hematológicas bajas en la sangre y reservas tisulares de hierro consumidas. El suministro de una inyección de hierro al nacer mejoró el crecimiento y el perfil de CBC antes y después del destete. La magnitud y el momento de las respuestas hematológicas máximas dependieron de la dosis.

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**Résumé - Effets de la dose de fer administrée aux porcelets nouveau-nés sur les paramètres hématologiques, les performances de croissance avant et après le sevrage et la teneur en minéraux des tissus après le sevrage**

**Objectif:** Évaluer l’effet de la dose de fer administrée à la naissance sur les performances de croissance des porcs, l’évolution du profil sanguin avant et après le sevrage et la concentration minérale tissulaire après le sevrage.

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**Matériels et méthodes:** Des porcs croisés (n = 70) ont été répartis en cinq groupes selon les doses de fer (0, 50, 100, 200, et 300 mg de fer) administrées par injection le jour 0. Le poids corporel et des échantillons de sang ont été prélevés au jour 0, 1, 2, 3, 4, 6, 8, 11, 17, 22, 23, 24, 25, 29, 38, et 52. Tous les échantillons sanguins ont été analysés pour une formule sanguine complète (CBC). Aux jours 22, 38, et 52, des tissus de trois porcs par traitement ont été obtenus pour l'analyse des oligo-éléments (Fe, Zn, Cu, et Mn).

**Résultats:** Les porcs ne recevant pas de fer à la naissance avaient la croissance la plus lente et le profil hématologique le plus bas, démontrant que l'anémie ferriprive (IDA) était induite. Les concentrations d'hémoglobine étaient augmentées dès le jour 6 et ont continué d'augmenter jusqu'au jour 17 pour les traitements de 200 et 300 mg de fer. Le poids corporel, les autres mesures hématologiques et la teneur en fer tissulaire étaient plus élevés chez les porcs ayant reçu une injection de fer à la naissance.

**Implications:** Les porcs n'ayant pas reçu d'injection de fer peu de temps après la naissance ont développé une IDA, ce qui a entraîné une croissance médiocre, de faibles valeurs hématologiques sanguines et une diminution des réserves de fer tissulaires. Fournir une injection de fer à la naissance a amélioré les performances de croissance avant et après le sevrage et le profil sanguin. L'ampleur et le moment des réponses hématologiques maximales dépendaient de la dose.

It is currently common practice to provide newborn piglets with supplemental iron usually through an intramuscular (IM) injection of an iron complex to prevent iron deficiency. Piglets are born with very low iron stores (approximately 50 mg of iron) and only receive approximately 1 mg of iron each day from sow milk.[1] Litter size and piglet growth have improved in current commercial swine production, which suggests the possibility that the traditional iron injection recommendation may not meet the requirements for modern piglets. It has previously been demonstrated that the standard 100 to 200 mg iron injection administered early in life is not sufficient to meet the individual requirements of all pigs, with the faster growing pigs at weaning having the greatest risk of becoming deficient.[2-4] Depending on the growth and metabolism of the pig, it has been suggested that a pig needs approximately 67 mg of iron per kg of body weight (BW) gain.[5] Others have suggested that under current commercial production conditions, where a pig has a normal growth of 5 to 6 kg in a 21-day period, around 310 to 380 mg of iron is required.[6] Therefore, pigs may start to become iron deficient right before weaning in production systems that only supplement 100 to 200 mg iron at birth leading to an iron gap, which is characterized by depleted iron stores before an adequate supply of iron can be absorbed from the nursery diet.[6,7] There has been additional work that shows the importance of higher hemoglobin concentrations at weaning and how it can lead to improved performance during the postweaning period.[8] It has been estimated that the economic impact of iron deficiency in the US swine industry is between $46 million to $335 million.[9] Thus, the objective of this experiment was to evaluate the effect of injectable iron dextran dosage administered at birth on pig growth performance, the course of the preweaning and postweaning blood profile, and postweaning tissue mineral concentration.

## Materials and methods

This experiment was conducted at the University of Kentucky Swine Research Center under protocols approved by the Institutional Animal Care and Use Committee of the University of Kentucky.

### Animals and experimental design

A total of 70 crossbred pigs [32 barrows and 38 gilts; (Yorkshire × Landrace) × Large White] from 7 litters were used. The experiment began in January 2019 and lasted for a total of 52 days. At birth (day 0) piglets were weighed and randomly allotted (random integer generator; Randomness and Integrity Services Ltd) within litters to 5 different iron dextran injection treatments (14 pigs/treatment) in a randomized complete block design using BW and sex as a factor. Treatments consisted of 0, 50, 100, 200, and 300 mg iron dextran (100 mg/mL; Henry Schein Animal Health) administered by IM injection in the right trapezius muscle on day 0. Iron injection treatments were administered by the same individual using a 5 mL syringe with a 20-gauge × 1-inch needle to minimize application variation and backflow. A total of 50 pigs from 5 litters that farrowed on the same day were used for blood sampling, these same pigs were used for each blood collection day. Pigs from the remaining 2 litters were used for measurement of tissue mineral content. On days 22 (weaning), 38, and 52, a total of 15 pigs (3 pigs/treatment) for each time point were euthanized. Pigs selected for tissue collection on day 22 consisted solely of the pigs not used for blood collection. On day 38 and 52, the pigs selected for tissue collection were selected from all remaining pigs based on best representation of the treatment BW average. After the day of iron administration, all personnel responsible for caring for the pigs and blood collection were blinded to pig treatment allotment.

### Housing and diets

Piglets were housed in individual farrowing crates (1.52 × 2.13 $m^2$) with their respective dam in an environmentally controlled room for the first 22 days of the experiment. On day 0 (within approximately 16 hours of birth), all pigs underwent litter processing (weighing, ear notching, needle teeth clipping, and tail docking), blood sampling, and then were administered the assigned iron dosage treatment. All male pigs were castrated on day 8 of the experiment.

The sow lactation diet was provided *ad libitum* and was formulated to supply an added 100 mg/kg iron as ferrous sulfate (Table 1). On day 22 all piglets were weaned to a nursery facility and 4 to 5 pigs were allotted per pen (1.22 × 1.22 $m^2$) based on BW and treatment. Pigs were penned by treatment to assure that the pigs that had received 0 mg iron at birth (and presumed to be anemic) were not bullied by pigs presumed to not be anemic. Each group of pigs were randomly allotted to pens (3 pens/treatment) located throughout the nursery room. The nursery diets fed post weaning were formulated to meet or exceed the nutrient requirements (NRC, 2012) of 7 to 25 kg growing pigs, which included an added 100 mg/kg iron as ferrous sulfate.

## Measurements and sample collection

Blood samples were taken on day 0, 1, 2, 3, 4, 6, 8, 11, 13, 17, 22, 23, 24, 25, 29, 38, and 52. Body weight was also recorded on these days and on day 44. Blood samples (3 mL) were collected by vena cava puncture into EDTA-containing tubes (Becton, Dickinson, and Company). Samples were later analyzed for a complete blood count (CBC) at the University of Kentucky Veterinary Diagnostic Laboratory using a hematological analyzer (Forcyte Veterinary Hematology Analyzer, Oxford Science). The CBC consisted of hemoglobin (Hb), hematocrit (HCT), red blood cell count (RBC), white blood cell count (WBC), mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), and mean corpuscular hemoglobin concentration (MCHC). Tissue samples (liver, spleen, and heart) were collected from 3 pigs/treatment on day 22, 38, and 52. All tissues were ground and mixed to a homogenous mixture, and a subsample was digested by a microwave digester using nitric acid and procedures recommended by the manufacturer (MARS 6; CEM Corporation). After digestion, tissue digests were appropriately diluted and analyzed for trace mineral content (Fe, Zn, Cu, and Mn) using flame atomic absorption spectrophotometry (Thermoelemental, SOLAAR M5; Thermo Electron Corp). Samples were submitted by code to the laboratories thereby blinding laboratory personnel to treatment identity.

**Table 1:** Composition of sow lactation and piglet nursery diets (as-fed basis)

| | | Nursery | |
|---|---|---|---|
| **Item** | **Sow lactation** | **Phase I** | **Phase II** |
| **Ingredient, %** | | | |
| Corn | 69.57 | 50.55 | 57.46 |
| Soybean meal, 48% CP | 27.00 | 28.50 | 32.50 |
| Grease, choice white | - | 2.00 | 2.00 |
| Fish meal (Menhaden) | - | 5.00 | 0.00 |
| Spray-dried animal plasma | - | 2.00 | 0.00 |
| Whey dried | - | 10.00 | 5.00 |
| L-Lysine•HCl | 0.04 | 0.07 | 0.24 |
| DL-Methionine | - | 0.05 | 0.13 |
| L-Threonine | - | 0.07 | 0.14 |
| Dicalcium phosphate | 1.60 | 0.33 | 0.97 |
| Limestone | 0.90 | 0.77 | 0.90 |
| Salt | 0.50 | 0.50 | 0.50 |
| Trace mineral premix* | 0.10 | 0.10 | 0.10 |
| Vitamin premix† | 0.10 | 0.04 | 0.04 |
| Santoquin‡ | 0.02 | 0.02 | 0.02 |
| Other§ | 0.17 | - | - |
| **Total** | **100.00** | **100.00** | **100.00** |
| **Calculated Composition** | | | |
| Metabolizable energy, kcal/kg | 3298.00 | 3423.00 | 3404.00 |
| Crude protein, % | 18.66 | 23.79 | 21.22 |
| SID Lysine, % | 0.87 | 1.35 | 1.23 |
| Calcium, % | 0.84 | 0.80 | 0.70 |
| STTD Phosphorus, % | 0.40 | 0.36 | 0.29 |

* Mineral inclusion per kg of all diets: 50 mg of Mn as manganous sulfate, 100 mg of Fe as ferrous sulfate, 125 mg of Zn as zinc sulfate, 18 mg of Cu as copper sulfate, 0.35 mg of I as calcium iodate, and 0.30 mg of Se as sodium selenite.

† Vitamin inclusion per kg of nursery diet: 9361 IU of vitamin A; 2342 IU of vitamin D3; 62.3 IU of vitamin E; 6.9 mg of vitamin K; 0.026 mg of vitamin B12; 20.9 mg of pantothenic acid; 4.16 mg of riboflavin, 0.23 mg of biotin; 0.17 mg of folic acid; 41.5 mg of niacin; 4.16 mg of vitamin B6; and 1.15 mg of thiamin.

‡ Santoquin (Monsanto) supplied 130 mg ethoxyquin/kg of diet.

§ Includes Chromax (a source of Cr), choline chloride (60%), and copper sulfate supplied at 0.05, 0.10, 0.02 % of the lactation diet (as-fed basis), respectively.

CP = crude protein; SID = standardized ileal digestible; STTD = standardized total tract digestible.

### Statistical analysis

Growth performance and tissue data were analyzed by analysis of variance for a randomized complete block design using PROC GLM of SAS (version 9.4, SAS Institute Inc). Models originally included the treatment and the litter of origin with the pig being the experimental unit. Because the litter of origin was not significant ($P > .10$), it was subsequently dropped from the model. All hematological data were subjected to repeated measures analysis to detect the effect of treatment, day, and treatment × day interaction using PROC MIXED of SAS with an autoregressive covariance structure. Data were evaluated for statistical outliers within each treatment and day using the Grubb's test outlier calculator (GraphPad Software) but were not detected. Orthogonal polynomial contrasts were used to further determine linear and quadratic treatment effects (ie, increasing iron dosage). All data are reported as least squares means with statistical differences being considered significant at $P < .05$ and a tendency at $P < .10$.

## Results

Early during the experiment (day 4), 1 pig from the 200 mg iron injection treatment group died resulting in growth performance means of 13 pigs for that treatment.

### Growth performance

Pigs that did not receive an iron injection at birth had the lowest numerical BW by day 8 that continued through day 52 (Table 2). The low BW is a function of a low cumulative average daily gain (ADG) of the control pigs. By week 2 (day 9-14) there was a quadratic increase in ADG as iron injection dosage increased. At week 3 (day 15-22) the differences in ADG between treatments were more noticeable. Average daily gain continued to be improved in a linear and quadratic fashion through weeks 4 and 5 (day 23-29 and 30-38; the first two weeks post weaning). There were no differences in ADG thereafter; however, the linear and quadratic increase ($P = .01$) remained for overall ADG (day 0-52). The improved ADG associated with increasing iron dosage resulted in statistically heavier BW seen first at weaning (day 22). The BW response to increasing iron dosage remained linear and quadratic ($P \leq .01$) from day 23 to day 52.

### Hematological measures

In addition to poor growth performance, pigs that received no iron injection had the lowest Hb concentration at all sampling times except for day 52 by which time it recovered (Figure 1). A treatment effect, day effect, and treatment × day interaction ($P < .001$; Figure 1) was observed for Hb concentrations. Both the 50 and 100 mg iron dosages had absolute Hb concentrations that peaked at day 6 whereas the Hb concentration for the 200 and 300 mg iron treatments peaked at day 17. Similarly, HCT, RBC, WBC, MCV, MCH, and MCHC were all impacted by the iron dosage as there was a treatment effect, day effect, and treatment × day interaction ($P < .01$; Figures 2 and 3).

The 0 mg iron dosage treatment had the lowest HCT and RBC values throughout the experiment except for day 52 by which time it recovered. However, for MCV and MCH measures these same pigs seem to begin to recover earlier around day 29. The 0 mg iron dose pigs showed elevated MCHC values leading up to day 11 whereupon they decline and then recover by day 38.

### Tissue mineral measures

A total of 3 pigs/treatment/sampling period were used to determine the mineral content of liver, spleen, and heart tissues. Liver iron content (Table 3) was higher in response to increasing iron dosage at weaning (day 22) and day 38 ($P = .004$ and $P = .02$, respectively). Also, at weaning, pigs in the 300 mg iron treatment had liver iron content about 17 times greater than the pigs not receiving iron. Liver zinc content also increased ($P = .01$) with increasing iron treatments at day 52.

Similarly, the spleen exhibited an increase in iron content ($P = .003$) at weaning. However, there was a decrease in spleen zinc content ($P = .03$) as iron dosage increased with a tendency ($P = .08$) to decrease quadratically with the 200 mg iron treatment having the largest reduction, which thereafter was increased (Table 4). At day 38, the relative weight of the spleen to the BW of the pig decreased ($P = .02$) as iron dosages increased. An increase ($P = .04$) in spleen iron content as iron dosage increased was observed again at day 52. Also, at day 52, there was a numerical decrease in spleen zinc content in pigs receiving 0 through 200 mg iron dosage but an increase observed for the 300 mg iron dosage treatment. Over the tissue collection periods of the experiment (days 22, 38, and 52), liver and spleen iron content continually increased over time for pigs receiving 0 to 200 mg iron dosages. However, the 300 mg iron dosage treatment was different as liver and spleen iron content decreased over time. Lastly, the liver and spleen zinc and copper content were much lower on day 52 for all treatments compared to the content at weaning.

There was an increase in the heart iron content ($P = .01$) as iron injection increased (Table 5). Moreover, there was a linear and quadratic decrease in the absolute ($P = .01$ and $P = .02$, respectively) and relative weight ($P = .001$ and $P = .01$, respectively) of the heart at weaning as iron dosages increased. The linear and quadratic effects of decreasing relative heart weight with increasing iron dosages continued to day 38 ($P = .01$ and $P = .004$, respectively), but there were no differences in heart size by day 52. The pigs receiving no iron had the greatest relative heart weights at both weaning and day 38.

## Discussion

Increasing iron dosages at birth resulted in increased growth performance during the preweaning and postweaning periods. The improved growth in the present experiment was mostly noticed during days 15 to 22, which was the week preceding weaning, and the first 2 weeks of the nursery period (days 23-38). The days leading up to weaning (days 17-21) have been shown to be important in regard to hematological measures declining below optimal levels after receiving a standard iron injection administered early in life.[2-4] It has also been observed that optimal iron status (Hb > 11 g/dL) at weaning may lead to improved growth performance in the subsequent nursery period.[8] The positive growth performance that may be associated with optimal iron status may be attributed to improved oxygen transport, immune function, vitality, and metabolism.[10] In the current experiment, the improved growth observed around weaning and after weaning was associated with an improvement in the iron status via increasing the iron dosage at birth.

A similar study[11] looking at administration of increasing amounts of injectable iron (0, 50, 100, 150, and 200 mg iron) at processing also resulted in linear and quadratic improvements ($P < .001$) in ADG from day 3 to 21 with the 100 mg

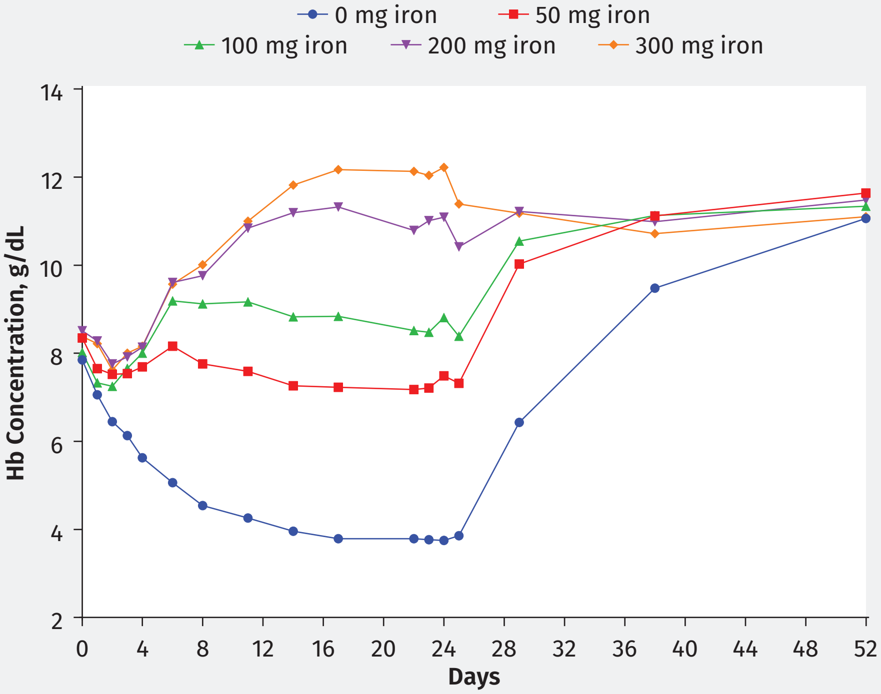
**Table 2:** Least squares means of individual preweaning and postweaning BW and ADG by iron dosage*

| | Iron dosage, mg | | | | | | P value | |
|---|---|---|---|---|---|---|---|---|
| | 0 | 50 | 100 | 200 | 300 | SEM | L | Q |
| **BW, kg** | | | | | | | | |
| d 0 | 1.45 | 1.45 | 1.44 | 1.46 | 1.49 | 0.05 | .51 | .73 |
| d 1 | 1.60 | 1.58 | 1.56 | 1.63 | 1.64 | 0.06 | .42 | .63 |
| d 2 | 1.76 | 1.74 | 1.71 | 1.77 | 1.86 | 0.07 | .20 | .26 |
| d 3 | 1.96 | 1.93 | 1.93 | 1.98 | 2.06 | 0.07 | .22 | .42 |
| d 4 | 2.13 | 2.10 | 2.11 | 2.15 | 2.25 | 0.08 | .15 | .40 |
| d 6 | 2.55 | 2.53 | 2.59 | 2.59 | 2.70 | 0.08 | .14 | .63 |
| d 8 | 2.97 | 3.01 | 3.08 | 3.05 | 3.19 | 0.10 | .10 | .92 |
| d 11 | 3.57 | 3.76 | 3.84 | 3.73 | 3.89 | 0.12 | .12 | .60 |
| d 14 | 4.20 | 4.55 | 4.59 | 4.51 | 4.64 | 0.15 | .11 | .31 |
| d 17 | 4.75 | 5.33 | 5.35 | 5.32 | 5.34 | 0.18 | .08 | .09 |
| d 22 | 5.48 | 6.69 | 6.62 | 6.67 | 6.63 | 0.25 | .01 | .01 |
| d 23 | 5.27 | 6.44 | 6.44 | 6.51 | 6.39 | 0.24 | .01 | .01 |
| d 24 | 5.44 | 6.93 | 6.87 | 6.93 | 6.77 | 0.26 | .01 | .001 |
| d 25 | 5.60 | 7.28 | 7.24 | 7.34 | 7.09 | 0.27 | .004 | < .001 |
| d 29 | 6.87 | 8.81 | 8.88 | 9.12 | 8.72 | 0.32 | .002 | < .001 |
| d 38 | 10.87 | 13.91 | 14.02 | 14.15 | 14.12 | 0.49 | < .001 | < .001 |
| d 44 | 14.90 | 17.53 | 18.03 | 18.37 | 18.20 | 0.65 | .002 | .01 |
| d 52 | 20.14 | 22.77 | 24.02 | 23.48 | 23.51 | 0.77 | .01 | .01 |
| **ADG, g** | | | | | | | | |
| d 0-8 | 189.7 | 195.5 | 205.0 | 199.2 | 212.3 | 7.92 | .06 | .89 |
| d 9-14 | 204.7 | 257.3 | 250.8 | 241.9 | 241.3 | 12.39 | .26 | .04 |
| d 15-22 | 160.1 | 266.7 | 253.4 | 271.2 | 247.1 | 18.96 | .01 | .001 |
| d 23-29 | 199.2 | 303.8 | 323.3 | 349.0 | 299.4 | 19.49 | .002 | < .001 |
| d 30-38 | 445.0 | 566.4 | 571.8 | 559.0 | 599.4 | 24.94 | < .001 | .05 |
| d 39-44 | 616.6 | 641.6 | 665.2 | 692.2 | 683.9 | 39.05 | .14 | .44 |
| d 45-52 | 748.2 | 747.9 | 855.1 | 730.0 | 758.2 | 40.13 | .76 | .34 |
| d 23-52 | 504.8 | 558.1 | 597.6 | 580.5 | 576.9 | 21.93 | .04 | .02 |
| d 0-52 | 366.6 | 418.5 | 442.8 | 430.9 | 432.4 | 14.72 | .01 | .01 |

* A total of 10 pigs/treatment were assigned to 1 of 5 iron dosages administered on day 0. All pigs were weaned on day 22. Day 44 and 52 means are representative of 8 pigs/treatment.

BW = body weight; ADG = average daily gain; L = linear; Q = quadratic.

**Figure 1:** Effects of iron dosage on preweaning and postweaning hemoglobin (Hb) concentration. Iron dosages were administered on day 0 in the form of iron dextran, all pigs were weaned on day 22. Data was subjected to ANOVA by repeated measures and reported as least squares means from 10 pigs/treatment on all days except day 52 (8 pigs/treatment). There was a treatment, day, and treatment × day interaction ($P < .001$).



iron dosage showing the greatest increase and no further improvement thereafter. Somewhat similar, increasing the injectable iron dosage at birth in the current experiment led to a linear increase during week 1, which was later observed again in week 3 alongside a quadratic response with the biggest improvement observed for the 200 mg iron dose.

Overall, pigs not receiving an iron supplement (0 mg iron) demonstrated the lowest growth performance which led to the lowest final BW. This poor growth performance from the 0 mg iron injection group was accompanied by lower CBC measures by day 4 for all measures except MCHC demonstrating that iron deficiency anemia (IDA) was induced as planned by the experimental design.

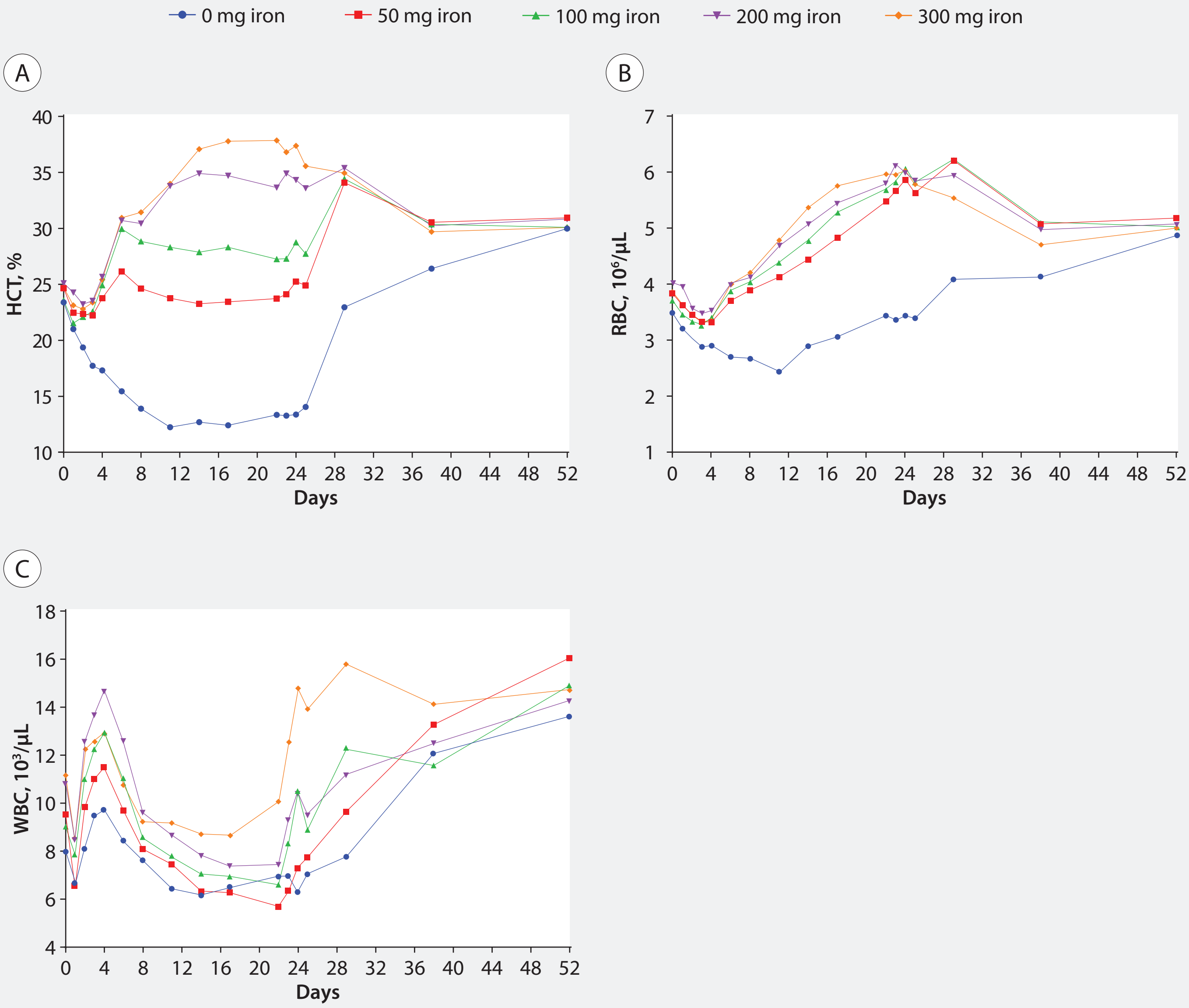
It is proposed that an IM injection of iron dextran is absorbed by the body relatively fast through the reticuloendothelial system due to the phagocytes in the liver, spleen, and bone marrow.[12] The absorbed iron is then reserved in storage sites and is subsequently transported to bone marrow for Hb synthesis, a process that can take several days in total. This may explain why there was an improvement in Hb and HCT in the current experiment at around day 4 and 6 that continued to increase until around weaning (day 22).

Pigs that received the 0 and 50 mg iron dosages were below the *Schlam's Veterinary Hematology*[13] reference range for Hb concentration (10-16 g/dL) until day 38 and day 29, respectively. Iron deficiency anemia is often defined as an Hb concentration below 9 g/dL.[3,4,13] In the current experiment, both the 0 and 50 mg iron injection treatments had Hb concentrations that were below this anemic classification for most of the experiment. On day 6 the pigs receiving 100, 200, and 300 mg iron dosages had Hb concentrations that surpassed the anemic status. Although pigs in the 100 mg iron treatment later dipped below 9 g/dL on day 14 which lasted until day 29, the pigs receiving 200 and 300 mg iron remained in the Hb reference range for the entirety of the study.

Mean corpuscular hemoglobin concentration is the Hb concentration within the red blood cell usually indicating the oxygen-carrying capacity of the blood. Different from all other CBC measurements for the control pigs, MCHC increased from day 6 to 11. However, the MCHC suddenly decreased from day 11 to 29. Data reported by Egeli et al[14] demonstrated that anemic pigs supplied with no iron at birth have higher MCHC values at day 21 than pigs that received an iron injection. This would explain and agree with the current experiment where an increase in MCHC was observed from day 6 to 11 in pigs that did not receive an iron injection. Given that total oxygen carrying capacity would be a function of the RBC and the MCHC, it is proposed that the lower RBC in pigs not receiving iron at birth may cause the body to compensate by loading the red blood cells with the hemoglobin that is present. From day 17 through 29 there was an increase in MCHC with increasing iron dosage, this improvement is simply explained by the other improvements in CBC measurements associated with increasing iron dosage that all contribute to an overall improved hematological profile. The elevated MCHC for the noninjected pigs is particularly interesting because it seems that these pigs are demonstrating a biological compensation for the lack of body iron until it is physically incapable of doing so (after day 11) where it then suddenly decreases. Thus, the initial response is to increase MCHC until such time that it is no longer possible and then it declines. While iron toxicity is always a concern when supplying greater doses of iron, within the conditions of the current experiment, supplying 300 mg of iron dextran at birth did not show any of the classical clinical signs of iron toxicity (lethargy, edema around injection site, muscle convulsions, and sudden death).

In the current experiment, the iron content of the liver, spleen, and heart at weaning increased as the injectable dosage at birth increased. The liver and spleen are major sites for ferritin and hemosiderin which are iron storage compounds that act as a reserve and are used for hemoglobin synthesis.[15] Thus why there was a linear response to iron dosages for liver and spleen iron content at weaning. Iron transport through the body is dependent on the transport protein transferrin. Transferrin delivers iron at a rate dependent on the pace of red blood cell production which is dependent on the overall iron status of the individual.[16] This concept may explain why in the present study there were greater concentrations of iron in tissues of those pigs receiving greater iron dosages.

**Figure 2:** Effects of iron dosage on preweaning and postweaning A) hematocrit (HCT), B) red blood cell count (RBC), and C) white blood cell count (WBC). Iron dosage treatments were administered on day 0 in the form of iron dextran, all pigs were weaned on day 22. Data were subjected to ANOVA by repeated measures and reported as least squares means from 10 pigs/treatment on all days except day 52 (8 pigs/treatment). There was a treatment, day, and treatment × day interaction ($P < .01$) for HCT, RBC, and WBC.



At weaning, the heart was larger for pigs receiving no supplemental iron. Due to the low amount of Hb or oxygen in the blood of anemic pigs, it is proposed that the heart must compensate and increase output to deliver more blood and oxygen to tissues. These results are supported by Dallman[15] who described that severe anemia leads to cardiac hypertrophy as observed at weaning in the current experiment.

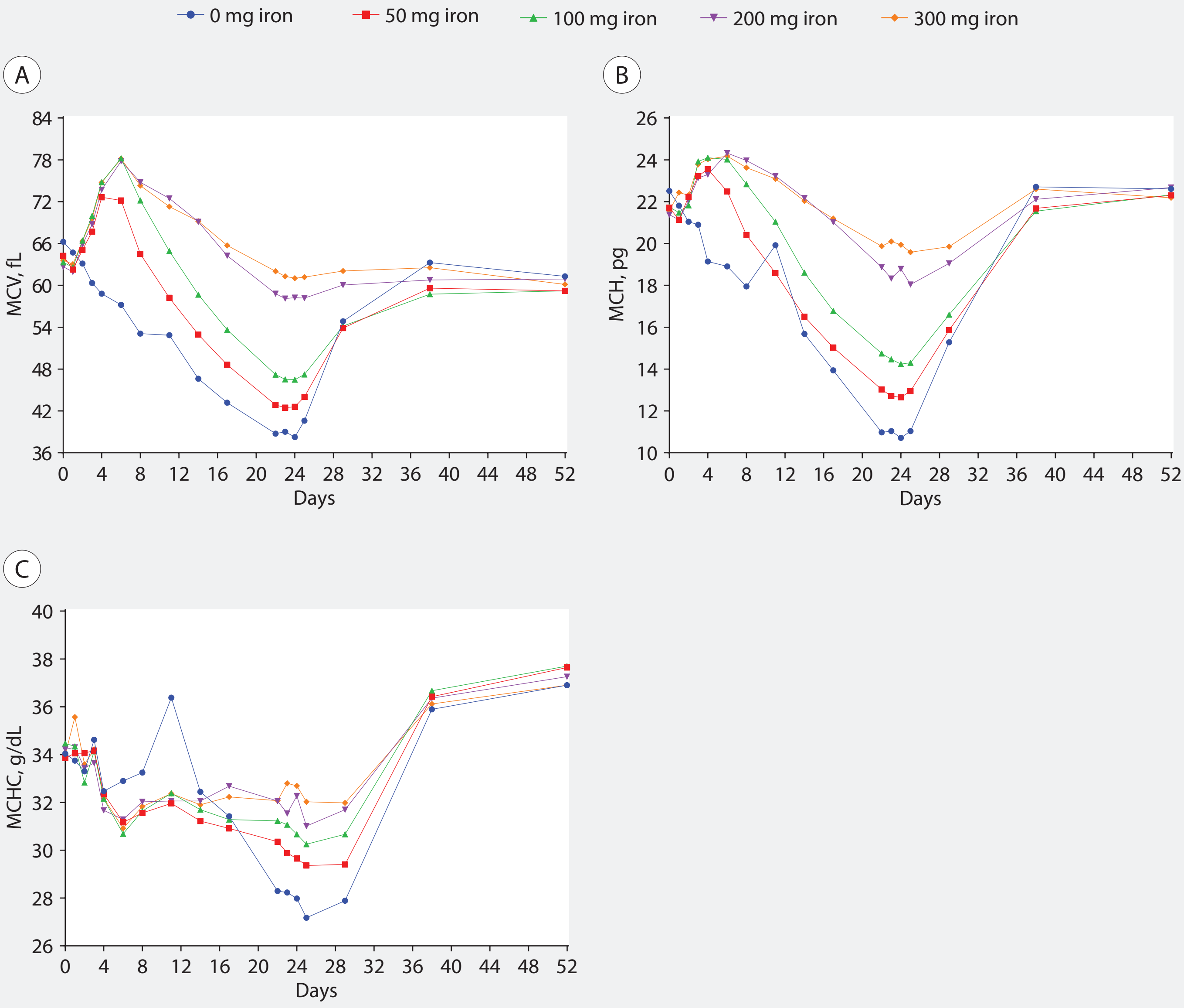
Also, at weaning, the zinc content of the spleen was reduced as iron dosage increased. Iron and zinc have been known to have competitive interaction for cellular transport especially when there are elevated iron levels.[17] Camaschella and Pagani[18] demonstrated that with higher iron concentrations in the body, zinc transporter protein 14 (ZIP14) will transport iron into hepatocytes and other cells. This could also explain the trend for a decrease in liver zinc content observed at day 38, which later increased by the end of the experiment (day 52). The liver and hepatocytes may still be processing the higher iron concentrations observed at weaning, but once the iron concentrations are under control (observed at day 38) the liver and hepatocytes can then start to compensate for the lower zinc concentrations leading to the increase in zinc concentrations by day 52.

## Implications

Under the conditions of this study:

- With no supplemented iron injection, piglets develop IDA shortly after birth.
- An iron injection at birth improves overall growth and CBC profile of piglets.
- Iron dosage impacts the magnitude and timing of peak hematological responses.

**Figure 3:** Effects of iron dosage on preweaning and postweaning A) mean corpuscular volume (MCV), B) mean corpuscular hemoglobin (MCH), and C) mean corpuscular hemoglobin concentration (MCHC). Iron dosage treatments were administered on day 0 in the form of iron dextran, all pigs were weaned on day 22. Data were subjected to ANOVA by repeated measures and reported as least squares means from 10 pigs/treatment on all days except day 52 (8 pigs/treatment). There was a treatment, day, and treatment × day interaction ($P < .01$) for MCV, MCH, and MCHC.



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### Conflict of interest

None reported.

### Disclaimer

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**Table 3:** Least squares means of liver mineral content[*] by iron dosage[†]

| | Iron dosage, mg | | | | | | *P* value | |
|---|---|---|---|---|---|---|---|---|
| | 0 | 50 | 100 | 200 | 300 | SEM | L | Q |
| **D 22** | | | | | | | | |
| BW, kg | 6.39 | 6.61 | 5.03 | 6.20 | 6.04 | 0.53 | .71 | .37 |
| Liver WT, g | 193.03 | 223.70 | 159.17 | 213.15 | 214.97 | 18.46 | .44 | .48 |
| Liver WT, % BW | 3.03 | 3.40 | 3.18 | 3.43 | 3.58 | 0.21 | .13 | .91 |
| Fe | 95.8 | 143.0 | 204.5 | 402.9 | 1652.5 | 348.73 | .004 | .15 |
| Zn | 287.8 | 247.6 | 296.9 | 211.5 | 276.0 | 23.75 | .47 | .26 |
| Cu | 413.6 | 415.6 | 482.8 | 448.1 | 413.7 | 56.34 | .99 | .43 |
| Mn | 7.1 | 6.4 | 6.3 | 6.4 | 8.6 | 1.21 | .36 | .25 |
| **D 38** | | | | | | | | |
| BW, kg | 9.73 | 14.94 | 14.30 | 13.84 | 14.51 | 1.16 | .08 | .09 |
| Liver WT, g | 367.40 | 602.57 | 536.17 | 538.03 | 570.47 | 67.93 | .20 | .26 |
| Liver WT, % BW | 3.72 | 3.99 | 3.74 | 3.89 | 3.91 | 0.25 | .72 | .94 |
| Fe | 380.1 | 627.1 | 586.5 | 610.8 | 654.2 | 57.61 | .02 | .13 |
| Zn | 146.0 | 137.8 | 117.2 | 100.8 | 107.4 | 17.26 | .08 | .35 |
| Cu | 159.6 | 73.9 | 99.9 | 121.9 | 85.7 | 29.48 | .38 | .58 |
| Mn | 5.8 | 6.1 | 7.3 | 5.7 | 6.6 | 0.35 | .49 | .38 |
| **D 52** | | | | | | | | |
| BW, kg | 21.19 | 22.27 | 24.49 | 22.53 | 22.61 | 2.38 | .80 | .52 |
| Liver WT, g | 803.00 | 806.83 | 870.63 | 865.17 | 828.57 | 90.84 | .78 | .60 |
| Liver WT, % BW | 3.80 | 3.63 | 3.56 | 3.84 | 3.66 | 0.11 | .99 | .69 |
| Fe | 742.4 | 796.5 | 819.6 | 786.0 | 913.4 | 90.74 | .27 | .81 |
| Zn | 114.5 | 129.5 | 132.6 | 182.1 | 180.2 | 17.78 | .01 | .58 |
| Cu | 35.5 | 25.2 | 35.1 | 33.3 | 30.1 | 3.81 | .80 | .90 |
| Mn | 4.2 | 5.5 | 3.3 | 2.8 | 3.1 | 0.67 | .06 | .54 |

[*] Mineral content is reported as mg/kg of tissue as measured on a dry matter basis.

[†] Pigs were administered 1 of 5 iron dosages on day 0. All pigs were weaned on day 22. A total of 3 pigs/treatment were used for tissue analysis at day 22, 38, and 52.

BW = body weight; WT = weight; L = linear; Q = quadratic.

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**Table 4:** Least squares means of spleen mineral content* by iron dosage†

| | Iron dosage, mg | | | | | | *P* value | |
|---|---|---|---|---|---|---|---|---|
| | **0** | **50** | **100** | **200** | **300** | **SEM** | **L** | **Q** |
| **D 22** | | | | | | | | |
| BW, kg | 6.39 | 6.61 | 5.03 | 6.20 | 6.04 | 0.53 | .71 | .37 |
| Spleen WT, g | 15.63 | 18.43 | 16.83 | 25.15 | 17.67 | 3.37 | .39 | .25 |
| Spleen WT, % BW | 0.25 | 0.28 | 0.33 | 0.40 | 0.28 | 0.06 | .37 | .09 |
| Fe | 483.6 | 665.1 | 983.0 | 1189.3 | 1149.8 | 134.59 | .003 | .10 |
| Zn | 83.5 | 81.7 | 73.4 | 62.7 | 72.8 | 4.53 | .03 | .08 |
| Cu | 7.3 | 8.2 | 6.0 | 6.3 | 6.2 | 0.96 | .17 | .58 |
| Mn | 1.47 | 1.35 | 1.34 | 1.07 | 1.56 | 0.27 | .99 | .22 |
| **D 38** | | | | | | | | |
| BW, kg | 9.73 | 14.94 | 14.30 | 13.84 | 14.51 | 1.16 | .08 | .09 |
| Spleen WT, g | 31.17 | 41.13 | 34.03 | 33.63 | 34.67 | 4.19 | .91 | .70 |
| Spleen WT, % BW | 0.32 | 0.27 | 0.24 | 0.24 | 0.24 | 0.02 | .02 | .07 |
| Fe | 581.9 | 692.1 | 643.4 | 713.4 | 644.9 | 84.13 | .66 | .42 |
| Zn | 56.0 | 67.5 | 59.4 | 56.1 | 56.5 | 6.60 | .58 | .72 |
| Cu | 4.4 | 4.5 | 3.8 | 3.7 | 3.7 | 0.40 | .16 | .49 |
| Mn | 1.17 | 1.08 | 1.06 | 1.07 | 1.14 | 0.07 | .93 | .22 |
| **D 52** | | | | | | | | |
| BW, kg | 21.19 | 22.27 | 24.49 | 22.53 | 22.61 | 2.38 | .80 | .52 |
| Spleen WT, g | 54.13 | 82.53 | 82.37 | 74.30 | 84.90 | 15.80 | .38 | .53 |
| Spleen WT, % BW | 0.28 | 0.37 | 0.33 | 0.33 | 0.39 | 0.08 | .51 | .98 |
| Fe | 774.7 | 1382.6 | 1125.7 | 1308.5 | 1571.7 | 194.75 | .04 | .68 |
| Zn | 63.6 | 44.6 | 61.0 | 48.1 | 66.0 | 7.05 | .60 | .13 |
| Cu | 3.7 | 4.6 | 4.5 | 3.8 | 5.4 | 0.41 | .07 | .41 |
| Mn | 1.07 | 0.70 | 1.04 | 0.87 | 0.98 | 0.11 | .99 | .42 |

* Mineral content is reported as mg/kg of tissue as measured on a dry matter basis.

† Pigs were administered 1 of 5 iron dosages on day 0. All pigs were weaned on day 22. A total of 3 pigs/treatment were used for tissue analysis at day 22, 38, and 52.

BW = body weight; WT = weight; L = linear; Q = quadratic.

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**Table 5:** Least squares means of heart mineral content* by iron dosage†

| | Iron dosage, mg | | | | | | *P* value | |
|---|---|---|---|---|---|---|---|---|
| | **0** | **50** | **100** | **200** | **300** | **SEM** | **L** | **Q** |
| **D 22** | | | | | | | | |
| BW, kg | 6.39 | 6.61 | 5.03 | 6.20 | 6.04 | 0.53 | .71 | .37 |
| Heart WT, g | 57.50 | 46.07 | 32.97 | 42.90 | 38.67 | 3.46 | .01 | .02 |
| Heart WT, % BW | 0.90 | 0.70 | 0.66 | 0.70 | 0.65 | 0.03 | .001 | .01 |
| Fe | 163.19 | 190.56 | 341.71 | 283.96 | 379.09 | 50.11 | .01 | .49 |
| Zn | 56.60 | 61.33 | 72.03 | 65.09 | 53.32 | 7.64 | .68 | .13 |
| Cu | 10.55 | 12.48 | 13.84 | 11.79 | 10.72 | 1.18 | .65 | .11 |
| Mn | 1.33 | 1.42 | 1.49 | 1.03 | 1.21 | 0.16 | .13 | .95 |
| **D 38** | | | | | | | | |
| BW, kg | 9.73 | 14.94 | 14.30 | 13.84 | 14.51 | 1.16 | .08 | .09 |
| Heart WT, g | 72.47 | 86.23 | 77.67 | 73.43 | 81.33 | 7.25 | .86 | .97 |
| Heart WT, % BW | 0.75 | 0.58 | 0.54 | 0.53 | 0.56 | 0.03 | .01 | .004 |
| Fe | 222.72 | 281.58 | 247.58 | 243.68 | 259.03 | 28.29 | .75 | .76 |
| Zn | 59.29 | 60.09 | 55.09 | 47.95 | 55.48 | 2.99 | .08 | .10 |
| Cu | 11.95 | 13.83 | 13.97 | 11.83 | 13.47 | 0.68 | .82 | .67 |
| Mn | 1.29 | 1.35 | 1.21 | 1.08 | 1.23 | 0.11 | .34 | .39 |
| **D 52** | | | | | | | | |
| BW, kg | 21.19 | 22.27 | 24.49 | 22.53 | 22.61 | 2.38 | .80 | .52 |
| Heart WT, g | 111.40 | 108.47 | 121.20 | 118.00 | 115.23 | 12.89 | .74 | .67 |
| Heart WT, % BW | 0.55 | 0.49 | 0.49 | 0.53 | 0.51 | 0.05 | .88 | .64 |
| Fe | 294.68 | 357.50 | 345.50 | 380.64 | 320.40 | 33.82 | .65 | .12 |
| Zn | 53.81 | 47.37 | 46.79 | 52.34 | 49.40 | 3.44 | .88 | .51 |
| Cu | 16.16 | 14.65 | 14.08 | 15.87 | 14.87 | 1.46 | .87 | .69 |
| Mn | 1.58 | 1.68 | 1.35 | 1.59 | 1.43 | 0.10 | .29 | .83 |

* Mineral content is reported as mg/kg of tissue as measured on a dry matter basis.

† Pigs were administered 1 of 5 iron dosages on day 0. All pigs were weaned on day 22. A total of 3 pigs/treatment were used for tissue analysis at day 22, 38, and 52.

BW = body weight; WT = weight; L = linear; Q = quadratic.

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