Supplemental iron dextran injections: Influence on hemoglobin concentrations and piglet growth

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Summary

This case study assessed effectiveness of protocols for iron dextran administration on hemoglobin (Hb) concentrations in pigs and evaluated the effect of supplemental iron dextran injections prior to weaning on subsequent body weights (BW). Whole blood samples and body weights were collected from piglets at 14 days of age in four farms and at 27 days of age in the fifth farm. For Farms 1 to 3, six piglets per litter were matched by BW to provide a pair of heavy weight (HW) piglets, medium weight (MW) piglets, and light weight (LW) piglets in each litter. For Farms 4 and 5, MW piglets were not included. One piglet from each pair was injected intramuscularly with 200 mg iron dextran immediately after blood collection (treatment pigs). The other piglet in each pair served as the control. At 3 weeks after weaning, pigs were weighed and whole blood samples were collected to determine Hb concentrations. At 14 days of age and after weaning, the results were inconsistent among the farms. Prior to recommending supplemental iron injections for pigs, one must evaluate the existing on-farm protocol for iron administration.

Keywords: swine, pigs, iron dextran, hemoglobin, body weights

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Resumen - Inyecciones complementarias de hierro dextrano: Influencia en las concentraciones de hemoglobina y crecimiento del lechón

Este caso valoró la eficacia de los protocolos de administración de hierro dextrano en las concentraciones de hemoglobina (Hb por sus siglas en inglés) en cerdos y evaluó el efecto de las inyecciones complementarias de hierro dextrano antes del destete en subsecuentes pesos corporales (BW por sus siglas en inglés). Se recolectaron muestras completas de sangre y pesos corporales de lechones a los 14 días de edad en cuatro granjas y a los 27 días de edad en una quinta granja. En las granjas 1 a 3, se organizaron seis lechones por camada por BW para tener un par de lechones de peso pesado (HW por sus siglas en inglés), lechones de peso medio (MW peso medio por sus siglas en inglés), y lechones de peso ligero (LW por sus siglas en inglés) en cada camada. De las granjas 4 y 5, no se incluyeron lechones de MW. Se inyecto intramuscularmente (IM por sus siglas en inglés) un lechón de cada par con 200 mg de hierro dextrano inmediatamente después de la recolección de sangre (cerdos de tratamiento). El otro lechón en cada par sirvió como control. A las 3 semanas después del destete, se pesaron los cerdos y se recolectaron muestras completas de sangre para determinar las concentraciones de Hb. A los 14 días de edad y después del destete, los resultados fueron inconsistentes entre las granjas. Antes de recomendar inyecciones complementarias de hierro para los cerdos, se debe evaluar el protocolo existente en cada granja para la administración de hierro.

Résumé - Injections de supplément de fer dextran: Influence sur les concentrations d'hémoglobine et la croissance des porcelets

Cette étude visait à juger l'efficacité des protocoles d'administration de fer dextran sur les concentrations d'hémoglobine (Hb) chez les porcs et à évaluer l'effet d'injections de supplément de fer dextran avant le sevrage sur les poids corporels (PC) subséquents. Des échantillons de sang complet ont été prélevés et les poids corporels notés chez des porcelets âgés de 14 jours dans quatre fermes et à 27 jours d'âge sur une cinquième ferme. Pour les fermes 1 à 3, six porcelets par portée ont été jumelés par PC afin de fournir une paire de porcelets de poids lourds (PLo), de porcelets de poids moyen (PM), et des porcelets de poids léger (PLe) pour chaque portée. Pour les fermes 4 et 5, des porcelets de PM n'ont pas été inclus. Un porcelet de chaque paire fut injecté par voie intramusculaire (IM) avec 200 mg de fer dextran immédiatement après le prélèvement de sang (porcs traités). L'autre porcelet de chaque paire servait de témoin. Trois semaines après le sevrage, les porcs étaient pesés et des échantillons de sang complet prélevés afin de déterminer les concentrations de Hb. Quatorze jours après le sevrage, les résultats étaient inconstants parmi les fermes. Avant de recommander des injections de supplément de fer pour des porcs, une évaluation doit être faite du protocole d'administration de fer actuellement en vigueur sur la ferme.

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Hemoglobin (Hb) contains iron, which is essential for the transfer of oxygen to tissues. Piglets are born with limited iron stores[1] and sow's milk fails to provide sufficient iron to meet the

demands of rapidly growing piglets. Thus, supplemental iron typically is given to piglets within the first 5 days after birth. This iron injection is intended to prevent iron deficiency anemia.

The timing, dosage, and number of injections of iron dextran are highly variable in the pork industry. Some farms use a single injection of 200 mg of iron dextran, while other farms use 100 or 150 mg of iron dextran. Others give 150 mg on the first day after birth and a subsequent 150 mg when pigs are 5 to 7 days of age. Thus, there is little consistency in the administration of iron dextran to the piglets. The rationales for the various iron injection schemes often are based on convenience of administration and cost, with limited consideration for the distinct possibility of anemia. Few veterinarians evaluate Hb concentration in piglets as a routine diagnostic test. Recently, it was shown that Hb status was associated with post-weaning weight gains[2] and that large piglets in a litter are at greater risk of iron deficiency anemia than are smaller piglets.[3] Hence, large piglets may require greater iron dextran supplementation. Moreover, in one investigation,[4] when an additional 100 mg of iron was administered at 10 days of age, Hb concentrations were higher through 14 days post weaning. Therefore, the influence of some protocols for iron dextran administration on Hb concentrations in pigs in commercial farms were evaluated in the cases in this study.

Materials and methods

Farms

All animals were raised and managed on commercial farms in North Carolina. Each farm was Pork Quality Assurance Plus certified and followed the animal care standards of the National Pork Board.[5] An Institutional Animal Care and Use Committee protocol was not required.

The case series involved five commercial sow farms (2000 to 3600 sows per farm) and their respective off-site nursery facilities (Table 1). Two farms (Farm 1 and Farm 2) injected piglets with 200 mg iron dextran at processing (3 to 5 days of age). Farm 3 used 150 mg iron dextran at the time of processing. Farm 4 and Farm 5 used 150 mg iron dextran when pigs were 1 day of age, and then injected a second time with the same dose at processing (approximately at 5 to 7 days of age). The five farms used Uniferon (Pharmacosmos Inc, Watchung, New Jersey) for the iron dextran injections, which were administered intramuscularly (IM). Weaning age was approximately 21 days and 28 days on farms 1 to 3 and farms 4 and 5, respectively.

Iron injection treatments and sampling protocols

For farms 1, 2, and 3, blood samples (5 mL) were collected from the jugular vein or anterior vena cava into EDTA tubes (Becton, Dickinson and Company, Franklin Lakes, New Jersey) from 120, 84, and 72 piglets, respectively, at 14 days of age. All piglets in each litter were weighed, and six piglets per litter were sampled and matched by body weight to provide a pair of heavy weight (HW) piglets, medium weight (MW) piglets, and light weight (LW) piglets in each litter. The first piglet of a pair that approached the investigators received 200 mg iron dextran (Uniferon) IM (treatment pigs; TMT pigs) immediately after the blood collection. The remaining pigs (control pigs; CON pigs), paired by weight with TMT pigs, were not injected with iron. At approximately 3 weeks after weaning, all pigs were weighed and whole blood samples were collected in EDTA tubes.

On the basis of the preliminary results for the first three farms and requests from the farm management to minimize the number of pigs in the cases, the MW piglets were not included in the trials on farms 4 and 5. For Farm 5, the initial blood collection, supplemental iron injection (200 mg), and initial body weight determinations were delayed until the pigs were 27 days of age. Pigs were weaned at 28 days of age on Farm 5 and thus a supplemental iron injection was convenient on the day prior to weaning.

All blood samples were stored in coolers with ice and transported to the research laboratory at the College of Veterinary Medicine, North Carolina State University. The samples were analyzed for Hb concentrations with a HemoCue Hb 201+ instrument (HemoCue America, Brea, California) within 6 hours after collection. The HemoCue Hb 201+ was previously validated as a reliable device to assess Hb in arterial blood of pigs[6] and venous blood of other mammalian species.[7,8] In brief, the samples were allowed to return to room temperature and then

Table 1: Summary of farms and experimental protocols for injection of pigs with iron dextran in five farms

Farm	No. animals	Iron injection protocol	Age at weaning (days)	Age at first blood sample (days)	Age at second blood sample (days)
1	120	200 mg at processing	21	14	44
2	84	200 mg at processing	21	14	44
3	72	150 mg at processing	21	14	44
4	80	150 mg at 1 day of age, 150 mg at processing	28	14	44
5	80	150 mg at 1 day of age, 150 mg at processing	28	27	52

* Whole blood samples and body weights were collected from pigs at 14 days of age in farms 1-4 and at 27 days of age in Farm 5. Additional blood samples and body weights were collected when pigs were 44 or 52 days of age. For farms 1 to 3, six pigs per litter were matched by body weight to provide a pair of heavy weight (HW) pigs, medium weight (MW) pigs, and light weight (LW) pigs in each litter. For farms 4 and 5, MW pigs were not included. One pig from each pair was injected intramuscularly with 200 mg iron dextran immediately after blood collection (treatment pigs) at 14 or 27 days of age. The other pig in each pair served as the control. Piglets were typically processed at 3-5 days of age on farms 1-3 and at 5-7 days of age on farms 4 and 5.

gently rolled to thoroughly mix them. Following the manufacturer's instructions, a drop of blood was placed on a plastic film using a pipette, and the microcuvette was filled with care to avoid air bubbles. Then the microcuvette was placed in the microcuvette holder of the instrument and Hb concentration determined.

Statistical analyses

For the initial analysis, the independent variables included farm, weight class, sex, and treatment. All variables were categorical. To examine the effect of supplemental iron injection on Hb concentrations and body weights, two separate multiple linear regression models initially were used (R Core Team R, Vienna, Austria). To test for multicollinearity, a variance inflation factor was calculated. When all main effects and second-order interaction terms were included, the variables in the model were highly correlated and the model suffered from multicollinearity. Since there were significant ($P < .05$) interactions among farm, treatment, and weight class, the simple effects of treatment were examined for certain combinations of all other factors.[9] Simple effects compare means when there is a statistically significant interaction and the average of the simple effects is the main effect. Because of the interactions, the means across the farms, sexes, and weight classes could not be compared in the statistical analyses; rather, the simple effects of treatment for certain combinations of the factors (sex and weight class within each farm) were examined.

Results

A summary of Hb status and pigs' weights is given in Table 2. It was evident that the

Table 2: Mean body weights (kg) and hemoglobin concentrations (g/L) in pigs from five sow farms*

				Body weight (kg)				Hemoglobin concentrations (g/L)			
Farm	Treatment group	Weight class	N	Day 14 (27)	SE	Day 44 (52)	SE	Day 14 (27)	SE	Day 44 (52)	SE
1	Iron	Light	20	3.8	0.2	9.9	0.5	94.4	2.2	97.2	3.5
		Medium	20	4.6	0.2	12.1	0.5	89.8	2.1	94.0	3.0
		Heavy	20	5.5	0.2	13.8	0.4	88.6	2.2	99.2	3.8
	Control	Light	20	3.7	0.2	9.9	0.4	96.3	1.8	91.6	2.7
		Medium	20	4.6	0.2	11.7	0.4	92.5	1.8	90.4	3.3
		Heavy	20	5.2	0.2	12.3	0.4	88.9	1.8	94.3	3.3
2	Iron	Light	14	3.8	0.2	10.0	0.5	98.4	3.0	101.9	3.8
		Medium	14	4.5	0.2	11.3	0.6	88.5	4.4	111.9	2.9
		Heavy	14	5.1	0.1	12.1	0.3	91.3	3.3	98.5	4.5
	Control	Light	14	3.8	0.2	10.0	0.6	95.3	2.9	102.1	3.7
		Medium	14	4.6	0.2	10.7	0.5	91.7	3.2	100.6	3.0
		Heavy	14	5.2	0.1	11.9	0.7	87.1	2.4	101.8	4.9
3	Iron	Light	12	4.1	0.1	12.6	0.5	84.8	9.8	113.6	4.9
		Medium	12	4.6	0.1	13.7	0.7	89.3	8.7	115.0	4.1
		Heavy	12	5.2	0.1	15.4	0.7	91.3	8.9	118.1	2.4
	Control	Light	12	4.1	0.2	11.2	0.4	95.3	7.7	114.0	5.0
		Medium	12	4.6	0.1	13.8	0.5	97.4	8.4	105.9	4.0
		Heavy	12	5.1	0.1	14.2	0.7	92.1	8.0	107.3	4.4
4	Iron	Light	20	3.7	0.1	13.6	0.5	114.9	3.0	113.6	3.2
		Heavy	20	5.1	0.2	16.2	0.6	115.4	2.5	115.8	2.3
	Control	Light	20	3.7	0.1	13.2	0.8	118.9	2.1	111.5	3.2
		Heavy	20	5.3	0.2	16.1	0.7	119.1	1.9	110.7	2.9
5	Iron	Light	20	6.8	0.2	14.4	0.5	118.9	3.5	107.4	3.7
		Heavy	20	8.7	0.2	16.9	0.5	111.5	3.4	109.7	3.4
	Control	Light	20	6.7	0.2	14.4	0.6	122.5	2.9	106.3	2.1
		Heavy	20	8.7	0.2	17.0	0.5	114.8	3.2	110.3	2.3

* Study described in Table 1. Pigs were assigned at 14 days of age to either the control or iron injection groups. Pigs were matched by body weight (light, medium, heavy) between the control and treatment groups. For farms 4 and 5, the medium weight pigs were not included. For Farm 5, the blood collection days were days 27 and 52. Day numbers represent days when pigs were weighed or had blood samples collected.

SE = standard error.

assignment of pigs to their respective weight classes was consistent for each farm. At 14 days of age, Hb concentrations were numerically lower in the HW pigs than in the LW pigs in farms 1 and 2, but not in the other three farms.

As shown in Table 3, the supplemental iron injection in Farm 1 pigs was associated with greater body weights in most pigs, with the exception of the female MW pigs. There were also higher Hb concentrations in the male pigs, but not necessarily the female pigs. The consistent increase in body weights observed in Farm 1 was not evident in the other farms. In Farm 2, the male MW pigs had greater body weights and Hb concentrations in the TMT pigs than in the CON pigs. These differences were not apparent in the different classes of pigs. Either body weight or Hb was greater following treatment, but not both.

Four of the weight classes (MW male, HW male, LW female, and HW female) had greater weight gains after the iron supplementation than in the CON pigs in Farm 3. In addition, at 44 days of age, four of the weight classes responded to the iron injection with Hb concentrations higher than those of the CON pigs. As evident in Farm 1, greater Hb concentration did not necessarily occur concomitantly with a greater weight gain.

Iron supplementation was associated with greater body weights at 3 weeks after weaning in the male LW pigs in Farm 4, but not in the female pigs. In fact, the treated female LW pigs gained 1.85 kg less than their pair-matched CON pigs. Although the iron injection resulted in 16.2 g per L more Hb in the female HW pigs than in the CON female HW pigs, there was no corresponding increase in body weight. A similar result was apparent in the male HW pigs in Farm 5. In Farm 5, only the female LW pigs benefited with greater body weights from the supplemental iron injection at 27 days of age.

Discussion

The assignments of pigs to weight class (LW, MW, HW) were successful on all farms. Thus, there was an appropriate distribution of pig weights at the onset of the cases. It is important to note that farms 1 to 3 weaned pigs at 21 days of age, while farms 4 and 5 weaned pigs at 28 days of age. Consequently, it was anticipated that body weights would be different among the farms at 3 weeks after weaning.

The HemoCue Hb 201+ provides a convenient method to assess Hb concentrations for on-farm use or within a veterinary clinic. However, this single assessment of Hb concentrations must be viewed with caution. Despite the use of Hb concentration as an indicator of iron status and anemia, other blood parameters, such as serum iron and total iron binding capacity, may be more sensitive in detecting iron deficiency.[3] In addition, this instrument was shown to underestimate the Hb concentrations in pigs at 3 or 25 days of age.[10]

In the current study, Hb concentrations were generally lower at 14 days of age in the HW pigs than in the LW pigs. This should not be surprising, as the HW pigs represent the fastest growing pigs with the greatest demand for iron.[3,11] This inverse relationship, ie, HW pigs with low Hb and LW pigs with high Hb, creates a perplexing issue when one tries to interpret the influence of supplemental iron on subsequent pig growth. Weaning weight is one of the most important factors in pig growth after weaning.[12,13] Consequently, one needs to use caution when interpreting the influence of supplemental iron on subsequent growth. Pre-planned

Table 3: Simple effects for each farm, sex, and weight class*

Farm	Sex	Weight class	N	Body weight (kg)	Hb concentrations
1	Male	Light	15	0.64	7.1
		Medium	20	0.79	13.3
		Heavy	22	0.85	4.4
	Female	Light	25	0.87	-19.1
		Medium	20	0.05	-4.2
		Heavy	18	1.50	5.6
2	Male	Light	14	-0.14	0.3
		Medium	18	2.36	14.0
		Heavy	16	0.65	-7.7
	Female	Light	14	0.22	-13.9
		Medium	10	-0.25	14.8
		Heavy	12	0.14	-7.4
3	Male	Light	13	0.24	-11.4
		Medium	13	1.59	23.6
		Heavy	12	1.06	-1.0
	Female	Light	11	2.58	37.1
		Medium	11	-1.61	9.0
		Heavy	12	1.24	15.2
4	Male	Light	19	1.75	2.1
		Heavy	17	0.14	1.6
	Female	Light	21	-1.85	8.0
		Heavy	23	-0.05	16.2
5	Male	Light	21	-0.75	5.5
		Heavy	31	-0.11	20.2
	Female	Light	19	0.73	3.5
		Heavy	9	-0.08	-1.9

* Study described in Table 1. The simple effects compare the difference due to treatment with specific combinations of the factors (sex and weight class within each farm). It can be concluded, for example, for the Farm 1 male-light situation, the average weight gain (over the 30 days) of TMT pigs was 0.64 kg more than that of the CON pigs, whereas the Hb concentrations were 7.1 g/L greater in TMT pigs than in CON pigs.

TMT = treatment ; CON = control; Hb = hemoglobin.

differences in body weight among the two or three classes at the onset of the study continued into the nursery phase of production. The pigs stayed in the same weight categories throughout the study. The most notable weight gains were in the female HW pigs (Farm 1), male MW pigs (Farm 2), and female LW pigs (Farm 3) treated with supplemental iron. Evidently, greater weights with supplemental iron injections were not consistent among the three farms that weaned piglets at 21 days of age. Furthermore, it would have been beneficial to determine the long-term influence of the iron injections on weight gains into the finishing phase of production. Among the farms, the duration of time in the nursery facilities was variable and it would have been difficult to follow all pigs through to finishing.

In Farm 3, the use of a single injection of 150 mg iron dextran at processing resulted in low Hb concentrations in pigs at 14 days of age. This observation is consistent with the results of recent studies.[2,3,12] In contrast, in Farm 4, the two separate injections of iron resulted in Hb concentrations comparable to previously reported values[3,12] in pigs. Interestingly, the Hb concentrations were similar between the two farms by 44 days of age. Thus, it can be inferred that the additional iron injection in Farm 3 had a beneficial influence on Hb concentrations in MW male pigs and all female pigs.

Farm 5 weaned pigs at 28 days and the supplemental iron injections were given at 27 days of age. Despite the apparent tendency in Hb concentrations at 27 days of age, ie, lower Hb in the HW pigs than in LW pigs, the only notable increase in Hb concentrations was observed in the male HW pigs at 52 days of age. The reason for the influence of sex is speculative, particularly in view of the fact that this observation was not consistent among the farms and weight classes.

Overall, it is evident that the supplemental iron injection failed to consistently increase Hb concentrations and body weights among the five farms. Unfortunately, the composition of the nursery diets was unavailable; however, none of the farms used high concentrations of zinc oxide (ZnO) in the feed. High concentrations of ZnO (> 2000 mg per kg) recently were noted as a potential cause of anemia in pigs after weaning.[3] Hemoglobin concentrations are useful markers for iron status; however, other biomarkers of iron metabolism likely would shed additional light on the usefulness of supplemental iron injections.

Farms 1 and 2 used similar protocols for iron injections, while the protocols for other three farms differed. Farms 4 and 5 used the same routine injections at processing and 5 days later; however, the timing of the supplemental injection differed. In view of these inconsistencies, it should not be surprising that the pigs in each farm responded somewhat differently. Before making a broad, general recommendation on supplemental iron injections to improve post-weaning weight gains or Hb status of pigs, one must evaluate the on-farm protocol for iron injections of piglets. In the on-farm conditions of the present series of cases, it was apparent that increased Hb concentrations are not necessarily associated with greater body weights at 3 weeks after weaning.

Implications

- A two-dose scheme of iron dextran injections in the first week of life appears to meet the iron requirements of piglets.
- Under the conditions of these cases, there was a lack of consistency in dose and number of injections of iron dextran. The influence of pre-weaning supplemental iron injections and (or) Hb concentrations is confounded by the differences in body weights and sex at weaning.
- Practitioners need to evaluate existing protocols, weight gains, and Hb concentrations for each farm prior to recommending a supplemental iron injection prior to weaning.

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Conflict of interest

None reported.

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