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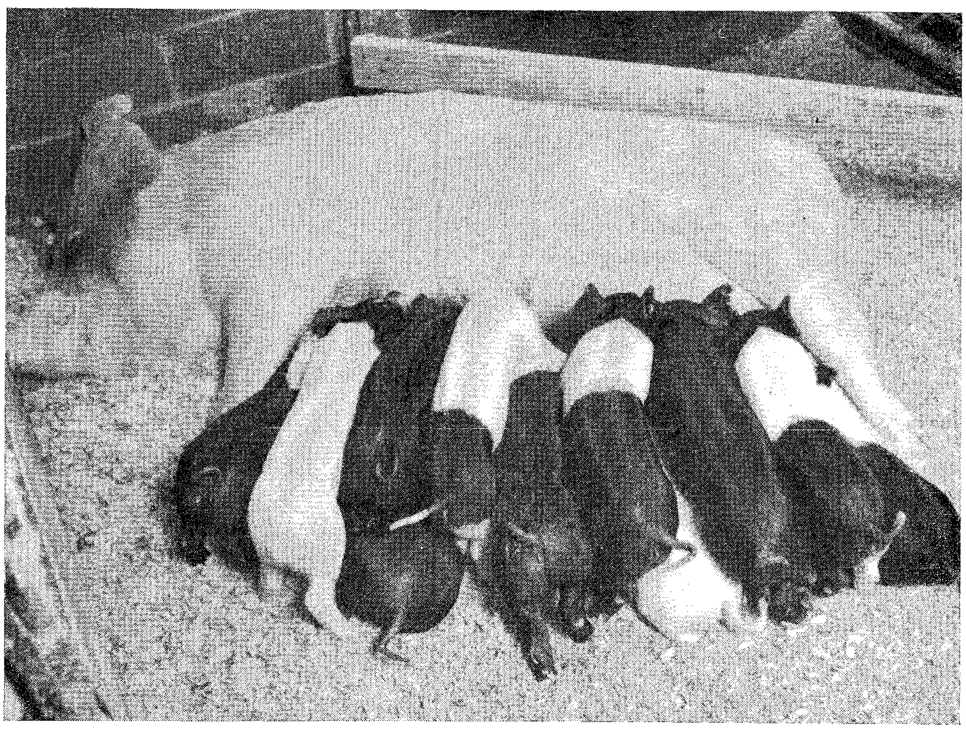

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Injectable Iron as a Preventative for Nutritional Anemia in the Young Pig

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MORE THAN FOUR DECADES have passed since anemia was associated with iron deficiency in the young pig (26).[1] Shortly thereafter, it was demonstrated that feeding iron salts to the sow did not delay the onset of anemia in baby pigs (9, 14, 15, and 16). A greater incidence of anemia was reported in pigs reared indoors than in those raised out of doors (9). An outside factor was associated with the prevention of anemia in young pigs reared out of doors and on concrete (9 and 14). However, it was soon demonstrated that pigs reared out of doors and on concrete developed anemia as rapidly as those reared indoors (13 and 35).

Daily doses of 25 mg. iron sulfate and 5 mg. copper sulfate prevented anemia from birth to weaning in young pigs (15 and 16), as did larger doses administered weekly (17). Swabbing the sow's udder with solutions containing iron and copper salts also prevented anemia in young pigs (14, 17, and 35). The role of soil in providing iron to protect pigs against anemia was also demonstrated (7, 8, 12, and 19).

From the early 1930's to the 1950's, swine producers and others concerned with nutritional anemia in young pigs sought a method that would greatly reduce the work required to prevent anemia by daily swabbing of the sow's udder with a saturated ferrous sulfate (copperas) solution, daily or weekly administration of iron salts or tablets to individual pigs, or provision of fresh uncontaminated sod. Following the discovery that the

[1] Numbers in parentheses refer to citations given on pages 23-24.

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iron-dextran compound Imferon was satisfactory for intramuscular treatment of iron-deficiency anemia in man (5 and 30), investigators tested this product in young pigs. In 1955, several researchers (2, 4, and 25) reported that iron-dextran effectively prevented iron deficiency anemia in young pigs. Kernkamp (20), working at the University of Minnesota, made one of the initial reports in the United States on the effectiveness of Imferon in maintaining high hemoglobin levels in pigs reared indoors to 3 weeks of age.

Subsequent studies demonstrated that iron-dextran was an effective hematinic for prevention and treatment of iron-deficiency anemia in the very young pig. The practice of using injectable iron compounds to combat nutritional anemia in young pigs is now widely accepted. However, compounds other than iron-dextran are being used; their efficacies may not be well established. In addition, the amounts of iron reported necessary to prevent anemia in pigs from 3-4 to 21-35 days of age vary among investigators. Differences in experimental conditions or in response criteria may be partially responsible for these variations.

As injectable iron compounds were developed, there was some tendency to include vitamin B_{12}, folic acid, and other B vitamins in the preparations. The amounts of vitamin B_{12} and folic acid required in such materials are not established; their real value in making such preparations more effective hematinics is not confirmed. Presumably, these vitamins were included because of their reported roles in hematapoiesis.

Objectives of this study were to:

1. Determine the optimum dosage of injectable iron for preventing iron-deficiency anemia in suckling pigs, as well as the feasibility of injecting the total dosage into the 3-4-day-old pig.

2. Compare the effectiveness of treating the 3-4-day-old pig versus the 7-day-old pig.

3. Obtain information on the influence of the chemical nature of the iron preparation on hemoglobin values and growth responses in suckling pigs.

4. Determine the value of simultaneously injecting vitamin B_{12}, folic acid, or a combination of the two with iron-dextran in increasing hemoglobin values and growth in the young pig.

MATERIALS AND METHODS

In this study, 710 pigs from 71 litters were used in a series of five investigations. All sows farrowed in farrowing stalls in a concrete-floored barn. They remained in the stalls for from 24 to 48 hours after farrowing. Sows and litters then were transferred from stalls to nursing pens with access to outside concrete runways. Initially, the pigs had access to the sow's ration. When they were 7 days old, a palatable high-energy pig starter formulated to be adequate in known required nutrients and containing an antibiotic was provided. Fresh water was provided at all times. Pens were bedded with wood shavings.

Pigs used in experiments I, II, III, and IV were weaned at 5 weeks of age. Pigs used in experiment V were weaned at 3 weeks. All pigs were weighed individually at birth and at weekly intervals until weaning. Blood samples for hemoglobin determinations were withdrawn from the anterior vena cava at each weighing. In experiment III, initial blood samples were obtained just prior to iron injection when pigs were 3-4 or 7 days old. Blood withdrawal and injection techniques were not aseptic. Intramuscular injections were made into the ham. Subcutaneous injections were made on the medial side of the front or rear flank. Hemoglobin determinations were made by the acid hematin method of Newcomer (27) in experiments I, II, and III and by the direct photoelectric method of Evelyn (11) in experiments IV and V.

All pigs were randomly assigned from within litters to treatments, except that entire litters were used when a copperas treatment was included. Summaries of data include only pigs completing the experiment. Duncan's (10) multiple range test was used to test for significant differences between means in experiments I, II, III, IV (trial 2), and V following statistical treatment of data by analysis of variance (32). In experiment IV (trial 1) and the factorial portion of experiment III (treatments 1 through 4b), treatment effects were subdivided into appropriate sources of variation.

The error term for testing significance of mean squares was obtained by dividing the within-treatment mean square resulting from analysis of variance on individual observations by the harmonic mean of the number of pigs per treatment. Regression coefficients, calculated on a within-experiment basis, were used to adjust 35-day weights for differences in initial weights.

EXPERIMENT I

In this experiment, 217 pigs from 23 litters of Yorkshire, Berkshire, and Berkshire x Yorkshire breeding were used. Pigs from 21 litters received an intramuscular injection of 100 mg. iron from iron-dextran[2] when 3-4 days old. They received subsequent treatments by intramuscular injection at 21 days as follows:

[2] The source of iron-dextran used in this study was always Armidexan, Armour Laboratories, Kankakee, Ill.

Treatment 1. No additional iron.

Treatment 2. 50 mg. iron.

Treatment 3. 100 mg. iron.

Pigs from the remaining two litters were administered iron by daily swabbing of the sow's udder with a saturated copperas solution from the time pigs were 3 days old until they were 35 days old. This copperas solution was prepared by dissolving 1 pound crude ferrous sulfate in 3 quarts water. The copperas treatment was included to compare the response of pigs treated with an injectable iron to that of pigs treated by an anemia preventative of established reliability.

EXPERIMENT II

In this study, 73 pigs from six litters of Hampshire and Hampshire x Yorkshire breeding were used. Pigs from four litters were given intramuscular injections of iron-dextran to provide the following amounts of iron:

Treatment 1. 100 mg. iron at 3-4 days.

Treatment 2. 150 mg. iron at 3-4 days.

Treatment 3. 200 mg. iron at 3-4 days.

Treatment 4. 100 mg. iron at 3-4 days; 100 mg. iron at 21 days.

As in experiment I, two complete litters were administered iron by daily swabbing of the sow's udder with a saturated copperas solution from the time pigs were 3 days until they were 35 days old.

EXPERIMENT III

Fourteen litters consisting of 132 pigs of Yorkshire, Berkshire, Berkshire x Yorkshire, and Hampshire x Yorkshire breeding were used in this experiment. Pigs on treatments 1a through 4b were administered iron-dextran intramuscularly as indicated below. In addition, pigs in groups designated b were simultaneously administered 30 mcg. injectable vitamin B_{12}.[3]

Treatments 1a and 1b. 100 mg. iron at 3-4 days.

Treatments 2a and 2b. 150 mg. iron at 3-4 days.

Treatments 3a and 3b. 100 mg. iron at 7 days.

Treatments 4a and 4b. 150 mg. iron at 7 days.

A ferric ammonium citrate compound[4] providing 80 mcg. vitamin B_{12} per 100 mg. iron was injected subcutaneously to provide pigs with the following amounts of iron:

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[3] The source of vitamin B_{12} used in this study was always Redisol, Merck Sharp and Dohme, Research Laboratories, Rahway, N.J.

[4] The source of ferric ammonium citrate used in this study was always Rubrafer, E. R. Squibb & Sons, New Brunswick, N.J.

Treatment 5. 100 mg. iron at 3-4 days.

Treatment 6. 150 mg. iron at 3-4 days.

Treatment 7. 100 mg. iron at 7 days.

EXPERIMENT IV

Trial 1. Treatments 1a through 4b of experiment III were repeated. However, in groups designated b, 60 mcg. vitamin B_{12} were administered intramuscularly and simultaneously with the iron dosage. Thirteen litters providing 118 pigs of Yorkshire, Hampshire, and Hampshire x Yorkshire breeding were used.

Trial 2. Four litters of Yorkshire and Hampshire pigs were used. All pigs were administered 150 mg. iron intramuscularly from iron-dextran when 3-4 days old. In addition, they were simultaneously injected with vitamins as follows:

Treatment 1. no vitamins.

Treatment 2. 60 mcg. vitamin B_{12}.

Treatment 3. 15 mg. folic acid.[5]

Treatment 4. a combination of treatments 2 and 3.

EXPERIMENT V

Eleven litters providing 126 pigs of Yorkshire and Yorkshire x Hampshire breeding were used. Treatments were as follows:

Treatments 1 and 3. 100 mg. iron from a dextrin-ferric oxide complex at 3-4 days.

Treatments 2 and 4. 100 mg. iron from a dextrin-ferric oxide complex at 7 days.

Treatment 5. 44 mg. iron from peptonized iron[6] at 3-4 days.

Treatment 6. 44 mg. iron from peptonized iron at 7 days.

Treatment 7. 88 mg. iron from peptonized iron at 7 days.

Treatment 8. 100 mg. iron from iron-dextran at 3-4 days.

Treatment 9. 200 mg. iron from iron-dextran at 3-4 days.

Treatment 10. 150 mg. iron from iron-dextran at 3-4 days.

The dextrin-ferric oxide preparations were from two sources, one compound for treatments 1 and 2[7] and another compound for treatments 3 and 4[8]. Dextrin-ferric oxide and iron-dextran were injected intramuscularly; peptonized iron was injected subcutaneously.

[5] The source of folic acid used in this study was always Folvite, American Cyanamid Company, Princeton, N.J.

[6] The source of peptonized iron used in this study was always PA-12, Specifide, Inc., Indianapolis, Ind.

[7] Pigdex, American Cyanamid Company, Princeton, N.J.

[8] Pig Iron, Dr. Salsbury's Laboratories, Charles City, Ia.

RESULTS AND DISCUSSION

Negative controls were not used in this study. It is a well established principle that pigs nursing their dams and being reared on concrete show an inevitable decline in hemoglobin content of the blood during the 3-10-day postnatal period. This decline persists until average hemoglobin values reach very low levels when untreated pigs are 18-25 days of age (see figure 1).

Kernkamp (19) expressed a normal hemoglobin curve for confined untreated pigs as a percent decline from the hemoglobin level at birth. He fixed the initial hemoglobin level at zero; then he established that hemoglobin levels of untreated pigs at 1, 2, 3, 4 and 5, and 6 weeks of age were, respectively, 30, 40, 51, 49, and 43 percent below the initial level. He also suggested that a hemoglobin level 30 percent or more below the initial level constituted anemia.

EXPERIMENT I

Average hemoglobin levels and weights of pigs at weekly intervals throughout the study are presented in table 1. Pigs treated with 100 mg. iron from iron-dextran maintained normal hemoglobin levels until 21 days

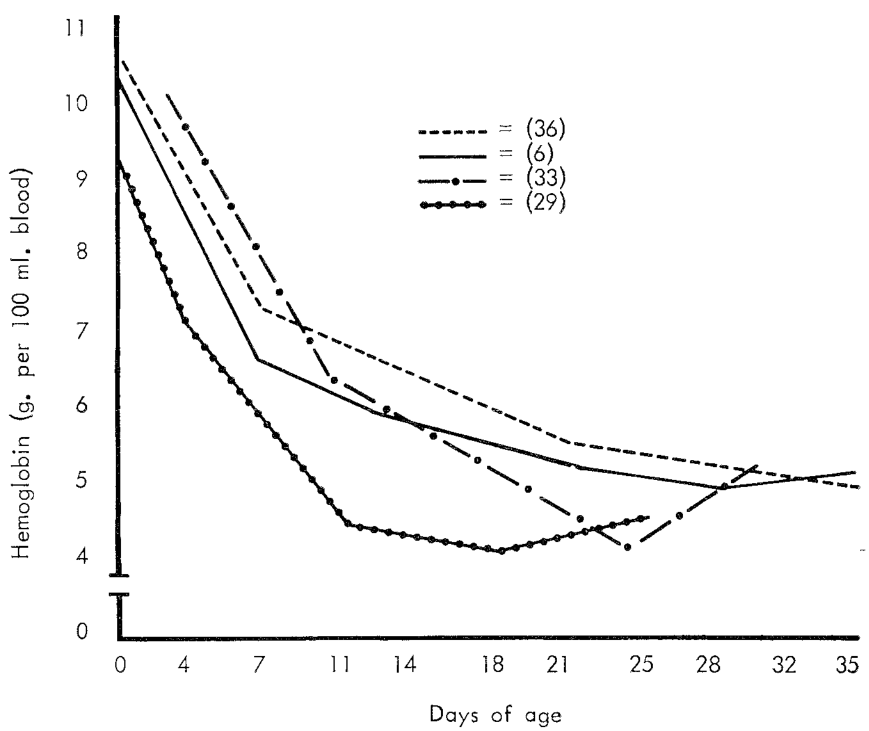

Figure 1. Curves demonstrating the decline in hemoglobin content of blood with age of nursing pigs not given supplemental iron.

Table 1. Effects of supplemental iron-dextran and of daily swabbing of the sow's udder with copperas on hemoglobin content of the blood and weights of nursing pigs—experiment I

Treatment number	1	2	3	
		Iron-dextran		Copperas
Iron, 3-4 days, mg.	100	100	100	
Iron, 21 days, mg.	0	50	100	
Number pigs	56	60	55	21
Birth:				
Average hemoglobin, g./100 ml.	9.8	10.2	9.7	8.7
Average weight, lb.	2.6	2.5	2.4	2.8
7 days:				
Average hemoglobin, g./100 ml.	9.5	9.4	9.2	8.4
Average weight, lb.	5.1	5.1	5.1	4.9
14 days:				
Average hemoglobin, g./100 ml.	10.5	10.4	10.4	10.0
Average weight, lb.	8.5	8.4	8.4	8.5
21 days:				
Average hemoglobin, g./100 ml.	9.7	9.5	9.4	10.1
Average weight, lb.	11.7	11.7	11.5	11.8
28 days:				
Average hemoglobin, g./100 ml.	8.4	9.3	9.9	10.1
Average weight, lb.	15.5	15.5	15.3	13.8
35 days:				
Average hemoglobin, g./100 ml.	7.7[a]	8.9[b]	9.7[c]	9.7[c]
Average weight, lb.	19.0	20.0	20.0	18.3
Average adjusted weight, lb.*	18.8[d]	20.2[a,e]	20.2[a,e]	17.8[b]

* Adjusted 35-day weights (final weights of pigs adjusted ± 0.27 pound for each deviation of ± 0.1 pound from mean birth weight for all pigs, 2.56 pounds).

a,b,c Means within line and having different superscript letters differ significantly ($P < 0.01$).

d,e Means within line and having different superscript letters differ significantly ($P < 0.05$).

old. However, when no supplemental iron was administered at 21 days, hemoglobin levels declined to 7.7 g. per 100 ml. blood at 35 days of age. Approximately one-fourth of the pigs on treatment 1 showed less than 7 g. hemoglobin per 100 ml. blood at 35 days. They also exhibited moderate physical symptoms of anemia—roughened hair coats, listlessness, and mild diarrhea.

Pigs treated with a total of 150 mg. iron had significantly ($P<0.01$) higher hemoglobin levels at 35 days of age than pigs given only 100 mg. Furthermore, they did not show physical symptoms of anemia. However, they did not exhibit maximum hemoglobin values because pigs administered a total of 200 mg. iron or given iron through copperas showed significantly ($P<0.01$) greater hemoglobin levels at 35 days of age. Effects of the separate treatments on the hemoglobin levels are presented in figure 2.

Average adjusted 35-day weights of pigs administered 50 or 100 mg. additional iron at 21 days were significantly ($P<0.05$) greater (1.4 pounds)

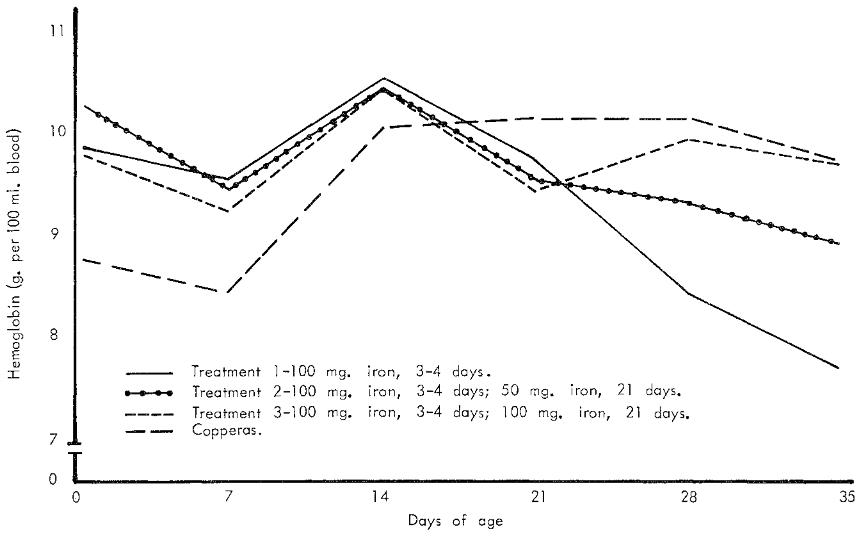

Figure 2. The influence of iron treatments on the average hemoglobin content of blood of suckling pigs—experiment I.

than those of pigs treated with only 100 mg. iron. Although a weight difference of 1.4 pounds may not be of great economic importance, it indicates that growth might have been slightly limited by an iron deficiency. Pigs nursing dams whose udders were swabbed daily with copperas solution weighed significantly ($P<0.01$) less at 35 days than those treated with either 150 or 200 mg. total iron from iron-dextran. The difference was probably the result of the two litters given copperas being afflicted with severe diarrhea—not due to anemia—when 21-28 days old.

Overall mortality was 11 percent. Differences in mortality cannot be attributed to anemia or to treatment. All deaths occurred before pigs were 21 days old. They resulted from such causes as crushing by the sow or physical injuries which necessitated destruction of the pigs.

EXPERIMENT II

Average weights and hemoglobin values are presented in table 2. Statistical analysis showed that pigs administered 150 or 200 mg. iron from iron-dextran at 3-4 days of age had significantly ($P<0.01$) greater hemoglobin levels at 21 days than pigs treated with only 100 mg. Pigs administered 150 or 200 mg. iron also showed significantly ($P<0.01$) greater concentrations of hemoglobin at 35 days than pigs given only 100 mg. iron. Furthermore, pigs treated with 200 mg. iron had significantly ($P<0.01$) higher hemoglobin levels than pigs treated with 150 mg.

One-third of the pigs given the smallest amount of iron showed from 5.8-6.8 g. hemoglobin per 100 ml. blood. This level, according to Kernkamp (19), indicated anemia. Therefore, administration of 100 mg. iron from iron-dextran did not maintain satisfactory hemoglobin levels in pigs nursing their dams and being reared on concrete to 35 days of age.

Sheffy (31) demonstrated that anemic pigs had a low resistance to disease. When anemic 5-7-week-old pigs were injected with modified hog cholera virus, they became stunted and died. On the other hand, pigs with normal hemoglobin levels became immune and continued to grow. Anders and Schipper (1) reported that varying degrees of vaccination-induced shock followed administration of hog cholera antiserum to anemic suckling pigs. The necessity of administering sufficient iron to assure that all pigs have satisfactory hemoglobin levels at 5-6 weeks of age cannot be overlooked.

Adjusted 35-day weights were unaffected by the amount of iron or time of administration. This finding is in contrast with results of experiment I in which pigs administered 150 or 200 mg. iron from iron-dextran were significantly ($P<0.05$) heavier at 35 days than pigs given 100 mg. iron at 3-4 days. However, Maner et al. (23) were unable to demonstrate

Table 2. Effects of level and time of dosage with iron-dextran and of daily swabbing of the sow's udder with copperas on hemoglobin content of the blood and weights of nursing pigs—experiment II

Treatment number	1	2	3	4	
	Iron-dextran				Copperas
Iron, 3-4 days, mg.	100	150	200	100	
Iron, 21 days, mg.	0	0	0	100	
Number pigs per treatment	10	13	10	10	23
Number deaths per treatment	2	1	2	0	2
Birth:					
Average hemoglobin, g./100 ml.	10.2	9.3	9.8	9.2	9.7
Average weight, lb.	3.1	2.8	2.8	2.8	2.8
7 days:					
Average hemoglobin, g./100 ml.	8.6	8.5	8.4	8.4	8.4
Average weight, lb.	6.3	5.5	5.5	5.2	5.3
14 days:					
Average hemoglobin, g./100 ml.	10.0	10.5	10.8	10.0	10.0
Average weight, lb.	10.3	9.2	9.1	9.0	8.6
21 days:					
Average hemoglobin, g./100 ml.	8.7[a]	10.4[b]	10.5[b]	9.2[a]	11.0[b]
Average weight, lb.	14.0	12.9	12.6	12.8	11.7
28 days:					
Average hemoglobin, g./100 ml.	8.0	9.8	10.9	9.9	11.5
Average weight, lb.	17.8	16.4	16.2	16.4	14.4
35 days:					
Average hemoglobin, g./100 ml.	7.4[a]	9.0[b]	10.2[c]	10.2[c]	11.9[d]
Average weight, lb.	19.9	18.9	19.6	19.6	17.0
Average adjusted weight, lb.*	19.3	18.9	19.6[e]	19.8[e]	17.2[f]

* Adjusted 35-day weights (final weights of pigs adjusted $\pm$ 0.28 pound for each deviation of $\pm$ 0.1 pound from mean birth weight for all pigs, 2.83 pounds).

[a,b,c,d] Means within lines and having different superscript letters differ significantly ($P < 0.01$).

[e,f] Means within lines and having different superscript letters differ significantly ($P < 0.05$).

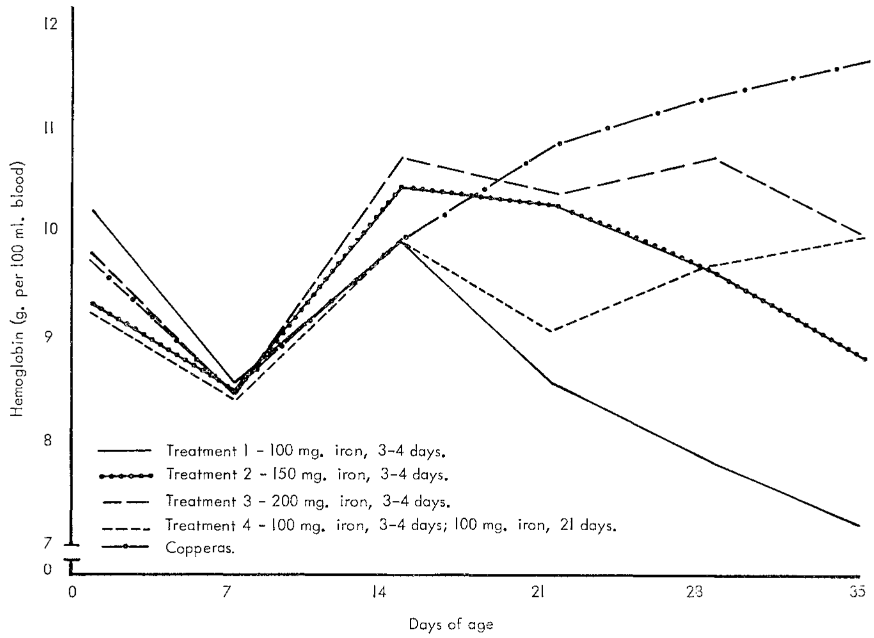

Figure 3. The average hemoglobin values of suckling pigs as affected by iron treatments—experiment II.

significant differences in weights of 24-day-old suckling pigs administered 100 or 200 mg. iron from iron-dextran. Likewise, Wahlstrom and Juhl (34) were unable to demonstrate significant differences in weights of 28-day-old pigs administered 100 or 150 mg. iron from iron-dextran.

Administration of 200 mg. iron from iron-dextran in a single dose to the 3-4-day-old pig or as a divided dose (100 mg. at 3-4 days and 100 mg. at 21 days) failed to result in significant differences in hemoglobin levels or weights at 35 days. There was no evidence that administering the total 150 or 200 mg. iron from iron-dextran to 3-4-day-old pigs resulted in toxicity. Maner et al. (23) did not observe symptoms of toxicity in baby pigs injected intramuscularly with as much as 500 mg. iron from iron-dextran during the 1st and 2nd weeks of life. Figure 3 clearly indicates the effects of inadequate iron on hemoglobin levels of suckling pigs. This figure, like others in this presentation, demonstrates the postnatal decline in hemoglobin levels as an inevitable physiological decrease due to the occurrence of erythropoietic adjustments (18).

Pigs nursing dams whose udders were swabbed with copperas were significantly ($P<0.05$) lighter in weight at 35 days than pigs administered 200 mg. iron from iron-dextran. These pigs did not have severe diarrhea as did pigs receiving this treatment in experiment I. The result may have been due to sampling error arising from the use of only two litters. Possibly, the copperas solution may have been so objectionable that milk consumption was reduced.

EXPERIMENT III

Data in table 3 show the effects of level of injectable iron and time of administration on hemoglobin levels during the nursing period. As in previous experiments, administration of 150 mg. iron from iron-dextran resulted in higher hemoglobin levels at 3, 4, and 5 weeks than administration of 100 mg. iron. Hemoglobin concentrations in blood of pigs injected with 150 mg. iron were not maximum at 35 days. Nevertheless, they were more nearly normal than were those of pigs treated with 100 mg. iron. Moreover, no pig injected with 150 mg. iron showed a sufficiently low hemoglobin level to be classified as anemic by Kernkamp's (19) criteria.

Delaying injection of iron-dextran from 3-4 days until pigs were 7 days old did not result in significantly higher hemoglobin levels when pigs were 3, 4, and 5 weeks old. Pigs treated at 7 days of age showed significantly ($P<0.01$) lower hemoglobin levels at 7 and 14 days than those treated when 3-4 days old.

The average hemoglobin levels of pigs treated when 7 days old were subnormal at 7 days; hemoglobin levels of some pigs indicated anemia. This situation in itself might not have been serious since pigs were administered an injectable iron compound. But, the possibility of reduced resistance to secondary infection due to subnormal hemoglobin levels as a consequence of delaying initial treatment cannot be overlooked. Kernkamp et al. (21) delayed administration of 150 mg. iron from iron-dextran until pigs were 7, 14, or 21 days old. They reported no deleterious effects due to delayed injection. They found prompt regeneration of hemoglobin as well as normal hemoglobin levels, hematocrit values, and weights of pigs at 56 days of age.

Adjusted 35-day weights of pigs treated with iron-dextran were not significantly affected by amount of iron or time of treatment. But, pigs administered 150 mg. iron averaged 0.9 pound heavier at 35 days than pigs given 100 mg. iron. The simultaneous administration of 30 mcg. vitamin B_{12} with the injectable iron also did not affect 35-day weights or hemoglobin values at any weekly interval during the experiment.

Treatment of pigs with 100 mg. iron from ferric ammonium citrate when 3-4 days old resulted in average adjusted 35-day weights that were significantly ($P<0.05$) lower than those of pigs administered 150 mg. iron from iron-dextran. The 35-day hemoglobin levels of pigs treated with 100 or 150 mg. iron from ferric ammonium citrate were significantly ($P<0.01$) lower than those of pigs treated with equivalent amounts of iron from iron-dextran (see figure 4). These findings suggest that the pig cannot fully utilize iron from ferric ammonium citrate for synthesis of hemoglobin, perhaps because of high urinary losses of iron (28).

Pigs treated with ferric ammonium citrate, particularly at the level of only 100 mg. iron, showed anemia symptoms at 4 weeks. At 5 weeks, many of these pigs were decidedly anemic as evidenced by subnormal hemoglobin levels, roughened hair coats, and lack of thrift; some stunting of growth may have occurred. Kernkamp et al. (21) found injectable ferric ammonium citrate to be much less effective in increasing hemoglobin level and growth rate of young pigs than iron-dextran or iron-dextrin.

Table 3. Effects of amount of iron, chemical nature of iron compound, and time of administration on hemoglobin content of the blood and weights of nursing pigs—experiment III

Treatment number	1a	1b	2a	2b	3a	3b	4a	4b	5	6	7
Iron compound				Iron-dextran					Ferric	ammonium	citrate
Iron, 3-4 days, mg.	100	100	150	150	0	0	0	0	100	150	0
Iron, 7 days, mg.	0	0	0	0	100	100	150	150	0	0	100
Vitamin B_{12}, mcg.	0	30	0	30	0	30	0	30	80	120	80
Number pigs per treatment	12	10	11	13	11	8	9	11	9	9	10
Number deaths per treatment	2	2	1	0	1	2	1	1	3	4	2
Birth:											
Average weight, lb.	3.1	2.9	3.0	2.8	2.8	2.9	2.9	2.6	2.8	3.0	3.1
3-4 days:											
Average hemoglobin, g./100 ml.	9.5	9.1	8.8	9.3	......	......	......	......	8.8	8.8	......
Average weight, lb.	3.8	3.3	3.4	3.3	......	......	......	......	3.3	3.6	......
7 days:											
Average hemoglobin, g./100 ml.	9.2^{a}	9.5^{a}	9.6^{a}	9.4^{a}	7.3^{b}	7.9^{b}	8.0^{b}	7.5^{b}	8.8	9.2	8.4
Average weight, lb.	5.4	4.7	4.6	4.6	4.9	5.0	5.0	4.8	4.0	4.7	5.1
14 days:											
Average hemoglobin, g./100 ml.	10.3	10.2	10.3	10.9	9.4	10.0	9.8	9.4	8.9	9.5	8.2
Average weight, lb.	8.6	7.5	7.6	7.2	7.9	7.8	8.2	7.7	6.2	7.3	7.7
21 days:											
Average hemoglobin, g./100 ml.	9.4	10.0	10.4	10.8	9.7	10.2	10.4	10.5	7.7	8.7	7.0
Average weight, lb.	11.9	10.5	10.7	10.5	11.0	10.7	11.5	10.8	9.0	10.2	10.8
28 days:											
Average hemoglobin, g./100 ml.	8.2	8.9	9.7	9.9	8.4	9.0	9.7	9.4	6.6	7.5	6.6
Average weight, lb.	15.8	14.4	14.6	14.3	14.8	14.0	15.8	14.8	11.9	13.6	14.0
35 days:											
Average hemoglobin, g./100 ml.	$7.5^{a,c}$	$7.8^{a,c}$	$8.5^{a,d}$	9.1^{d}	$7.6^{a,c}$	$8.2^{a,d}$	$8.7^{a,d}$	$8.5^{a,d}$	6.1^{b}	$6.8^{b,c}$	6.1^{b}
Average weight, lb.	20.0	18.4	19.1	19.1	19.0	18.2	20.9	18.1	14.9	17.4	17.0
Average adjusted weight, lb.*	18.9	18.2	18.7	$19.4^{e,g}$	$19.5^{e,g}$	18.1	$20.8^{e,g}$	$19.5^{e,g}$	15.5^{f}	$17.1^{e,f,h}$	16.2^{f}

* Adjusted 35-day weights (final weights of pigs adjusted ± 0.45 pound for each deviation of ± 0.1 pound from mean birth weight for all pigs, 2.90 pounds).

^{a,b,c,d} Means within lines and having different superscript letters differ significantly ($P < 0.01$).

^{e,f,g,h} Means within lines and having different superscript letters differ significantly ($P < 0.05$).

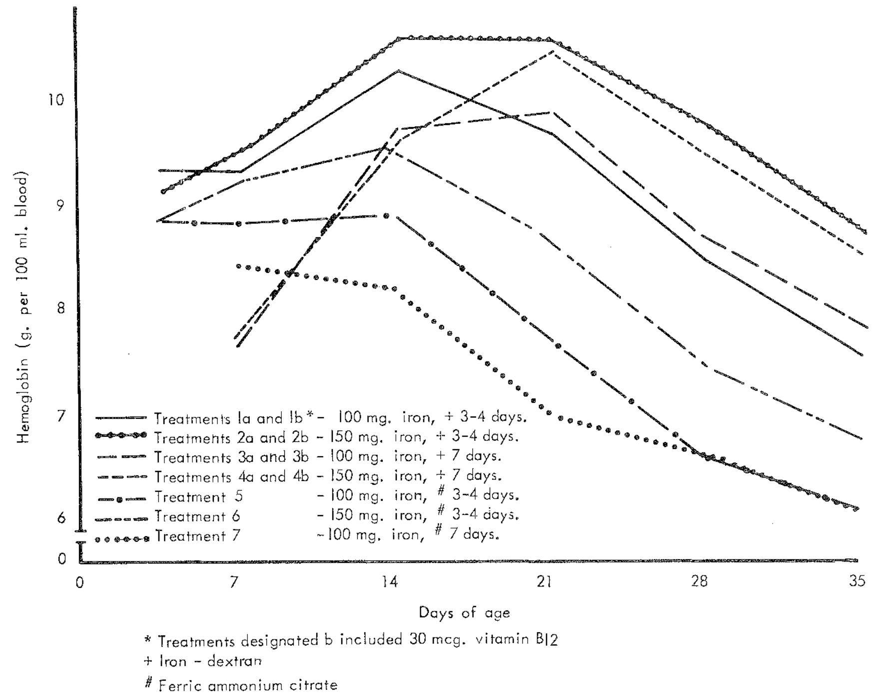

Figure 4. **The effect of various iron-dextran and ferric ammonium citrate treatments on the average hemoglobin content of the blood—experiment III.**

Approximately 24 percent of the pigs treated with ferric ammonium citrate died following treatment. Most pigs that died showed symptoms of toxicity by vomiting within a few minutes after administration of the compound. This condition often was followed by convulsive seizures, stiffness, apparent paralysis, and, finally, death. The rapidity of the reaction may indicate that the iron was released more rapidly than it could be removed from the circulatory system by the reticuloendothelial system or the kidney. Toxicity may have been due to a high level of ionic iron in the blood. Severity of postinjection reactions with subsequent death losses was greater when pigs were treated when 3-4 days old than when 7 days old.

EXPERIMENT IV

TRIAL 1. Average hemoglobin values and weights of pigs at weekly intervals throughout the experiment are shown in table 4. Results essentially agreed with those of the corresponding portion of experiment III. Administration of 150 mg. iron from iron-dextran resulted in significantly ($P<0.01$) higher hemoglobin values at 3, 4, and 5 weeks than did the 100 mg. treatment. Greater differences in hemoglobin values due to amount of iron administered resulted in this experiment than in experiment III.

Table 4. Effects of amount of iron*, time of dosages, and vitamin B_{12} on hemoglobin content of the blood and weights of nursing pigs—experiment IV, trial 1

Treatment number	1a	1b	2a	2b	3a	3b	4a	4b
Iron, mg.	100	100	150	150	100	100	150	150
Day of administration	3-4	3-4	3-4	3-4	7	7	7	7
Vitamin B_{12}, mcg.	0	60	0	60	0	60	0	60
Number pigs per treatment	13	15	15	13	15	13	13	11
Number deaths per treatment	1	1	1	0	0	2	1	4
Birth:								
Average hemoglobin, g./100 ml.	10.0	9.4	10.0	9.6	10.0	9.2	9.7	9.8
Average weight, lb.	2.8	2.5	2.6	2.6	2.8	2.6	2.6	2.8
7 days:								
Average hemoglobin, g./100 ml.	9.2	8.2	8.5	8.7	7.3	7.1	7.4	7.2
Average weight, lb.	4.6	4.7	4.5	4.4	4.7	4.4	4.5	5.0
14 days:								
Average hemoglobin, g./100 ml.	10.8	10.3	10.8	10.9	9.8	10.1	10.1	10.3
Average weight, lb.	7.6	7.7	7.5	7.1	7.6	7.0	7.3	7.8
21 days:								
Average hemoglobin, g./100 ml.	10.5	10.0	11.4	11.4	10.6	10.8	11.5	11.7
Average weight, lb.	11.0	11.1	10.7	10.1	10.6	9.9	10.5	10.7
28 days:								
Average hemoglobin, g./100 ml.	9.1	9.0	10.9	10.8	9.9	10.3	11.3	10.8
Average weight, lb.	14.8	14.3	14.0	13.5	13.9	12.9	13.4	14.6
35 days:								
Average hemoglobin, g./100 ml.	8.3	8.1	10.0	10.0	9.2	9.1	10.4	10.0
Average weight, lb.	18.2	17.4	17.9	17.1	17.8	16.4	17.6	19.0
Average adjusted weight, lb.†	17.5	18.3	18.1	17.5	17.2	16.5	17.7	18.2

* All pigs treated with injectable iron-dextran.

† Adjusted 35-day weights (final weights of pigs adjusted ± 0.43 pound for each deviation of ± 0.1 pound from mean birth weight for all pigs, 2.65 pounds).

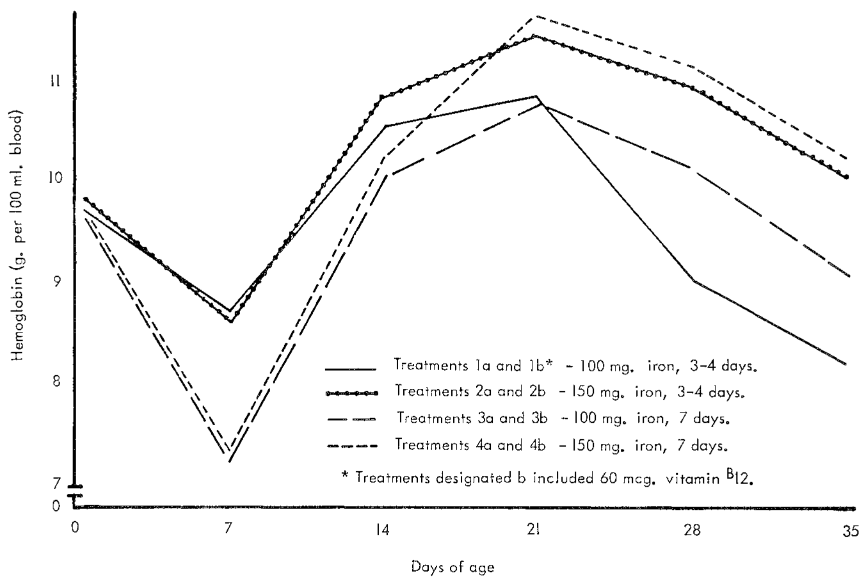

Figure 5. The effect of various iron-dextran treatments on the average hemoglobin content of the blood of suckling pigs—experiment IV, trial 1.

As demonstrated in experiment III, pigs treated at 7 days of age had significantly ($P<0.01$) lower hemoglobin concentrations in the blood at 7 and 14 days than did pigs treated initially when 3-4 days old. Figure 5 shows the slightly higher hemoglobin levels at 28 and 35 days when pigs were treated initially at 7 days. It also illustrates that the earlier administration may partially have alleviated the inevitable postnatal decline in hemoglobin concentrations (18).

In this experiment, as contrasted to the previous one, pigs treated at 7 days of age showed significantly ($P<0.01$) but not substantially greater hemoglobin levels when 4 and 5 weeks old than did pigs treated with equivalent amounts of iron at 3-4 days of age. Neither amount of iron nor time of treatment significantly affected 35-day weights. However, pigs treated with 150 mg. iron averaged 0.5 pound heavier than pigs treated with 100 mg. Simultaneous injection of 60 mcg. vitamin B_{12} with the iron dosage did not affect either hemoglobin values or pig weights at 35 days.

Deaths apparently were due to crushing by the sow—not to treatment. Hemoglobin levels for pigs that died during the 1st week were normal; pigs that died later had hemoglobin levels ranging from 7.1 to 10.6 g. per 100 ml. blood at 1 week of age.

TRIAL 2. Average hemoglobin values and weights presented in table 5 indicate that injection of 60 mcg. vitamin B_{12}, 15 mg. folic acid, or a combination of the two simultaneously with 150 mg. iron from iron-dextran did not significantly affect hemoglobin levels or weights of pigs at 35 days. However, the comparison's validity can be questioned due to the possible bias introduced because of death by overlaying of five pigs assigned to the treatment administered iron-dextran alone.

Table 5. **Influence of iron-dextran in combination with vitamin B_{12}, folic acid, or a combination of the two on hemoglobin content of the blood and weights of nursing pigs—experiment IV, trial 2**

Treatment number	1	2	3	4
Iron, 3-4 days, mg.	150	150	150	150
Vitamin B_{12}, mcg.	0	60	0	60
Folic acid, mg.	0	0	15	15
Number pigs per treatment	6	11	10	9
Number deaths per treatment	5	1	1	1
Birth:				
Average hemoglobin, g./100 ml.	9.9	10.0	9.9	9.4
Average weight, lb.	3.1	3.1	3.0	2.9
7 days:				
Average hemoglobin, g./100 ml.	8.8	8.9	8.7	8.7
Average weight, lb.	5.2	5.3	5.0	5.0
14 days:				
Average hemoglobin, g./100 ml.	11.1	10.7	10.7	10.8
Average weight, lb.	8.7	8.6	8.3	8.0
21 days:				
Average hemoglobin, g./100 ml.	11.3	11.0	11.3	11.6
Average weight, lb.	11.9	12.1	11.4	11.2
28 days:				
Average hemoglobin, g./100 ml.	11.0	10.9	11.3	11.0
Average weight, lb.	14.9	14.7	14.0	14.0
35 days:				
Average hemoglobin, g./100 ml.	10.0	9.9	10.0	10.0
Average weight, lb.	19.7	19.2	18.2	18.5

Results concerning administration of vitamin B_{12} agree with those reported for the two previous experiments. Furthermore, Ullrey et al. (33) were unable to show that injection of 5 ml. of a hemopoietic vitamin solution simultaneously with administration of 100 mg. iron from iron-dextran significantly affected hemoglobin levels and weights of pigs at 31 and 35 days of age. The vitamin solution provided 1 g. nicotinamide, 100 mg. pyridoxine, 10 mg. folic acid, and 100 mcg. vitamin B_{12}.

EXPERIMENT V

Except for pigs from three litters, all pigs used in this experiment were weaned at 21 days of age. The summary in table 6 shows results for all pigs to 21 days and only for the numbers of pigs indicated to 35 days of age.

Figure 6 shows the effects of treatment when pigs were 3-4 days old in alleviating the inevitable physiological decline in hemoglobin levels previously cited (18). It also illustrates the wide differences in hemoglobin levels resulting from the use of various sources and levels of injectable iron.

Treatment with 44 mg. iron from peptonized iron at 3-4 or 7 days

Table 6. Effects of chemical nature of iron compound, amount of iron, and time of administration on hemoglobin content of the blood and weights of suckling pigs—experiment V

Treatment number	1	2	3	4	5	6	7	8	9	10
	Colloidal ferric oxide*				Peptonized iron†			Iron-dextran		
Iron, mg.	100	100	100	100	44	44	88	100	200	150
Day of administration	3-4	7	3-4	7	3-4	7	7	3-4	3-4	3-4
Number pigs per treatment‡	11(3)	12(4)	13(4)	12(3)	13(3)	11(3)	10(3)	12(3)	10(2)	10(3)
Number deaths per treatment	2	1	0	1	0	2	2	0	2	2
Birth:										
Average hemoglobin, g./100 ml.	9.5	9.8	10.0	9.3	9.6	9.7	9.3	9.5	10.1	9.7
Average weight, lb.	2.9	2.9	2.7	2.7	2.8	2.8	2.9	2.8	2.9	2.7
7 days:										
Average hemoglobin, g./100 ml.	8.4	7.3	8.4	6.5	8.3	6.8	7.1	8.4	8.6	8.3
Average weight, lb.	5.4	5.2	5.1	5.0	5.3	5.2	5.3	5.0	5.3	5.0
14 days:										
Average hemoglobin, g./100 ml.	9.5	9.4	9.2	8.7	7.9	7.4	9.1	10.2	11.2	10.9
Average weight, lb.	9.2	8.2	8.3	8.3	8.7	8.6	8.9	8.4	8.8	8.5
21 days:										
Average hemoglobin, g./100 ml.	8.7[a,d,e]	9.2[a,d]	8.7[a,d,e]	8.5[a,d,e]	6.9[b,d,e]	6.7[b,d,e]	8.3[a,d,e,g]	9.8[a,f,h]	11.6[c]	11.0[c]
Average weight, lb.	12.6	11.1	11.5	11.8	11.6	11.9	12.7	11.8	11.8	11.9
28 days:										
Average hemoglobin, g./100 ml.	7.2	7.1	7.5	7.7	5.7	5.5	6.9	8.0	10.6	9.3
Average weight, lb.	16.7	16.1	15.7	15.3	16.6	16.5	16.1	17.5	17.7	17.3
35 days:										
Average hemoglobin, g./100 ml.	6.4	6.3	6.4	6.6	5.5	5.2	5.7	7.6	9.6	8.1
Average weight, lb.	21.1	20.5	20.8	21.0	20.0	20.9	21.2	23.1	23.7	23.2

* Dextrin-ferric oxide; colloidal ferric oxide stabilized with low viscosity dextrin and supplying 50 mg. iron per ml.
† Supplied 44 mg. iron and 2 mcg. vitamin B_{12} per ml.
‡ Numbers in parentheses show number of pigs continuing from 21 to 35 days of experiment.
[a, b] Means having different superscript letters differ significantly ($P < 0.01$).
[c, d] Means having different superscript letters differ significantly ($P < 0.01$).
[e, f] Means having different superscript letters differ significantly ($P < 0.05$).
[g, h] Means having different superscript letters differ significantly ($P < 0.01$).

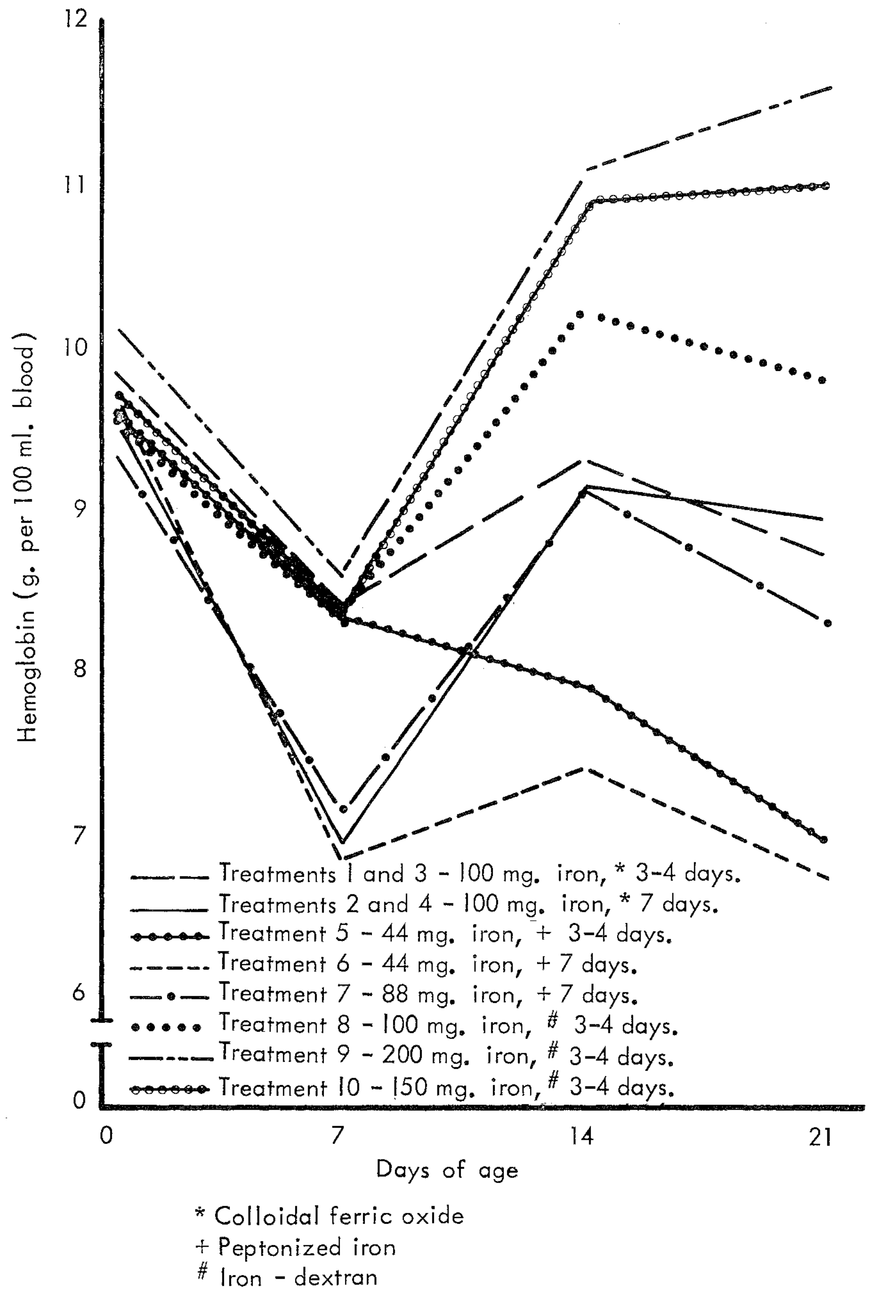

Figure 6. The effect of various injectable iron preparations on the average hemoglobin content of the blood of suckling pigs—experiment V.

of age resulted in significantly lower hemoglobin levels at 21 days than shown by pigs on any of the other eight treatments. One-half of the pigs administered this level of iron also showed visible symptoms of anemia when 21 days old—pale skin and pale mucous membranes of the mouth.

This anemic condition could have resulted in reduced resistance of the pigs to infection and predisposal to disease according to the concept of Sheffy (31). Furthermore, immunity is lowest at about 3 weeks of age because active antibody production is quite low and the concentration of colostrum-acquired antibodies has declined to a low level (3).

The average hemoglobin level of 21-day-old pigs treated with 88 mg. iron from peptonized iron at 7 days was significantly ($P<0.01$) less than that of pigs treated with 100 mg. iron from iron-dextran at 3-4 days of age. Three of the four groups of pigs treated with 100 mg. iron from dextrin-ferric oxide had significantly ($P<0.05$) lower concentrations of hemoglobin in the blood at 21 days than pigs treated with equivalent iron from iron-dextran. Indications were that 88 mg. iron from peptonized iron or 100 mg. iron from dextrin-ferric oxide supported adequate hemoglobin levels in nursing pigs weaned at 3 weeks of age but not in pigs weaned at 5 weeks. There was relatively less depression in hemoglobin levels at 3 weeks contrasted to the very low hemoglobin levels and symptoms of anemia showed by the few pigs that were continued on experiment to 35 days of age. In addition, other pigs were terminated from the experiment at 21 days of age but not weaned. These pigs showed evidence of anemia at 28 days.

Assuming a base of 5.5 g. hemoglobin per 100 ml. blood at 21 days of age for untreated pigs, pigs administered 100 mg. iron from iron-dextran utilized this iron approximately 26 percent more efficiently for hemoglobin synthesis than pigs treated with equivalent iron from dextrin-ferric oxide when 3-4 days old or 88 mg. iron from peptonized iron at 7 days of age.

Results with dextrin-ferric oxide were similar to those reported by Linkenheimer et al. (22). Available data indicate that the suckling pig must receive 200 mg. iron from dextrin-ferric oxide to maintain hemoglobin levels to 35 days of age that are comparable to those administered 150 mg. iron from iron-dextran. However, data obtained in this experiment do not permit the conclusion that a total dosage of 200 mg. iron from dextrin-ferric oxide can be administered to the 3-4-day-old pig.

Average unadjusted 21-day weights of pigs on various treatments were not significantly different. Death losses, nearly one-half of which occurred in one litter, were within normal limits and, apparently, not associated with treatment. This finding conflicts slightly with those of Wahlstrom and Juhl (34) who reported 37-percent mortality in pigs treated with peptonized iron. However, they injected 44 mg. iron intramuscularly at 1 day of age and again at 21 days.

SUMMARY

Seven hundred and ten suckling pigs from 71 litters were used in five experiments to obtain information on the roles of the following in preventing iron-deficiency anemia in suckling pigs:

- The amount of iron from injectable iron compounds.
- Time of administration of injectable iron.
- The value of vitamin B_{12}, folic acid, or a combination of the two when administered simultaneously with iron-dextran.
- Effects of chemical nature of the injectable iron compound.

Administration of 100 mg. iron from iron-dextran supported normal hemoglobin levels to 21 days of age. However, some pigs treated with this amount of iron showed hemoglobin levels and physical characteristics at 35 days indicative of anemia. Administration of 150 or 200 mg. iron from iron-dextran resulted in significantly higher hemoglobin levels in 35-day-old pigs than did the 100 mg. treatment. Treatment with 150 mg. iron from iron-dextran supported satisfactory hemoglobin levels until pigs were 35 days of age. Pigs administered 150 mg. iron from iron-dextran averaged 9 g. or more hemoglobin per 100 ml. blood in all but one experiment. None of the pigs showed hemoglobin levels indicative of anemia. Higher, but apparently no more satisfactory, hemoglobin levels were obtained when pigs were administered 200 mg. iron from iron-dextran compared to 150 mg.

A total dosage of either 150 or 200 mg. iron from iron-dextran was safely administered to the 3-4-day-old pig. Dividing the dosage to supply 100 mg. iron at 3-4 days and the remaining 50 or 100 mg. at 21 days did not result in greater hemoglobin levels or pig weights at 35 days. Generally, pigs administered 150 or 200 mg. iron from iron-dextran were heavier at 35 days than pigs treated with 100 mg. However, the difference in adjusted 35-day weights was significant in only one experiment.

Delaying administration of 100 or 150 mg. iron from iron-dextran from 3-4 to 7 days of age did not result in significantly greater hemoglobin levels or weights of 35-day-old pigs in one experiment. In a second experiment, however, significant but not substantial increases in 35-day hemoglobin values resulted with no apparent effect on weights. The simultaneous injection of 30 or 60 mcg. vitamin B_{12}, 15 mg. folic acid, or a combination of the two with 100 or 150 mg. iron from iron-dextran did not affect 35-day hemoglobin levels or weights. Delaying administration of iron from 3-4 to 7 days resulted in pigs showing significantly lower hemoglobin values when 7 and 14 days old than pigs treated at 3-4 days.

Chemical nature of the iron preparation was important in evaluating effectiveness of the compound and in determining proper levels of the injectable iron compound to administer. Iron from ferric ammonium citrate, peptonized iron, and dextrin-ferric oxide appeared to be less efficiently utilized for hemoglobin synthesis than equivalent amounts of iron from iron-dextran. At 35 days of age, pigs treated with ferric ammonium

citrate showed significantly lower and less satisfactory hemoglobin levels and substantially lower weights than pigs treated with equivalent amounts of iron from iron-dextran. Some pigs also showed evidence of toxicity following administration of ferric ammonium citrate.

Suckling pigs treated with 44 mg. iron from peptonized iron showed lower hemoglobin levels at 21 days of age than pigs treated with 100 mg. iron from iron-dextran. Dextrin-ferric oxide, administered to provide 100 mg. iron to the 3-4 or 7-day-old pig, maintained satisfactory hemoglobin levels in pigs weaned at 21 days. Apparently, 200 mg. iron from dextrin-ferric oxide are required to maintain satisfactory hemoglobin levels in pigs nursing their dams to 35 days of age.

2M—5-66

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