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Effect on type dietary protein and form of iron in creep feed diets on utilization of iron by nursing pigs

Cecillia Kim Bensinger

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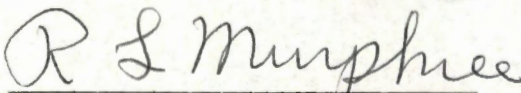
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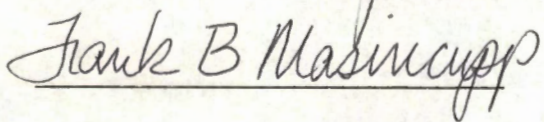
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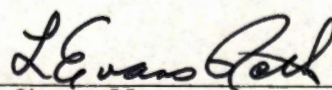
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Vice Chancellor
Graduate Studies and Research

EFFECT ON TYPE OF DIETARY PROTEIN AND FORM OF IRON

IN CREEP FEED DIETS ON UTILIZATION

OF IRON BY NURSING PIGS

A Thesis

Presented for the

Master of Science

Degree

The University of Tennessee, Knoxville

Cecillia Kim Bensinger

March 1979

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ABSTRACT

In two experiments nursing piglets (142 Duroc x Yorkshire x
Duroc pigs from the October 1977 farrowing and 160 Duroc x Yorkshire
x Duroc pigs from the January 1978 farrowing), respectively, were
utilized to determine the effect of type of dietary protein and
form of iron in creep feed diets on utilization of iron by nursing
pigs. The litters were randomly allotted to one of four treatment
rations. In experiment one the treatment rations consisted of corn
+ soybean meal + ferrous sulfate (control ration) (1); corn + soy-
bean meal + ferric citrate (2); corn + soybean meal + dried whey pro-
duct + ferrous sulfate (3); corn + soybean meal + dried whey product
+ ferric citrate (4). In experiment two the treatment rations (1)
and (2) were the same as in experiment one. Treatment ration (5)
consisted of corn + dried skim milk + ferrous sulfate, and (6) consis-
ted of corn + dried skim milk + ferric citrate. One-half the piglets in
each litter were injected with 100 mg of iron from iron dextran
intramuscularly at three days of age.

In experiment one the pigs were weighed and bled at 3, 4, and
5 weeks and in experiment two at 5, 6, and 7 weeks. At 10 days of
age feeding of the experimental rations was initiated, and throughout
the trial creep feed consumption was recorded. At each bleeding
various hematological analyses were performed on each blood sample.

The results of this study indicate that source of protein and
form of iron can affect iron utilization in the nursing pig. In
experiment one pigs receiving soybean meal protein had greater hemo-
globin and hematocrit values, serum iron concentrations, and total

iron binding capacities. In experiment one hematocrit values were higher for those pigs receiving ferric citrate, but hemoglobin, serum iron and total iron binding capacity values were less at each of the three bleeding periods.

There was no significant difference on average pig weight due to source of protein or form of iron at any of the three weigh periods. Pigs receiving dried whey product and those receiving ferric citrate consumed less total creep feed per pig weaned than those receiving soybean meal or ferrous sulfate.

In experiment two hemoglobin and hematocrit values, serum iron, total iron binding capacity and percent saturation of trans- ferrin values of pigs receiving soybean meal as a source of protein were higher at each of the three bleeding periods. Those pigs re- ceiving ferrous sulfate had higher hematocrit, hemoglobin, serum iron and total iron binding capacity values at all three time periods. The hemoglobin, hematocrit and serum iron concentrations were signifi- cantly higher by the seventh week.

The average weekly weights were significantly greater for pigs receiving dried skim milk as the major source of dietary protein. Form of iron had no significant effect; however, pigs receiving ferric citrate were heavier.

Pigs receiving dried skim milk in the diet consumed more total creep feed per pig weaned than did pigs receiving soybean protein. In both experiment one and two, pigs receiving ferric citrate con- sumed less total creep feed per pig weaned than pigs receiving ferrous sulfate.

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CHAPTER I

INTRODUCTION

Since 1923 when McGowan and Crichton discovered that anemia
in nursing pigs reared in confinement was caused by an iron deficiency,
there has been much time and effort spent in developing an efficient
and economic means of supplying this iron to the rapidly developing
piglet.

Until 20 years ago, when British workers developed an effective
injectable form of iron, methods of preventing anemia were numerous
and ineffective. The injectable iron compound led the way for more
efficient and economical methods of preventing baby pig anemia. Not
only have researchers worked with parenteral supplements, they also
have developed ways of orally administering iron.

As early as 1926, workers demonstrated the differing availability
of iron compounds fed to piglets, and it has also been recently re-
ported that other dietary constituents -- fats, carbohydrates, and
proteins -- have an effect on iron utilization.

Many early weaning diets contain milk products as the source
of protein. Because milk protein is expensive, it would be economi-
cally desirable to use less expensive soybean protein. The avail-
ability of zinc, calcium, phosphorus and magnesium is less in baby
pig diets containing soybean protein than in diets containing
casein or milk protein. However, the availability of iron is higher
in diets containing soybean protein than that in casein diets of similar

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levels of iron. It has been reported that the caseins of milk are capable of sequestering iron in such a fashion as to render it nutritionally unavailable to the organism.

Evidence is accumulating indicating that the iron requirements of pigs can be influenced by many dietary factors. There is need for further research to determine the effects of these dietary factors on oral iron utilization by the baby pig and in preventing iron deficiency anemia in baby pigs. Therefore, the objective of this study was to determine the effect of type of dietary protein -- soybean meal and milk protein -- and form of iron -- ferrous sulfate and ferric citrate -- in creep feed diets on utilization of iron by nursing pigs.

CHAPTER II

LITERATURE REVIEW

I. GENERAL INTRODUCTION

Iron is one of the most abundant elements on our planet. Iron is also a metal of remarkable biological versatility. Since its presence in the tissue was first demonstrated in 1713, and related to tissue combustion 30 years later, we have constantly added to our knowledge of iron's biological importance. Although iron is a micromineral and is found in minute amounts in the living organism, it is "complexed" with a variety of protein molecules. The way it combines with each of the various proteins confers specific function to that protein. If the protein is globin, the resultant molecule is hemoglobin. Other proteins combine with iron to form enzymes as catalase, peroxidase, Warburg's respiratory enzyme, and the cytochromes, which by virtue of their iron content, perform the essential function of electron transport in tissue respiration. Therefore iron metabolism is the means by which iron changes the biological behavior of molecules that have affinity for it (Finch, 1969). There is still confusion in defining the cellular function of many of the iron-containing enzymes, but their ability for oxygen-hydrogen ion exchange is of biological importance.

Iron in the body is in a dynamic state. It is distributed in several pools: (1) the circulatory pool, i.e., hemoglobin (60%-70%)

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and plasma transferrin (0.1%); (2) stored, i.e., ferritin and hemo-siderin (25%); (3) stored in myoglobin (3%-7%); and (4) as a consti-tuent of respiratory enzymes (0.1%) (Szabuniewicz and McCrady, 1977).

Iron turnover in the body of an adult male is 10% to 15% a year or less than 1 milligram per day. The body secretes only minimal quantities or iron. Three-fourths is lost through the gastrointestinal tract, one-fourth through epithelial desquamantion, sweat, bile, and urine. There is no specific iron excretion mechanism, therefore iron balance is maintained primarily by adjusting the amount of iron ab-sorbed. Even though iron is consumed in sufficient quantities, the answer to the anemia problem is related to the difficulty of converting dietary iron to an available form for absorption.

McGowan and Critchon (1923) were the first to recognize iron deficiency anemia in baby pigs raised in confinement, and since then the problem has been to provide the necessary iron for the baby pigs by the most efficient means. Miller (1973) stated that the need for supplemental iron by the nursing pig to prevent anemia continues to be one of the most important facets of confinement rearing of swine. It has been recognized for over 40 years since Wisconsin researchers recognized that one could prevent anemia in baby pigs by access to the iron in the soil. It has been nearly 20 years since British researchers introduced an effective injectable iron preparation. Still, the search goes on for better ways of meeting the iron needs of the nursing pig.

II. THERAPEUTIC USES OF IRON

The history of the increasing importance of anemia parallels
efforts to produce pork more efficiently. To control diseases and
parasites in swine, there has been maximal use of concrete which
could be thoroughly cleaned and disinfected. In doing this the prob-
abilities of anemia were unwittingly increased because the baby pigs
were separated from their natural source of antianemic elements in the
soil (Smithwick et al., 1967). There are basically four reasons why
anemia rapidly develops in nursing pigs reared in confinement unless
they receive supplemental iron either orally or by injection. These
are: (1) low body storage of iron in the newborn pig; (2) low iron
content of sow's colostrum; (3) elimination of contact with soil iron;
and (4) tremendous growth rate of nursing pig (Miller, 1973). Anemia
can also result from abnormal situations such as hemorrhage at birth,
vitamin B_6, copper deficiency, or anemia due to parasites.

The newborn pig has a comparatively lower amount of tissue iron
than other species, but as he matures he has the highest level of tis-
sue iron (Miller, 1973). Venn et al. (1947) reported that the newborn
piglets contained about 50 mg of iron at birth. Thorin-Tolling (1975)
reported about 30 mg in the neonatal pig, but positively correlated
the iron content to body weight. The greatest tissue iron deposit
at birth was found in the liver (about 4-5 mg) but the greatest per-
cent of iron was found in the hemoglobin pool (Thorin-Tolling, 1975).
The liver store of iron is readily available if no iron supplementa-
tion except the sow's milk is given; however, the amounts of iron in

the liver at birth are insufficient to make a significant contribution to the requirements of the body during the following three weeks (Venn et al., 1947), which is the period of rapid growth and development. To grow at a normal rate without becoming anemic the piglet must retain about 7 mg of iron per day (Venn et al., 1947). If it is assumed that during the first week piglets feed 12 times a day and on each occasion consume 35 g of milk containing 0.2 mg of iron per 100 g, then each obtained 0.84 mg of iron per day or 5.9 mg per week. Within the next 14 days the milk may supply 1.2 mg iron per day or a total of 23 mg in the first three weeks of life. In order to grow reasonably and maintain a normal hemoglobin level (8 g/100 ml) (Hart et al., 1929) the piglet is required to absorb and retain approximately 250-350 mg of iron during this time. Piglets on cleaned concrete sites had 78 mg of iron in the first three weeks (Venn et al., 1947), so the major part of the iron must come from some other source, such as the soil or an oral or parenteral supplement.

Sow's milk meets all the mineral requirements for the piglet except in the iron content during the first three weeks of life. Colostrum contains only 10 ppm iron and milk 5 ppm. This is not adequate to meet the 100 ppm requirement of the nursing pig (Miller, 1973). Attempts by workers (Venn et al., 1947; Hart et al., 1929; Miller et al., 1967) to increase the iron content of sow's milk by supplemental iron in the gestation or lactation ration or by intramuscular injection of the sow with iron have proven unsuccessful. Veum et al. (1965) and Miller et al. (1964) found that the iron content of sow's milk did not

increase when the lactation ration was supplemented with iron in the form of ferrous fumerate. However, other researchers (Chaney and Barnhart, 1963; Catron et al., 1963; Hansard et al., 1963) have data that support milk iron levels were increased by feeding ferrous fumerate in the lactation ration. In these studies the piglets nursing the iron supplemented sows had hemoglobin and hematocrit values that were similar to blood values of piglets injected with 100 mg iron dextran and considered normal. Miller (1976) suggests that most of the iron obtained by the baby pigs nursing sows receiving high levels of iron is obtained from the sow's feed and feces. Workers at the University of Florida, Gainesville, have examined the effect of an intramuscular injection of the dam with iron dextran in conjunction with peak movements of an iron-containing progesterone-induced glyco-protein (PIG) across the placenta on iron stores of prenatal and neo-natal piglets. The data suggest that treatment of the dam with iron at the time of peak movement of PIG into the fetal allantois fluid has a positive effect on iron stores of the prenatal and neonatal piglet (Ducsay et al., 1977). This new information may lead to a more economic method of preventing baby pig anemia by increasing the amount of iron in the fetal and neonatal pig.

There can be no doubt that pigs reared in modern farrowing facilities need supplemental iron and that attempts to meet this need indirectly through the sow are inadequate. Currently acceptable means of meeting this need are by direct oral or intramuscular administration to the pig (Miller, 1973).

The oral iron requirement of baby pigs is currently listed as

80 ppm in the diet or as 7 mg of absorbed iron daily (Miller, 1973). It is well known that the efficiency of utilization of orally administered iron is dependent upon many exogenous factors, as well as the state of iron depletion, and that much more must be supplied per os than is necessary for normal metabolic function (Ullrey et al., 1960). The chemical form of iron is very important as some forms are almost totally unavailable to the pig.

As early as 1926, Mitchell and Schmidt demonstrated that dietary ferric chloride and ferric ammonium citrate produced greater improvement in hemoglobin levels of anemic rats than did ferric oxide or ferrous carbonate. Also, Elvehjem and Hart (1929) demonstrated that anemia in chickens fed a milk diet was corrected by feeding ferric chloride ($FeCl_3$) or ferrous sulfate ($FeSO_4 \cdot 7H_2O$), but not by feeding ferric oxide (Fe_2O_3). Based on hemoglobin and red blood cell regeneration in anemic rats, Nesbit and Elmslie (1960) found ferrous carbonate to be relatively unavailable when compared to ferrous sulfate (Ammerman and Miller, 1972). In 1970, Fritz et al. determined the relative biological availability of iron from various sources. Compounds classified as good sources of iron when compared with the standard ferrous sulfate ($FeSO_4 \cdot 7H_2O$) included ferric ammonium citrate, ferric chloride, ferric sulfate, ferrous ammonium sulfate and ferrous fumerate. Iron sources yielding a poor response included ferric oxide, ferric orthophosphate, ferrous carbonate and sodium iron pyrophosphate.

In 1961, Pickett et al. demonstrated that iron from ferric oxide was relatively unavailable to young pigs. Ferrous sulfate ($FeS_4 \cdot H_2O$) yielded the highest gains and hemoglobin values while

iron carbonate was intermediate. Ferric ammonium citrate was equal to ferrous sulfate for baby pigs based on hemoglobin values and weight ginas (Harmon et al., 1967). Harmon et al. (1969) compared a feed grade ferrous carbonate with USP grade ferrous sulfate as iron sources for young pigs. The ferrous carbonate which provided 130 ppm iron failed to promote hemoglobin levels higher than the hemoglobin levels obtained with the 18 ppm iron contained in the basal diet. About 35 to 50 ppm iron as ferrous sulfate increased hemoglobin levels and body weight gains over those from the basal diet. Ammerman et al. (1974) compared feed grade ferrous carbonate to reagent grade ferrous sulfate. Pigs were weaned at 14 to 21 days and were not given iron injections or oral iron treatments prior to tests. In one of the two experiments with swine no significant differences in growth, hemoglobin or hematocrit were observed between pigs fed ferrous sulfate and those fed ferrous carbonate ore most soluble in 0.4% hydrochloric acid. The two carbonate ores containing the highest percentage of iron soluble in either 0.4% HCl or 2.0% citric acid produced higher hemoglobin and hematocrit values than the basal diets. These certain ferrous carbonate ores may have value as iron sources for baby pigs. In studies with rats by the same workers, ferrous carbonate ores gave consistently lower hemoglobin responses than were obtained with ferrous sulfate. Harmon et al. (1974) found that iron dextran administered orally in the newborn pig supported hemoglobin and hematocrit values as effectively as that injected. Also in the same study blood values of pigs on coated vs uncoated steel slats support the thesis that significant quantities of iron can be obtained by the pigs from steel

components of the pen. The process of oxidation of steel is multi-staged and some chemical forms or iron may be biologically available.

Iron dextran, which has been widely accepted as an injectable hematinic, is just as effective when given in the first 12 hours of life in maintaining hemoglobin and hematocrit values through a 28-day lactation (Harmon et al., 1974). Results from Ullrey et al. (1960) suggested that 125 ppm of oral iron in form of ferrous sulfate to be adequate for the baby pig.

Several studies have shown the effectiveness of injecting iron dextran intramuscularly. Maner et al. (1959) found that an intra-muscular injection of 100 mg (2 ml) iron at 3 and 10 days of age pro-duced body weight and hemoglobin and hematocrit levels at 24 days equal to those of pigs whose dam's udders were sprayed daily with a ferrous sulfate solution. One single injection of 100 mg iron at three days produced significantly lower hemoglobin and hematocrit levels than two injections, but higher values than oral tablets sup-plying 584 mg iron given at 3 and 10 days of age. Zimmerman et al. (1959) reported that a single 2 ml intramuscular injection of iron dextran administered at two days yielded higher hemoglobin values than weekly oral doses of an iron-copper paste. In another experiment by the same workers, a 2 ml injection of iron dextran on the day of birth was compared with fresh soil provided daily or an iron-copper paste given twice weekly. At five weeks of age the average hemo-globin values of the pigs receiving soil and paste were slightly higher than the iron-dextran group but the iron dextran injected pigs grew faster. In a third experiment, 2 ml of iron dextran injected

on the day of birth was required for maximum rate of growth when pigs were weaned at three weeks of age; however, pigs nursing until eight weeks of age required 100 mg of iron on the day of birth followed by 100 mg at three weeks of age for maximum growth.

In 1962, Kernkamp et al. observed that iron dextran or dextrin injections increased hemoglobin levels and growth rate significantly greater than pigs receiving injections of ferric ammonium citrate + vitamin B_{12}. Ullrey et al. (1961) also reported lower blood values and weights in pigs receiving 150 mg of iron as ferric ammonium citrate. Kernkamp et al. (1962) also reported that a single injection of iron dextran promptly overcame iron deficiency anemia in baby pigs treated on the 7th, 14th, or even 21st postnatal day, and those pigs receiving delayed injections were comparable to those treated at one week of age in hemoglobin and hematocrit levels and weight. Danielson and Noonan (1975) reported that the amount of iron required to support maximum weight gain was less than for increased hemoglobin levels. Their results also indicated that males responded better to the iron injection than did females in respect to body weight gain.

III. IRON METABOLISM

Piglet anemia comprises two phases: It starts from physiological anemia which is caused by a rapid increase of plasma volume due to absorption of intact maternal colostrum, and is followed by iron deficiency anemia which occurs based on an imbalanced demand-supply relationship caused by an increasing iron requirement to meet the requirement for rapid growth in the nursing period. These two

phases occur successively in a very short time (Furogouri, 1975).
Anemia of the suckling pig is of the hypochromic, microcytic type,
and the clinical signs are quite characteristic. The pigs may be
well developed and apparently well nourished, but they show poor
growth, listlessness, rough haircoat, wrinkled skin, and drooping
ears and tail. They may also exhibit labored respiration, dyspnea,
fatigue, pale skin, and pale mucous membranes. The mortality rate
may be high. Some pigs may appear healthy but die suddenly. Sur-
viving pigs are permanently unthrifty. The anemia occurs more fre-
quently when the litters are housed on concrete floors than when the
pigs are reared on pasture, since vegetation and soil normally provide
sufficient amounts of iron and copper (Szabuniewicz and McCrady, 1977).

Pigs are born with a limited store of iron; sow's milk only
supplies one-seventh of the daily requirement. The newborn pig can
obtain only 1 mg of iron per day from sow's milk or total of 21 mg
during the first three weeks of life. To grow normally and maintain
its normal hemoglobin the pig must absorb and retain a total of about
300 mg of iron. Normally there is adequate iron in the diet of the
adult animal and human being. However, only a small part (ca 10%)
of the iron present in the ingesta is absorbed from the digestive
tract (Szabuniewicz and McCrady, 1977). Many dietary components may
affect iron absorption. Those improving absorption, by increasing
solubility, are sugars (fructose and sorbitol), several amino acids,
and a number of other organic acids including ascorbic, succinic,
lactic, pyruvic and citric. Those components depressing absorption
are phosphates, phytates, oxalate, pancreatic secretions and

bicarbonate. Other factors that may play a role in iron absorption
(extraluminal) are erythropoiesis, body iron stores and anemia
(Szabuniewicz and McCrady, 1977).

The iron compounds of the body may be divided into two groups:
the iron porphyrin or heme compounds, and the nonheme-iron compounds.
The heme compounds consist primarily of blood hemoglobin (60%-70% of
the total body iron), myoglobin (3%-5%), and heme enzymes (0.1%). Of
the nonheme-iron compounds, ferritin is the most significant, accounting
for 15% of the total iron in the body. The essential function of the
heme compounds when they are associated with particular proteins is to
make oxygen available to the cell, while nonheme compounds primarily
function as storage forms for iron (MacGuire, 1977).

Iron is utilized chiefly in the synthesis of hemoglobin. The
iron-bearing portion of hemoglobin includes iron molecules in the
ferrous state, incorporated in a porphyrin ring, as an iron proto-
porphyrin, or heme. Heme is then combined with a protein, globin, to
form hemoglobin (Szabuniewicz and McCrady, 1977). While absorption
can occur from the stomach and from any portion of the intestinal
tract, it seems to be greatest in the duodenum and to decrease progres-
sively in the more distal segments. The reason for this gradient is
unknown; the duodenum may have a greater intrinsic ability to absorb
iron than does the remaining intestine, or complex insoluble compounds
of iron less capable of being absorbed may simply be formed as the iron
proceeds distally in the intestinal tract (Moore and Dubach, 1962).
Ferrous iron is much more efficiently absorbed than ferric iron and in
all probability the ferric form must always be reduced before

absorption can occur under physiological conditions. Iron is absorbed
directly into the blood stream rather than by way of the lymphatics.
The ionic iron in the intestinal lumen absorbs to specific receptors in
the brush border of the mucosal cell. From these receptors iron is
transferred to the cytoplasm of the mucosal cell by an active process
involving energy. The absorbed iron appears to be present in the cytosol
in small molecular weight form possibly chelated to amino acids, and
in equilibrium with iron poor ferritin present there. On reaching the
serosal surface of the cell, the ferric iron circulates in the plasma
where it is bound to transferrin. Transferrin or siderophilin, is
a plasma protein, $\beta1$-globulin, which selectively binds and transports
iron in the plasma. Normally one-third of the transferrin in the
plasma is bound to iron. The iron in the mucosal cell not transported
to plasma is retained in the cell as ferritin. The iron-protein
complex ferritin is formed from ferric iron and the protein apoferritin.
Ferritin is stable and is one of the mechanisms of iron storage in the
body. Ferritin occurs in significant amounts in the liver, spleen,
and bone marrow and in lesser amounts in other tissues. Ferritin is
retained in the cell until sloughed off at the tip of the villus and
thus returns to the lumen of the gut. Once incorporated into the
body, either through intestinal absorption or parenteral administra-
tion, iron is almost totally retained. Since there is no specialized
mechanism for iron excretion, iron homeostasis is unique in that it
is regulated primarily by absorption (Linder and Munro, 1977). There-
fore the body loses minimal amounts of iron through the feces, urine,
bile, sweat, blood and epithelial desquamation.

Miller (1973) reported that much of the oral iron is not absorbed across the intestinal wall, but is excreted in the feces. That which is absorbed from the small intestine appears first as plasma iron (transferrin) which is transported to the bone marrow for the production of red blood cell hemoglobin, muscle myoglobin which aids in muscle metabolism, iron enzymes in many tissues, and storage iron in the liver, spleen and bone marrow. Intramuscularly injected iron is rapidly picked up from the muscle by nearby lymph nodes and more slowly released to the plasma and tissue.

Forbes and Reina (1972) have defined three distinct age phases of gastrointestinal absorption:

(1) Infantile -- Iron absorption almost complete. Attempts to alter the level of absorption by dietary manipulation have proven unsuccessful.

(2) Transition (age 21 to 23 days) -- rats GI tract rapidly acquires the ability to reject most of iron presented to it.

(3) Adult -- percent absorption decreases.

The duodenum is the principal site of iron absorption in the newborn and adult pigs. The iron absorptive system is fully functional at birth and no fundamental development for iron absorption was observed in the neonatal period. Iron metabolism in the newborn piglet changes rapidly from fetal to adult pattern and signs of iron deficiency such as reduced concentrations of iron in the liver and plasma appear at three days of age. The data suggest that the intestinal mechanisms for iron absorption develop rapidly in the neonatal period. Forbes and Reina (1972), and Furugouri and Kawabata (1976)

suggested that the maturation process may be due to the loss by the intestine of pinocytotic capacity at around the 18th day, but since low molecular iron chelates may prevent pinocytosis activity from influencing intestinal absorption of iron, it seems likely that iron transfer from lumen to carcass did not coincide with the changes associated with protein absorption taking place in the pig intestinal mucosa during the neonatal period. Accordingly, it is possible that active absorption of iron in the neonate is not due to the intestinal capacity of pinocytosis. Furugouri and Kawabata (1976) suggested that the surfaces of the mucosa cell rather than the serosal site of the mucosal cell regulates iron transfer from lumen to carcass.

The physiochemical properties of iron and the size of the dose strongly affect the rate of intestinal absorption of iron. Also important are the intraluminal factors controlling iron absorption which are solubility of the iron salts and the molecular weight of the iron chelates or polymers which are formed with increasing pH in the medium of the intestinal lumen. Bothwell (1958) reported the most important physiologic factors related to variations in iron absorption are total body stores and the rate of erythropoiesis (Furugouri and Kawabata, 1975). Matrone et al. (1960) reported that iron need is largely a function of (1) rate of growth and (2) turnover of red blood cells. Matrone et al. (1960) and Furugouri and Kawabata (1975) reported that iron depletion did not enhance the percentage absorption of iron from the intestine. Since it was established that erythropoiesis in nursing pigs is very active (Furugouri, 1974), this fact may be due to the enhancement of iron absorption resulting from great physiological iron

need for extremely rapid growth in piglets. However, even in severe anemia there is a small amount of nonheme-iron in the liver. Matrone et al. (1960) have also reported in piglets that there is a minimum iron need required by the tissues.

Bothwell and Finch in 1962 demonstrated that 70% to 85% of iron present in plasma is used for red cell production within two weeks in normal subjects. In nursing piglets, however, radioiron incorporation into red cells after the intravenous injection of ^{59}Fe labeled plasma increased rapidly with a plateau in concentration after four days suggesting ready transfer to hemoglobin synthesis. Only a small amount of ^{59}Fe was detected in nonheme-iron in the liver of piglets. These results indicate that utilization efficiency of dosed iron for red cells is remarkably high in the nursing period (Furugouri and Kawabata, 1975). Furugouri and Kawabata (1975) reported that daily oral doses of 8 mg of iron maintained normal hemoglobin levels, but nonheme-iron in the liver was very low.

Nonheme-iron in the liver was markedly increased with the elevated dose indicating that the incorporation of iron into hemoglobin has preference over the accumulation of nonheme-iron in the liver. Once the iron demand of heme synthesis is satisfied, nonheme-iron depots increase. However, piglets receiving the elevated level of iron and those receiving 8 mg per os daily had similar rates of intestinal absorption of iron. Therefore, it appears intestinal absorption of iron in nursing pigs is not largely affected by the elevated dose. Additionally, considering the rate of iron absorption in the

gastro-intestinal tract and any need to store iron in the liver, the daily iron requirement for ingested iron will be approximately 24 mg (Furugouri and Kawabata, 1975).

IV. INFLUENCE OF DIETARY PROTEIN

The amount of iron absorbed from the diet depends on the physiological state of the organism, the nature of the food source of iron and the other dietary components consumed with the iron. There is considerable information on iron availability from individual food sources and factors that generally enhance or inhibit iron absorption (Bowering et al., 1976), but the influence of macro nutrients -- protein, fat, and carbohydrate -- on iron absorption have received relatively little attention. There is some evidence that amino acids and some sugars, in particular fructose, form soluble chelates of iron in the intestinal lumen that favor iron absorption (Bowering et al., 1976). Most of the studies have attempted to assess the effect of a single meal rich in either protein, fat, or carbohydrates on iron absorption.

The nature of the carbohydrate in the diet does modify the availability of dietary iron. Amine and Hegsted (1975) reported that iron utilization was greatest with diets containing lactose, less with diets containing sucrose, and least with diets containing starch. However, the effect of carbohydrate was not uniform when iron sources of differing availability were tested. Chang and Varnell (1977) reported the lowest active transport of ^{59}Fe across the intestinal wall as well as the lowest intestinal accumulation of radioiron occurred in rats

fed either dextrin or fructose, while rats fed cornstarch, sucrose or sucrose plus lactose exhibited the highest active transport and the highest accumulation of radioiron in the intestinal tissue. From a study by Derman et al. (1977) using Indian women, it was observed that ascorbic acid is capable of improving iron absorption from a cereal source (Maize-meal porridge).

The results of studies in which rats were fed test diets for several weeks suggested that increased fat levels enhanced iron absorption (Amine and Hegsted, 1975; Kaufman et al., 1958). Kaufman et al. (1958) observed very high levels of liver iron in rats fed diets low in protein (10% casein) and high in saturated fat (60% lard). Amine and Hegsted (1975) reported that iron utilization and iron absorption were greater when the fat in the diets fed was supplied as coconut oil (a saturated fat) than as corn oil. Bowering et al. (1977) reported that increasing both the fat level and changing to a more saturated fat source indicated a small but significant increase in iron absorption when compared to the control diet containing 5% corn oil. Hitchcock et al. (1977) observed greater weight gains and higher hematocrit and hemoglobin values in nursing pigs fed a diet containing 2.5% corn oil than in pigs fed a diet containing 2.5% dry flaked animal tallow. Pigs receiving 5% corn oil or tallow had lower hematological values than pigs receiving 2.5% corn oil or tallow in their diet.

Protein also has been observed to effect iron absorption. Klavins et al. (1962) reported that the amount of iron absorbed as determined by total body iron analysis was influenced by the protein

content of the diet. Rats fed 15% or more protein absorbed more iron than rats fed 5% and 10% protein. The total liver iron values of rats fed 5% and 10% protein were significantly less than the values of rats fed an 18% protein diet. The lower amounts of liver iron indicate that iron was not deposited in the livers of animals on low protein diets to the same degree as livers of animals fed diets containing 18% protein (Klavins et al., 1962). In addition, they cited research indicating that the amount of iron incorporated in the blood, mucosa of the small intestine, and adrenal glands was greater in animals fed diets containing 20% and 40% casein than in animals fed 5% casein. Klavins also stated that the type of protein in the diet influenced the absorption of iron for there was from two to four times greater uptake of iron in the blood when casein was fed than when egg albumin was used (Klavins et al., 1962). Roehm and Mayfield (1963) reported an impairment in the iron-transport system and hemopoietic processes when rats were fed the diets with 3.5% protein from whole ground wheat, with or without added lysine, but not when they were fed a similar diet with 3.5% protein from beef. Hitchcock et al. (1974) reported the baby pig utilizes iron contained in soy diets better than that in casein diets of similar levels of iron. Steinke and Hopkins (1978) reported that the high iron content (0.18 mg/g protein) coupled with the bioavailability of the isolated soybean protein make the isolated soybean protein a good dietary source of iron.

It has been demonstrated that amino acids in general are effective chelating agents and certain of them will increase the absorption of iron, the inference being that iron is carried in as an iron-amino

acid chelate and that for absorption of iron, amino acids are necessary. If this is true, then as the protein decreases, the amino acids in the gastro-intestinal tract decrease and there are fewer vehicles present for the transport of iron through the gastro-intestinal mucosa. This does not account for the fact that when additional iron is added to the diet of animals receiving 5% protein, the amount of iron absorbed is equal to that found in animals given a 20% protein diet without an iron supplement. As dietary protein decreases there is significant depletion of body protein, and apoferritin synthesis in the duodenum is decreased, and therefore, less iron can be taken up by the mucosal cells. Also in protein deficiency there is a decrease in transferrin, the iron carrying protein, and this may decrease the amount of iron available for deposition in the tissues as well as for hemoglobin synthesis. Although all the factors involved are not known, it is evident that protein plays an important role in the absorption of iron from the gastro-intestinal tract. It appears that approximately 15%-18% protein is necessary for adequate iron absorption, and that with lower amounts of protein the absorption of iron is impaired (Klavins et al., 1962).

CHAPTER III

EXPERIMENTAL PROCEDURES

Iron deficiency anemia is a major problem in young suckling swine raised in confinement if no supplemental iron is supplied. This study was designed to determine the effect of type of dietary protein and form of iron in creep feed diets on utilization of iron by nursing pigs. The two sources of protein used in the diets were soybean meal and milk protein. The iron was supplied in the form of ferrous sulfate ($FeSO_4 \cdot 7H_2O$) and ferric citrate ($FeC_6H_5O_7 \cdot 5H_2O$). All sows were farrowed in the farrowing barn at the University of Tennessee Blount Farm, and the piglets remained in confinement through-out the course of study, from birth to seven weeks of age.

I. GENERAL PROCEDURE

The study was conducted in two experiments. In the first experiment, Yorkshire x Duroc sows bred to a Duroc boar and their litters (142 Duroc x Yorkshire x Duroc pigs) from the fall farrowing, October 1977, were used during the experiment. In experiment two, Yorkshire x Duroc sows bred to a Duroc boar and their litters (160 Duroc x York-shire x Duroc pigs) from the winter farrowing, January 1978, were used.

At the beginning of each experiment, the litters were randomly allotted to one of four treatment rations. All treatment rations were formulated to contain 18% crude protein, provided 100 ppm iron, and

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each ration was a combination of one of the two sources of protein --
soybean meal or milk protein -- and one of the two sources of iron --
ferrous sulfate or ferric citrate. In experiment one the treatment
rations consisted of corn + soybean + ferrous sulfate (control ration)
(1); corn + soybean meal + ferric citrate (2); corn + soybean meal +
dried whey product + ferrous sulfate (3); corn + soybean meal + dried
whey product + ferric citrate (4). In experiment two the treatment
rations (1) and (2) were the same as in experiment one. Treatment
ration (5) consisted of corn + dried skim milk + ferrous sulfate and
(6) consisted of corn + dried skim milk + ferric citrate. In both
experiments one-half the pigs in each litter were given 1 cc of iron
dextran containing 100 mg of iron by intramuscular injection in the ham
at three days of age.

The baby pigs were bled from the anterior vena cava. In experi-
ment one, the pigs were bled at 3, 4, and 5 weeks and in experiment
two at 5, 6, and 7 weeks. Approximately 1 ml of blood was immediately
placed in heparinized tubes, and all the hematological parameters in-
volving whole blood were determined on the day the blood was drawn.
About 8 ml of blood were placed in empty test tubes, allowed to clot,
centrifuged, and the serum was collected and frozen at -20°C until
serum iron and total iron-binding capacity determinations were performed
at a later date. Each pig was weighed at each bleeding. Creep feed
consumption was measured weekly throughout the trial, and the total
amount of creep feed consumed per litter was recorded.

II. FACILITIES

The farrowing barn had 24 farrowing crates that were seven feet long and six feet wide, with two and a half feet of aluminum slats at the back. The sow's portion of the crate was two feet wide and enclosed with steel bars. The piglets were provided with two feet of space on each side of the sow's enclosure. The slat openings were one inch wide behind the sow and three-eights inch wide on the piglets' side of the crate. In December the slats were replaced with one inch steel rods spaced three-fourths inch wide across the back of the crate. A 25 pound capacity aluminum creep feeder was attached to the side partition of the piglets' portion of the crate. An automatic waterer and feed trough were located in front of the sow.

The crates were cleaned three times a week and the water troughs were cleaned once a week. Since the experiments were conducted in fall and winter, the farrowing barn was heated by heat lamps suspended about 18 inches above the floor in each crate.

III. MANAGEMENT

The sows were wormed prior to being moved into the farrowing barn and were washed and placed in farrowing crates approximately three days before farrowing. After farrowing, the pigs had their needle teeth clipped, birth weight recorded and they were earnotched for identification.

Routine management practice at the University of Tennessee Blount Farm normally required that all pigs receive an intramuscular

iron injection at approximately 3 and 21 days of age. Since this experiment involved iron administration, only one-half the piglets received iron injections at three days of age.

The pigs were vaccinated against erysipelas, and at four weeks of age males were castrated. When scouring was noted to occur in a litter, each pig was orally dosed with 2 ml of Swinenex (Semed Veterinary Pharmaceuticals, Bristol, Tennessee).

IV. FEEDING

All sows were fed the Blount Farm lactation ration (16% protein). The composition of this ration is presented in Table I. The sows were not fed for approximately 12 hours before or 24 hours after farrowing to reduce the stress involved with farrowing. The sows were hand-fed twice daily.

At 10 days of age, feeding of the experimental rations was initiated. The composition of the experimental rations are presented in Table II. They were fed ad libitum and feeders were checked and cleaned daily to insure pigs had access to fresh feed at all times, and feed was added as necessary. The creep feed was weighed at each feeding for all litters. The pigs remained on treatment rations until the third and final bleeding. The pigs were then fed the control ration until weaning at eight weeks of age.

V. PROCEDURES AND METHODS OF ANALYSES

The following observations were collected at each weighing period: weight, hemoglobin concentration, hematocrit, serum iron, total iron binding capacity and creep feed consumption.

TABLE I. Composition of Sow Lactation Ration

Ingredient	International Reference No.	Pounds
No. 2 Yellow corn (9%)	4-02-931	558
Soybean meal (44%)	5-04-604	74
Alfalfa meal (17%)	1-00-023	149
Wheat bran	4-05-190	149
Limestone	6-02-632	15
Dicalcium phosphate	6-01-080	30
Salt		7
UT-vitamin-mineral premix[a]		15
Antibiotic		3
Total		1000

[a]The University of Tennessee vitamin mineral premix contains 400,000 USP units of Vitamin A, 100,000 IC units of Vitamin D-3, 500 international units of Vitamin E, 400 mgs of Riboflavin, 1,000 mgs of d-Panthothenic acid, 2,000 mgs of Niacin, 1,500 mgs of choline chloride, 1.2% zinc, 1.2% iron, .8% manganese, .12% copper, .02% cobalt, .008% iodine, and .001% selenium per pound.

TABLE II. Composition of Experimental Rations

Ingredient	International Reference No.	Soy Protein		Dried Whey Product		Dried Skim Milk	
		Sulfate	Citrate	Sulfate	Citrate	Sulfate	Citrate
Corn	4-02-931	690.5	690.5	550	550	597	597
Soybean meal (44%)	5-04-604	268	268	223.5	223.5	-	-
Dried whey product (16%)	4-01-186	-	-	200	200	-	-
Dried skim milk (34%)	5-01-175	-	-	-	-	371.5	371.5
Dicalcium phosphate	6-01-080	15	15	10	10	10	10
Limestone	6-02-632	10	10	5	5	5	5
Salt		5	5	-	-	5	5
UT-Exp premix H-2[a]		5	5	5	5	5	5
Iron premix		5[b]	5[c]	5[b]	5[c]	5[b]	5[c]
Antibiotic[d]		1.5	1.5	1.5	1.5	1.5	1.5
Total		1000.0	1000.0	1000.0	1000.0	1000.0	1000.0

[a]Provided the following per pound of premix: Vitamin A, 300,000 IU; Vitamin D, 60,000 IU; Vitamin E, 500 IU; Vitamin K compound, 200 mg; riboflavin, 300 mg; nicotinic acid, 1,600 mg; D-pantothenic acid, 1,200 mg; choline, 10,000 mg; Vitamin B_{12}, 1.8 mg; zinc, 6,800 mg; manganese, 3,400 mg; iodine, 250 mg; copper, 900 mg; selenium, 9.08 mg; antioxidant, 4,500 mg.

[b]Provided 134.409 g of $FeSO_4 \cdot 7H_2O$ to give a total of 100 ppm Fe in the diet.

[c]Provided 161.969 g of $FeC_6H_5O_7 \cdot 5H_2O$ to give a total of 100 ppm Fe in the diet.

[d]Pfizer TM 50, 50 g/ton.

All the glassware used was washed free of iron with repeated applications of 50% hydrochloric acid and double distilled water.

Hemoglobin concentration was determined by using the cyano-methemoglobin method of Crosby et al. (1954).

Hematocrit was determined according to the micromethod described by McGovern et al. (1955). Blood samples were centrifuged for five minutes at 10,000 r.p.m. in an International "Micro-capillary" centri-fuge.

The methods used for the determination of serum-iron concentra-tion, total iron binding capacity and percent saturation of transferrin with iron were adapted from Olson and Hamlin (1969). Percent satura-tion of transferrin with iron was calculated by dividing serum iron concentration by total iron binding capacity.

VI. STATISTICAL ANALYSIS

The data were analyzed by the General Linear Model (GLM) pro-cedure and Duncan's Means Separation procedure of the Statistical Analysis System (SAS) (Barr et al., 1978).

The GLM procedure is a multivariate regression analysis which determines differences by significant F test. The Duncan's Means Separation procedure was used to determine significance when signifi-cant interactions were obtained in the analysis.

The independent variables used in the regression analysis were sex, injection, source of protein, form of iron, and replication. The de-pendent variables were weight, hemoglobin, hematocrit, serum iron, total iron binding capacity, and percent saturation of transferrin.

CHAPTER IV

RESULTS AND DISCUSSION

I. EXPERIMENT ONE

Measurements of hematocrit and hemoglobin were performed at
each bleeding. These parameters were used as indicators of anemia
in the piglets. The effect of the source of protein and the form of
iron on hematocrit and hemoglobin values of October farrowed pigs
are presented in Table III.

The effects of source of protein and form of iron in experiment
one on hematocrit values were similar. The hemoglobin concentrations
at three weeks in pigs receiving soybean protein as the source of pro-
tein were greater than those of pigs receiving the dried whey product
in their diet. Pigs receiving ferrous sulfate as the form of iron had
higher hemoglobin concentrations at three weeks than did pigs that
received ferric citrate. Hemoglobin concentrations during weeks
4 and 5 were not significantly affected by either of the main effects
in this experiment. Harmon et al. (1967) reported that ferric ammonium
citrate and ferrous sulfate were equal in availability for baby pigs
based on hemoglobin values and weight gains which may partially ex-
plain the lack of significant effect of protein source or form of
iron in this experiment. In addition, pigs receiving the dried whey
product diet did not receive dried whey product as the sole source
of supplemental protein. Dried whey product was added at 20% of the diet
and soybean oil meal supplied the remainder of the supplemental protein.

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TABLE III. Effect of Source of Protein and Form of Iron on Hematocrit and Hemoglobin (Experiment One)

	Protein			Iron	
	SBM[1]	DWP[2]		Sulfate	Citrate
Hematocrit, %					
3 weeks	28.9	28.0		28.0	28.8
4 weeks	29.9	30.3		29.8	30.4
5 weeks	31.9	31.4		31.5	31.7
Hemoglobin, g/100 ml					
3 weeks	9.59	7.69		9.12	7.90
4 weeks	9.49	9.64		9.45	9.69
5 weeks	10.52	10.44		10.47	10.48

[1]Soybean meal.

[2]Dried whey product.

The means for serum iron concentration, total iron binding
capacity and percent saturation of transferrin are presented in Table
IV.

Serum iron concentrations were not significantly affected by
either source of protein or form of iron in this experiment. By five
weeks, pigs receiving soybean protein as the source of protein and
ferrous sulfate as the form of iron had larger serum iron concentra-
tions. Steinke and Hopkins (1978) reported that soybean protein was
a good dietary source of protein which would require less iron sup-
plementation than a casein diet.

Total iron binding capacity was significantly affected by
the addition of dried whey product to the diet and by form of iron
in this experiment. The addition of dried whey product to the diets
of nursing pigs resulted in significant decreases in total iron binding
capacity at three and four weeks of the experiment and lower but non-
significant binding capacity at five weeks. The use of ferric citrate
as the form of iron in diets of nursing pigs resulted in significantly
lower (P < .01) total iron binding capacities at four and five weeks
of the experiment. In 1970, Fritz et al. determined that ferrous
sulfate and ferric citrate were iron sources of similar biological
availability. The results of this study do not totally agree with
the results obtained by Fritz et al. since significant differences
in total iron binding capacity were detected.

The percent saturation of transferrin was calculated by dividing
serum iron by the total iron binding capacity and was used to determine
the proportion of transferrin bound to iron in the unmodified serum.

TABLE IV. Effect of Source of Protein and Form of Iron on
Serum Iron, Total Iron Binding Capacity, and Percent
Saturation Transferrin (Experiment One)

	Protein			Iron	
	SBM[1]	DWP[2]		Sulfate	Citrate
Serum Fe, µg/100 ml					
3 weeks	169.0	175.4		162.2	182.6
4 weeks	192.8	205.4		201.1	198.2
5 weeks	282.0	265.7		285.1	260.8
Total iron binding capacity, µg/100 ml serum					
3 weeks	770.4^a	725.9^b		753.8	740.0
4 weeks	761.8^c	663.0^d		738.9^c	676.5^d
5 weeks	634.2	598.2		656.6^c	572.5^d
Percent saturation of transferrin, %					
3 weeks	22.9	26.0		22.0	27.0
4 weeks	28.4	32.1		28.9	32.1
5 weeks	42.5	43.8		43.1	43.4

[1]Soybean meal.

[2]Dried whey product.

[a,b]Means in the same row under a common heading with different superscripts are significantly different (P < .05).

[c,d]Means in the same row under a common heading with different superscripts are significantly different (P < .01).

There were no significant differences in the mean percent saturation of transferrin due to protein source or form of iron. Pigs receiving the dried whey product and ferric citrate had a larger percent saturation of transferrin throughout the experiment.

The piglets were weighed weekly and the average pig's weights are presented in Table V. There were no significant differences in average pig weight due to source of protein or form of iron at any of the three weigh periods. These results may be partially attributable to the fact that dried whey product was added to the diet at the rate of 20% of the diet and did not constitute the entire source of supplemental protein. In addition, the iron level of 100 ppm utilized in this experiment was adequate to meet all the pigs' requirements even though differences might have been observed in hematological parameters indicating small differences in availability of iron.

Creep feed consumption per pig weaned for experiment one is presented in Table VI. Pigs receiving dried whey product in the diet consumed less total creep feed per pig weaned than did pigs receiving all soybean protein in the diet. This result was unexpected since pigs usually will consume more of a diet containing some type of milk product than one that does not have a milk product added to the diet. However, since dried whey is a by-product of the cheese manufacturing industry, it possibly contained high levels of salt, lactose, or some other factor that may have affected palatability or consumption of the dried whey product containing diets. Pigs receiving ferric citrate as the form of iron in the diet consumed less total creep feed per

TABLE V. Effect of Protein and Source of Iron on Weight (Experiment One)

	Protein			Iron	
	SBM[1]	DWP[2]		Sulfate	Citrate
Weight, lb					
3 weeks	12.0	12.8		12.6	12.2
4 weeks	16.0	16.5		16.4	16.1
5 weeks	20.5	21.5		21.3	20.6

[1]Soybean meal.

[2]Dried whey product.

TABLE VI. Effect of Protein Source and Form of Iron on Creep
Feed Consumption (Experiment One)

	Protein			Iron	
	SBM[1]	DWP[2]		Sulfate	Citrate
Creep feed consumed per pig weaned, lb					
3 weeks	.24	.25		.25	.24
4 weeks	1.03	1.25		1.01	.78
5 weeks	1.85	1.44		1.84	1.45
Total	3.12	2.51		3.12	2.51

[1]Soybean meal.

[2]Dried whey product.

pig weaned than did pigs which received ferrous sulfate for which there is no apparent explanation.

II. EXPERIMENT TWO

In experiment two, hematocrit values (Table VII) were not significantly affected by source of protein even though pigs receiving dried skim milk had lower values at each bleeding. Pigs receiving ferric citrate had lower hematocrit values at all three time periods. The difference in hematocrit due to form of iron approached significance ($P < .053$) at six weeks and by the seventh week pigs receiving ferrous sulfate had significantly higher ($P < .05$) hematocrit values than did pigs receiving ferric citrate as the form of iron.

The hemoglobin concentrations of pigs in experiment two are presented in Table VII. Hemoglobin concentrations were significantly lower ($P < .05$) for pigs receiving dried skim milk than for pigs receiving soybean protein at six weeks and by form of iron at week 7 ($P < .01$). Ferric citrate did not support hemoglobin values as high as that of ferrous sulfate. This agrees with previous research indicating that ferric citrate as a form of iron was not as available as ferrous sulfate. Steinke and Hopkins (1978) demonstrated that rats could use soybean protein with less iron supplementation than casein protein, indicating that soybean protein was a more available source of iron. The results of this experiment may be due to the more available form of iron ferrous sulfate and the pigs consuming more creep feed.

The average serum iron concentrations, total iron binding

TABLE VII. Effect of Source of Protein and Form of Iron on Hematocrit and Hemoglobin (Experiment Two)

	Protein			Iron	
	SBM[1]	DSM[2]		Sulfate	Citrate
Hematocrit, %					
5 weeks	31.2	30.7		31.6	30.3
6 weeks	30.8	29.6		31.0	29.4
7 weeks	32.0	31.2		32.8[a]	30.6[b]
Hemoglobin, g/100 ml					
5 weeks	10.49	10.09		10.51	10.09
6 weeks	10.91[a]	10.20[b]		10.71	10.36
7 weeks	11.11	11.02		11.63[c]	10.60[d]

[1]Soybean meal.

[2]Dried skim milk.

[a,b]Means in the same row under a common heading with different superscripts are significantly different ($P < .05$).

[c,d]Means in the same row under a common heading with different superscripts are significantly different ($P < .01$).

capacity and percent saturation of transferrin are presented in Table VIII. Source of protein had a signifcant (P < .01) effect on serum iron and percent transferrin saturation during week 7. Hitchcock et al. (1974) reported that the baby pigs utilized iron contained in soy diets better than that in casein diets of similar levels of iron. Therefore, higher values due to soybean protein might be expected. Dried skim milk as the major source of protein decreased serum parameters at all measurement times.

Form of iron had significant effects on serum parameters. Pigs receiving ferric citrate had lower values for serum iron, total iron binding capacity and percent saturation of transferrin at all measurement times. Significant effects were observed in serum iron at week 6 (P < .05) and week 7 (P < .01); total iron binding capacity at week 6 (P < .01) and week 7 (P < .055); percent transferrin saturation at week 7 (P < .05).

The average piglet weight at each measurement period is presented in Table IX. The average weekly weights at weeks 5, 6, and 7 were significantly greater for pigs receiving dried skim milk as the major source of dietary protein than for pigs receiving soybean protein as the major source of dietary protein. Form of iron had no significant effect on average pig weight at weeks 5, 6, and 7. These findings were as expected that pigs receiving some form of milk protein in the diet usually consume more feed and gain better than pigs receiving starter rations without some source of milk protein and since requirement levels of iron were provided regardless of the form of iron, one would not expect large differences in weight or gain unless one form

TABLE VIII. Effect of Source of Protein and Form of Iron on
Serum Iron, Total Iron Binding Capacity, and Percent
Saturation Transferrin (Experiment Two)

	Protein		Iron	
	SBM[1]	DSM[2]	Sulfate	Citrate
Serum Fe, μg/100 ml				
5 weeks	289.8	275.3	294.8	270.2
6 weeks	446.7	438.6	493.5[a]	393.9[b]
7 weeks	475.8[c]	321.8[d]	451.0[c]	341.6[d]
Total iron binding capacity, μg/100 ml serum				
5 weeks	500.8	494.5	501.7	493.4
6 weeks	515.5	514.0	534.7[c]	491.5[d]
7 weeks	585.8	560.6	587.3	558.9
Percent saturation of transferrin, %				
5 weeks	55.1	53.0	56.7	51.4
6 weeks	82.1	81.5	87.6	76.4
7 weeks	79.5[c]	55.5[d]	75.0[a]	59.5[b]

[1]Soybean meal.

[2]Dried skim milk.

[a,b]Means in the same row under a common heading with different superscripts are significantly different ($P < .05$).

[c,d]Means in the same row under a common heading with different superscripts are significantly different ($P < .01$).

TABLE IX.   Effect of Source of Protein and Form of Iron on
            Weight (Experiment Two)

	Protein		Iron	
	SBM[1]	DSM[2]	Sulfate	Citrate
Weight, lb				
5 weeks	16.0[a]	17.7[b]	16.4	17.3
6 weeks	18.7[a]	20.5[b]	19.2	20.0
7 weeks	22.3[c]	24.9[d]	23.0	24.2

[1]Soybean meal.

[2]Dried skim milk.

[a,b]Means in the same row under a common heading with different
superscripts are significantly different (P < .05).

[c,d]Means in the same row under a common heading with different
superscripts are significantly different (P < .01).

or iron was much less available to the animal and an iron deficiency was created.

Creep feed consumption per pig weaned for experiment two is presented in Table X. Pigs receiving dried skim milk in the diet consumed more total creep feed per pig weaned than did pigs receiving soybean protein as the major protein source. This result agrees with previous observations that pigs usually will consume more of a diet containing some type of milk product than one that does not have a milk product added to the diet.

Pigs receiving ferric citrate as the form of iron in the diet consumed less total creep feed per pig weaned than did pigs which received ferrous sulfate. This result agrees with the findings of experiment one for which there is no readily apparent explanation.

TABLE X.   Effect of Protein Source and Form of Iron on Creep
Feed Consumption (Experiment Two)

	Protein			Iron	
	SBM[1]	DSM[2]		Sulfate	Citrate
Creep feed consumed per pig weaned, lb					
5 weeks	.86	1.97		1.34	1.49
6 weeks	1.40	.98		1.54	.84
7 weeks	1.65	1.73		1.98	1.40
Total	3.71	5.06		5.08	3.68

[1]Soybean meal.

[2]Dried skim milk.

CHAPTER V

SUMMARY

In two experiments nursing piglets (142 Duroc x Yorkshire x Duroc pigs from the October 1977 farrowing and 160 Duroc x Yorkshire x Duroc pigs from the January 1978 farrowing), respectively, were utilized to determined the effect of type of dietary protein and form of iron in creep feed diets on utilization of iron by nursing pigs. The litters were randomly allotted to one of four treatment rations. In experiment one the treatment rations consisted of corn + soybean meal + ferrous sulfate (control ration) (1); corn + soybean meal + ferric citrate (2); corn + soybean meal + dried whey product + ferrous sulfate (3); corn + soybean meal + dried whey product + ferric citrate (4). In experiment two the treatment rations (1) and (2) were the same as in experiment one. Treatment ration (5) consisted of corn + dried skim milk + ferrous sulfate and (6) consisted of corn + dried skim milk + ferric citrate. One-half the piglets in each litter were injected with 100 mg of iron from iron dextran intramuscularly at three days of age.

In experiment one the pigs were weighed and bled at 3, 4, and 5 weeks and in experiment two at 5, 6, and 7 weeks. At 10 days of age, feeding of the experimental rations was initiated, and throughout the trial creep feed consumption was recorded. At each bleeding various hematological analyses were performed on each blood sample.

The results of this study indicate that source of protein and

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form of iron can affect iron utilization in the nursing pig. In
experiment one pigs receiving soybean meal protein had greater hemo-
globin and hematocrit values, serum iron concentrations and total
iron binding capacities. In experiment one hematocrit values were
higher for those pigs receiving ferric citrate, but hemoglobin, serum
iron and total iron binding capacity values were less at each of the
three bleeding periods.

There was no significant difference on average pig weight due
to source of protein or form of iron at any of the three weigh periods.
Pigs receiving dried whey product and those receiving ferric citrate
consumed less total creep feed per pig weaned than those receiving
soybean meal or ferrous sulfate.

In experiment two hemoglobin and hematocrit values, serum
iron, total iron binding capacity and percent saturation of trans-
ferrin values of pigs receiving soybean meal as a source of protein
were higher at each of the three bleeding periods. Those pigs re-
ceiving ferrous sulfate had higher hematocrit, hemoglobin, serum iron
and total iron binding capacity values at all three time periods.
The hemoglobin, hematocrit and serum iron concentrations were signifi-
cantly higher by the seventh week.

The average weekly weights were significantly greater for pigs
receiving dried skim milk as the major source of dietary protein.
Form of iron had no significant effect; however, pigs receiving ferric
citrate were heavier.

Pigs receiving dried skim milk in the diet consumed more total
creep feed per pig weaned than did pigs receiving soybean protein. In

both experiment one and two, pigs receiving ferric citrate consumed less total creep feed per pig weaned than pigs receiving ferrous sulfate.

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VITA

Cecillia Kim Bensinger was born in Lewisburg, Tennessee on April 19, 1953. She attended Hardison Elementary School and graduated from Marshall County Senior High School, Lewisburg, Tennessee in 1971. In September of 1971, she entered the University of Tennessee at Martin and graduated with a Bachelor of Science degree in Agriculture in 1976.

Beginning in September of 1976, she began a graduate program in Animal Science at the University of Tennessee, Knoxville, on a graduate research assistantship. She received her Master of Science degree in Animal Science in March of 1979.