

A STUDY OF THE CALCIUM AND PHOSPHORUS
REQUIREMENTS OF ARTIFICIALLY
REARED YOUNG PIGS

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INTRODUCTION

Calcium and phosphorus have for many years been recognized as nutrients essential for skeletal formation, growth and maintenance of various physiological functions in swine. Previous to the development of rations for early weaned pigs, there was little interest or incentive in studying the calcium and phosphorus needs of very young pigs. Even today the quantitative requirements for these two major minerals for young pigs have not been well established. However, some recent events have stimulated interest in this area of work. Some have suggested that specific-pathogen-free (SPF) pigs may have higher calcium and phosphorus requirements than conventionally reared pigs. The typical turbinate bone degeneration symptomatic of that found in atrophic rhinitis has been produced experimentally by inadequate or imbalanced calcium and phosphorus levels. These observations, coupled with increased efficiencies in production and more rapid growth rates, have given rise to the question: "Are current swine formulations meeting the calcium and phosphorus needs of swine?".

The present investigation was undertaken to study some of the effects of various dietary calcium and phosphorus levels upon the well-being of young SPF pigs as measured by body weight gain, feed consumption, feed efficiency, deter-

mination of various blood components and slaughter information.

LITERATURE REVIEW

Calcium and Phosphorus Functions

The essentiality of calcium and phosphorus has been known for many years and the importance of these two elements can hardly be over-emphasized. The total amounts of calcium and phosphorus in the body are large, calcium constituting nearly 2 percent of the weight of an animal and phosphates about 3 percent (Ruch and Fulton, 1960). Nearly all of the calcium and a large portion of the phosphorus is found in the skeleton.

A dry bone is composed of from 30 to 40 percent organic substances and 60 to 70 percent inorganic substances. This latter material is composed of two highly insoluble calcium salts: $\text{Ca}_3(\text{PO}_4)_2$, 85 percent and CaCO_3 , 10 percent; and a small amount of magnesium. The calcium to phosphorus ratio is about 2 to 1 (Tuttle and Schottelius, 1961).

The explanation generally advanced for the deposition of calcium in growing bone is the "phosphatase theory." Part of the blood phosphorus is held in combination with organic materials to form esters of phosphoric acid. Phosphatase hydrolyzes these esters, which releases the phosphoric acid and gives rise to an excess of phosphate ions. A series of complex reactions between the phosphate ions and the soluble

calcium in the growing matrix leads to the formation and precipitation of insoluble tricalcium phosphate

The soft tissues, although they contain little calcium, contain fairly large amounts of phosphate. Most of this is in the form of labile organic combinations, as nucleotides, phospholipids, phosphocreatine and phosphorylated intermediates of carbohydrate metabolism.

Calcium ions function in the activation of lipase and adenosine triphosphatase activity of myosin and promote the conversion of trypsinogen to trypsin (Fruton and Simmonds, 1961). These ions also promote the conversion of prothrombin to thrombin in the blood clotting mechanism and are important in the preservation of the integrity of cell membranes and in the normal activity of excitable tissues. Calcium ions play a specific and essential role in complement fixation, an immunological reaction. These ions are required in the union of the C'1 portion of the complement with erythrocytes and antibody (Carpenter, 1965).

Calcium Studies

Results of analyses of sow's milk by Hughes and Hart (1935), Jylling (1960), Braude et al. (1947) and Perrin (1955) revealed that post-colostral calcium concentration increases throughout lactation but that for the first three weeks of lactation, when sow's milk is usually the sole source of the baby pig's nutrients, the concentration of calcium in sow's milk consistently falls within the range of

0.8 to 1.0 percent, on a dry matter basis. Freese (1958), as cited by Miller et al. (1964b), found that about 95 percent of this calcium was retained; this high percentage retention is perhaps a result of the presence of lactose (Lengemann et al., 1959). On this basis the available calcium content of the dry matter in sow's milk is therefore in the range of 0.76 to 0.95 percent. Lenkeit and Freese (1959), using four baby pigs, observed that pigs on milk substitutes utilized both calcium and phosphorus better than pigs on sow's milk.

Since a level of calcium in a synthetic diet higher than that found in the dry matter of sow's milk has been shown to be inadequate for optimum skeletal development, it follows that the calcium supplied by the synthetic diet must have been of lower availability, since intakes of synthetic diet and sow's milk dry matter appear to have been about equal (Blair, 1963; Blair et al., 1963). Results of digestibility studies carried out by Livingstone et al. (1962) confirm that calcium as CaHPO_4 is of lower availability than the calcium of sow's milk, being about 73 percent retained when it formed 1.6 percent of the diet.

Results of Blair and Benzie (1964) suggest that for satisfactory skeletal development the available calcium content of the dietary dry matter should be at least 1.3 percent when the diet contained 1.5 percent phosphorus and 331 I.U. vitamin D per kg. A calcium level of 1.6 percent gave the most rapid skeletal development.

This estimate of the calcium requirement is higher than estimates suggested by most other workers, although Freese (1958), cited by Miller et al. (1964b), recommended that sow's milk substitutes should contain 1.3 percent calcium in readily available form. Rutledge et al. (1961) gave diets containing 0.4, 0.6, 0.8 and 1.0 percent calcium (mainly in the form of limestone) and 0.6 percent phosphorus to 16 crossbred pigs over the age period from 3 to 9 weeks. Rate of gain, feed efficiency and serum calcium and phosphorus levels were not significantly influenced by calcium content of the ration. During this relatively long experimental period nutrient requirements would undoubtedly change, but 0.8 percent calcium was suggested as a minimum requirement on the basis of the ash and calcium contents and the density of the femur, and the breaking strength and radiographic density of the humerus. No details of the availability of the calcium were given, and since the breaking strength, the ash and calcium contents, and the density of the bones were significantly increased with an increase in dietary calcium level from 0.4 to 1.0 percent, the requirement for maximum rate of bone calcification would appear to be at least 1.0 percent.

This suggests a higher calcium requirement than 0.8 percent listed for the 4.5 kg. pig (N.R.C., 1964).

Dudley et al. (1961) studied the response of young pigs to dietary calcium (mostly from CaCO_3) levels ranging from 0.1 to 2.0 percent. It was found that 0.2 percent calcium

supported maximum rate of weight gain and maximum efficiency of food conversion, but the ash content of the femur increased with all increments of calcium up to 2.0 percent of the diet.

Based on body composition, Mitchell and McClure (1937) estimated that the dietary calcium requirement would be 0.53 percent for a 13.5 kg. pig and 0.20 percent for a 90 kg. pig.

In a lengthy article, Dunlop (1935) indicated that a dietary calcium level of 0.45 percent and a calcium to phosphorus ratio of 1 to 1.3 was optimal for pigs between 13.6 and 90.7 kg.

In extensive studies, Aibel et al. (1941) tested various levels of dietary calcium (from 0.1 to 1.0 percent) with phosphorus kept constant at 0.3 percent. They concluded that 0.41 percent calcium was "definitely adequate" for normal development of the 18.1 kg. pig. However, their conclusion is surprising because both daily gain and feed conversion were superior with calcium at 0.6, 0.8 and 1.0 percent, respectively.

Golding et al. (1922) fed 0.338 percent calcium to pigs from 53 to 198 days of age and observed impaired growth and diminishing bone calcium.

Wintrobe (1939), Lehrer et al. (1949), Miller et al. (1954) and Johnson et al. (1948) obtained satisfactory rates of growth of very young pigs fed synthetic milk diets in which calcium constituted 0.94, 0.74, 0.80 and 0.91 percent,

respectively, of the total solids portion of the diet.

Whiting and Bezeau (1958) found that increasing dietary calcium from 0.40 to 0.86 percent, with phosphorus held at 0.46 percent, did not influence body weight gains or the appetite of pigs of various body weights.

Chapman et al. (1955b) found it necessary to provide 0.8 percent of calcium in the diet to insure optimal rate of gain and skeletal development for pigs from 11.3 to 45.4 kg. live weight. Pigs above 45.4 kg. needed 0.7 percent calcium.

Miller et al. (1960) studied the calcium requirement of forty baby pigs receiving a synthetic milk diet. Levels of 0, 0.4, 0.6, 0.8, 1.0, 1.2 and 1.6 percent calcium were studied with the dietary phosphorus level held constant at 0.5 percent. A level of 0.6 percent calcium supported maximum growth and feed efficiency but for maximal bone ash and specific gravity and for maximal breaking strength at least 0.8 percent calcium was required.

Lloyd et al. (1961) obtained maximal rate of gain and apparent carbohydrate digestibility in 2 to 8 week old pigs receiving a natural diet containing 1.2 percent of calcium. However, they compared only dietary levels of calcium (from 1.2 to 4.0 percent of the complete diet), while phosphorus was maintained at 1.0 percent.

Dudley et al. (1961) fed a total of 96 fourteen day old pigs a purified diet for 5 or 6 weeks to investigate the response of the baby pig to levels of dietary calcium. After 2 to 4 weeks on test, pigs fed 0.1 percent calcium

and 0.8 percent phosphorus developed weak legs and pasterns, impaired locomotion and seizures of calcium tetany which frequently culminated in death. For maximum efficiency and rate of gain, the calcium requirement was no higher than 0.2 percent. However, the ash content of the femur increased with all increments of calcium up to 2.0 percent of the diet.

Miller et al. (1962b) reported on the calcium requirement of baby pigs from 2 to 6 weeks of age. Using a synthetic milk diet, the phosphorus level was maintained at 0.5 percent, and calcium concentration varied from 0 to 1.6 percent of the dietary solids. A level of 0.4 percent calcium appeared adequate to effect normal growth, feed efficiency and blood clotting time. In one trial 0.8 percent of dietary calcium was necessary to support normal levels of serum calcium, phosphorus and alkaline phosphatase. These workers also noted that optimal skeletal development, as measured by bone density, ash content, breaking strength and the absence of rachitic symptoms, occurred in pigs consuming 1.0 percent of calcium. Maximal calcium retention also occurred at this dietary level. About 95 percent of the calcium excreted by pigs receiving 0.8 percent or less of dietary calcium was present in the feces, but pigs consuming higher levels of calcium excreted a considerable amount in the urine. Calcium content of the liver, heart and kidneys was directly related to calcium intake. Levels of serum phosphorus and alkaline phosphatase were inversely related to

dietary and serum calcium levels. They concluded that the minimal calcium requirement of the baby pig lies between 0.8 and 1.0 percent.

Hoefler et al. (1959) studied weaning pigs up to 112 days and determined their growth response to various levels of calcium, with and without zinc supplement. Levels of 0.3, 0.5 and 0.7 percent calcium were used. Zinc supplementation improved average daily gains at all calcium levels but in each case pig performance on the lower calcium level was superior to the next higher one.

A total of 202 pigs weaned at 2 weeks of age was used in a series of five experiments conducted by Combs and Wallace (1962) to study the influence of dietary calcium levels. When calcium was increased by increments of 0.10 from 0.40 to 0.80 percent and the phosphorus content was held constant at 0.44 percent, daily gains tended to decrease linearly with increasing calcium. In all experiments the most efficient utilization of feed occurred with the low calcium rations.

Newman et al. (1967) fed weanling pigs diets calculated to contain 0.2, 0.4, 0.6 and 0.8 percent calcium and 0.45 percent phosphorus. Feed intake and average daily gain increased linearly up to 0.6 percent dietary calcium. Femur calcium increased with level of calcium intake up to 0.6, while femur phosphorus increased linearly through the 0.8 percent level. Maximum bone density was attained in pigs receiving 0.6 percent calcium, whereas breaking strength was

increased linearly through the 0.8 percent level. Plasma calcium at sacrifice was somewhat reduced in pigs receiving the 0.2 percent calcium diet.

Phosphorus Studies

Results of the phosphorus analyses of sow's milk (Hughes and Hart, 1935; Perrin, 1955; Jylling, 1960) show that post-colostral phosphorus concentration increases during lactation but for the first 3 weeks the concentration of phosphorus in the dry matter of sows' milk is consistently near 0.60 percent.

Bethke et al. (1933) asserted that 0.6 percent represented the minimal requirement of swine for phosphorus, irrespective of the amount of calcium present. Freese, as cited by Miller et al. (1964b), recommended 1.1 percent phosphorus in a liquid milk diet from which 73 percent of the phosphorus was retained. Wintrobe (1939) obtained good growth and normal health in 2 to 23 day old pigs fed artificial diets containing 0.52 percent phosphorus.

Based on body composition, Mitchell and McClure (1937) estimated that the 13.5 kg. pig has a phosphorus requirement of 0.37 percent of the complete diet.

Aubel et al. (1936) fed swine on low phosphorus rations and reported the following symptoms: loss of appetite, poor utilization of feed, and failure to make normal growth and to develop bone and muscle normally, and lowering phosphorus retention. Increasing phosphorus levels beyond 0.5 percent

did not increase phosphorus retention. Dietary phosphorus levels less than 0.5 percent resulted in reduced calcium retention, whereas increasing phosphorus levels above 0.5 percent did not affect calcium balance. These workers stated that for optimal utilization of calcium and phosphorus, the dietary phosphorus level should be about 0.5 percent.

Chapman et al. (1955b) concluded that pigs below 45.4 kg. of body weight required a phosphorus level of 0.6 percent and pigs above 45.4 kg. needed 0.5 percent.

Lucas and Lodge (1961), as cited by Miller et al. (1964b), have chosen 0.90, 0.65 and 0.45 percent as "preferred estimates" of phosphorus requirements for the 4.5, 9.0 and 13.6 kg. pig, respectively.

Miller et al. (1961c), using levels of 0.2, 0.4, 0.5, 0.6, 0.7 and 0.8 percent phosphorus, observed that growth rate and feed consumption were depressed only in baby pigs receiving the lowest phosphorus level. Levels of femur ash, calcium, phosphorus, specific gravity and breaking strength were all positively related to levels of dietary phosphorus intake.

Zimmerman et al. (1961) gave diets containing 0.2 to 0.8 percent phosphorus to young pigs and found an increase in ash content, breaking strength and radiographic density of the bones with increasing phosphorus level.

Miller et al. (1964b) conducted studies with 32 baby pigs in two trials to determine their dietary phosphorus requirement. Using a synthetic milk diet, the calcium level

was maintained at 0.8 percent, and phosphorus concentration varied from 0.2 to 0.8 percent of dietary solids. A dietary phosphorus level of 0.4 percent appeared adequate to effect normal growth and economy of food utilization. A 0.5 percent level was adequate to maintain normal concentrations of serum calcium, inorganic phosphorus and alkaline phosphatase and to provide ample skeletal development. However, to obtain maximal strength of bone and to insure the absence of rachitic lesions, it appeared necessary to provide baby pigs with 0.6 percent dietary phosphorus.

Combs et al. (1962) studied the phosphorus requirement of pigs between the ages of 2 and 7 weeks and concluded that 0.44 percent of dietary phosphorus adequately met the minimal requirement of young pigs fed a fortified corn-soybean meal diet.

Miller et al. (1964d) conducted calcium and phosphorus balance studies on 29 baby pigs receiving a synthetic milk diet containing phosphorus levels of 0.3, 0.4, 0.5, 0.6, 0.7 and 0.8 percent with dietary calcium held constant at 0.8 percent. Growth rate, feed intake and mineral retention were greatly depressed in pigs receiving 0.2 percent phosphorus. Increasing the dietary phosphorus levels to 0.5 percent resulted in increased percentage of inorganic phosphorus in the blood and a marked increase in thirst and a corresponding excretion of urine. The increased water intake and excessive urine output were associated with enlarged kidneys, which were pale in color. Under the condi-

tions of the experiment, the phosphorus intake was considered adequate when the ration contained from 0.27 to 0.30 percent of phosphorus.

Newman et al. (1964b) fed corn-soybean meal rations containing 0.6 percent calcium and 0.35, 0.45, 0.55 and 0.65 percent phosphorus at 0 and 10 percent levels of tallow to 64 weanling pigs. The level of phosphorus had no effect on growth, feed consumption, feed efficiency or carcass traits. Femurs from pigs fed 0.35 percent phosphorus contained significantly more fat. Percent femur ash, calcium and phosphorus increased linearly with increasing increments of dietary calcium. Femur breaking strength and specific gravity were significantly increased at the 0.45 percent level, but no further increase was observed at the higher dietary phosphorus levels.

Vandepopuliere et al. (1959) investigated techniques for measuring the phosphorus adequacy of both a semi-synthetic and a practical type ration with 135 pigs weaned at 2 weeks of age. Results of two trials where phosphorus levels of 0.24, 0.36, 0.48, 0.60 and 0.72 percent were fed with calcium to phosphorus ratios of 1.2 to 1 and 2 to 1 showed that bone density and ash exhibited a linear response; growth was non-linear and was significantly influenced by the calcium-phosphorus ratios. Results of a factorial experiment where phosphorus levels of 0.40, 0.44 and 0.48 percent were fed with calcium-phosphorus ratios of 0.9:1, 1.2:1 and 1.5:1 indicated similar results to the response criteria

Van Zante et al. (1967) fed 90 pigs from 3 to 9 weeks of age on corn-soybean rations containing 0.35, 0.45 or 0.55 percent phosphorus. In one trial, pigs fed the highest phosphorus level had significantly faster and more efficient gains and percent calcium, phosphorus and ash of the metatarsal bone was higher than pigs fed the lowest level.

Newman et al. (1967) fed weanling pigs diets containing 0.35, 0.45, 0.55 and 0.65 percent phosphorus with calcium held constant at 0.65 percent of the total diet. Differences in feedlot performance and carcass characteristics were not significant. Femur ash, calcium and phosphorus increased in a linear fashion with increases of dietary phosphorus. Femur specific gravity and breaking strength was greatest at 0.45 percent phosphorus. Plasma inorganic phosphorus was highest in pigs fed the 0.35 percent phosphorus diet. Plasma calcium was remarkably constant at all levels of phosphorus intake.

Noland et al. (1964), using 110 weanling pigs, fed three levels (0.15, 0.25 and 0.35 percent) of inorganic phosphorus. Inorganic calcium was maintained at 0.6 percent. There was no difference in bone ash, calcium or magnesium among the pigs. Bone phosphorus was highest in the pigs fed 0.15 percent phosphorus.

Johnson et al. (1962) obtained satisfactory performance of finishing pigs (68 to 90.7 kg. live weight) with only 0.1 percent added phosphorus to a corn-soybean meal ration. The fact that a low calcium level plus adequate dietary vitamin

D may allow appreciable utilization of phytin phosphorus probably explains these satisfactory results (Taylor, 1965).

Calcium-Phosphorus Studies

Calcium and phosphorus requirements for swine have been established using a variety of criteria, i.e., growth rate, feed conversion, total ash, calcium and phosphorus content of various bones, breaking strength, specific gravity, blood clotting time and levels of calcium, phosphorus and alkaline phosphatase in blood serum.

Carroll and Krider (1956) stated the following as general clinical symptoms of a dietary calcium deficiency: slow or interrupted growth, reduced appetite, poor hair and skin condition, lameness and stiffness, weakened bone structure and in severe cases reduced serum calcium and tetany. The phosphorus deficiency symptoms listed were: slow or interrupted growth, reduced appetite, lameness and stiffness, weakened bone structure and reduced inorganic blood phosphorus.

The calcium and phosphorus of a bone are in a state of dynamic equilibrium with the calcium and phosphorus of the blood. Comar et al. (1952) used an autoradiographic technique to study the distribution of calcium and phosphorus in the bones of growing pigs. Radioactive solutions were injected intra-arterially and the pigs were slaughtered at intervals varying from 2.5 minutes to 60 days after the injection. Autoradiograms were made from longitudinal sections

of the bone. Within 2.5 minutes there was evidence of calcium⁴⁵ accumulation in the region of the epiphyseal plates. In one hour there was a large amount of radio-activity in the bone. After 60 days practically all the radioactivity had disappeared from the trabecular regions, but the endosteal region of the shaft showed the presence of some calcium⁴⁵.

With an optimal dietary intake of calcium and phosphorus, apposition of bone exceeds resorption in the growing animal; this is the basis of skeletal growth. If serum calcium is lowered, either directly from a dietary calcium deficiency or secondarily from a dietary phosphorus excess, resorption exceeds apposition and generalized osteitis fibrosa results.

Manners and McCrea (1963a) gave a detailed picture of the changes occurring in bone development in sow-reared piglets during the suckling period. There was a tendency for the proportion of ash in the bones of the piglets to decrease over the first week of life. Using suckled pigs, Manners and McCrea (1963b) found that the proportion of ash in fat-free body tissue decreased over the period from birth to 7 days of age, owing largely to a decrease in calcium concentration. From this research it would appear that the sow-reared pig is short of bone forming minerals during the first week of life when both bone weight and body weight show maximum relative increase.

Linear measurements of skeletal growth of young pigs

which nursed or were raised on purified diets have been reported by Blair et al. (1963) and Blair and Benzie (1964), and of normal and under-nourished pigs by McCance and Ford (1961) and Dickerson and McCance (1961).

Pullar (1960) reported a study of bone composition of pigs intended to provide standards for the diagnosis of rickets. He reported that bone ash content was relatively low in cases of rickets and that an animal may tolerate a considerable degree of mineral depletion before clinical signs or obvious pathological changes appear.

McCrea and Tribe (1956) stated that bones of the fore limb showed a greater proneness to distortion on artificial diets low in bone forming minerals.

Dickerson (1962) has published data on the composition of the humerus of pig foetuses at 46, 65 and 90 days of gestation and of newborn, 20 to 45, 65 to 143 days old and of 1 year old pigs. There was no regular change in the calcium to phosphorus ratio in pig bone with age.

The early (1912-1935) published calcium and phosphorus work dealt almost exclusively with pigs between weaning and market weights. It was not until the development of artificial rearing techniques and purified diets that baby pigs requirements were published with any degree of confidence.

Eggert et al. (1959) reported that bone ash was significantly lower for rations low in total calcium and phosphorus.

Zimmerman et al. (1961) used 144 baby pigs (fed from 3

to 7 weeks of age) to study the calcium and phosphorus requirements in a high milk product ration. A 4 X 4 factorial arrangement of 0.50, 0.65, 0.80 and 0.95 percent calcium and 0.4, 0.5, 0.6 and 0.7 percent total phosphorus was used. Phosphorus at 0.40 percent was inadequate for maximum gains regardless of the calcium level. Increasing the phosphorus level significantly improved feed efficiency, and conversely, increasing the level of calcium significantly decreased the same. Thus, feed utilization became progressively poorer as the calcium-phosphorus ratio widened. The treatment effects on percent metatarsal ash were additive and significant. Percent ash was increased by increasing levels of either phosphorus or calcium.

Zimmerman et al. (1963) stated that a maximum calcium level of 0.8 percent and a minimum phosphorus level of 0.6 percent was needed to assure maximum performance and adequate skeletal development with 3 to 7 week old pigs fed a high milk product diet. However, the highest levels of calcium (1.0 percent) and phosphorus (0.7 percent) in the ration produced the maximum responses in metatarsal calcification. Calcium and phosphorus appeared to independently influence feed efficiency. High calcium levels (above 0.8 percent) reduced the efficiency, while phosphorus up to approximately 0.6 percent of the ration improved the efficiency of ration utilization.

Menshan et al. (1963), using a semi-purified diet, studied the response of 76 baby pigs to graded levels of calcium

The phosphorus level was maintained at 0.45 or 0.70 percent while the calcium level varied from 0.11 to 0.7 percent. A dietary level of 0.45 percent calcium and 0.45 percent phosphorus appeared inadequate based on average daily gain, feed efficiency and bone ash determinations. There were no significant differences in serum inorganic phosphorus level or on phosphorus as a percent of femur ash.

Blair and Benzie (1964) attempted to determine the dietary levels of calcium and phosphorus that would promote satisfactory bone development in pigs weaned at approximately 10 days of age. Synthetic diets formulated to provide adequate amounts of the known nutrients and containing 0.4, 0.8, 1.2 and 1.6 percent calcium and 0.6, 0.9, 1.2 and 1.5 percent phosphorus were given to pigs of 3.6 and 11.3 kg. live weight. Raising the calcium and phosphorus levels caused a significant increase in the dry fat-free weight, ash content and radiographic density of the bones studied. Width of distal epiphysical cartilage of the ulna was significantly decreased by increasing the levels of calcium and phosphorus. These workers concluded that the 3.6 to 11.3 kg. pig requires at least 1.3 percent available calcium and a value somewhat less for phosphorus (1 to 1.2 percent). This estimate is higher than that suggested by other workers; however, few experiments have included phosphorus in excess of 1.0 percent of the ration.

Using 16 individually fed pigs, Zimmerman et al. (1960) investigated the influence of various calcium levels (0.52

to 1.05 percent) used in combination with either 0.52 percent or 0.70 percent phosphorus in diets containing a high percentage of milk products. The performance of pigs 2 to 6 weeks of age was observed in three experiments. In general, calcium levels exceeding 0.8 percent caused a depression of growth and a lower feed utilization. However, bone calcification increased with each added increment of calcium in two of the three experiments. In one experiment, where calcium balance was determined on pigs 3 to 4 weeks of age, the total retention of calcium was made maximal with a ration containing 0.88 percent calcium.

Blair (1963) reported that 22.7 kg. pigs showed signs of leg weaknesses when they were fed synthetic diets containing 0.87 percent calcium and 0.92 percent phosphorus from 3.6 to 11.3 kg. and 0.65 percent calcium and 0.67 percent phosphorus from 11.3 to 22.7 kg. He stated that more suitable results were obtained when the diets were modified to contain about 1.0 to 1.2 and 0.8 to 1.0 percent calcium for the 3.6 to 11.3 and 11.3 to 22.7 kg. pig, respectively.

Kellerman et al. (1943) reported satisfactory growth of weanling pigs (10.0 to 12.7 kg.) fed rations containing 0.73 percent calcium and 0.41 percent phosphorus.

Calcium to Phosphorus Ratio

The early work on calcium and phosphorus requirements emphasized the desirability of providing a favorable ratio of these two major mineral elements. The general consensus

of interpretations of research at that time was that the most desirable calcium to phosphorus ratio was 1.5:1 to 2:1. Even today the data on present calcium requirements have tended to approximate 1.5 times the phosphorus level.

A large excess of either calcium or phosphorus interferes with the absorption of the other. With an excess of either one, the other tends to become tied up as the insoluble tricalcium phosphate, which is not absorbed by the gut. This explains why it is important to have a suitable ratio between calcium and phosphorus (Cunha, 1957). When the supply of vitamin D is adequate, less favorable ratios can be tolerated.

In general, calcium to phosphorus ratios of 1.6 to 1.0 or wider adversely influence gains (Zimmerman et al., 1961, 1963). However, Lloyd et al. (1961) stated that under the condition of their experiment, a critical calcium to phosphorus ratio appeared to exist between 2 to 1 and 3 to 1. This suggested ratio is quite wide, especially when compared to the narrow ratio of 1.5 to 1.0 indicated by Bohstedt (1955). Combs et al. (1962) reported that with an adequate phosphorus level (0.44 percent) the optimum calcium to phosphorus ratio was 0.9 to 1.0

Livingstone et al. (1962) found that calcium and phosphorus from synthetic diets were retained in the ratio of 1 to 1.1.

Without stating exact ratios, it is safe to conclude that the phosphorus requirement is somewhat less than the

calcium requirement, but will depend on the dietary level of calcium and vitamin D and on the relative availability of the minerals. In more recent years much less emphasis has been placed on exact ratios in investigations on specific calcium and phosphorus requirements.

Calcium and Phosphorus Recommendations

For growth, calcium and phosphorus requirements decrease with advancing development (weight gain). The National Research Council's 1953 and 1959 calcium, phosphorus and vitamin D requirements for swine are presented in Tables I and II, respectively.

TABLE I

1953 NATIONAL RESEARCH COUNCIL'S REQUIRED LEVELS OF DIETARY CALCIUM, PHOSPHORUS AND VITAMIN D¹

Pig Weight (kg.)	Calcium(Ca) (% of diet)	Phosphorus(P) (% of diet)	Ca:P ratio	Vitamin D (I.U. per kg.)
11.3	0.80	0.60	1.33:1.0	198.4
22.7	0.65	0.45	1.44:1.0	198.4
45.4	0.65	0.45	1.44:1.0	198.4
68.0 to 113.4	0.55	0.33	1.67:1.0	198.4
Breeding stock	0.60	0.40	1.50:1.0	198.4

¹N.R.C. (1953).

TABLE II

1959 NATIONAL RESEARCH COUNCIL'S REQUIRED
LEVELS OF DIETARY CALCIUM, PHOSPHORUS
AND VITAMIN D¹

Pig Weight (kg.)	Calcium(Ca) (% of diet)	Phosphorus(P) (% of diet)	Ca:P ratio	Vitamin D (I.U. per kg)
4.5	0.70	0.60	1.17:1.0	220
11.3	0.65	0.50	1.30:1.0	198
22.7	0.65	0.50	1.30:1.0	198
45.4 to 90.7	0.50	0.40	1.25:1.0	132
Breeding stock	0.60	0.40	1.50:1.0	132

¹N.R.C. (1959).

The current National Research Council calcium, phosphorus and vitamin D recommendations for various weights of swine are presented below (Table III).

TABLE III

1964 NATIONAL RESEARCH COUNCIL'S REQUIRED
LEVELS OF DIETARY CALCIUM, PHOSPHORUS
AND VITAMIN D¹

Pig Weight (kg.)	Calcium(Ca) (% of diet)	Phosphorus(P) (% of diet)	Ca:P ratio	Vitamin D (I.U. per kg)
4.5	0.80	0.60	1.33:1.0	220
11.3	0.65	0.50	1.30:1.0	198
22.7	0.65	0.50	1.30:1.0	198
34.0 to 102.0	0.50	0.40	1.25:1.0	132
Breeding stock	0.60	0.40	1.50:1.0	220

¹N.R.C. (1964).

Although the calcium requirement has not been appreciably altered since 1953 (Table I), dietary phosphorus recommendations have tended to increase, resulting in ratios narrower in calcium to phosphorus ratios.

Calcium-Phosphorus and Disease Interrelationships

Some have suggested that SPF pigs may have higher than normal requirements for calcium and phosphorus. However, that does not seem to be the case. Seerley et al. (1963) reported SPF pigs did not require more calcium or phosphorus than current recommendations (N.R.C., 1964), and the use of higher levels gave poor feed efficiency.

The incidence of atrophic rhinitis or a condition resembling atrophic rhinitis, a respiratory disease of swine, has been linked to improper calcium and phosphorus levels by Cornell University workers (Brown et al., 1965; Brown et al., 1966; Krook et al., 1965; Pond et al., 1965).

The summary of Pond et al. (1965) is quoted "Three experiments with virus pneumonia-free Yorkshire pigs involving a total of approximately 200 animals have clearly established that atrophic rhinitis can be produced by feeding diets low in calcium or imbalanced in calcium and phosphorus. The symptoms, in addition to typical clinical signs of turbinate bone atrophy include generalized bone lesions showing osteoclastic and osteolytic resorption and replacement of osseous tissue with fibrous tissue, resorption of the tubular cancellous bone of the nasal turbinates, hypertrophy of

the parathyroid gland and reduced bone ash. Atrophic rhinitis is apparently one manifestation of generalized osteitis fibrosa. These experimentally-produced lesions are identical to those seen in field cases of atrophic rhinitis."

Sows fed low-calcium and calcium-phosphorus-imbalanced diets during gestation and lactation were not affected in terms of reproductive efficiency. They produce normal litters with normal preweaning growth rate. However, histologically, there was evidence of less osseous formation of the tubular cancellous bone in the turbinates in pigs from low-calcium sows. Post weaning weight gains were reduced severely in pigs fed low calcium-low phosphorus or low calcium-low phosphorus diets. There was no difference in weight gains among pigs fed diets containing 0.8 percent calcium and 0.6 percent phosphorus and those fed 1.2 percent calcium and 1.0 percent phosphorus, but the higher levels of calcium and phosphorus promoted higher ash content of bones and better integrity of the nasal turbinates. These workers suggest that dietary calcium be increased above the level currently recommended for growing-finishing pigs.

The beneficial effects in the control of atrophic rhinitis by an antibiotic-sulfa drug combination has been attributed to the improved calcium absorption by Pond et al. (1965).

These reports have evoked considerable argument among researchers, since normal turbinates (scroll-shaped bones in the nasal cavity) are found in herds fed lower calcium and

phosphorus levels than recommended by Brown et al. (1966), and since the antibiotic-sulfa drug combination is known to eliminate the nasal organisms associated with atrophic rhinitis.

Peo et al. (1967) stated that there did not appear to be any relationship between calcium and/or phosphorus and atrophic rhinitis in swine, within the levels studied.

Storts and Koestner (1965) reported a diet deficient in calcium and phosphorus (0.26 percent calcium and 0.14 percent phosphorus) caused severe skeletal lesions after 6 weeks when fed to 1 day old pigs. Growth rates were slow, and the first signs of trouble were noted at about 4 weeks of age. These included weakness, lack of appetite, with some pigs being unable to rise. Approximately 50 percent of the pigs had bone fractures, with many others showing abnormal bowing of the legs. Radiographic evidence of bone demineralization was pronounced. Postmortem autopsy at 6 weeks of age demonstrated that all bones were thin, soft, and pliable. Microscopic bone lesions were typical of fibrous osteodystrophy secondary to hyperparathyroidism. Bone densities were 50 percent of normal values. Correction of the deficient diet caused marked clinical improvement and restoration of normal bone density.

A second group of 15 day old pigs was fed the same basal diet, except that it was deficient only in calcium (0.26 percent calcium and 0.60 percent phosphorus). At 6 to 8 weeks of age, these pigs had radiographic evidence of de-

creased mineralization of bones. However, bone densities returned to near normal after 16 to 18 weeks of age, without a change in diet. Histologically, all pigs had mild lesions characteristic of fibrous osteodystrophy. All pigs fed either type of deficient diet had, in addition, hyperplasia of the thyroid glands. The parathyroid glands which were examined from the calcium deficient group in the 2nd trial, had evidence of mild hyperactivity. No important changes were detected in the blood serum levels of calcium, phosphorus or alkaline phosphatase.

Calcium and Phosphorus Availability

In establishing exact requirements for both calcium and phosphorus, it should be recognized that the forms in which these minerals exist in the ration may influence the efficiency of their utilization. The values presuppose that the individual elements are not present to any significant extent in forms or under conditions that are abnormal in terms of poor utilization, as may be true for phytate phosphorus or poorly utilized inorganic phosphates. According to recent observations, allowance should be made for failure of the young pig to utilize a fair portion of the diet.

The availability of the calcium and phosphorus in feeds and in inorganic supplements to these feeds has attracted considerable attention since advances in radioactive procedures have permitted differentiation between the unabsorbed fractions and the absorbed and re-excreted fractions of the

feces.

A number of experiments on the availability of calcium and of phosphorus from inorganic materials discloses that within species differences are mainly small and of doubtful significance. This generalization applies to several species and to a wide range of common mineral supplements (Chapman et al., 1955a; Creech et al., 1956; Hansard et al., 1957; Long et al., 1957; Plumlee et al., 1958; Tillman and Brethour, 1958; Bethke et al., 1930).

An exception is soft phosphate with colloidal clay, which is significantly less effective as a source of phosphorus than materials such as bonemeal or mono-, di- and tricalcium phosphates (Chapman et al., 1955a; Motzok et al., 1956; Plumlee et al., 1958; Harmon et al., 1965).

Dudley et al. (1959) and Harmon et al. (1964) obtained lower mean blood inorganic phosphorus values when soft phosphate replaced dicalcium phosphate as the only dietary phosphorus source.

Investigations (Chapman et al., 1955a; Gobble et al., 1956; Plumlee et al., 1958; Aldinger et al., 1959; Noland et al., 1964; Van Zante et al., 1967) have demonstrated the excellent usage of dietary phosphorus from dicalcium phosphate by pigs.

Combs and Wallace (1962) observed a significant reduction in dry matter and crude protein digestibility when gypsum (calcium sulfate) rather than oyster shell or ground limestone supplied the supplementary calcium.

Eggert et al. (1959) observed no difference between rations containing calcium sulfate or ground limestone as the supplemental calcium source.

Casein phosphorus is fully as available as phosphorus from inorganic phosphate sources (Bunkfeldt and Steenbock, 1943).

The relative unavailability of the phosphorus of phytate compared with the availability of phosphorus from several inorganic sources has been demonstrated (Scott et al., 1962; Green et al., 1964; McGinnis et al., 1944).

Spitzer and Phillips (1945) have shown that 58 percent of the phosphorus in soybean meal is in the form of phytin or phytate which was readily available to the rat due to the activity of intestinal phytase.

In pigs, appreciable phytate hydrolysis can occur in the stomach as a result of the presence of plant phytases of dietary origin and of the favorable pH for their action (Moore and Tyler, 1955). Considerable phytate hydrolysis can occur in the large intestine of the pig when calcium phosphate but not calcium carbonate is used. It is suggested that the higher intestinal pH induced by the latter is not conducive to bacterial hydrolysis of phytate in the large intestine.

Dietary vitamin D has been shown to be particularly essential in the utilization of phytate phosphorus (Krieger et al., 1940).

Intestinal phytase activity in the very young pigs has

not been measured. Data by Miller et al. (1965c) suggested that perhaps the intestinal phytase activity of baby pigs reared with purified diets is inadequate to digest the phytate present in isolated soy protein, thus failing to make available for absorption the phosphorus of phytate as well as the cations which it effectively binds (calcium and magnesium).

Calcium and Phosphorus Absorption and Retention

Thompson (1964) has presented a simplified flow scheme for mineral absorption and excretion connected with the intestinal tract. The amounts of calcium and phosphorus which can be absorbed from the gastrointestinal tract are limited by the solubility of the salts; the absorption of these substances is then influenced by the ratio of dietary calcium and phosphorus, as well as by other factors such as the amount or kind of carbohydrate, vitamin D, and other nutrients present.

Lengemann et al. (1957) demonstrated the marked effect of milk in increasing calcium absorption in rats and cattle but observed no such effect in rabbits.

In seeking an explanation of the enhancement of calcium absorption by milk, Wasserman et al. (1956) confirmed the influence of lactose and also showed the L-lysine and L-arginine markedly stimulated the absorption of calcium⁴⁵ in the rat.

Subsequently, it was demonstrated that in the normal

rat the effects of L-lysine and lactose were additive and in the rachitic rat the effects of L-lysine and vitamin D were also additive (Wasserman et al., 1957). In contrast, neither arginine, lysine or skim milk increased calcium⁴⁵ absorption in the rachitic chick, although treatment with vitamin D promoted nearly complete absorption.

The increased calcium absorption is the major physiological action of vitamin D as shown further by the isotopic studies with chicks (Keane et al., 1956) and with calves (Conrad and Hansard, 1957). The latter workers demonstrated that massive doses of vitamin D not only increased absorption but decreased endogenous fecal calcium losses and promoted deposition of radiocalcium in areas of new bone growth.

Kline et al. (1932) reported that calcium absorption was enhanced in chicks by an acid pH in the intestine and that vitamin D or lactose increased acidity throughout the intestine, whereas ultraviolet irradiation of the chick increased acidity only in the proximal portion of the intestine.

Ali and Evans (1966) stated that, except in the cecum, lactose increased both relative and absolute absorption of calcium by rate. These workers stated that the distal half of the small intestine was the main site of calcium absorption.

The mechanism of impaired calcium utilization in the absence of vitamin D is not presently known, but several theories have been offered (Dowdle et al., 1960; Migicovsky

and Jamieson, 1955; Nicolaysen and Eeg-Larson, 1953).

Dowdle et al. (1960) reported that the active transfer of calcium from mucosal to serosal surfaces was enhanced by dietary vitamin D as well as by ultraviolet irradiation of the animal.

Taylor and Wasserman (1965) demonstrated a calcium binding capacity in the intestinal mucosa of rachitic chicks which was associated with the protein or polypeptide fractions of the tissue homogenates. The capacity to bind calcium was dependent upon vitamin D₃. In the live chick the effect of vitamin D₃ on calcium absorption was demonstrated in 5 to 10 hours.

Wasserman and Taylor (1966) reported a good correlation between the concentration of the vitamin D₃-induced calcium-binding factor and the rate of calcium absorption.

Wasserman et al. (1966) observed that administration of vitamin D to chicks and rats significantly increased the intestinal transfer of calcium⁴⁷ from the intestinal lumen to blood plasma, and plasma to lumen. The influx effect was not due to transmural electropotential differences resulting from vitamin D administration or to differences in plasma calcium concentrations. The major direct effect of vitamin D probably is not on a unidirectionally-oriented calcium transport system, but rather on an increased diffusional permeability of the intestinal membrane. The latter is perhaps manifested as change in membrane structure and/or an effect on calcium carrier synthesis.

Norman (1966) concluded that vitamin D was necessary to induce synthesis of appropriate enzyme systems or to cause the alteration of membrane structure necessary for calcium absorption. A turn-over time of 24 to 36 hours was suggested for the biochemical machinery associated with vitamin D₃-mediated calcium absorption.

Miller et al. (1965b) reported decreased calcium retention when pigs were not given supplemental vitamin D.

Combs et al. (1966b) showed in one instance that negative calcium digestion coefficients were obtained in the absence of supplemental vitamin D, and in another trial calcium digestibility was similar for the vitamin D supplemented and unsupplemented groups.

Whiting and Bezeau (1958) observed that the apparent absorption of calcium was not influenced by the presence of dietary vitamin D. These investigators also reported a significant vitamin D X dietary zinc interaction and postulated that this interaction could partially explain why supplemental vitamin D had increased calcium absorption in some instances and not in others.

The contrasting results regarding the influence of vitamin D on calcium digestion may be partially explained by the multiplicity of functions reported for vitamin D. Wasserman (1962) found that vitamin D increased the absorption of magnesium, barium, cobalt and strontium, as well as calcium. Worker and Migicovsky (1961) observed that zinc absorption was enhanced by vitamin D. These varied roles

coupled with the complexity and interactions of mineral elements have undoubtedly contributed to the diversity found in the literature regarding the need for vitamin D.

Digestion and metabolism experiments were conducted by Whiting and Bezeau (1958) using pigs of various weights (9.1, 15.9, 24.9 and 36.3 kg.) to determine the effects of supplementing a ration with calcium, zinc and vitamin D on the apparent absorption and retention of calcium and phosphorus. The levels of calcium, zinc and vitamin D in the rations were 0.40 and 0.86 percent, 34 and 140 ppm, and 0 and 1,764 I.U. per kg., respectively. The apparent absorption and retention of calcium were not influenced by the addition of calcium or vitamin D, but were increased by the addition of zinc. The apparent absorption of phosphorus was decreased by the addition of calcium or vitamin D. The retention of the phosphorus consumed was not affected by calcium, zinc or vitamin D.

Combs et al. (1966b) found that phosphorus digestion was not significantly influenced by the presence or absence of supplemental vitamin D or by ultraviolet irradiation. Similar results were obtained with respect to vitamin D (Combs et al., 1966a), whereas an increase (Miller et al., 1965b) and a decrease (Whiting and Bezeau, 1958) in phosphorus absorption have been found with dietary vitamin D supplements.

Combs et al. (1966b) stated that calcium did not significantly affect apparent phosphorus digestibility or per-

cent bone ash.

The influence of tartaric and citric acids in increasing calcium absorption in both normal and rachitic rats has been cited (Underwood, 1959).

Dudley et al. (1961) fed baby pigs a purified ration from 2 to 7 or 8 weeks of age and reported that the nature of the dietary carbohydrate (glucose or lactose) did not influence the occurrence of deficiency symptoms or influence the calcium requirement. However, in one trial the femur ash at all levels of calcium was higher when lactose was the carbohydrate source.

Restriction of calcium intake increased the efficiency of calcium absorption and retention by the rat (Hansard and Plumlee, 1954; Marz, 1962; Evans and Ali, 1966). Similar results have been reported in the pig (Walker and Jones, 1963; Newman et al., 1967). The latter workers observed that although percent calcium absorption was increased on low calcium diets, the absolute amounts absorbed were greater at the higher levels of calcium intake.

Hurwitz and Bar (1965), in a radioactive tracer study with hens, reported that percentage calcium absorption was not influenced significantly by its dietary level. Percentage absorption of phosphorus, however, was depressed by an increased dietary calcium level.

Hansard et al. (1961), using radio-chemical procedures, studied the absorption, excretion and utilization of calcium by 42 pigs at different ages. Calcium absorption and reten-

tion was greatest in pigs 15 days of age and decreased rapidly to 5 months. Daily endogenous fecal calcium losses and the subsequent requirements for maintenance increased with animal age to 5 months.

Research by Hendricks et al. (1967) revealed that less calcium and less phosphorus were absorbed and retained by baby pigs fed isolated soybean protein than pigs receiving casein.

A report by Combs and Wallace (1962) showed that the digestibility of protein and ether extract was significantly reduced by a high dietary calcium level in one trial but not in another.

Studies by Hart and Steenbock (1913) with adolescent pigs demonstrated that the addition of magnesium salts to a basal bran diet resulted in increased urinary calcium elimination and a negative calcium balance. However, data obtained with the rat (Forbes, 1963) and with the baby pig (Miller et al., 1965d) have demonstrated that increasing the level of dietary magnesium did not significantly affect daily calcium or phosphorus retention.

The relationship of dietary fat to calcium and phosphorus utilization has been reported for the rat (Calverley and Kennedy, 1949; Cheng et al., 1949; Haldi et al., 1939 and French, 1942). High dietary levels of fat have increased fecal calcium in monogastric animals through the formation of insoluble soaps (Boyd et al., 1932 and Fedde et al., 1960).

Newman et al. (1964a), using 16 weanling pigs, fed corn-soybean meal rations containing 0.2 percent and 0.8 percent calcium with and without 10 percent stabilized tallow. Level of calcium had no effect on carcass traits or on digestibility of dry matter, energy, protein or nitrogen-free extract. Digestion coefficients were greater for calcium, phosphorus, TDN and fat at the lower calcium level. Their data indicate an inhibition of fat digestion by calcium while calcium digestibility was not materially affected by added fat.

Newman et al. (1967) reported that 10 percent added tallow had no consistent effect on calcium digestibility, although apparent digestibility of phosphorus was significantly decreased by tallow and by increased calcium levels.

Excess Calcium and Phosphorus Levels

The influence of a high dietary calcium level on zinc utilization has been the subject of many reports. Ellis (1953) demonstrated that swine parakeratosis was aggravated by high levels of calcium and phosphorus. The ability of supplemental zinc to prevent or cure parakeratosis has been established (Kernkamp and Ferrin, 1953).

From the investigations on parakeratosis, it appears that: (1) signs of zinc deficiency may arise in growing pigs fed rations containing 30 to 40 ppm zinc, or less; (2) these signs can be overcome or prevented by zinc supplementation at the rate of 40 to 100 ppm and partially overcome by

smaller amounts; (3) severity of the symptoms increases as the ration calcium level increases.

The suggestion of Lewis et al. (1957a,b) that excess dietary calcium reduced zinc absorption through binding by calcium phosphates in the intestine is not supported by the work of Beardsley (1958). This investigator found that calcium reduced zinc retention in the baby pig primarily by increasing its elimination in the urine.

The early studies of parakeratosis showed that calcium aggravated the syndrom and led to the concept that calcium is an antagonist to zinc. This generalization is not valid. It has been demonstrated in pigs (Oberleas et al., 1962), chicks (O'Dell et al., 1964) and rats (Likuski and Forbes, 1965; Oberleas et al., 1966) that when the diet contained phytate, excess calcium accentuated zinc deficiency symptoms, but not in the absence of phytate.

In vitro studies by Oberleas et al. (1966) have shown that zinc phytate is highly insoluble at the pH encountered in the small intestine and the presence of calcium ions produces an even more insoluble complex composed of calcium-zinc-phytate. It is believed that the formation of this highly insoluble complex in the intestinal tract accounts for the low availability of zinc when both calcium and phytate are present in the ration.

In addition to the increased incidence of parakeratosis associated with high calcium rations (Lewis et al., 1956; Luecke et al., 1957; Roberts et al., 1961; Hoefer et al.,

1959; Tucker and Salmon, 1955; Valee, 1959; Pond and Jones, 1964) excessive calcium in the rations of young pigs has impaired both rate and efficiency of gain (Combs et al., 1960; Stephens et al., 1964; Conrad and Beeson, 1957).

Combs et al. (1966b) obtained decreasing rates of gain with increasing levels of dietary calcium. In contrast, Foster et al. (1964) reported that a variation in calcium and phosphorus levels did not affect rate of gain of growing-finishing pigs. Virginia workers (Carter et al., 1959) noted a depression in average daily gains of pigs weighing 11.3 to 88.4 kg. when dietary calcium was increased from 0.60 to 1.25 percent. Phosphorus was maintained at 0.40 percent.

Eggert et al. (1959) and Zimmerman et al. (1963) showed the depressing effect of high calcium intakes on blood serum chlortetracycline levels of pigs. Harms and Waldroup (1961) found that calcium interfered with the utilization of tetracycline antibiotics by the chick.

Excessively high calcium intakes as well as low (under 0.8 percent) ration calcium levels have been postulated to be causitive or contributing factors involved in tail-biting in finishing pigs (Gadd, 1967).

Miller et al. (1959) observed that increasing the dietary calcium level from 0.60 to 1.25 percent had no significant effect on rib, muscle or blood copper concentration. The addition of calcium decreased the copper, iron, manganese, molybdenum and zinc content of both the liver and

kidney.

High levels of dietary calcium and phosphorus markedly affect magnesium requirements. O'Dell et al. (1958) observed that high intakes of phosphorus accentuated magnesium deficiency symptoms in guinea pigs and rats as much or more than high levels of calcium. There was a derangement of the calcification process, expressed as widespread tissue calcification and failure of teeth and bones to calcify normally.

Experiments dealing with the general problem of the influence of high intakes of calcium have been reviewed by Davis (1959) and Whedon (1959).

Summary

Reported studies have shown calcium and phosphorus requirements to vary from 0.30 to 1.20 percent and from 0.40 to 0.90 percent, respectively, with the reported requirement differences largely dependent upon the response criteria selected. However, it appears that a calcium level of 0.8 percent and a phosphorus level of 0.6 percent is adequate to assure maximum performance and adequate skeletal development of baby pigs. However, optimal skeletal development, as measured by bone density, ash content and breaking strength, occurs in pigs consuming 1.0 percent of calcium and 0.7 percent of phosphorus.

MATERIALS AND METHODS

General Procedure

In a preliminary experiment it was extremely difficult to keep all of the colostrum-deprived pigs alive long enough to measure dietary effects. For example, it was not unusual to lose approximately one-half of the baby pigs. Therefore, the results reported here were obtained from pigs which received pasteurized sow colostrum during their first 24 hours of life and an antibiotic in their feed throughout the experimental period.

One trial was conducted using 59 purebred Yorkshire baby pigs (27 males and 32 females) and 20 purebred Hampshire pigs (11 males and 9 females). These pigs were maintained under Specific-Pathogen-Free (SPF) conditions, thus minimizing the confounding effects of certain environmental conditions on the response of the pigs to the dietary treatments.

In brief, the pigs were obtained by allowing gravid gilts and sows to give birth under aseptic conditions. The pigs were collected in sterile plastic bags as they emerged from the birth canal and were then placed in a pre-sterilized plastic isolator. They were transported to the Swine Nutrition Laboratory, fed pasteurized sow colostrum, placed in

individual cardboard rearing units and fed a common liquid diet. At 2 weeks of age they were moved to individual, open-topped, solid-sided metal pens and assigned to four experimental calcium-phosphorus purified diets. The period from 14 to 21 days of age was used to adjust the pigs to their respective rations. The experimental period was 42 days (21 to 63 days of age). Individual weekly feed consumption was recorded and live body weights were taken at weekly intervals. Blood samples were obtained when the pigs were 21, 42 and 63 days of age. Six pigs from three rations and 7 pigs from one ration were sacrificed and various response measurements were obtained.

Animal Rooms and Equipment

The Swine Nutrition Laboratory consisted of three separate temperature controlled rearing rooms, the incubator room, nursery room and grower room, where the pigs were housed from birth to 2, 2 to 6 and 6 to 9 weeks of age, respectively.

Incubator Room. The incubator facilities consisted of one room 6.1 X 3.05 X 2.68 m., and an anteroom 3.05 X 3.05 X 2.68 m., for changing clothes and preparing the liquid diet (Table V). Forty individual disposable cardboard incubators¹ were used to hold the pigs during the first 2 weeks of life (Figure 1). The incubators were designed to provide each baby pig with dry, heated, filtered and sterilized air.

¹Fort Dodge Container Corporation, Fort Dodge, Iowa.

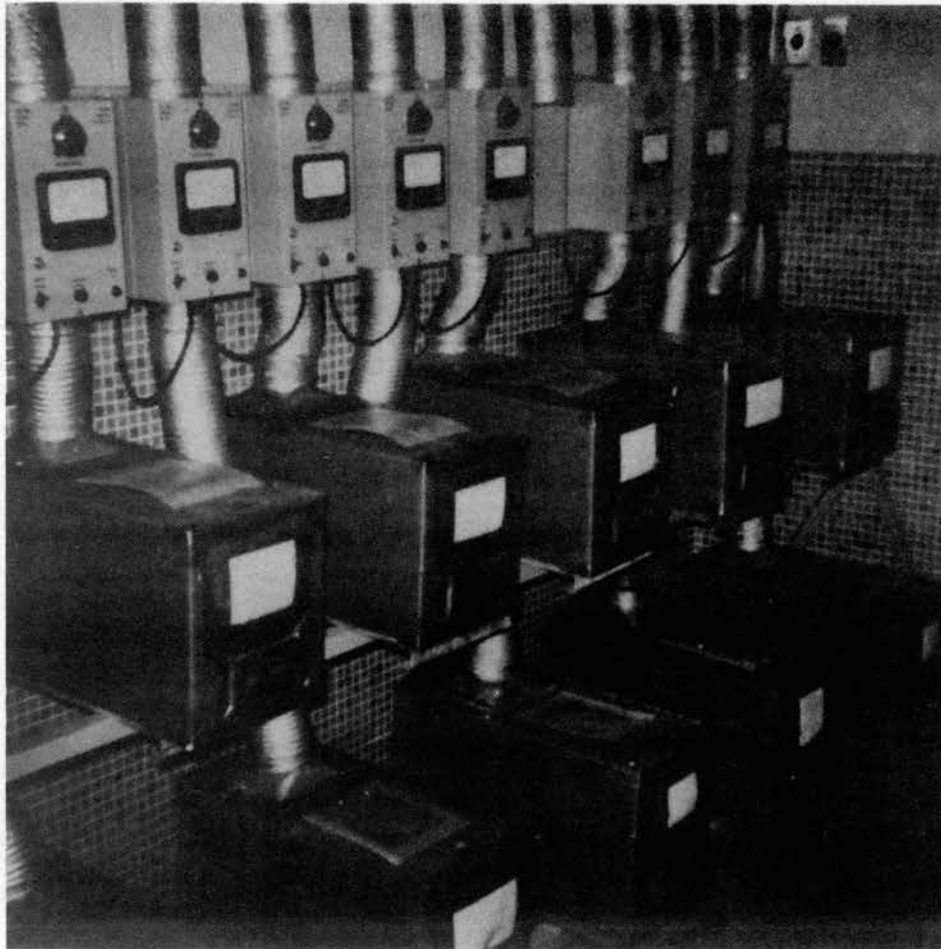


Figure 1. View of the Incubator Room with the Individual Cardboard Incubators in Place

Inside dimensions were 54.61 X 26.67 X 21.12 cm. Each had an observation port on top, an opening for a feed tray in front and was connected to an overhead air input supply system by plastic tubes. The input air was sterilized² and dehumidified², forced over thermostatically controlled heaters, taken through three or four layers of cotton filters which covered four square openings (11.05 X 11.05 cm.) in the top of the incubator and passed out of the incubator and into the room at the rate of 141.58 dm³ per minute. This air was exhausted from the building and was not recycled through the incubators. The air outflow port of the incubators located in the front of the boxes consisted of a series of 25 holes (17.78 mm. diameter) in a five X five arrangement and covered with 3 or 4 layers of cotton.

The interior of each incubator was kept at a positive air pressure. This minimized the possibility of air-borne contamination.

A grill of 6.35 mm. wire mesh raised 53.34 mm. above the bottom of the incubator kept the piglet free of its urine and feces. Each incubator was quilon coated on the inside liner to help condition the cardboard to withstand moisture.

A closely fitted metal feeding tray was taped into place in such a way that it could not be overturned. This tray measured 14.73 X 10.16 X 4.57 cm.

²Kathabar, Surface Combustion Division, Midland-Ross Corporation, Toledo, Ohio.

Each incubator had a 17.78 X 12.70 cm. observation opening in the top which was covered with a plexi-glass material after the pig was placed inside to prevent the piglet from jumping out.

Temperature within the incubator room itself was maintained at 26°C. Control of the temperature within the incubator boxes was of vital importance. Provision of an environmental situation that reduces heat loss and provides an immediately available supply of dietary energy is critical to the survival of newborn pigs. This was achieved by putting a 100°C. thermometer into each rearing chamber. Each heater was thermostatically controlled so that the temperature in the box could be adjusted to any required level between 25° and 70°C.

Feed trays and wire mesh bottoms were steam sterilized (121°C. for 30 minutes) before being placed within the incubators. The units were allowed to stand at least 30 minutes before air flow was started through the filters. The cotton air filters were dipped in 1.0 percent mercuric chloride, then dried. Thus, if they became wet, the germicide would be activated and kill vegetative bacteria.

The use of the individual cardboard incubators for control of physical factors such as temperature, humidity, air-flow, sound and light. The methods used in handling the animals were designed to provide isolation from each other, from other pigs, from the caretakers and to keep immediate surroundings as clean as possible. Meticulous attention to

detail was essential. The personnel who cared for the pigs changed clothes in the anteroom, putting on clean overalls and rubber boots. They also stepped into a disinfectant¹ pan when entering and leaving the incubator room.

Nursery Room. The nursery room was 9.37 X 3.05 X 3.05 m. and contained 36 solid-sided open-topped pens. Each pen had a perforated, reinforced, galvanized expanded metal floor, was 76.2 X 45.7 X 58.4 cm. in size and was equipped with an adjustable self-feeder and an automatic watering device. The feeder was fitted with a heavy lip around the inside to minimize feed loss. A waste feed tray (34.92 X 25.40 X 13.97 cm.) was located under the self feeder.

Grower Room. The grower room contained 25 pens similar to those described for the nursery. Each pen was 11.76 X 59.69 X 56.69 cm. in dimensions and provided adequate space for each pig up to 9 weeks of age. Figure 2 shows a pig in one of these pens.

All Rooms. A room adjacent to the nursery provided space for storage of the purified ration ingredients, ration preparation and mixing, and some ration storage.

All laboratory rearing rooms had concrete floors and the incubator and nursery rooms contained no windows, thus eliminating the entrance of sunlight.

The air conditioning of each room was controlled individually and conditions could be varied as desired. Venti-

¹Nolvasan-S, Fort Dodge Laboratories, Fort Dodge, Iowa.



Front View



Rear View

Figure 2. Two Views of a Nine Week Old Pig in a Grower Room Pen

lation is required to remove the moisture produced by the animals as well as to control temperature and odors. Normally, the temperature in all rooms was maintained at 26°C. and 35 percent relative humidity. To avoid contamination, the pressure in all of the rooms was maintained slightly higher than the environmental pressure so that air swept out of the rooms when doors were opened.

The rooms were prepared for the pigs a few days before the pigs were to enter the facilities. The rooms and pens were aseptically cleaned with a detergent and disinfectant¹ and thoroughly steamed. Then all the equipment necessary to last throughout the experiment was put in place, i.e., incubators were assembled and placed in the incubator room. The rooms were sealed, dampers in the air conditioning system were closed, relative humidity increased by allowing steam to flow into the rooms, and the rooms fumigated with formaldehyde gas.² After 12 hours the dampers were opened and the rooms aerated for at least 8 hours before use.

Ultraviolet germicidal lamps were used as aids in cleaning the air in the incubator and nursery rooms. These were located on the wall approximately 50.8 cm. down from the ceiling and were complete with a shield to protect the pigs from any direct ultraviolet rays. All lamps were cleaned each week by wiping with an alcohol solution.

¹Klenzade XY-12, Klenzade Products, Beloit, Wisconsin.

²Formaldegen, Vineland Poultry Laboratories, Vineland, New Jersey.

Pig Isolator. The rigid molded acrylic isolator¹ used for collection and transport of the baby pigs was similar to the one in Figure 3.

The complete isolator consisted of the basic housing which was in two sections. Both top and bottom sections were molded of 10.28 mm. clear acrylic with radiused crevice-free edges and corners for easy cleaning and decontamination. The flat working area was 121.92 cm. X 76.2 cm. The overall outside dimensions were 137.2 cm. X 91.44 cm. The top and bottom were both 38.1 cm. high and hinged so that either side could be opened or the top completely removed.

Shoulder length (81.29 cm.) black neoprene gloves were attached to the isolator by means of three complete turns of plastic film tape and a clamp so arranged that only neoprene was exposed to the interior of the isolator.

Both the air input and the output ports were fitted with four layers of Owens-Corning Fiberglas PF 115 filter material, and the plastic structure was maintained under a positive air pressure to reduce or eliminate the migration of contaminants through any leaks in the wall.

A 1/30 horsepower, 3,020 rpm, 140 cfm., detachable electric centrifugal blower motor² forced fresh air into the unit through the four layers of filter material, and the exhaust air passed through a similar filter arrangement which

¹The Germfree Laboratories, Incorporated, Miami, Florida.

²Model 20610, Dayton Electric Mfg. Co., Chicago, Illinois.

prevented the back flow of air into sterile environment, which could occur during a sudden withdrawal from the gloves.

The air filter units (input and output) were sterilized in an autoclave at 150°C. for 90 minutes and attached to the isolator. The interiors of the isolator were cleaned with copious detergent, dried, sprayed with a 1.0 percent mercuric chloride solution and allowed to dry. This was followed by formaldehyde gas¹ sterilization.

Three ordinary burlap feed sacks were sterilized by autoclaving and placed on the isolator floor to permit firm footing for the pigs.

Leaks were detected by swabbing the suspect area with a soap or detergent solution while the isolator was under air pressure. The air pressure had to overcome the capillary force of the solution in the leak in order to form a leak. Leakage at the inner door of the sterile lock was often found. It is granted that the pressure that was used in the isolator may not have been great enough to detect minute leaks, thus complete tightness may not have been achieved. No effective protection could be provided against pinholes that may have developed while the isolator was in use.

Isolator space was quite adequate for a large litter. Good visibility and ease of transportation were also definite advantages of the isolator. Two distinct disadvantages

¹Formaldegen, Vineland Poultry Laboratories, Vineland New Jersey.

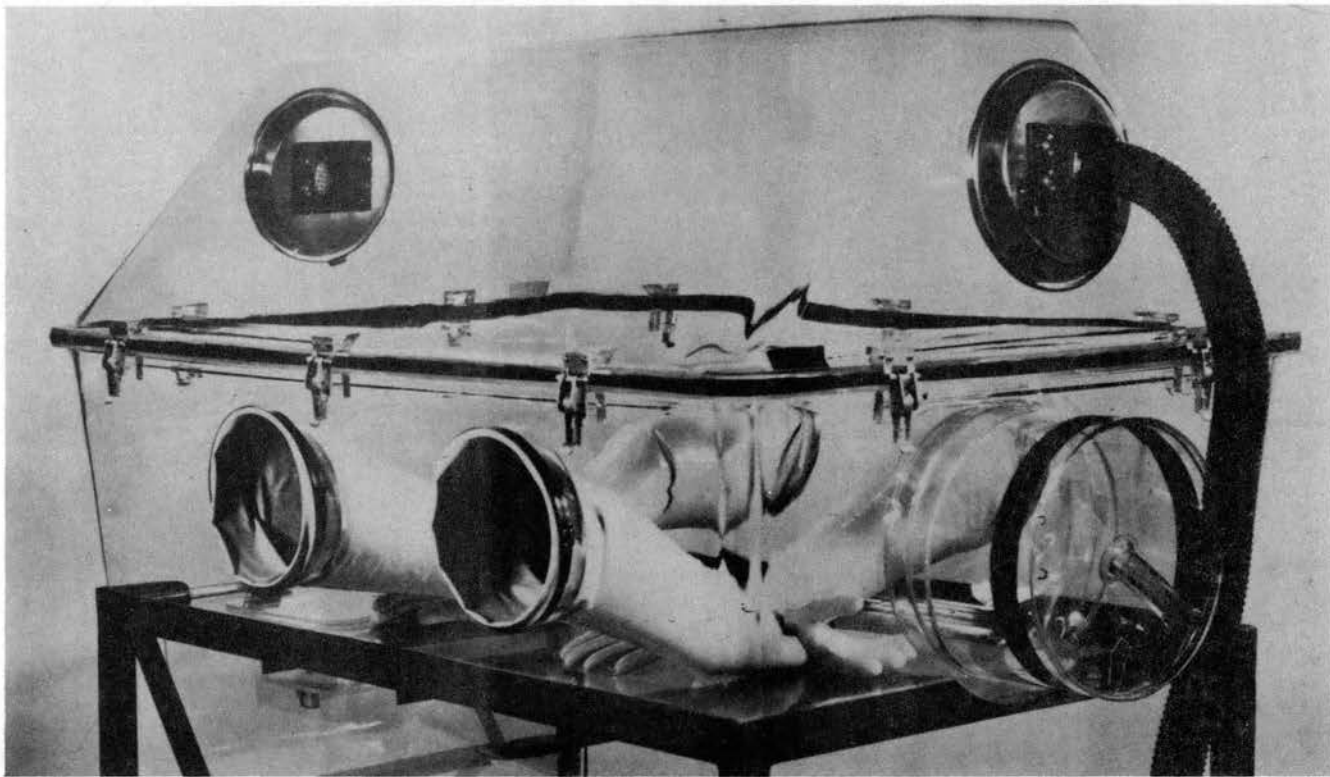


Figure 3. A Rigid Molded Acrylic Isolator Similar to the One
Used in this Study

were the lack of a germicide trap and lack of an air heater.

Pig Diets

Sow Colostrum. Approximately 200 ml. of sow colostrum (Table IV) were given each pig during the first 24 hours of life. This colostrum was obtained just prior to or during parturition or within 12 hours after onset of parturition. Since the sows were confined in farrowing crates, 25 to 50 ml. of colostrum were obtained without great difficulty and without injected oxytocin. However, to obtain larger volumes, the sow was restrained by placing a running noose on the upper jaw, and approximately 20 U.S.P. units of pituitary oxytocic principle¹ were injected into an ear vein to accomplish colostrum let-down. After a few seconds, colostrum was obtained from several teats by manual extrusion. Up to 900 ml. were obtained from a single milking. The colostrum from several sows was pooled, pasteurized (61.7-62.8°C., 30 minutes), cooled and frozen at -17°C. until needed.

Liquid Milk Diet. The liquid diet fed the pigs from birth to 2 weeks of age consisted of pasteurized, homogenized cows' milk fortified with dried whole milk, dried whey, minerals and vitamins (Table V). Whole milk and whey were added in sufficient quantity to approximately double the dry matter content of the cows' milk. The liquid diet was formulated to resemble sow's milk rather than cows' milk in

¹P.O.P., Armour Pharmaceutical Company, Kankakee, Illinois.

TABLE IV
 ANALYZED CHEMICAL COMPOSITION
 OF POOLED SOW COLOSTRUM

Nutrient	Liquid ^a	Dry
	%	%
Total solids	23.44	--
Crude protein (N X 6.38)	13.63	58.15
Fat	7.80	33.28
Solids-not-fat	15.64	66.72
Nitrogen-free extract	1.20	5.12
Ash	0.814	3.47
Calcium	0.118	0.50
Phosphorus	0.134	0.57

^aAll mean values are based on six determinations.

TABLE V
COMPOSITION OF THE FORTIFIED
COWS' MILK DIET

Ingredient	Composition
Homogenized cow milk	1 gal. (3,785.3 ml.)
Dried whole milk	515.87 gm.
Dried whey (50% lactose)	260.04 gm.
Mineral mixture ^a	20.00 ml.
Sulfamethazine-antibiotic ^b	1.50 gm.
Citric acid	5.00 gm.
Niacin	16.67 mg.
Vitamin K	3.039 mg.
Vitamin D ₂	100 I.U.
Vitamin E	10.7 mg.

^a21.47 gm. FeSO₄ · 7H₂O

15.19 gm. ZnSO₄ · 7H₂O

9.27 gm. MnCl₂ · 4H₂O

18.07 mg. KI

With 2.5 ml. of concentrated HCl, diluted with distilled water to 1 liter volume.

^bAureo S-P 250, a trademark for a premix of chlortetracycline (44.1 gm. per kg.), sulfamethazine (4.4%) and penicillin (22.05 gm. per kg.), American Cyanamid Company, Princeton, New Jersey.

order to provide more energy per unit volume. The analyzed and calculated chemical composition of the diet is presented in Tables VI and VII, respectively. On a dry matter basis, calcium and phosphorus content was 1.319 and 0.983 percent, respectively. Antibiotic material was added at a level to supply 132.3 mg. of active antibiotic per kg. of dry matter.

The liquid diet was prepared in the following manner: One and one-half l. of cows' milk were poured into a 2 liter Waring Blender, the remaining ingredients were added in appropriate quantities (Table V) and the resultant mixture was homogenized by constant stirring at low speed (15,500 rpm) for approximately 5 minutes.

The remaining 2.29 l. of milk were added and the diet was blended for an additional 5 minutes. Although foaming of the mixture occurred, the foam disappeared on cooling or on warming while stirring. It was not considered necessary to homogenize the final preparation since the diet formed a rather stable emulsion upon mixing.

The liquid diet was placed in a pasteurization unit¹ and heated at a temperature of 61.7°C., for 30 minutes. This heat process assured a negative phosphatase test which is the officially recognized test for satisfactory pasteurization. It was then rapidly cooled, transferred to sterile containers and either maintained at 2°C. until fed or frozen

¹Home Health milk and cream pasteurizer, Model PA-52A, two gallon size, Waters Conley Company, Inc., Rochester, Minnesota.

TABLE VI
 ANALYZED CHEMICAL COMPOSITION OF THE
 FORTIFIED COWS' MILK DIET

Nutrient	Liquid ^a	Dry
	%	%
Total milk solids	25.02	--
Crude protein (N X 6.38)	6.23	24.90
Fat	4.90	19.58
Solids-not-fat	20.12	80.42
Nitrogen-free extract	11.86	47.40
Ash	2.03	8.11
Calcium	0.330	1.319
Phosphorus	0.246	0.983

^aAll mean values are based on six determinations.

TABLE VII
CALCULATED NUTRIENT COMPOSITION OF THE
FORTIFIED COWS' MILK DIET

Nutrient	Quantity ^a
Energy, kcal.	
Digestible energy	4,971.20
Metabolizable energy	4,083.70
Minerals, mg.	
Magnesium	880.39
Iron	195.38
Zinc	55.56
Manganese	44.54
Copper	11.31
Iodine	0.22
Vitamins ^b	
Vitamin A	9,513.20
Vitamin D	230.20
Vitamin E	8.60
Vitamin K	2.45
Choline	2,988.33
Inositol	621.79
Pantothenic acid	26.86
Niacin	24.50
Riboflavin	12.99
Pyridoxine	6.44
Thiamine	3.77
Folic acid	0.34
Biotin	0.33
B ₁₂	0.33

^aAll quantities are expressed as amount per kg. of diet dry matter.

^bVitamins A and D are stated in International Units (I.U.) while all other vitamins are expressed in mg.

at -17°C . until needed. The frozen diet was thawed by permitting it to stand at 20°C ., followed by heating and shaking just prior to dispensing to the piglets.

Since the diet was not subjected to the high temperature of steam sterilization, heat labile vitamins were not supplemented.

Feedings were at 4 hour intervals, starting at 6 a.m. and ending at 10 p.m. Although a less frequent feeding schedule would have reduced labor and time required to feed the pigs, it was felt that five times a day feeding would restrict the quantity of diet consumed at a given time and accomplish a greater total daily dietary intake without complicating management because overfeeding diarrhea would be absent or at least minimal. With this regimen scouring was a minor problem and was usually controlled by limiting the dietary intake of the pig until the diarrhea was alleviated. When diarrhea was severe, a single oral treatment with neomycin sulfate¹ gave excellent results.

The fortified milk diet was heated to approximately 38°C . during the initial 14 days of the test by placing it in a water bath maintained at 53°C ; thereafter, it was fed at room temperature.

Tray feeding was initiated approximately 4 to 6 hours after the final pigs had been collected. The pigs started with a quantity of 20 to 25 ml. of diet per feeding. The

¹Liquid Biosol-M, The Upjohn Company, Kalamazoo, Michigan.

volume of diet allotted to each pig was determined on an individual basis. This was estimated at each feeding by considering the condition of the animal and how well its previous feeding was consumed. By this technique, the feedings of the pigs appeared to be essentially on an ad libitum basis. The pigs obtained their water requirements from the liquid diet that was used. The quantity of diet was increased approximately 5 ml. at successive 6 a.m. feedings except for individual pigs that did not consume the previous feeding. Normally, pigs were consuming approximately 280 ml. per feeding at 14 days of age. Dietary consumption by individual pigs was accurately measured and recorded. General observations were made at the time of feeding. No water or dry feed was available to the pigs during this period.

The pigs were fed by an operator who wore a freshly laundered cap, gown, mask, and gloves which had been soaking in a disinfectant solution.

Calcium-Phosphorus Purified Rations. The physical compositions of the calcium-phosphorus purified diets are presented in Table VIII and the proximate analysis and analyzed calcium and phosphorus levels for each diet are presented in Table IX. On the basis of the literature, two dietary calcium-phosphorus levels (Diets A and B) below the current recommendation (N.R.C., 1964) were chosen. Diet C was formulated to meet current requirements (N.R.C., 1964), while Diet D provided a higher calcium and phosphorus

TABLE VIII
PERCENTAGE COMPOSITION OF EXPERIMENTAL
PURIFIED DIETS

Ingredient ^a	Diet A	Diet B	Diet C	Diet D
	%	%	%	%
Casein ^b	25.58	25.58	25.58	25.58
Corn starch ^c	46.15	45.41	44.66	43.92
Glucose monohydrate ^d	11.30	11.30	11.30	11.30
α-Cellulose ^e	6.00	6.00	6.00	6.00
Corn oil ^f	5.30	5.30	5.30	5.30
Mineral mixture ^g	3.00	3.00	3.00	3.00
Dicalcium phosphate ^h	1.1785	2.0116	2.8450	3.6783
Calcium carbonate ⁱ	0.3403	0.2528	0.1650	0.0773
Water with water soluble vitamins ^j	1.00	1.00	1.00	1.00
Fat soluble vitamins ^k	0.0041	0.0041	0.0041	0.0041
Antioxidant ^l	0.0125	0.0125	0.0125	0.0125
Sulfamethazine-antibiotic ^m	0.125	0.125	0.125	0.125
Total	99.99	99.996	99.992	99.997
Kcal. per kg. feed (calculated) ⁿ	3,450.8	3,422.2	3,391.3	3,362.6

^aIngredients are expressed on an air dry feed basis.

^bBorden's New Zealand Lactic Casein, 83.9% crude protein by analysis, The Borden Company, New York, New York.

^cCorn Products Company, Argo, Illinois.

^dCerelose 2001, Corn Products Company, Argo, Illinois.

^eSolka-Floc, BW-100, Brown Company, Berlin, New Hampshire.

^fMazola, Corn Products Company, Argo, Illinois.

^gSee Table X. Supplied 50 ppm of supplemental zinc as well as other minerals.

^hContained 21.62% elemental calcium and 18.34% elemental phosphorus by analysis.

ⁱContained 39.89% elemental calcium by analysis. Courtesy Calcium Carbonate Company, Quincy, Illinois.

^jSupplied 3 mg. thiamine, 6 mg. riboflavin, 40 mg. niacin, 30 mg. pantothenic acid, 2 mg. pyridoxine, 13 mg. para-aminobenzoic acid, 80 mg. ascorbic acid, 130 mg. inositol, 1.3 gm. choline, 260 mcg. folic acid, 50 mcg. biotin and 100 mcg. cyanocobalamin per kg. of total ration. Courtesy Hoffman-Taff, Inc., Springfield, Missouri.

^kSupplied 10 mg. alpha-tocopherol, 1.5 mg. vitamin A, 40 mcg. 2 methyl. 1,4 naphthoquinone and 12.5 mcg. of vitamin D₂ per kg. of total ration.

^lSantoquin liquid, 1,2 dihydro-6-ethoxy,2,2,4, trimethyl quinoline, Monsanto Chemical Company, St. Louis, Missouri.

^mAureo S·P 250, a trademark for a premix of chlortetracycline (44.1 gm. per kg.), sulfamethazine (4.4%) and penicillin (22.05 gm. per kg.), American Cyanamid Company, Princeton, New Jersey.

ⁿMetabolizable energy values of Diggs *et al.* (1965).

TABLE IX
 CHEMICAL COMPOSITION OF THE PURIFIED DIETS
 FED FROM THREE TO NINE WEEKS OF AGE

Analysis	Diet A	Diet B	Diet C	Diet D
Proximate analysis, %				
Dry matter	89.78	89.84	90.12	90.10
Crude protein (N X 6.25)	21.60	21.43	21.55	21.62
Ether extract	5.90	5.81	5.75	5.85
Crude fiber	3.39	3.00	3.64	3.14
Nitrogen-free extract	55.58	55.70	54.72	54.48
Ash	3.31	3.90	4.46	5.01
Minerals, %				
Calcium	0.368	0.569	0.776	0.952
Phosphorus	0.279	0.443	0.610	0.729
Phosphorus to calcium ratio	1:1.32	1:1.28	1:1.27	1:1.31

level. The calcium and phosphorus levels in this study were selected so as to maintain a constant calcium to phosphorus ratio of 1.30:1.

The protein, cellulose, fat, calcium-phosphorus free basal mineral mixture (Table X) and vitamin portions of the diet were held constant in all rations and only the quantity of cornstarch, calcium carbonate and dicalcium phosphate were altered to form the four dietary treatments studied.

The rations were formulated to be adequate in all other nutrients, particularly vitamin D and zinc, and they contained 125 gm. of active antibiotic per ton. The vitamin levels used in making the diets are shown in Table VIII. These were based on the best estimates of the requirements of the pig as determined from the current literature.

The rations were mixed for 15 minutes in a horizontal mixer and stored in large plastic containers with lids and identifying labels.

Experimental Pigs

Collection and Handling. A total of 11 Yorkshire and 5 Hampshire pregnant gilts and sows were obtained from the Oklahoma State University purebred herd. Older sows were preferable to gilts since the former were likely to have larger litters and their performance was better known. However, on occasions, gilts were used as an aged sow was not always available when laboratory space was available.

In every case the selected dam had been maintained on a

TABLE X
COMPOSITION OF BASAL MINERAL MIXTURE^a

Mineral	Percent
NaHCO ₃ ^b	50.00
K ₂ SO ₄ ^b	23.37
MgCO ₃ ^b	4.10 (1,861.4) ^c
FeSO ₄ · 2H ₂ O ^b	1.40 (635.6)
ZnSO ₄ · H ₂ O ^b	0.80 (363.2)
MnSO ₄ · H ₂ O	0.30 (136.2)
CuSO ₄	0.20 (90.8)
CoCO ₃	0.20 (90.8)
KI	0.004 (1.816)
Cerelose ^d	19.63
Total	100.004

^aFed at the level of three percent of the purified diets.

^bSupplied through the courtesy of Calcium Carbonate Company, Quincy, Illinois.

^cGrams.

^dCerelose 2001, Corn Products Company, Argo, Illinois.

ration (Table XI) designed to meet their nutritive needs as defined by the N.R.C. (1964). This ration contained 0.80 percent calcium and 0.62 percent phosphorus and was hand-fed at the level of 2.7 kg. per head per day, fed in equal portions twice daily (a.m. and p.m.). It provided more calcium (0.20 percent) and phosphorus (0.22 percent) than is currently recommended by the National Research Council (1964).

At 110 days of gestation, selected gilts and sows were taken to clean surroundings and retained in farrowing crates.

Two methods were used to determine the proper time to initiate a frequent to constant observation of the pregnant gilts and sows. One was to commence observation on the 112th or 113th day of gestation. The other method was to await the appearance of milk in the udder. Neither method was, in itself, completely reliable, due to possible errors in breeding date records and due to normal variation from one sow to another. The average gestation length for the 16 gilts and sows was 114.5 days with a range of 111 to 118 days.

The newborn pigs were protected from bacterial contamination and respiratory infection by catching each pig in a sterilized plastic bag as the sow farrowed normally. The buttocks of the sow were first cleansed with a mild antiseptic¹ and the bag opening was held against the buttocks. After a pig dropped into a bag, the top was immediately

¹Nolvasan-S, Fort Dodge Laboratories, Inc., Fort Dodge, Iowa.

TABLE XI
 PERCENTAGE COMPOSITION OF THE SOW
 RATION FED DURING GESTATION

Ingredients	Quantity
	%
Milo, Western Yellow, ground (8.0%) ^a	79.62
Soybean meal (50%)	10.43
Tankage (60%)	2.50
Alfalfa meal (17%)	5.00
Dicalcium phosphate (28% Ca-18%P)	1.25
Calcium carbonate (38% Ca)	0.50
Salt (trace mineral)	0.50
Zinc sulfate ^b	0.02
Vitamin B ₁₂ supplement ^c	0.012
Vitamin B supplement ^d	0.06
Total	99.89
Calculated chemical composition	
Crude protein	13.93
TDN	76.44
Calcium	0.80
Phosphorus	0.62

^aCrude protein content chemically determined.

^bZinc added to supply a supplement of 50 ppm in diet.

^cContained 88.2 mg. B₁₂ per kg. of supplement.

^dContained riboflavin, 4.4 gm.; pantothenic acid, 8.8 gm.; nicotinic acid, 19.8 gm.; and choline chloride, 198 gm. per kg. of supplement.

closed and the bag immersed in an antiseptic solution¹ and the pig was passed via the entrance lock directly into the body of the previously described isolator unit.

Working through the rubber gloves, thoroughly drenched with a two percent tincture of iodine, each pig was quickly dipped into a germicidal solution¹. Hemostats were used to prevent loss of blood from the umbilical cord. The nose and the mouth of the pig were wiped free of membranes and mucus. Each pig was dried vigorously by wiping with sterile paper towels, and encouraged to breathe as needed by a gentle massage.

Oxytocin² was administered into a marginal auricular vein in cases where farrowing was slow and/or difficult.

After the entire litter had been collected and treated, the pig laden isolator was transported from the procurement location to the Swine Nutrition Laboratory, which is isolated from the swine barn and from personnel in contact with the swine herd.

Before removing the pigs from the isolator, personnel donned sterile 30.5 cm. rubber gloves, freshly laundered coveralls, disposable masks, and thoroughly disinfected rubber boots. A 10 percent formalin solution was sprayed around the entrance lock before the front plastic cover was

¹Nolvassan-S, Fort Dodge Laboratories, Inc., Fort Dodge, Iowa.

²P.O.P., Armour Pharmaceutical Company, Kankakee, Illinois.

removed. Only one pig was removed at a time and the operators' gloves were dipped in a disinfectant solution¹ before handling each pig.

Pigs were removed at random from the isolator unit. Sex, birth weight (to the nearest gm.) and strength score were recorded. The umbilical cord was ligated approximately 2.5 cm. from the navel, severed and the proximal end was swabbed with a 2 percent solution of tincture of iodine. Needle teeth were not clipped and no iron injections were given.

Each baby pig received 25 ml. of pasteurized sow colostrum (composition shown in Table IV). A 35 cc. plastic syringe with an attached 4.0 mm. diameter flexible rubber tube was used to feed the colostrum. Pigs were held vertically by their head and their jaws were forced open with the fingers and thumb. The rubber tube was directed down the esophagus to the stomach. After the tube was in place, the colostrum was put into the stomach by pressing the plunger on the syringe. The danger of getting colostrum into the lungs was minimized by placing the end of the tube near the entrance of the stomach. Occasionally there was a very weak lethargic member of a litter which was fed 20 ml. of a 20 percent glucose solution.

This method of pig collection rather than hysterectomy or caesarean section was used in order to save the sow and

¹Nolvasan-S, Fort Dodge Laboratories, Inc., Fort Dodge, Iowa.

minimize the expensive equipment and trained personnel required. A definite advantage was that full-term pigs were obtained.

Each pig was then assigned to an individual disposable cardboard rearing unit located in the incubator room. The rearing units were kept at 33°C. for 3 or 4 days, then the temperature was gradually reduced to 27°C.

Assignment to Treatment. At 14 days of age, the pigs were removed from their individual sealed incubator boxes and transported to the nursery room (Figure 4) where they remained for 4 weeks. The pigs were weighed to the nearest 50 gm., individually identified (ear notched) and placed in individual metal pens. Pigs were randomly assigned to each treatment diet, with the restriction that treatment groups were balanced as nearly as possible, with respect to breed, litter and sex.

Adjustment of the pigs to the purified diet in dry meal form was facilitated by means of mixing the dry meal with reducing quantities of the fortified milk ration in shallow metal feed trays. Two sets of feeding trays were used; while one set was in use, the other set was soaking in a detergent and disinfectant solution¹. Before use the trays were rinsed with hot water direct from a hose. After feeding, the used trays were thoroughly cleaned with hot water and placed in the disinfectant solution to soak until the

¹Nolvassan-S, Fort Dodge Laboratories, Fort Dodge, Iowa.



Figure 4. View of a 14 Day Old Pig in a Nursery Room Pen, Consuming the Liquid Diet

next feeding.

By the end of the third day (17 days of age) the animals had become satisfactorily adjusted to the dry diet and the environment and the liquid milk starter diet was completely eliminated. The animals were fed ad libitum, with the feeders refilled as necessary.

Data Obtained and Chemical and Statistical Analysis

All pigs were weighed at birth, at 2 weeks of age and weekly thereafter to 9 weeks of age. Weekly feed consumption was recorded. The pigs were observed daily for their well-being and records were kept of any physical abnormalities.

Blood samples were collected by anterior vena cava puncture as described by Carle and Dewhirst (1942) at 3, 6 and 9 weeks of age. A 10 ml. plastic disposable syringe fitted with a 38.1 mm. 20-gauge needle was used to withdraw approximately 6 ml. of blood. To 1.5 ml. of the blood sample was added 0.05 ml. of a heparin solution (2.5 mg. per ml.). Clotting time, hemoglobin, hematocrit (packed cell volume) and red and white cell counts were determined immediately.

The remaining blood sample (approximately 4.5 ml.) was placed in a plastic centrifuge tube and clotting time was determined. The sample was then held at 20°C. for approximately 40 minutes to promote syneresis. Separation of serum and clot was completed in a refrigerated centrifuge at 3,000

rpm for 15 minutes. The resulting serum was removed by decantation, placed in sterile stoppered vials and stored at -17°C . for the later determination of levels of serum calcium, inorganic phosphorus and alkaline phosphatase.

Hemoglobin was determined using the cyanmethemoglobin method of Cannon (1958). Hematocrit was determined in duplicate according to the micromethod described by McGovern et al. (1955). Blood samples were centrifuged for 5 minutes at 10,000 rpm in an International "Hemacrit" centrifuge.

Erythrocytes were counted in duplicate from a single filling of a "zero error" pipette using Hayem's solution as the diluting fluid. Cells were counted in a hemocytometer with Neubauer ruling using a National Bureau of Standards certified cover glass. Acceptable duplicate counts differed no more than 8 to 9 percent of the lower count.

Total leukocyte counts were made in duplicate on a hemocytometer with a National Bureau of Standards certified cover glass from one dilution with a "zero error" Hellige pipette. Turk's acetic acid solution was used for the diluting fluid. Counts were considered acceptable if the higher value was no greater than 110 percent of the lower value.

Mean corpuscular volume (M.C.V.), mean corpuscular hemoglobin (M.C.H.) and mean corpuscular hemoglobin concentration (M.C.H.C.) were calculated by the following equations as described by Wintrobe (1961):

$$\text{M.C.V. } (\mu^3) = \frac{\text{vol. packed red blood cells, ml. per 1,000 ml.}}{\text{red blood cells, millions per mm}^3}$$

$$\text{M.C.H. (micro-mcg.)} = \frac{\text{hemoglobin, gm. per 1,000 ml.}}{\text{red blood cells, millions per mm}^3}$$

$$\text{M.C.H.C. (\%)} = \frac{\text{hemoglobin, gm. per 100 ml.} \times 100}{\text{vol. packed red blood cells, ml. per 100 ml.}}$$

Blood clotting time was recorded as the time interval, in minutes, between the appearance of blood in the syringe and the time at which a fibrin thread first appeared.

Serum calcium was determined in duplicate by atomic absorption spectrophotometry using a Perkin-Elmer Model 303 with a Boling (total consumption) burner and an air-acetylene flame. Serum phosphorus determinations were made by the method of Tauskey and Shorr (1953). Serum calcium to phosphorus ratios and the product of calcium times phosphorus were calculated.

Serum alkaline phosphatase activity was determined by a colorimetric procedure developed by Klein *et al.* (1960), as outlined by General Diagnostics (1965). Phosphatase activity was expressed in Klein-Babson-Read units per 100 ml. serum. One unit is defined as the amount of enzyme that will liberate 1.0 mg. of phenolphthalein in 30 minutes at 37°C. Since 0.2 ml. of serum was used in the assay, the activity, expressed as units per 100 ml. of serum, was numerically equal to one-half the amount (in mcg.) of phenolphthalein released.

Morbidity and mortality were recorded and cause of death diagnosed by consulting personnel in the Veterinary

Pathology Department.

At the conclusion of the experiment, six pigs (four males and two females) from Rations A, C and D and seven pigs (four males and three females) from Ration B were selected at random and sacrificed by exsanguination. Feed, but not water, was withdrawn approximately 8 hr. prior to slaughter.

Various bones, organs and glands were removed, blotted to remove excess blood and weighed. The thyroid gland was excised by blunt dissection and freed of connective tissue under a dissecting microscope. The heart was dissected free from the great veins. The arteries were severed at the point of emergence from the heart, where their color changes from deep red to white and blood was removed before weighing. The lungs were dissected free of the pleural membranes, the trachea and the chief bronchi. The liver was freed of the gall bladder and cystic duct. The spleen was removed and closely trimmed at the hilus. The adrenals were excised before removing the kidneys and trimmed free of vascular and connective tissue. The kidneys were removed from their retroperitoneal position, and the arteries, veins and ureters were cut at the hilus. The renal capsule was left intact, and the two kidneys were weighed separately. The stomach was dissected free from the esophagus and duodenum, emptied of its contents, washed with water and blotted dry. All these organs were weighed immediately after excision and the heart, liver and left kidney, along with a sample of

hair taken from the back region, were stored in sealed polyethylene bags at -17°C . until analyzed for calcium and phosphorus. Suitable aliquots for analysis were obtained after thawing, chopping and homogenizing the entire organ.

The right humerus, femur, ulna-radius and eighth rib were dissected free of muscles, ligaments and periosteum, weighed and the maximum length was measured by means of a vernier scale slide-calipers. The length of the humerus (paralleled to the axis) was from head to condyle. The femur length was from head to condyle while the ulna-radius was measured from the proximal end of the ulna to the distal end of the ulna-radius. The 8th rib was measured from the head directly to the sternal extremity. Diameter or width of each of the bones was recorded and the specific gravity was found using the equation by Whiteman et al. (1953). These four bones were then stored in polyethylene bags at -17°C . until analyzed for calcium and phosphorus.

Bones were extracted with 95 percent ethanol for 16 hours, followed by a petroleum ether extraction for an additional 3 hours. The fat-free bones were dried, ground and along with the left kidney, liver, heart and hair were ashed at 550°C . for 16 hours, with the ashes being dissolved in 4 N HCL to a known volume.

Bone and tissue phosphorus were analyzed in duplicate by the method of Fiske and Subbarow (1925). Calcium analysis was performed in duplicate using a Perkin-Elmer Atomic Absorption Spectrophotometer by methods set forth by the

manufacturer.

A cross-section of the snout through the first maxillary premolar teeth was grossly examined by Veterinary Pathology personnel for any evidence of nasal turbinate atrophy and then scored.

The remainder of the pigs were moved to another location and placed on a fortified milo-soybean meal ration (Table XII) containing 16 percent protein, 0.80 percent calcium and 0.68 percent phosphorus. This ration was fed ad libitum from 9 weeks of age to slaughter weight. At that time 2 pigs from Diets B, C and D and 3 pigs from Diet A were slaughtered and a gross examination was made of nasal turbinates and lungs.

Proximate analysis values were determined by the methods of A.O.A.C. (1960).

Analyses of variance and calculation of standard errors were conducted according to the methods outlined by Steel and Torrie (1960). Duncan's new multiple range test was used to make comparisons among treatment means. Mean comparisons were conducted only when analysis of variance for the various criteria were significant ($P < .10$).

TABLE XII
 PERCENTAGE COMPOSITION OF THE RATION FED FROM
 NINE WEEKS OF AGE TO APPROXIMATELY
 91 KG. LIVE WEIGHT

Ingredients	Quantity
	%
Milo, Western Yellow, ground (8.0%) ^a	73.48
Soybean meal (50%)	18.47
Alfalfa meal (17%)	5.00
Dicalcium phosphate (28% Ca-18% P)	1.85
Calcium carbonate (38% Ca)	0.50
Salt (trace mineral)	0.50
Zinc sulfate ^b	0.02
Vitamin B ₁₂ supplement ^c	0.012
Vitamin B supplement ^d	0.06
Antibiotic ^e	0.02
Total	99.912
Calculated chemical composition	
Crude protein	15.96
TDN	76.26
Calcium	0.80
Phosphorus	0.68

^aCrude protein content chemically determined.

^bZinc added to supply a supplement of 50 ppm in diet.

^cContained 88.2 mg. B₁₂ per kg. of supplement.

^dContained riboflavin, 4.4 gm.; pantothenic acid, 8.8 gm.; nicotinic acid, 19.8 gm.; and choline chloride, 198 gm. per kg. of supplement.

^eAurofac 40, contained 88.2 gm. of chlortetracycline per kg., American Cyanamid Company, Princeton, New Jersey.

RESULTS AND DISCUSSION

Preliminary Trial 1

In a preliminary trial to evaluate the liquid diet and management procedures for the non-maternal rearing of pigs, 28 pigs (16 males and 12 females) were collected from four first-litter gilts. Without the aid of sow colostrum and antibiotics, only 42.9 percent of the pigs reached 9 weeks of age (Table XIII). Most deaths occurred during the third week of life. A two-year English survey (Jennings, 1959) of 26,684 pigs showed an average death rate of 21.1 percent from birth to 8 weeks. Of that mortality, 77.4 percent occurred during the first week.

There was a high incidence of scours in this preliminary trial and all pigs that died had some degree of diarrhea. E. coli was a direct cause of two deaths and was probably a contributing factor in seven others. E. coli has been the most frequently found causative organism of intestinal infections such as scours (Smith, 1965). It has been reported (Anonymous, 1960) that 43.6 percent of 1,742 pre-weaning deaths was due to alimentary or bacterial type of infections. E. coli was identified as the cause of 57.7 percent of the mortality from these infections.

Death losses did not tend to be confined to any particu-

TABLE XIII
 SUMMARY OF RESULTS OF PRELIMINARY TRIAL I:
 SURVIVAL OF PIGS DEPRIVED OF SOW
 COLOSTRUM AND ANTIBIOTICS

Item	Sex		Combined
	Males	Females	
<u>Number of pigs</u>			
Started at birth	16	12	28
Surviving to 9 weeks	6	6	12
Dying (Birth to 9 weeks)	10	6	16 ^a
<u>Survival, %</u>	37.5	50.0	42.9
<u>Birth wt., kg.</u>			
All pigs	1.156	1.152	1.152
Survivors to 9 weeks	1.392	1.206	1.297
Non-survivors	1.011	1.102	1.048
<u>Age at death, days</u>	18.6 (2-35) ^b	18.2 (12-22) ^b	18.4

^aNecropsy of 13 of the pigs by the Oklahoma State University Veterinary Pathology Department revealed the following as causes of death; 4 bacterial septicemia, 3 gut edema, 3 bronchopneumonia, 2 E. coli enteritis and 1 polyarthritis and omphalophlebitis.

^bRange.

lar litter. Although pig numbers were small, more difficulty was encountered in the rearing of males than females.

Rearing of pigs under carefully controlled laboratory conditions would appear to offer a means of removing many of the environmental influences assumed to be competitively disadvantageous to the smaller pigs. However, in this preliminary trial there was an apparent high degree of association between birth weight and survival, particularly among the males. Vestal (1938) stated that, under usual swine production conditions, approximately 35 percent of baby pigs weighing 0.91 kg. or less did not survive to weaning. England et al. (1961) reported only one death loss during the first 5 days for 90 artificially reared pigs, involving 27 that weighed 0.91 kg. or less at birth.

England and Chapman (1962) reported that the correlation between birth weight and 56-day weight was 0.13 for artificially reared pigs while Blunn et al. (1954) found a corresponding correlation of 0.53 for conventionally reared swine. The plausible explanation for the lower association between birth weight and 56-day weight in the artificially reared pigs is the greater degree of environmental adequacy for the pigs of low birth weights.

The development of the pig from birth depends partly on its weight and viability and partly on its intake of nutrients from sow's colostrum and milk and from supplementary feed (Lodge, 1966).

The composition of sow colostrum and milk has been stud-

ied by Braude et al. (1947), Hughes and Hart (1935), Bowland et al. (1948), Heidebrecht et al. (1950), Sheffy et al. (1952), Morgan and Lecce (1964), Earle and Stevenson (1965), Perrin (1954), Pond et al. (1962), Davis et al. (1951), Luecke et al. (1947), Washam (1966), Barnhart et al. (1954), Braude et al. (1945-46), Blair et al. (1964), deMan and Bowland (1963) and Whittlestone (1952). The secretion of the mammary gland which follows parturition is known as colostrum. The colostrual stage in the sow lasts for some 5 days during which time the milk composition changes rapidly to that of "normal" milk. In reality, however, there is no such thing as "normal" milk because its composition changes continually throughout lactation.

The compositions of colostrum, "normal" sow's milk and the liquid diet used in this study are compared in Table XIV.

Dry matter and total protein concentration in the sow's mammary secretions follow the same general trends throughout the sow's lactational period. With the initiation of nursing, total protein concentration drops precipitously. Dry matter concentration, on the other hand, undergoes a less dramatic change, apparently as a result of increasing fat and lactose concentrations (Perrin, 1954). There is a gradual increase in both total protein and dry matter in late lactation (Morgan and Lecce, 1964). These trends in dry matter and total protein concentrations are probably the reciprocal of the total volume curve.

Of particular significance to the survival of the new-

TABLE XIV

COMPARISONS OF SOW COLOSTRUM¹, SOW MILK¹ AND THE LIQUID DIET
 FED FROM BIRTH TO TWO WEEKS OF AGE

	Colostrum Milk	Normal Milk	Colostrum as a % of Normal Milk	Liquid Diet
Dry matter, %	25.70	19.26	133	25.02
Fat, %	5.31	6.75	79	4.90
Crude Protein, %	15.25	6.50	235	6.23
Nitrogen-free extract, %	2.96	4.90	60	11.86
Mineral matter, %	0.67	0.96	70	2.03
Calcium, %	0.064	0.25	26	0.330
Phosphorus, %	0.082	0.16	51	0.246
Vitamin A, I.U./gm. of fat	77.5	18.5	419	
Vitamin C, mg./100 ml.	26.2	12.2	215	
Thiamine, mcg./100 ml.	96.8	67.7	143	
Riboflavin, mcg./100 ml.	44.9	45.7	98	

¹Hughes and Hart (1935); Braude et al. (1947); Bowland et al. (1948).

born pig is the supply and, perhaps, the composition of colostrum; under practical conditions consumption of colostrum is an essential prerequisite of survival.

It is not known how much colostrum the piglet must consume to obtain sufficient antibodies against pathogenic organisms, but it seems possible that in the cases of prolonged farrowing and very small piglets at birth which may not suck for several hours, that some piglets within litter may nurse too late to obtain the required intake of antibodies, thus some deaths within a few days of birth may be related to inadequate intake of colostrum.

Sow's milk is highly nutritious, with a crude protein content increasing from about 25 to 33 percent of the dry matter as lactation advances. Sow's milk is particularly rich in fat, contains approximately 7.0 percent on an as-is basis and 36 percent on a dry matter basis. Fat is the most variable of the major constituents of milk and the most readily influenced by feeding. The fat percentage, although very erratic, tends to reach a peak around the third week of lactation (Lodge, 1959) and has been reported as high as 17.2 percent (Perrin, 1955). This increase to a high fat level in the diet, together with the known decrease in the immunity of the pig to disease around this time, may be of considerable significance in the incidence of piglet scours which frequently occurs at about three weeks of age.

Apart from its inadequate iron level, sow's milk appears to be an ideal feed for young pigs, allowing an efficiency of feed conversion on a dry matter basis of some .08 kg.

feed per kg. of gain. Its failure is quantitative rather than qualitative; thus, an average yield of 45.4 kg. of milk per piglet suckled, or 9.1 kg. of dry matter in an eight weeks lactation period would, at an efficiency of conversion of 0.8, be sufficient to allow a weight gain of 11.3 kg. With an average birth-weight of 1,362 gm., this would allow the production of 12.7 kg. pigs at eight weeks of age. An 18.1 kg. pig at this same age must have consumed approximately 10.9 kg. of creep feed at an efficiency of feed conversion of about 2:1.

It was evident from Preliminary Trial I that it would be quite difficult to rear baby pigs without the aid of sow colostrum, even when they were kept in apparently good environmental conditions. This finding was not totally unexpected.

Early attempts to raise baby pigs taken at birth were unsuccessful (Weybrew et al., 1949; Bustad et al., 1948; Cartwright et al., 1950; Cartwright et al., 1949; Barrick et al., 1954; Cartwright et al., 1951; Curtin et al., 1952; Catron et al., 1953; McRoberts and Hogan, 1944) and colostrum was considered essential for survival. Most pigs developed diarrhea around the fourth day and if unmedicated, 60 to 100 percent died. Bellis (1957) summarized this situation with the statement that "mortality increases with progressively earlier weanings, and the survival of piglets that have not received colostrum is very unlikely....."

Many investigators (Brambell, 1958; Rutquist, 1958;

Nordbring and Olsson, 1957; Lecce and Matrone, 1960, 1961; Lecce et al., 1961a,b; McCance and Widdowson, 1959; Bauriedel et al., 1954; Wadill et al., 1962; Wellmann and Engel, 1964b; Danilevsky, 1960; Jakobsen and Moustgaard, 1950; Kono-patkin, 1964a; Long et al., 1964; Olsson, 1960) have shown that the baby pig is born with a deficiency in albumin, beta and gamma globulin, and it is the function of colostrum to supply this missing protein to the newborn pig.

Rapid maturation of the serum protein profile occurs in nursing pigs (Lecce and Matrone, 1960; Sharpe, 1966; Miller et al., 1961a; Pavlovic et al., 1961) and pigs hand fed colostrum (Widdowson and McCance, 1956; McCance and Widdowson, 1956, 1957; Hardy, 1965) while pigs fed other diets such as cow's milk experience a delay in their serum protein maturation process and are susceptible to diarrhea, bacteraemia and death (Lecce and Reep, 1962; Lecce and Matrone, 1961; Lecce et al., 1961a).

Antibody-rich-colostral-globulin is believed to cross unaltered from the gut to the serum of the nursing pig within the first 24 to 36 hours after birth (Earle, 1935; Nelson, 1932; Barrick et al., 1954; Hoerlein, 1952; Brambell, 1958; Speer et al., 1959; Bruner et al., 1949; Asplund et al., 1962; Kaeberle and Segre, 1964; Wellman and Engel, 1964a; Coggins, 1964; Lecce et al., 1964; Payne and Marsh, 1962).

Chamberlain et al. (1965) found that only negligible amounts of gamma globulin were absorbed by baby pigs 60 hours old. However, the period of antibody absorption can be ex-

tended to 86 hours (Lecce and Morgan, 1962) and 106 hours (Payne and Marsh, 1962) when pigs are starved.

Thus, the baby pig is given a ready supply of globulin before its own synthesizing mechanisms can fulfill this need. Further, colostrum globulin, carrying immune bodies, endows the offspring with passive immunity to infectious disease (Konopatkin, 1964b) accounting for the vigor in the pigs fed colostrum.

No such resistance to disease is expected in the non-immune, colostrum-free pigs; hence, the difficulty in raising them (Kenworthy and Crabb, 1963; Owen and Bell, 1964; Owen et al., 1961). However, Young and Underdahl (1953) demonstrated that pigs deprived of colostrum could be raised if elaborate techniques were employed to give the pig a complete sterile environment. Perry (1966) successfully raised colostrum-deprived pigs which were caught in sterile towels at birth and fed different concentrations of cow's milk solids in an automatic feeding device.

A number of workers (Brown et al., 1961; Miller et al., 1961a; Olsson, 1959; Nordbring and Olsson, 1957; Miller et al., 1957; Miller et al., 1962a; Hoerlein, 1957; Young and Underahl, 1949, 1950; Harmon et al., 1959; Segre and Kaeberle, 1962a,b; Aiken and Blore, 1964; Dardiri et al., 1966; Murdock and Jungk, 1957; Sharpe, 1965, 1966; Segre and Myers, 1964; Wide and King, 1962; Cochrane et al., 1964; Michna, 1965) have reported extensive studies of colostrum acquired immunity and active antibody production in baby pigs using

several different antigens.

Preliminary Trial 2

In view of the literature and the low survival rate obtained in the colostrum-deprived pigs (Preliminary Trial 1), another preliminary trial was conducted to gain experience in the artificial rearing technique and to attempt to provide information on the quantity of pasteurized sow colostrum that would be needed to obtain an acceptable survival level.

A total of 110 baby pigs was collected and assigned, by litter, to receive one of five different pasteurized sow colostrum quantities. These amounts were equally spaced and ranged from 40 to 280 ml.

A summary of the results from Preliminary Trial 2 is presented in Table XV. Statistical analysis was not performed on the data; however, maximum survival, 2 week and 3 week weights were attained by supplying 280 ml. of colostrum. The influence of level of colostrum on body weight is graphically presented in Figure 5.

The pigs receiving 40 ml. of colostrum were less thrifty in appearance and had a slightly higher incidence of scours than other colostrum groups.

In agreement with Preliminary Trial 1, males were much more difficult to rear than females. Of the 18 deaths occurring from birth to 3 weeks of age, 15 or 83.3 percent were males.

TABLE XV

SUMMARY OF RESULTS OF PRELIMINARY TRIAL 2: SURVIVAL AND
WEIGHT GAIN OF BABY PIGS SUPPLIED DIFFERENT
QUANTITIES OF PASTEURIZED SOW COLOSTRUM

Item	Quantity of Sow Colostrum, ml.				
	40	100	160	220	280
<u>Number of pigs</u>					
Started at birth	9	25	27	24	25
Surviving to 2 weeks	7	20	23	22	23
Surviving to 3 weeks	7	18	21	21	23
<u>Survival, %</u>					
2 weeks	77.8	80.0	85.2	91.7	92.0
3 weeks	77.8	72.0	77.8	87.5	92.0
<u>Pig wt., kg.</u>					
Birth	1.220	1.329	1.138	1.181	1.265
2 weeks	1.937	2.199	1.905	2.091	2.476
3 weeks	2.512	3.111	2.712	3.090	3.330

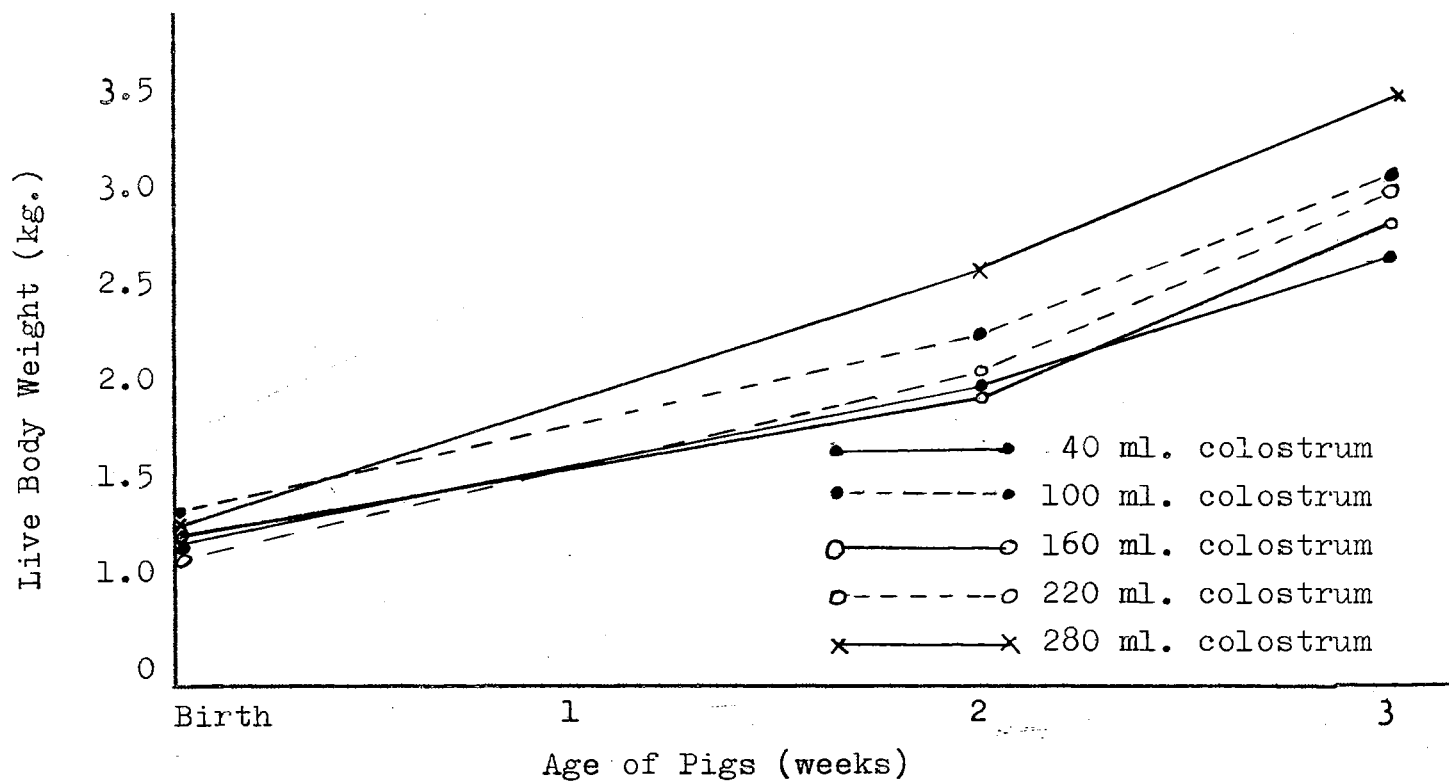


Figure 5. Influence of Pasteurized Sow Colostrum Intake on Live Body Weights of Baby Pigs from Birth to Three Weeks of Age

General Observations -- Preliminary Trials 1 and 2

No major inherent difficulties were encountered in using the collection and artificial rearing technique, but it was apparent that close attention to detail was essential. Obviously, technical efficiency improved as experience was gained. Certainly some losses in the preliminary trials could be attributed to lack of experience.

The fecal material (stools) of pigs kept in the cardboard incubators was normal in consistency for the first few days except that they were dark in color. By 7 to 10 days the fecal material had become rather soft and semifluid in consistency, and it became somewhat more solid by the 14th day. The accumulation of urine and excrement in the incubators during the first 2 weeks of life was not excessive.

At approximately 2 weeks of age, a moderate degree of scales and flakey skin was observed on a majority of the pigs. This flakiness covered almost the entire body surface. This condition was alleviated by brushing the pigs followed by a light application of oil. Thus, this condition was not thought to be nutritionally caused but rather it resulted from the inability of the pigs to rub off their dying tissue when in the confines of the isolation boxes. Some difficulty was encountered with sore feet. This condition was more prevalent as the pigs approached 2 weeks of age.

Calcium-Phosphorus Study

In light of the information derived from Preliminary

Trials 1 and 2, it was decided to provide each baby pig with pasteurized sow colostrum during its first day of life. The quantity provided was dependent upon the amount of colostrum available; however, every attempt was made to see that each newborn received at least 200 ml.

To obtain pigs for the calcium-phosphorus study, a total of 88 baby pigs were collected and successfully reared to 14 days of age. Table XVI summarizes the performance of these pigs to 14 days of age. Fourteen day weights were satisfactory but obviously heavier pigs would have been more desirable. Attempts to obtain faster growth rates while the pigs were in the individual incubators increased the incidence of nutritional scours. The liquid diet was palatable and feed utilization was quite good.

At two weeks of age the pigs were assigned to one of the purified rations and adjusted to that ration during the following seven days. Nine of the 2 week old pigs had varying degrees of diarrhea, were weak and as a result were doing poorly. These nine pigs were removed and only 79 pigs were assigned to the calcium-phosphorus study.

The number of pigs allotted to each ration, together with their respective survival rates to 9 weeks of age, are presented in Table XVII. Survival for each treatment group was considered excellent, although four pigs, of equal sexes, died during the experimental period; two pigs on Ration A and one each on Rations B and C. All pig mortalities were submitted to the Veterinary Pathology Department for exami-

TABLE XVI

SUMMARY OF THE PERFORMANCE OF NONMATERNALLY REARED
BABY PIGS: BIRTH TO 14 DAYS OF AGE

Item	
<u>Number of pigs</u>	
Birth	88
14 days	88
<u>Pig weight, kg.</u>	
Birth	0.57
14 days	2.16
<u>Strength score^a</u>	
Birth	2.4
14 days	2.2
<u>Liquid diet consumed,</u>	
Birth to 14 days, l.	3.62
<u>Feed utilization^b</u>	0.42

^a1=very strong, 2=strong, 3=average, 4=weak and 5=very weak.

^bKg. body weight gain per liter of liquid diet.

TABLE XVII

SURVIVAL OF PIGS FED DIFFERENT LEVELS OF CALCIUM (Ca) AND
PHOSPHORUS (P) FROM THREE TO NINE WEEKS OF AGE

Dietary Ca, %	0.368	0.569	0.776	0.952
Dietary P, %	0.279	0.443	0.610	0.729
Ration Designation	A	B	C	D
Number of pigs started (3 wk.)	18(9) ^a	20(10)	19(9)	22(10)
Number of pigs finished (9 wk.)	16(8) ^a	19(9)	18(9)	22(10)
Number of deaths (3 to 9 wk.) ^b	2 ^c	1 ^d	1 ^e	0
Survival, %	88.9	95.0	94.7	100.0

^aNumber of males.

^bNecropsy by the Oklahoma State University Veterinary Pathology Department.

^cOne female pig died at thirty-seven days of age due to osteodystrophy and polysecre-sitis and one male pig hemorrhaged to death at fifty-two days of age due to rupture of an umbilical hernia.

^dOne male pig hemorrhaged to death at thirty-one days of age due to rupture of an umbilical hernia.

^eOne female pig died suddenly at thirty-eight days of age due to bacterial septicemia.

nation.

Only one death, a female on Ration A, could be directly related to the ration treatment. Post-mortem examination of this pig revealed that the bones were very soft and although they would bend quite easily, it was very difficult to break them. There appeared to be some evidence of atrophy of the ventral turbinates. Microscopic examination showed very little osteoid tissue laid down in the turbinates. Cause of death was attributed to osteodystrophy and polyserositis.

With the exception of the pig described above, the pigs which died during the experimental period had grown well up until death. Data, for these pigs, to the last weekly weigh period prior to death has been included in appropriate treatment means.

The weekly body weight data from pigs receiving different dietary levels of calcium and phosphorus are presented in Table XVIII. No significant difference in weekly body weights were observed during the 6 weeks feeding period. However, without exception, heavier pigs were obtained at each weight period with each increment in dietary calcium and phosphorus. This growth data is graphically demonstrated in Figure 6. Differences between mean treatment body weights became greater with increased time on feed so that the greatest differences in weights were observed at the termination of the experiment. From this standpoint, it would have been interesting to have prolonged the experimental period by at least 3 weeks.

TABLE XVIII

WEEKLY LIVE BODY WEIGHTS OF PIGS FED DIFFERENT LEVELS OF CALCIUM (Ca)
AND PHOSPHORUS (P) FROM THREE TO NINE WEEKS OF AGE

Dietary Ca, % Dietary P, % Ration Designation	0.368 0.279 A	0.569 0.443 B	0.776 0.610 C	0.952 0.729 D	SE ^a	SD ^b
2 weeks, kg.	2.03(18) ^c	2.20(20)	2.11(19)	2.28(22)	0.11	NS
3 weeks, (initial), kg.	2.91(18) ^c	3.10(20)	3.05(19)	3.29(22)	0.16	NS
4 weeks, kg.	4.08(18) ^c	4.26(19)	4.31(19)	4.80(22)	0.25	NS
5 weeks, kg.	5.59(18) ^c	5.95(19)	6.11(19)	6.60(22)	0.40	NS
6 weeks, kg.	8.74(17) ^c	8.81(19)	9.06(18)	9.63(22)	0.60	NS
7 weeks, kg.	11.39(16) ^c	11.82(19)	11.93(18)	12.82(22)	0.73	NS
8 weeks, kg.	14.65(16) ^c	15.11(19)	15.76(18)	16.79(22)	0.90	NS
9 weeks (final), kg.	17.65(16) ^c	18.54(19)	19.69(18)	20.72(22)	1.04	NS

^aStandard error of treatment means. Calculated standard errors are based on twenty pigs per treatment mean.

^bSignificant differences. NS = non-significant ($P > .05$).

^cNumber of animals included in the treatment mean.

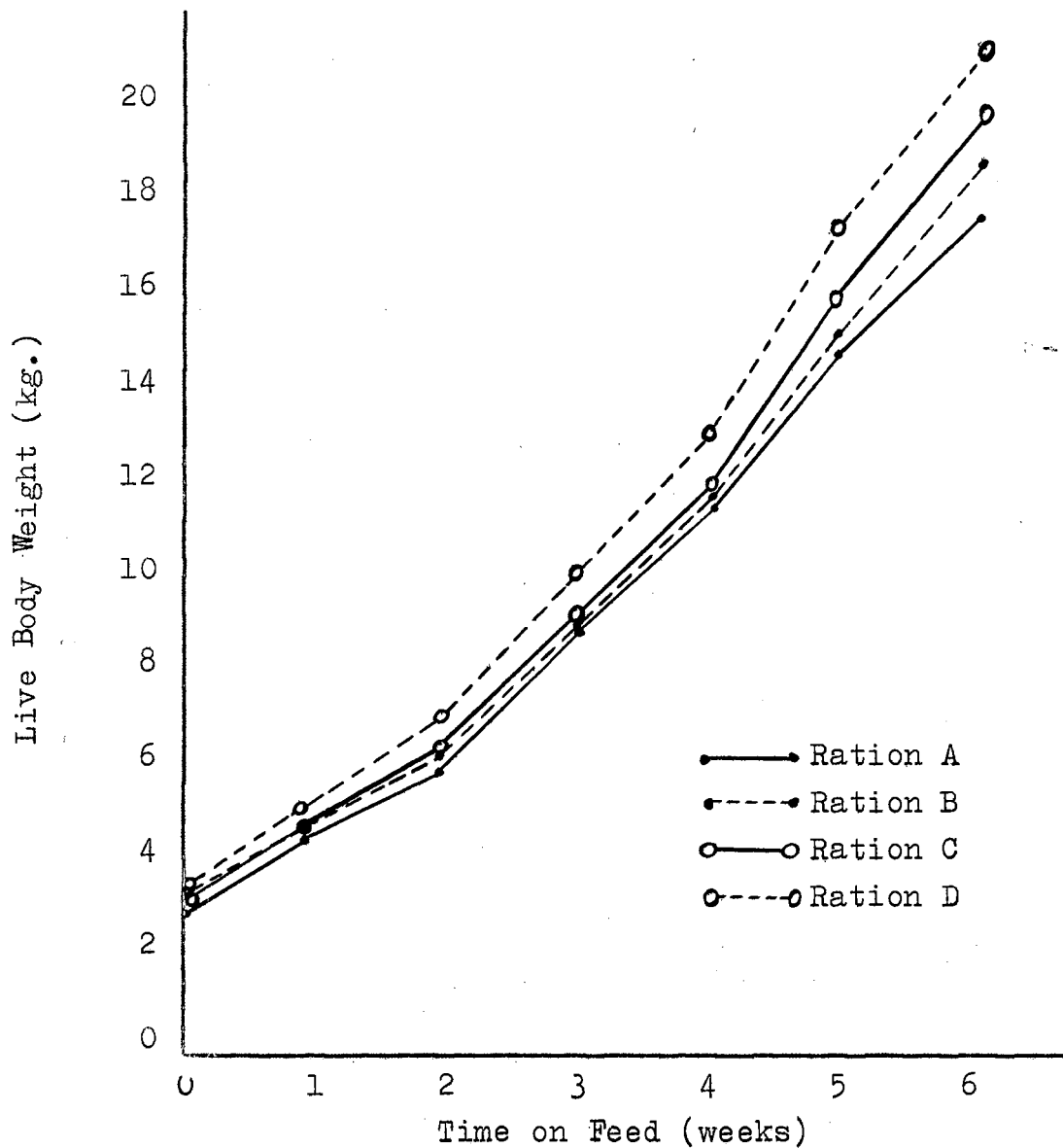


Figure 6. Growth Curves of Pigs Fed Different Levels of Calcium and Phosphorus from Three to Nine Weeks of Age

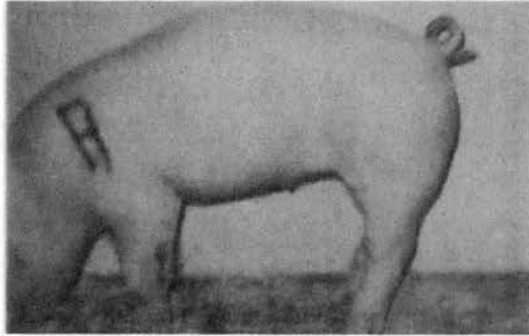
Growth rates in this study were appreciably higher than those reported for the suckling pig by Ashton and Crampton (1943b) but less than more recent values given by Wood and Groves (1965) for suckling pigs and by Catron et al. (1953) for nonmaternally reared pigs. Data on normal body weights have been compiled by Altman and Dittmer (1962) and body weight for age values from large populations of pigs are extant (Bell, 1964; Headley et al., 1961; Cameron et al., 1945; Ashton and Crampton, 1943a; Crampton, 1933).

Mean final body weights (9 weeks of age) were considered to be quite good, especially on Rations C and D. Littermate Yorkshire pigs which were fed the treatment rations are shown in Figure 7.

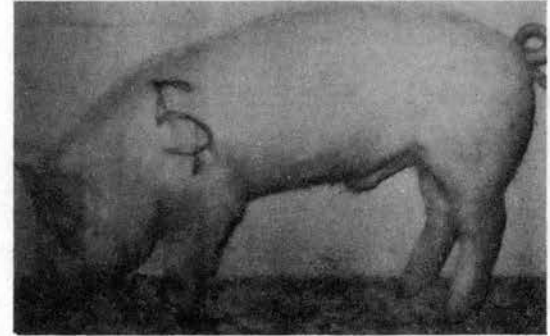
Kellerman et al. (1943), Combs et al. (1962) and Win-trobe (1939) obtained satisfactory growth rate of young pigs in which phosphorus constituted 0.41, 0.44 and 0.52 percent, respectively, of the total solids; but Zimmerman et al. (1961) found that phosphorus at 0.40 percent of the ration was inadequate for maximum gains and increasing the phosphorus level significantly improved feed utilization in baby pigs fed from 3 to 7 weeks of age.

Newman et al. (1964b), using rations containing 0.35, 0.45, 0.55 and 0.65 percent phosphorus, demonstrated that these levels of phosphorus had no effect on growth, feed consumption or feed utilization. Similar results were reported by Miller et al. (1961c).

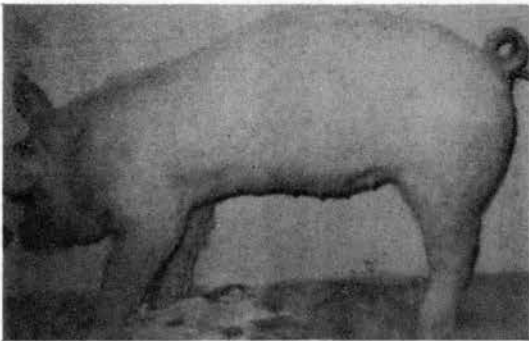
During the 6 weeks feeding period, females gained more



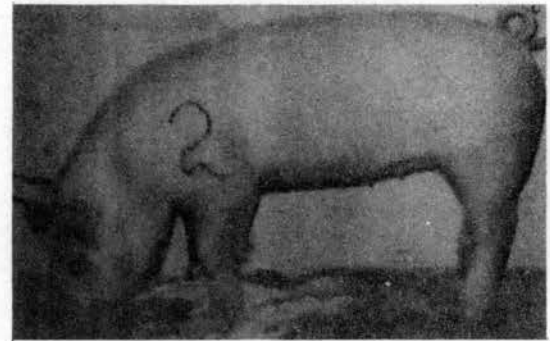
Ration A



Ration B



Ration C



Ration D

Figure 7. Side Views of Nine Week Old Litter-mate Yorkshire Pigs Fed Different Levels of Calcium and Phosphorus from Three to Nine Weeks of Age

weight (16.74 vs. 15.55 kg.), consumed more feed (29.04 vs. 27.17 kg.) and were slightly more efficient (1.75 vs. 1.80 kg. of feed per kg. of gain) than males. However, these differences were statistically nonsignificant.

Although not significant, both total body weight gain and total ration intake during the 6 weeks feeding period were increased with each increase in ration calcium and phosphorus (Table XIX).

Feed efficiencies, expressed as kg. of feed per kg. of body weight gain, did not differ significantly; however, pigs supplied Ration D were the most efficient over the entire experimental period.

Kellermen et al. (1943), Lehrer et al. (1949), Miller et al. (1954), Johnson et al. (1948) and Wintrobe (1939) obtained satisfactory growth rate of young pigs in which calcium constituted 0.73, 0.74, 0.80, 0.91 and 0.94 percent, respectively, of the total solids.

Dudley et al. (1961) reported that for maximum rate and efficiency of gain in young pigs, the calcium requirement was no higher than 0.2 percent of the diet. In contrast, Miller et al. (1960) stated that 0.6 percent calcium supported maximum growth and feed efficiency of baby pigs receiving a synthetic milk diet.

Combs and Wallace (1962) observed that when ration calcium was increased by increments of 0.10 from 0.40 to 0.80 percent, daily gains from 2 to 8 weeks of age tended to decrease linearly with increasing calcium.

TABLE XIX

TOTAL BODY WEIGHT GAIN. FEED CONSUMPTION AND FEED EFFICIENCY
OF PIGS FED DIFFERENT LEVELS OF CALCIUM (Ca) AND
PHOSPHORUS (P) FROM THREE TO NINE WEEKS OF AGE

Dietary Ca, %	0.368	0.569	0.776	0.952		
Dietary P, %	0.279	0.443	0.610	0.729	SE ^a	SD ^b
Ration Designation	A	B	C	D		
Number of pigs at 9 wk.	16	19	18	22		
Total wt. gain, kg.	14.66	15.50	16.67	17.43	0.98	NS
Total feed intake, kg.	25.76	27.76	29.03	29.46	1.66	NS
Feed efficiency ^c	1.784	1.882	1.758	1.697	0.655	NS

^aStandard error of treatment means. Calculated standard errors are based on twenty pigs per treatment mean.

^bSignificant differences. NS = non-significant ($P > .05$).

^cKg. of feed solids per kg. of body weight gain.

When the level of calcium was increased from 0.48 to 0.88 and to 1.32 percent in the diet of young pigs, Combs et al. (1966b) reported that average daily gain and efficiency were significantly decreased, but there was no significant effect on feed intake from 2 to 8 weeks of age.

In contrast, Lloyd et al. (1961) obtained maximal rate of gain in 2 to 8 week old pigs receiving a natural diet containing 1.2 percent of calcium.

Body weight gain, feed consumption and feed efficiency by weekly periods are presented in Table XX.

Total gain figures were not affected by dietary treatment until the period from 42 to 49 days when there appeared to be a slight positive relationship between ration calcium and phosphorus and gain. During the last week Ration A significantly depressed total gain.

Although nonsignificant at all periods, total feed intake tended to follow the same pattern as total gain.

Mean serum calcium levels (Table XXI) were considered to be normal, ranging from 11.0 to 12.55 mg. per 100 ml. The concentration of calcium in blood serum of the pig varies from 9 to 15 mg. per 100 ml. (Dukes, 1955). This serum calcium is composed of diffusible and non-diffusible portions. The former is ionized; the latter is protein bound. The ionized calcium is believed to be the physiologically active portion.

Serum calcium levels were not significantly affected by rations at 3 and 6 weeks on feed, although at 6 weeks serum

TABLE XX

WEEKLY BODY WEIGHT GAIN, FEED CONSUMPTION AND FEED EFFICIENCY OF PIGS
 FED DIFFERENT LEVELS OF CALCIUM (Ca) AND PHOSPHORUS (P)
 FROM THREE TO NINE WEEKS OF AGE

Dietary Ca, %	0.368	0.569	0.776	0.952		
Dietary P, %	0.279	0.443	0.610	0.729	SE ^a	SD ^b
Ration Designation	A	B	C	D		
21 to 28 days						
No. of pigs	18	20	19	22		
Total gain, kg.	1.12	1.28	1.26	1.52	0.15	NS
Gain/day, kg.	0.160	0.184	0.179	0.216	0.02	NS
Total feed solids, kg.	1.91	2.09	2.09	2.03	0.16	NS
Feed solids/day, kg.	0.273	0.299	0.299	0.289	0.02	NS
Feed efficiency ^c	1.54	1.90	1.91	1.82	0.31	NS
28 to 35 days						
No. of pigs	18	19	19	22		
Total gain, kg.	1.76	1.69	1.80	1.79	0.20	NS
Gain/day, kg.	0.251	0.241	0.257	0.256	0.03	NS
Total feed solids, kg.	3.11	3.07	3.06	3.27	0.28	NS
Feed solids/day, kg.	0.444	0.439	0.438	0.467	0.04	NS
Feed efficiency ^c	1.75	2.06	1.96	2.41	0.29	NS
35 to 42 days						
No. of pigs	17	19	18	22		
Total gain, kg.	3.00	2.86	2.68	3.04	0.28	NS
Gain/day, kg.	0.429	0.452	0.383	0.436	0.04	NS
Total feed solids, kg.	3.89	4.44	4.37	4.44	0.36	NS
Feed solids/day, kg.	0.555	0.634	0.625	0.634	0.05	NS
Feed efficiency ^c	1.35	1.86	1.68	1.69	0.16	NS

TABLE XX CONTINUED

42 to 49 days							
No. of pigs	17	19	18	22			
Total gain, kg.	2.66	3.01	2.87	3.18	0.20		NS
Gain/day, kg.	0.380	0.431	0.411	0.455	0.03		NS
Total feed solids, kg.	5.38	5.67	5.64	5.69	0.39		NS
Feed solids/day, kg.	0.769	0.810	0.839	0.813	0.06		NS
Feed efficiency ^c	2.34	2.20	1.97	1.79	0.23		NS
49 to 56 days							
No. of pigs	16	19	18	22			
Total gain, kg.	3.26	3.28	3.83	3.98	0.23		NS
Gain/day, kg.	0.466	0.468	0.547	0.568	0.03		NS
Total feed solids, kg.	5.60	6.10	6.37	6.56	0.38		NS
Feed solids/day, kg.	0.800	0.881	0.909	0.936	0.06		NS
Feed efficiency ^c	1.75	2.21	1.75	1.64	0.16		D<B*
56 to 63 days							
No. of pigs	16	19	18	22			
Total gain, kg.	2.66	3.28	3.73	3.93	0.32		(C&D)>A*
Gain/day, kg.	0.381	0.469	0.532	0.561	0.05		(C&D)>A*
Total feed solids, kg.	5.94	6.38	7.42	7.49	0.54		NS
Feed solids/day, kg.	0.849	0.912	1.059	1.069	0.08		NS
Feed efficiency ^c	1.59	1.92	1.84	2.13	0.22		NS

^aStandard error of treatment means. Calculated standard errors are based on twenty pigs per treatment mean.

^bSignificant differences. * $P < .05$, NS = non-significant ($P > .05$).

^cKg. of feed solids per kg. of body weight gain.

calcium was highest on Ration D, which contained the most calcium.

The maintenance of normal serum calcium concentrations even in pigs fed low dietary calcium levels might be explained by the function of parathyroid hormone in maintenance of serum calcium homeostasis (Sherwood et al., 1966; Harrison, 1966). Bronner and Aubert (1965) showed that blood calcium levels in rats remained constant over a wide range of daily calcium absorption amounts. Measure of the calcium deposition and resorption rates in bone showed that the former changed only little with increasing absorption; whereas, the latter decreased nearly linearly under the same conditions. Thus, calcium resorption from bone appeared to play the major role in regulating the blood calcium level.

If a pig is capable of maintaining normal serum calcium levels in the face of calcium deprivation, then obviously serum calcium is not an appropriate assessor of the calcium status of that pig.

Calcium values did not vary appreciably with age (time on feed); however, one pattern was consistent over all treatments; initial concentrations were lowest, 3 week values highest and final levels were intermediate.

Ullrey et al. (1967) studied the serum calcium levels of pigs at birth and at the following postpartum intervals: 24, 48 and 72 hours; 5 and 7 days; 2, 3 and 5 weeks; and 2, 3, 4 and 5 months. Mean calcium values did not vary appreciably with age, and ranged from 9.3 to 11.8 mg. per 100 ml.

Miller et al. (1962b, 1964b, 1965c) have published similar calcium values in adequately nourished young pigs reared on semipurified diets. Brown et al. (1966) fed swine rations containing 0.8 percent calcium and 0.8 percent phosphorus and found serum calcium values at birth and at 1, 2, 3 and 4 months of age which were similar to, or slightly higher than, the values of Ullrey et al. (1967).

The concentration of inorganic phosphorus (determined as phosphate but calculated as phosphorus) in blood serum can range from 5 to 8 mg. per 100 ml. (Dukes, 1955).

Mean values in this study ranged from 7.19 to 10.01 mg. per 100 ml. (Table XXI).

Ullrey et al. (1967) stated that serum inorganic phosphorus concentration was 5.3 mg. per 100 ml. at birth, rose to 11.6 mg. per 100 ml. at 2 weeks and then gradually declined to 7.1 mg. per 100 ml. at 5 months. This same trend has been reported in sheep (Long et al., 1965b). McClellan et al. (1966), working with miniature swine, have also observed a decrease in serum phosphorus from weaning at 6 weeks (11 mg. per 100 ml.) to an apparently stable concentration (5 mg. per 100 ml.) at 4 years of age.

Brown et al. (1966) reported mean serum inorganic phosphorus concentrations at birth of 3.2 mg. per 100 ml. and 5.2 mg. per 100 ml. at weaning (4 weeks). However, in another experiment reported in the same article, pigs had serum phosphorus values of 11.5, 8.0, 6.2 and 5.4 mg. per 100 ml. at 1 (weaning), 2, 3 and 4 months, respectively.

The initial levels of serum inorganic phosphorus in this study were lower in pigs assigned to rations A and B ($P < .05$). No explanation for this observation is apparent. The initial values for all treatment groups were somewhat lower than normal, indicating that probably some depletion had occurred during the adjustment period.

By 3 weeks, serum inorganic phosphorus concentrations were restored to normal levels, although Ration D still maintained a higher level than Ration A ($P < .01$). This statistical advantage had disappeared by 6 weeks on feed.

Serum inorganic phosphorus was maximum at 6 weeks in pigs receiving 0.610 percent dietary phosphorus (Ration C). This was in agreement with work by Miller et al. (1961c).

Work with sheep (Becker and Smith, 1950) and pigs (Miller et al., 1964b) has furnished evidence that serum inorganic phosphorus concentration is quite subject to dietary phosphorus intake. Despite these findings, Brown et al. (1966), using rations which varied considerably from the N.R.C. (1964) recommendations, containing 0.18 percent calcium and 0.35 percent phosphorus in one experiment and 0.35 percent calcium and 1.40 percent phosphorus in a second, found the same general trend in serum inorganic phosphorus concentration changes with age as that published by Ullrey et al. (1967).

Dudley et al. (1959), using four equal increments of dietary phosphorus from 0 to 0.3 percent, demonstrated a significant nonlinear regression of blood inorganic phosphorus

on phosphorus dosage, the increment of response increasing with each increment of dose. The linear regression on dose was 0.60, 1.78 and 1.01 mg. percent per 0.1 percent of phosphorus at 7 a.m., 10 a.m. and 4 p.m., respectively; these values differed significantly ($P < .01$).

The influence of ration calcium and phosphorus levels on serum calcium to serum inorganic phosphorus ratios are given in Table XXI. Ration A significantly ($P < .01$) raised the ratio at 3 weeks on feed, but at the 6 week sampling period this difference had disappeared and all ratios were very similar.

Serum calcium x inorganic phosphorus products in this study (Table XXI) varied somewhat with time, but mean treatment differences within sampling periods were small and statistically nonsignificant. There was, however, a slight elevation of the products with each increment in dietary calcium and phosphorus.

There is apparently a reciprocal relationship between serum calcium and serum phosphorus, so that the product of calcium x phosphorus is maintained at a fairly constant level. As the serum calcium level rises, the serum phosphorus level falls, and vice versa (Netter, 1965).

Dunlop (1935) observed that, in general, the concentration of blood inorganic phosphorus tended to vary inversely with the serum calcium. Harmon et al. (1964) found that serum calcium values were significantly decreased with increasing levels of dietary phosphorus.

Ullrey et al. (1967) reported that within-age correlations of serum calcium and phosphorus were generally low and nonsignificant. However, significant correlation coefficients of 0.80, 0.53 and 0.81 were obtained at 72 hours, 1 week and 3 weeks, respectively.

Serum alkaline phosphatase activity reflects various nutritional states (Combs et al., 1955; Hibbs et al., 1945; Correll and Wise, 1938), pathological conditions (Kay, 1930), and the concentration of this enzyme declines with age in dogs (Bodansky, 1934) and foals (Earle, 1952).

Mean serum alkaline phosphatase values are shown in Table XXI. Serum alkaline phosphatase activity is, in part, a reflection of osteoblastic activity. Therefore, it is not surprising that the initial values were quite high early in life and declined with age. According to Young and Underdahl (1948), activity is highest in the pig at birth, falls approximately one-half by the fifth day, levels off at about 28 days and gradually declines to 56 days post-partum.

Long et al. (1965a) determined serum alkaline phosphatase activity in the pig at intervals from birth to 4 weeks of age. Although appreciable variation was apparent within and between litters, the mean values declined abruptly between 2 and 7 days in four of five litters and continued to decline in those litters examined to 28 days. Serum alkaline phosphatase activity was negatively correlated (-.53) with body weight ($P < .05$). It would appear that as body mass increased, the phosphatase available for maintenance of serum

TABLE XXI

SERUM CALCIUM, PHOSPHORUS, CALCIUM TO PHOSPHORUS RATIOS, CALCIUM TIMES
PHOSPHORUS PRODUCTS AND ALKALINE PHOSPHATASE LEVELS OF PIGS
FED DIFFERENT LEVELS OF CALCIUM (Ca) AND PHOSPHORUS (P)
FROM THREE TO NINE WEEKS OF AGE

Dietary Ca, %	0.368	0.569	0.776	0.952	SE ^a	SD ^b	
Dietary P, %	0.279	0.443	0.610	0.729			
Ration Designation	A	B	C	D			
<u>Weeks on Feed</u>							
	Serum calcium, mg./100 ml.						
0	11.00(18) ^c	11.03(20)	11.20(19)	11.11(22)	0.35	NS	
3	12.55(17) ^c	12.35(19)	12.21(18)	12.28(22)	0.26	NS	
6	11.18(16) ^c	11.23(19)	11.41(18)	11.85(22)	0.25	NS	
	Serum inorganic phosphorus, mg./100 ml.						
0	7.19(18) ^c	7.33(20)	8.02(19)	8.01(22)	0.22	(C&D)>A&B*	
3	8.09(17) ^c	9.00(19)	9.25(18)	10.01(22)	0.42	D>A**	
6	8.29(16) ^c	8.57(19)	9.17(18)	8.85(22)	0.28	NS	
	Serum calcium to inorganic phosphorus ratio						
0	1.54(18) ^c	1.53(20)	1.43(19)	1.40(22)	0.06	NS	
3	1.66(17) ^c	1.38(19)	1.33(18)	1.28(22)	0.07	A>(B,C&D)**	
6	1.37(16) ^c	1.33(19)	1.27(18)	1.36(22)	0.05	NS	
	Serum calcium X inorganic phosphorus product						
0	79.12(18) ^c	80.56(20)	90.76(19)	89.10(22)	4.14	NS	
3	102.56(17) ^c	111.42(19)	113.81(18)	123.18(22)	6.12	NS	
6	93.73(16) ^c	96.39(19)	104.79(18)	104.82(22)	4.36	NS	
	Serum alkaline phosphatase, Klein-Babson-Read units						
0	28.09(18) ^c	29.13(20)	25.65(19)	25.62(22)	1.44	NS	
3	25.02(17) ^c	19.42(19)	17.16(18)	15.57(22)	1.27	A>(B,C&D)**	
6	16.47(16) ^c	12.91(19)	12.77(18)	10.59(22)	1.34	B>D** A>D**	

^aStandard error of treatment means. Calculated standard errors are based on twenty pigs per treatment mean.

^bSignificant differences. *P<.05, **P<.01, NS = non-significant (P>.05).

^cNumber of animals included in the treatment mean.

levels declined.

The rates of decline in serum alkaline phosphatase concentration with time was dependent upon the dietary treatment, becoming more rapid with each increase in ration calcium and phosphorus content (Figure 8).

The serum alkaline phosphatase level becomes elevated in baby pigs with dietary deficiencies of calcium (Miller et al., 1962b; Dunlop, 1935) and phosphorus (Miller et al., 1964b). Miller et al. (1961c) observed a significant decrease in serum alkaline phosphatase as dietary phosphorus was increased from 0.2 to 0.4 percent and from 0.4 to 0.6 percent.

Brown et al. (1966) stated that as a general rule, serum alkaline phosphatase was inversely related to serum calcium, although the increase-decrease was somewhat delayed compared to the decrease-increase of serum calcium. Dunlop (1935) found blood alkaline phosphatase to be at its lowest concentration when the dietary calcium to phosphorus ratio was 1:2.

Cabell and Earle (1964) noted an elevation in serum alkaline phosphatase in rats when additional dietary zinc was supplied. Similar findings have been reported in reproducing gilts (Hoekstra et al., 1965), in baby pigs (Miller et al., 1965a) and in weanling pigs (Roberts et al., 1961).

Increasing dietary protein (Hendricks et al., 1965) and substituting isolated soybean protein for casein (Hendricks et al., 1966) have resulted in increased serum alkaline

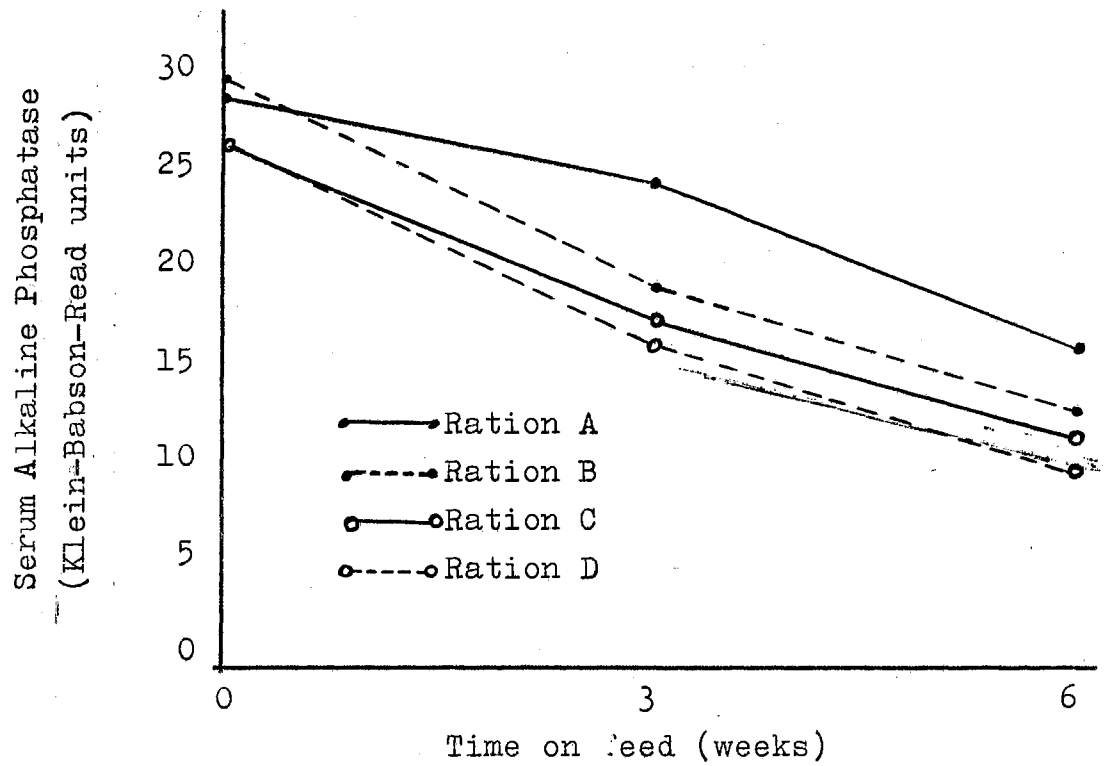


Figure 8. Influence of Dietary Calcium and Phosphorus Levels on Serum Alkaline Phosphatase Values

phosphatase concentrations.

At all sampling periods in this study, males had slightly higher serum alkaline phosphatase levels than females, but these differences were small and nonsignificant statistically. Combs et al. (1959) determined the phosphatase activity of 321 Duroc, Hampshire, Spotted Poland China and Cross-bred pigs when they were 1 and 7 days of age. Both sex and breed exerted a significant influence on the phosphatase activity of 1 day old pigs. The females in all breeds exhibited a higher activity than the males. When the pigs reached 7 days of age, phosphatase activity was considerably lower than initially; and breed, but not sex, still had a significant influence. Fletcher et al. (1956) and Kunkel et al. (1953), working with cattle, observed a similar situation regarding the effects of sex and breed on phosphatase activity.

The mean values of hematocrit, hemoglobin, erythrocyte population and leukocyte counts are presented in Table XXII. No significant differences were observed in these variables, nor were any trends immediately obvious.

Mean corpuscular volume (M.C.V.), mean corpuscular hemoglobin (M.C.H.) and mean corpuscular hemoglobin concentration (M.C.H.C.) values were calculated. As indicated by the results shown in Table XXIII, none of the parameters calculated were significantly influenced by dietary treatment.

The average M.C.V. value, averaged over treatments and sampling periods, was $65.9\mu\text{3}$. This is similar to values ob-

TABLE XXII

BLOOD HEMATOCRIT, HEMOGLOBIN, RED BLOOD CELL AND WHITE BLOOD
CELL COUNTS OF PIGS FED DIFFERENT LEVELS OF CALCIUM (Ca)
AND PHOSPHORUS (P) FROM THREE TO NINE WEEKS OF AGE

Dietary Ca, %	0.368	0.569	0.776	0.952			
Dietary P, %	0.279	0.443	0.610	0.729	SE ^a	SD ^b	
Ration Designation	A	B	C	D			
Weeks on Feed							
		Hematocrit, %					
0	31.1(18) ^c	31.1(20)	33.8(19)	32.5(22)	1.8	NS	
3	39.1(17) ^c	39.8(19)	37.4(18)	37.8(22)	2.1	NS	
6	35.9(16)	38.3(19)	35.8(18)	33.8(22)	2.2	NS	
		Hemoglobin, gm./100 ml. blood					
0	10.2(18) ^c	10.8(20)	10.4(19)	10.9(22)	0.5	NS	
3	11.5(17) ^c	12.4(19)	11.7(18)	12.2(22)	0.5	NS	
6	11.3(16) ^c	12.4(19)	11.8(18)	11.2(22)	0.5	NS	
		Red blood cells, millions/cubic mm.					
0	5.13(18) ^c	5.02(20)	5.71(19)	5.32(22)	0.35	NS	
3	5.51(17) ^c	5.68(19)	5.82(18)	6.02(22)	0.32	NS	
6	5.24(16) ^c	5.65(19)	5.23(18)	5.18(22)	0.22	NS	
		White blood cells, thousands/cubic mm.					
0	8.21(18) ^c	8.41(20)	8.23(19)	9.33(22)	0.85	NS	
3	8.64(17) ^c	9.94(19)	11.85(18)	10.82(22)	1.06	NS	
6	9.72(16) ^c	11.50(19)	9.11(18)	8.14(22)	1.15	NS	

^aStandard error of treatment means. Calculated standard errors are based on twenty pigs per treatment mean.

^bSignificant differences. NS = non-significant (P>.05).

^cNumber of animals included in the treatment mean.

TABLE XXIII

MEAN CORPUSCULAR VOLUME, MEAN CORPUSCULAR HEMOGLOBIN AND MEAN CORPUSCULAR HEMOGLOBIN CONCENTRATION OF PIGS FED DIFFERENT LEVELS OF CALCIUM (Ca) AND PHOSPHORUS (P) FROM THREE TO NINE WEEKS OF AGE

Dietary Ca, %	0.368	0.569	0.776	0.952	SE ^a	SD ^b	
Dietary P, %	0.279	0.443	0.610	0.729			
Ration Designation	A	B	C	D			
Weeks on Feed		Mean corpuscular volume, cubic microns					
0	63.22(18) ^c	64.01(20)	62.53(19)	60.66(22)	4.23	NS	
3	71.56(17) ^c	71.70(19)	67.14(18)	65.38(22)	4.08	NS	
6	68.21(16) ^c	68.39(19)	69.44(18)	60.86(22)	4.27	NS	
		Mean corpuscular hemoglobin, micro-mcg.					
0	20.68(18) ^c	22.18(20)	19.30(19)	20.95(22)	1.07	NS	
3	21.06(17) ^c	22.21(19)	21.28(18)	21.19(22)	1.02	NS	
6	21.57(16) ^c	22.18(19)	22.89(18)	22.40(22)	1.00	NS	
		Mean corpuscular hemoglobin concentration, %					
0	34.54(18) ^c	36.39(20)	31.97(19)	34.05(22)	1.75	NS	
3	30.64(17) ^c	31.71(19)	32.77(18)	33.46(22)	1.44	NS	
6	33.08(16) ^c	33.12(19)	34.66(18)	35.44(22)	1.59	NS	

^aStandard error of treatment means. Calculated standard errors are based on twenty pigs per treatment mean.

^bSignificant differences. NS = non-significant (P>.05).

^cNumber of animals included in the treatment mean.

served by Swenson et al. (1958), Waddill (1960) and Miller et al., 1961b).

Sex did not appear to influence M.C.V. significantly although the cells of males were more frequently larger than females. In contrast, observations in pigs (Miller et al., 1961b) and in humans (Anderson and Mugarage, 1936) have shown females to have higher M.C.V. values.

The M.C.H. value, averaged over treatments and sampling periods, was 21.5 μg . This value is slightly higher than values reported by Miller et al. (1961b) and Swenson et al. (1958).

The M.C.H.C., averaged over treatments and sampling periods, was 33.5 percent. This is slightly higher than values reported by Miller et al. (1961b).

At all sampling periods, females had slightly higher M.C.H.C. values than males, but these differences were small and nonsignificant. Similar observations were made by Miller et al. (1961b).

Since the calcium ion is essential for the conversion of prothrombin to thrombin in the blood clotting mechanism, it seemed that blood clotting time might be a sensitive measure of dietary calcium adequacy or at least a measure of circulating calcium.

The data presented in Table XXIV reveals no significant differences in blood clotting time between groups receiving different dietary calcium levels. Miller et al. (1962b) reported a prolonged clotting time in pigs receiving no die-

TABLE XXIV
 BLOOD CLOTTING TIME¹ OF PIGS FED DIFFERENT LEVELS OF
 CALCIUM (Ca) AND PHOSPHORUS (P) FROM THREE
 TO NINE WEEKS OF AGE

Dietary Ca, %	0.368	0.569	0.776	0.952	SE ^a	SD ^b
Dietary P, %	0.279	0.443	0.610	0.729		
Ration Designation	A	B	C	D		
Weeks on Feed						
0	1.54(18) ^c	1.48(20)	1.79(19)	1.45(22)	0.15	NS
3	1.59(17) ^c	1.50(19)	1.88(18)	1.60(22)	0.14	NS
6	1.37(16) ^c	1.42(19)	1.60(18)	1.28(22)	0.13	NS

¹Expressed in minutes.

^aStandard error of treatment means. Calculated standard errors are based on twenty pigs per treatment mean.

^bSignificant differences. NS = non-significant (P>.05).

^cNumber of animals included in the treatment mean.

tary calcium or 0.4 percent of calcium after 4 weeks; however, no significant differences were found between groups receiving higher levels of calcium, although pigs receiving 1.6 percent of calcium exhibited the shortest clotting time.

No attempt will be made to compare actual clotting time in this study to published values since even seemingly minimal variation in technique can make a considerable difference in the clotting time.

Mean slaughter weights were 17.31, 19.45, 22.09 and 22.44 kg. for Rations A, B, C and D, respectively (Table XXV).

Skeletal correctness scores (Table XXV) were given to grossly evaluate bone development and straightness and difficulty associated with standing and moving. No significant differences were observed; however, pigs assigned to Rations A and B had less desirable scores.

Hair and skin condition scores (Table XXV) were not significantly affected by the dietary treatments imposed. Nevertheless, pigs fed Ration A had rougher hair coats than did other treatment groups.

At slaughter, nasal turbinates were grossly observed for evidence of atrophy. Turbinates were scored (Table XXV) in an attempt to evaluate them from the standpoint of amount of bone present, volume and distortion of dorsal and ventral turbinates and deviation of the septum. Although not statistically significant, poorer scores were given to pigs supplied Rations A and B. The largest differences were apparent

TABLE XXV

INCIDENCE OF ATROPHIC RHINITIS (AR) AND VISUAL SCORES FOR SKELETAL CORRECTNESS,
 NASAL TURBINATES AND HAIR AND SKIN CONDITION OF NINE WEEK OLD PIGS FED
 DIFFERENT LEVELS OF CALCIUM (Ca) AND PHOSPHORUS (P) FROM
 THREE TO NINE WEEKS OF AGE

Dietary Ca, %	0.368	0.569	0.776	0.952		
Dietary P, %	0.279	0.443	0.610	0.729	SE ^a	SD ^b
Ration Designation	A	B	C	D		
Number of pigs	6	7	6	6		
Slaughter wt., kg.	17.31	19.45	22.09	22.44	1.58	NS
Pig condition scores						
Skeletal correctness ^c	2.9	2.9	2.3	2.5	0.1	NS
Hair and skin ^d	3.0	1.7	1.8	1.5	0.2	NS
Nasal turbinate atrophy ^e	1.8	1.7	1.3	1.2	0.3	NS
Number with AR ^f	0	0	0	0	0	NS
Incidence of AR, %	0	0	0	0	0	NS

^aStandard error of treatment means. Standard error when seven per treatment is the reported standard error times $\sqrt{6/7}$.

^bSignificant differences. NS = non-significant (P<.05).

^c1=normal, 2=slightly crooked, 3=moderately crooked, 4=very crooked and 5=severe crookedness.

^d1=normal, 2=slightly dry and rough, 3=moderately dry and rough, 4=very dry and rough and 5=severely dry and rough.

^e1=normal, 2=slight, 3=moderate, 4=moderately severe and 5=severe.

^fRepresents only those with gross evidence of nasal turbinate atrophy; microscopic and cultural examinations were not made. Gross examinations were performed by the Oklahoma State University Veterinary Pathology Department.

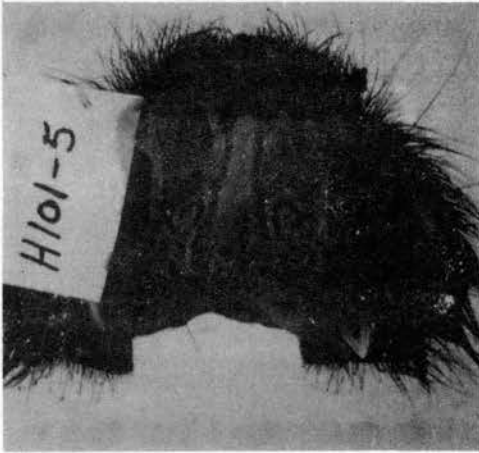
in the ventral turbinates. None of the pigs were diagnosed as having atrophic rhinitis. Figure 9 shows a nasal turbinate from each of the treatment groups.

Absolute bone diameter and length data are presented in Table XXVI. With the exception of femur diameter, no significant differences were observed. However, femur diameter, 8th rib length and ulna-radius length were maximum in pigs receiving Ration D. Maximum rib and ulna-radius diameters and humerus length were obtained in pigs provided Ration C.

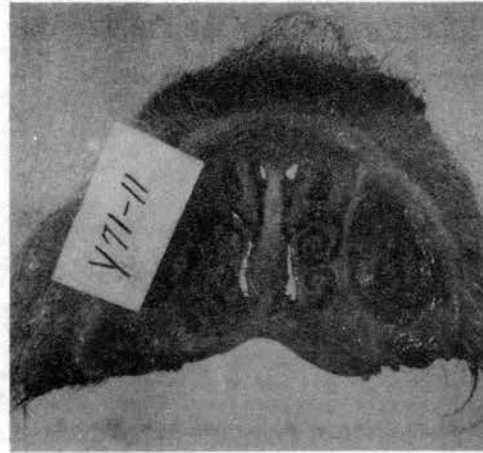
Since slaughter weights differed among treatment groups (Table XXV), it was considered advantageous to adjust for body weights by expressing bone diameters and lengths on a relative basis. That is, in mm. per kg. of slaughter body weight. No significant differences were found after calculating these relative bone values (Table XXVI). But there was a trend toward increased relative bone diameters and lengths with decreasing ration calcium and phosphorus.

Differences in absolute diameter to length ratios for all bones were surprisingly small and statistically nonsignificant (Table XXVI). This would tend to indicate that bone diameter and length increased at approximately the same rate regardless of the dietary treatment imposed and that dimensional bone growth tended to keep pace with the development of soft tissue.

Fresh weights of the 8th rib, humerus, femur and ulna-radius (Table XXVII), from which all soft tissue and periosteum had been removed, did not differ significantly. How-



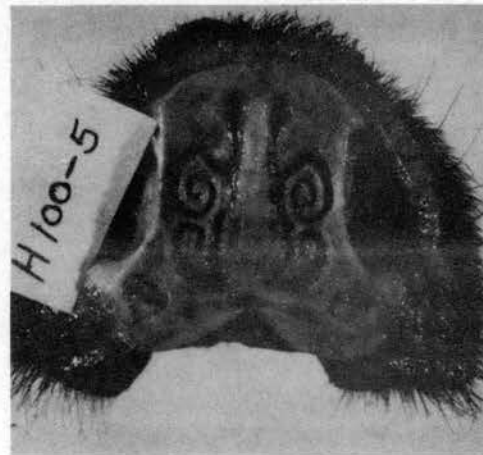
Ration A



Ration B



Ration C



Ration D

Figure 9. Cross-section of the Snout through the First Maxillary Premolar Teeth of Representative Pigs from Groups Fed Different Levels of Calcium and Phosphorus from Three to Nine Weeks of Age

TABLE XXVI

INFLUENCE OF DIETARY CALCIUM (Ca) AND PHOSPHORUS (P) LEVELS ON ABSOLUTE
AND RELATIVE BONE DIAMETER AND LENGTH OF PIGS
SACRIFICED AT NINE WEEKS OF AGE

Dietary Ca, %	0.368	0.569	0.776	0.952	SE ^a	SD ^b
Dietary P, %	0.279	0.443	0.610	0.729		
Ration Designation	A	B	C	D		
Number of pigs	6	7	6	6		
			Absolute diameter, mm.			
8th rib	5.6	5.9	6.4	6.3	0.4	NS
Humerus	12.8	13.2	13.9	14.3	0.6	NS
Femur	14.5	16.1	17.2	17.3	0.8	D>A**,B* C>A*
Ulna-radius	25.3	25.0	27.5	27.2	1.0	NS
			Absolute length, mm.			
8th rib	101.9	106.0	109.5	113.7	4.5	NS
Humerus	97.4	101.2	106.0	105.3	3.5	NS
Femur	109.2	111.7	116.7	121.9	4.1	NS
Ulna-radius	116.3	119.8	124.1	127.5	3.8	NS
			Relative diameter, mm./kg. live body wt.			
8th rib	0.33	0.32	0.29	0.29	0.02	NS
Humerus	0.74	0.71	0.64	0.65	0.05	NS
Femur	0.84	0.87	0.79	0.79	0.06	NS
Ulna-radius	1.48	1.36	1.26	1.23	0.09	NS
			Relative length, mm./kg. live body wt.			
8th rib	5.95	5.76	5.08	5.13	0.43	NS
Humerus	5.68	5.49	4.89	4.80	0.40	NS
Femur	6.36	6.05	5.39	5.50	0.43	NS
Ulna-radius	6.79	6.43	5.73	5.81	0.40	NS
			Ratio of absolute diameter to length			
8th rib	1:18.4	1:18.2	1:17.7	1:18.1	1.3	NS
Humerus	1:7.6	1:7.7	1:7.6	1:7.4	0.2	NS
Femur	1:7.6	1:7.0	1:6.8	1:7.0	0.3	NS
Ulna-radius	1:4.6	1:4.8	1:4.5	1:4.7	0.1	NS

^aStandard error of treatment means. Standard error when seven per treatment is the reported standard error times $\sqrt{5/7}$.

^bSignificant differences. *P<.05, **P<.01, NS = non-significant (P<.05).

ever, in all cases heavier bones were obtained with each successive increase in dietary calcium and phosphorus. This is graphically presented in Figure 10.

Chapman et al. (1962) found femur weight to be positively correlated with dietary phosphorus level and Miller et al. (1964b) reported that both femur and 8th rib fresh weights were increased through their highest phosphorus level (0.6 percent). Miller et al. (1962b) obtained maximum mean fresh 8th rib, femur and humerus weights whenever the ration calcium levels were 1.2, 1.2 and 1.6 percent, respectively.

Miller et al. (1967) reported that a calcium deficiency in young pigs resulted in significantly reduced actual femur and humerus weights. Although not significant, actual 8th rib weights were also appreciably decreased. A phosphorus deficiency resulted in significantly reduced actual femur and 8th rib weights.

Relative bone weights are compared in Table XXVII. Rib and femur relative weights were increased with each increment in ration calcium and phosphorus. Miller et al. (1967) reported that relative weights (percent of body weight) of femur and 8th rib were significantly greater in phosphorus deficient pigs than in control animals. This was in contrast to their findings in calcium and vitamin D deficiencies where relative bone weights were unaltered. Blood serum investigations (Miller et al., 1962b, 1964c) indicated a primary lack of calcium in these two nutrient deficiencies, while a dietary phosphorus deficiency (Miller et al., 1964b)

TABLE XXVII

INFLUENCE OF DIETARY CALCIUM (Ca) AND PHOSPHORUS (P) LEVELS ON
ABSOLUTE AND RELATIVE BONE WEIGHTS OF PIGS
SACRIFICED AT NINE WEEKS OF AGE

Dietary Ca, %	0.368	0.569	0.776	0.952			
Dietary P, %	0.279	0.443	0.610	0.729	SE ^a	SD ^b	
Ration Designation	A	B	C	D			
Number of pigs	6	7	6	6			
		Absolute weights, gm.					
8th rib	6.3	8.2	9.8	10.6	1.4	NS	
Humerus	66.1	71.3	84.6	88.3	7.4	NS	
Femur	73.9	84.6	98.6	102.0	9.2	NS	
Ulna-radius	50.8	53.1	63.9	67.3	5.7	NS	
		Relative weights, units ^c /kg. live body wt.					
8th rib	358.8	398.1	440.3	478.7	53.6	NS	
Humerus	3.8	3.7	3.8	4.0	0.2	NS	
Femur	4.2	4.4	4.5	4.6	0.3	NS	
Ulna-radius	2.9	2.7	2.9	3.0	0.2	NS	

^aStandard error treatment means. Standard error when seven per treatment is the reported standard error times $\sqrt{6/7}$.

^bSignificant differences. NS = non-significant ($P > .05$).

^cUnit for 8th rib is mg. and all other units reported are in gm.

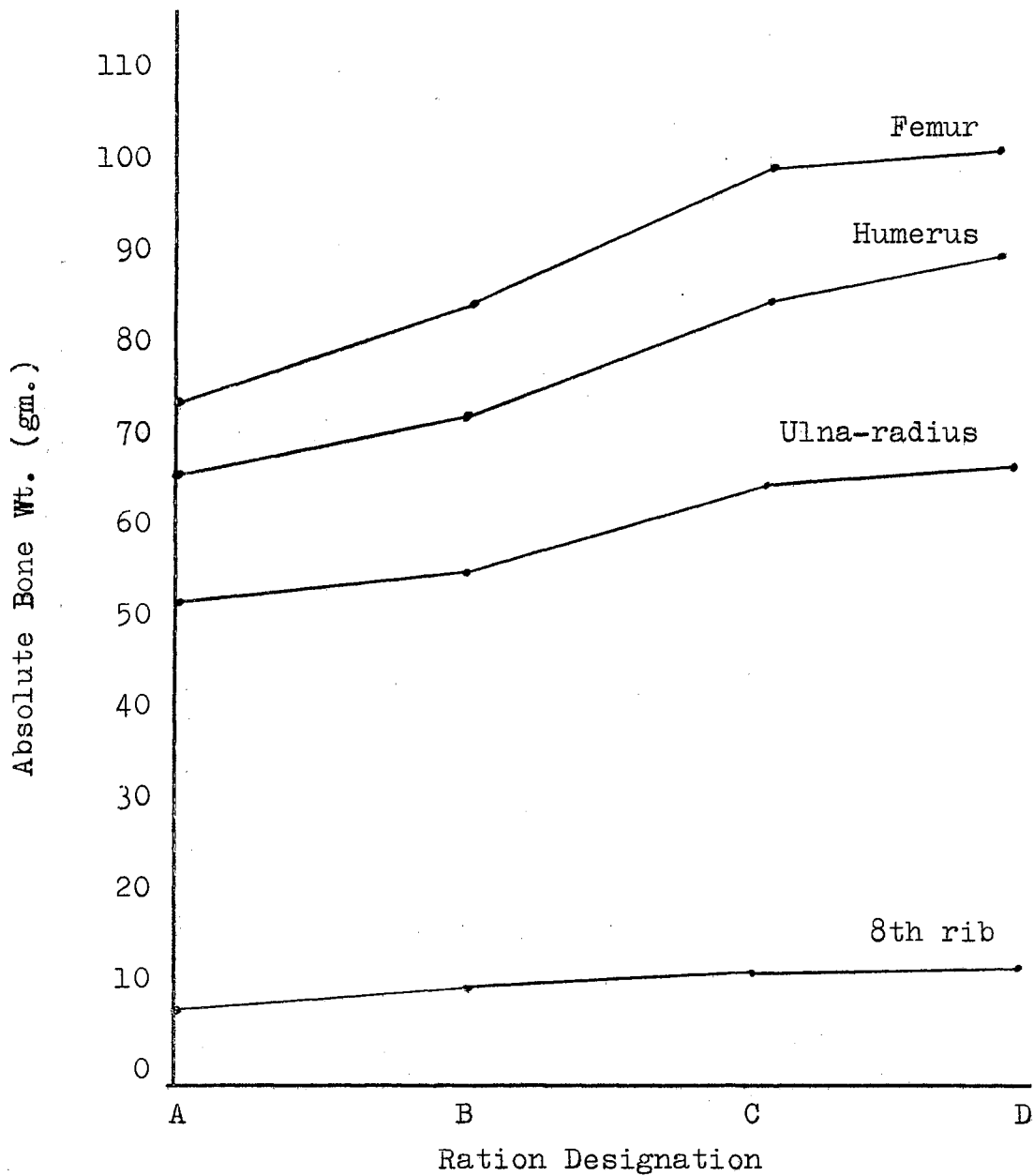


Figure 10. Influence of Ration Calcium and Phosphorus Levels on Fresh Bone Weights of Pigs Slaughtered at Nine Weeks of Age

indicated a primary lack of calcium in these two nutrient deficiencies, while a dietary phosphorus deficiency (Miller et al., 1964b) resulted in an apparent excess of usable calcium. It is apparent from these studies that phosphorus deficiency has a more direct effect upon the growth of non-skeletal tissue in the baby pig than does calcium deficiency.

Bone specific gravity was positively related to the ration calcium and phosphorus level (Table XVIII), but was not significantly increased by increasing calcium and phosphorus beyond 0.776 and 0.610 percent, respectively.

Without exception, specific gravity values were increased with successive increments of dietary calcium and phosphorus (Figure 11).

Miller et al. (1962b) found that humerus, femur and 8th rib specific gravity values were increased by dietary calcium levels up through 1.2 percent but these increases were not significant beyond 0.8 percent calcium. Newman et al. (1967) obtained maximum femur specific gravity values in pigs receiving 0.60 percent calcium.

Rib and femur specific gravity are positively related to dietary phosphorus levels (Miller et al., 1961c). Vandepopuliere et al. (1959) noted a linear response in bone density when phosphorus levels of 0.24, 0.36, 0.48, 0.60 and 0.72 percent were fed. However, Newman et al. (1964b, 1967) reported that femur specific gravity was not increased by dietary phosphorus levels above 0.45 percent. Miller et al. (1964b) obtained maximum humerus and 8th rib specific gravity

TABLE XXVIII

SPECIFIC GRAVITY OF BONES TAKEN FROM NINE WEEK OLD PIGS FED
DIFFERENT LEVELS OF CALCIUM (Ca) AND PHOSPHORUS (P)
FROM THREE TO NINE WEEKS OF AGE

Dietary Ca, %	0.368	0.569	0.776	0.952	SE ^a	SD ^b
Dietary P, %	0.279	0.443	0.610	0.729		
Ration Designation	A	B	C	D		
Number of pigs	6	7	6	6		
		Specific gravity				
8th rib	1.1118	1.1646	1.2090	1.2137	0.0182	A<C*, D**
Humerus	1.1390	1.1658	1.1875	1.2077	0.0075	A<B*, C** (A&B)<D**
Femur	1.1283	1.1576	1.1808	1.1973	0.0077	(A&B)<D** A<B*, C** B<C*
Ulna-radius	1.1542	1.1884	1.2065	1.2208	0.0080	A<(B, C&D)** B<C*

^aStandard error of treatment means. Standard error when seven per treatment is the reported standard error times $\sqrt{6/7}$.

^bSignificant differences. *P<.05, **P<.01.

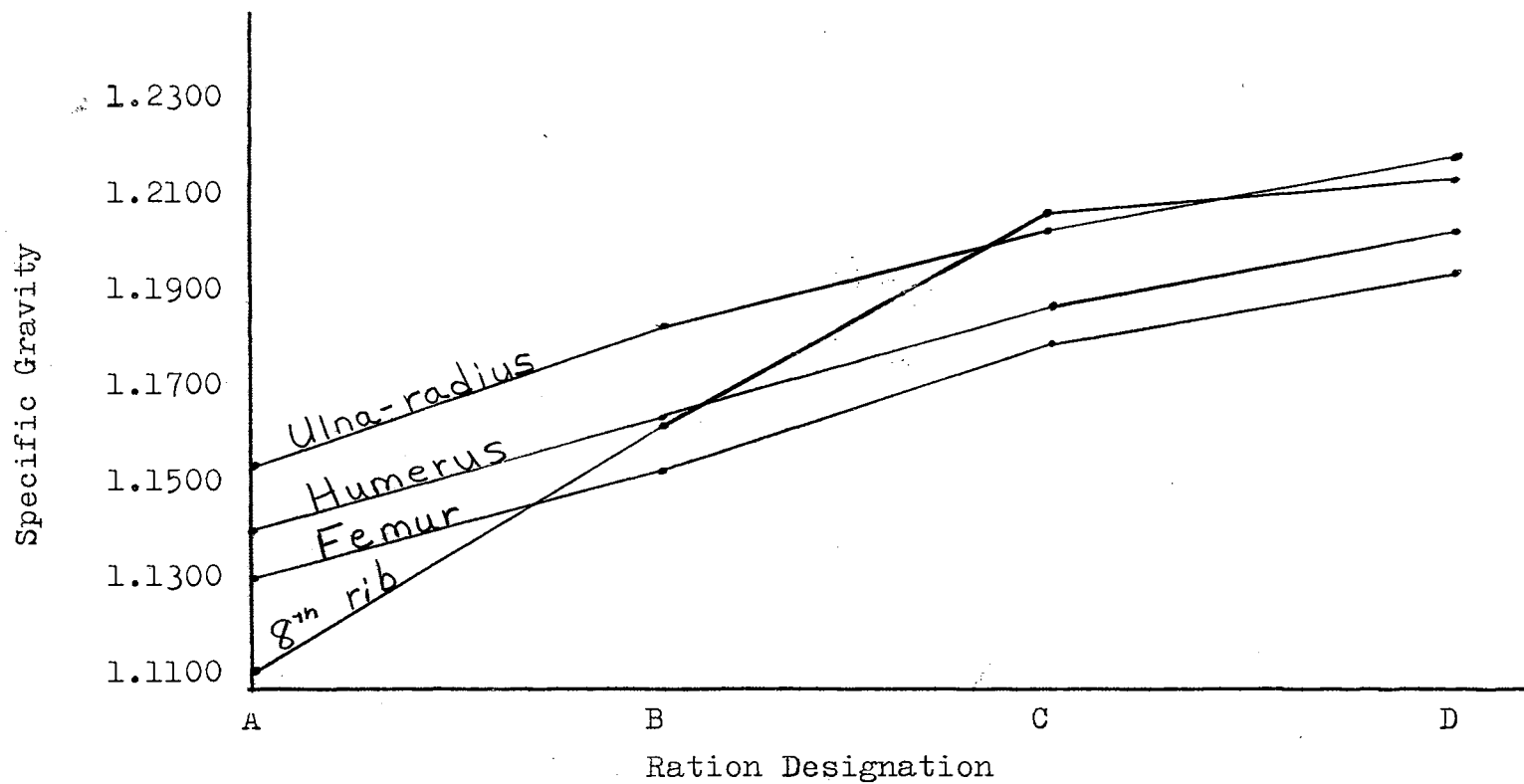


Figure 11. Influence of Ration Calcium and Phosphorus Levels on Specific Gravity of Bones from Pigs Slaughtered at Nine Weeks of Age

values when the dietary phosphorus levels were 0.5 and 0.6 percent, respectively.

Mean values of bone ash, calcium, phosphorus and bone calcium:phosphorus ratio are presented in Table XXIX. With the exception of humeral calcium and phosphorus, ash, calcium and phosphorus of each of the bones were increased with successive increases of dietary calcium and phosphorus; however, none of these values were significantly increased beyond values for pigs receiving Ration C (0.776 percent calcium and 0.610 percent phosphorus). Normal serum calcium and inorganic phosphorus levels (Table XXI) did not guarantee a normal state of bone nutrition.

Numerous workers have demonstrated that pig rations low in calcium and phosphorus will decrease bone ash values (Eggert et al., 1959; Vandepopuliere et al., 1959; Miller et al., 1960; Zimmerman et al., 1961; Blair and Benzie, 1964). Femur ash has been increased with all increments of calcium up to at least 2.0 percent (Dudley et al., 1961) and with increments of phosphorus to 0.80 percent (Miller et al., 1961c). However, Combs et al. (1966b) reported that neither femur nor fibula ash percent was significantly influenced by increasing dietary calcium from 0.48 to 1.32 percent.

The influence of calcium and phosphorus levels on actual and relative organ and gland weights is presented in Tables XXX and XXXI, respectively. With the exceptions of the thyroid and lungs, no significant differences were apparent in absolute weights. These two differences disappeared when

TABLE XXIX

ASH, CALCIUM AND PHOSPHORUS CONTENT OF BONES TAKEN FROM NINE WEEK OLD PIGS FED DIFFERENT LEVELS OF CALCIUM (Ca) AND PHOSPHORUS (P) FROM THREE TO NINE WEEKS OF AGE

Dietary Ca, ‰	0.368	0.569	0.776	0.952	SE ^a	SD ^b
Dietary P, ‰	0.279	0.443	0.610	0.729		
Ration Designation	A	B	C	D		
Number of pigs	6	7	6	6		
			8th rib ash ^c			
Ash, ‰	51.00	54.02	55.30	56.86	0.98	A<B*, C**, D**
Calcium, ‰	20.61	21.61	22.43	23.07	0.44	A<C*, D**
Phosphorus, ‰	8.97	9.11	10.15	10.73	0.31	(A&B)<C*, D**
Calcium/phosphorus	2.30	2.41	2.22	2.15	0.06	B>D*
			Humeral ash ^c			
Ash, ‰	59.88	63.14	66.26	67.44	1.09	A<B*, C**, D** B<D**
Calcium, ‰	23.59	26.22	26.87	26.85	0.73	A<B*, C**, D**
Phosphorus, ‰	10.07	11.76	12.65	12.34	0.33	A<B*, C**, D**
Calcium/phosphorus	2.34	2.23	2.13	2.18	0.04	A>C**, D*
			Femur ash ^c			
Ash, ‰	59.25	62.14	66.02	66.18	1.19	A<(C&D)** B<(C&D)*
Calcium, ‰	23.22	24.84	26.36	26.76	0.48	A<B*, C**, D** B<(C&D)*
Phosphorus, ‰	11.26	11.70	12.69	13.12	0.25	(A&B)<(C&D)**
Calcium/phosphorus	2.06	2.12	2.08	2.04	0.33	NS
			Ulna-radius ash ^c			
Ash, ‰	54.24	56.54	59.80	60.97	1.50	A<C*, D**
Calcium, ‰	21.71	23.16	23.91	24.49	0.66	A<(C&D)*
Phosphorus, ‰	10.04	10.43	11.00	11.11	0.42	NS
Calcium/phosphorus	2.17	2.22	2.18	2.21	0.06	NS

^aStandard error of treatment means. Standard error when seven per treatment is the reported standard error times $\sqrt{6/7}$.

^bSignificant differences. *P<.05, **P<.01, NS = non-significant (P>.05).

^cExpressed on a dry, fat-free basis.

TABLE XXX

INFLUENCE OF DIETARY CALCIUM (Ca) AND PHOSPHORUS (P) LEVELS
ON ABSOLUTE ORGAN AND GLAND WEIGHTS OF PIGS
SACRIFICED AT NINE WEEKS OF AGE

Dietary Ca, %	0.368	9.569	0.776	0.952	SE ^a	SD ^b
Dietary, P, %	0.279	0.443	0.610	0.729		
Ration Designation	A	B	C	D		
Number of pigs	6	7	6	6		
	Absolute weights, gm.					
Adrenals	1.5077	1.5857	1.7157	1.8766	0.2040	NS
Kidneys						
both	115.5	124.6	153.0	134.4	11.0	NS
left	57.2	61.3	77.8	68.3	6.0	NS
right	58.3	60.4	75.2	66.0	6.1	NS
Spleen	29.0	32.3	29.8	36.8	6.6	NS
Stomach	129.2	125.7	155.5	145.2	14.7	NS
Liver	480.5	506.7	578.3	568.0	47.6	NS
Heart	86.8	86.7	109.0	106.8	8.3	NS
Thymus	91.3	76.0	81.3	98.5	16.0	NS
Thyroid	1.8627	2.2900	2.5064	2.5406	0.1882	(C&D)>A*
Lungs	134.2	139.3	174.0	184.2	10.0	D>(A&B)** C>(A&B)*

^aStandard error of treatment means. Standard error when seven per treatment is the reported standard error times $\sqrt{6/7}$.

^bSignificant differences. *P<.05, **P<.01, NS = non-significant (P>.05).

TABLE XXXI

INFLUENCE OF DIETARY CALCIUM (Ca) AND PHOSPHORUS (P) LEVELS
ON RELATIVE ORGAN AND GLAND WEIGHTS OF PIGS
SACRIFICED AT NINE WEEKS OF AGE

Dietary Ca, %	0.368	0.569	0.776	0.952	SE ^a	SD ^b
Dietary P, %	0.279	0.443	0.610	0.729		
Ration Designation	A	B	C	D		
Number of pigs	6	7	6	6		
	Relative weights, units ^c /kg. live body wt.					
Adrenals	84.8	81.1	79.0	84.8	7.9	NS
Kidneys						
both	6.7	6.4	6.9	6.0	0.3	NS
left	3.3	3.1	3.5	3.0	0.2	NS
right	3.4	3.1	3.4	3.0	0.2	NS
Spleen	1.7	1.6	1.4	1.6	0.3	NS
Stomach	7.4	6.6	7.1	6.5	0.5	NS
Liver	27.6	26.2	26.0	25.4	0.1	NS
Heart	5.0	4.5	5.0	4.8	0.2	NS
Thymus	5.4	3.7	3.8	4.3	0.7	NS
Thyroid	109.1	118.2	117.9	114.2	9.8	NS
Lungs	7.8	7.2	8.0	8.3	0.4	NS

^aStandard error of treatment means. Standard error when seven per treatment is the reported standard error times $\sqrt{6/7}$.

^bSignificant differences. NS = non-significant ($P > .05$).

^cUnit for adrenals and thyroid is mg. and all other units reported are in gm.

weights were expressed on a relative basis (units per kg. of live body weight).

Data on normal body and organ weights of many species have been compiled (Altman and Dittmer, 1962) and organ data from young pigs reared along different growth curves have been published (Elsley, 1963).

Filer et al. (1966) have recently published data on the affects of dietary calcium and phosphorus upon organ weights in miniature swine.

Miller et al. (1967) reported that a calcium deficiency in young pigs resulted in significantly reduced actual liver weights and significantly increased relative weights of kidneys, heart and thyroid, as well as significantly increased actual and relative weights of adrenals. Cabell and Earle (1965) reported that relative rat kidney weights were lowered by high dietary levels of calcium.

A significant increase in relative kidney weight of calcium deficient pigs has been reported by Menahan et al. (1963). These workers observed increased relative adrenal weights of calcium deficient pigs; however, the degree of the deficiency was not as great as that in the study by Miller et al. (1967).

Storts and Koestner (1965) have shown an increase in relative thyroid weight of calcium deficient pigs.

Using baby pigs reared on synthetic milk diets which were either nutritionally complete or mono-nutrient-deficient, Miller et al. (1964a) studied the effects of calcium and

phosphorus deficiencies upon actual and relative organ weights. Reduced actual weights of the spleen, liver, and heart were observed. Relative weights of the spleen, liver, heart, thyroid and adrenals were increased, although only the adrenals and kidney weights were significantly increased. Specific dietary mineral levels used in investigation were not stated.

Miller et al. (1967) demonstrated that a phosphorus deficiency in baby pigs resulted in significantly reduced actual weights of liver, kidneys, heart, spleen, adrenals, and thyroid and significantly increased relative weights of each of the organs except the liver and thyroid.

Cabell and Earle (1965) observed that relative rat kidney weights were increased by high dietary levels of phosphorus.

Although not statistically significant, the calcium concentration of the left kidney (Table XXXII) was directly related to the level of dietary calcium. On a dry matter basis, the calcium content in the kidneys of pigs receiving the highest dietary calcium level (Ration D) was 22.4 percent higher than that present in the kidneys of animals fed the lowest calcium level. This increase in the calcium concentration of the left kidney, paralleling increments of dietary calcium, may be pertinent in view of the concern for the possible influence on renal function (Henneman, 1959).

Kidney phosphorus concentration was somewhat increased in pigs receiving the lowest level of calcium and phosphorus

TABLE XXXII

CALCIUM (Ca) AND PHOSPHORUS (P) CONTENT OF LEFT KIDNEY OF NINE WEEK
 OLD PIGS FED DIFFERENT LEVELS OF CALCIUM AND PHOSPHORUS
 FROM THREE TO NINE WEEKS OF AGE

Dietary Ca, %	0.368	0.569	0.776	0.952	SE ^a	SD ^b
Dietary P, %	0.279	0.443	0.610	0.729		
Ration Designation	A	B	C	D		
Number of pigs	6	7	6	6		
Left kidney						
Weight, fresh, gm.	57.2	61.3	77.8	68.3	6.0	NS
Dry matter, %	21.10	22.40	21.84	22.78	0.54	NS
Ash, dry basis, %	5.74	5.88	5.74	6.89	0.60	NS
Ca, fresh tissue mg./100 gm.	6.51	7.24	7.59	8.55	0.82	NS
Ca, dry basis mg./100 gm.	30.82	32.15	35.03	37.71	3.78	NS
Total left kidney Ca, mg.	3.75	4.46	5.87	5.52	0.52	(C&D)>A*
P, fresh tissue mg./100 gm.	302.50	295.37	273.28	300.19	8.50	(A&D)>C*
P, dry basis mg./100 gm.	1434.48	1321.04	1254.75	1325.10	48.29	A>C*
Total left kidney, P, mg.	173.12	180.54	212.69	204.33	18.30	NS
P/Ca, dry basis	47.63	40.87	36.61	38.69	2.91	NS

^aStandard error of treatment means. Standard error when seven per treatment is the reported standard error times $\sqrt{6/7}$.

^bSignificant differences. *P>.05, NS = non-significant (P>.05).

(Ration A). No explanation for this observation is apparent. However, Miller et al. (1964b) observed an elevated kidney phosphorus concentration in phosphorus-deficient pigs which received a ration containing 0.8 percent calcium and 0.2 percent phosphorus.

Calcium and phosphorus content of the heart and liver are presented in Tables XXXIII and XXXIV, respectively. No significant differences were found in any of the variables examined. Heart and liver calcium and phosphorus concentrations did not appear to bear the close relationship to the dietary intake of these two minerals observed in the kidney calcium and phosphorus analysis. However, total organ calcium and phosphorus for both heart and liver appeared to bear a direct relationship to dietary intake. These increases in total calcium are in agreement with the findings of Miller et al. (1962b).

Calcium content of the hair (Table XXV) did not differ significantly because of great variation within treatment groups.

Following termination of the calcium-phosphorus study (9 weeks of age), all pigs were provided a natural feedstuffs ration to approximately 91 kg. At that time, 3 pigs from Ration A and 2 pigs from Rations B, C and D were slaughtered and nasal turbinates were examined by the Veterinary Pathology Department. No significant differences were observed in skeletal, hair and skin, and nasal turbinate scores (Table XXXVI). None of the pigs were diagnosed as having atro-

TABLE XXXIII

CALCIUM (Ca) AND PHOSPHORUS (P) CONTENT OF HEART OF NINE WEEK OLD PIGS
 FED DIFFERENT LEVELS OF CALCIUM AND PHOSPHORUS
 FROM THREE TO NINE WEEKS OF AGE

Dietary Ca, %	0.368	0.569	0.776	0.952		
Dietary P, %	0.279	0.443	0.610	0.729	SE ^a	SD ^b
Ration Designation	A	B	C	D		
Number of pigs	6	7	6	6		
Heart						
Weight, fresh, gm.	86.8	86.7	109.0	106.8	8.3	NS
Dry matter, %	22.46	21.92	22.43	22.13	1.61	NS
Ash, dry basis, %	4.96	4.94	4.89	4.79	0.13	NS
Ca, fresh tissue mg./100 gm.	5.75	5.90	5.41	5.43	0.18	NS
Ca, dry basis mg./100 gm.	25.75	26.94	24.16	24.60	0.99	NS
Total heart Ca, mg.	5.00	5.06	5.91	5.83	0.50	NS
P, fresh tissue mg./100 gm.	251.94	251.73	258.43	249.87	9.80	NS
P, dry basis mg./100 gm.	1129.88	1150.77	1155.86	1133.65	57.89	NS
Total heart P, mg.	218.66	217.99	281.10	267.44	22.86	NS
P/Ca, dry basis	44.06	43.02	47.83	45.98	1.98	NS

^aStandard error of treatment means. Standard error when seven per treatment is the reported standard error times $\sqrt{6/7}$.

^bSignificant differences. NS = non-significant (P>.05).

TABLE XXXIV

CALCIUM (Ca) AND PHOSPHORUS (P) CONTENT OF LIVER OF NINE WEEK
 OLD PIGS FED DIFFERENT LEVELS OF CALCIUM AND PHOSPHORUS
 FROM THREE TO NINE WEEKS OF AGE

Dietary Ca, %	0.368	0.569	0.776	0.952		
Dietary P, %	0.279	0.443	0.610	0.729	SE ^a	SD ^b
Ration Designation	A	B	C	D		
Number of pigs	6	7	6	6		
Liver						
Weight, fresh, gm.	480.5	506.7	578.3	568.0	47.6	NS
Dry matter, %	28.21	28.31	28.97	27.95	0.32	NS
Ash, dry basis, %	5.18	4.99	5.04	5.31	0.10	NS
Ca, fresh tissue mg./100 gm.	7.78	7.62	7.27	8.37	0.63	NS
Ca, dry basis mg./100 gm.	27.68	26.95	25.12	29.99	2.36	NS
Total liver Ca, mg.	37.78	39.60	41.81	48.31	5.76	NS
P, fresh tissue mg./100 gm.	384.00	365.21	344.97	363.31	12.58	NS
P, dry basis mg./100 gm.	1358.33	1287.14	1186.67	1293.33	46.85	NS
Total liver P, mg.	1833.77	1871.47	1998.99	2058.42	184.38	NS
P/Ca, dry basis	50.46	48.52	47.50	45.91	3.13	NS

^aStandard error of treatment means. Standard error when seven per treatment is the reported standard error times $\sqrt{6/7}$.

^bSignificant differences. NS = non-significant (P>.05).

TABLE XXXV

CALCIUM (Ca) CONTENT OF HAIR OF NINE WEEK OLD PIGS FED
DIFFERENT LEVELS OF CALCIUM AND PHOSPHORUS FROM
THREE TO NINE WEEKS OF AGE

Dietary Ca, %	0.368	0.569	0.776	0.952	SE ^a	SD ^b
Dietary P, %	0.279	0.443	0.610	0.729		
Ration Designation	A	B	C	D		
Number of pigs	5 ^c	7	6	6		
<u>Hair</u>						
Dry matter, %	83.23	83.91	87.26	87.24	1.51	NS
Ca, dry basis mg./100 gm.	42.39	46.94	41.03	47.74	14.24	NS

^aStandard error of treatment means. Calculated standard errors are based on six pigs per treatment mean.

^bSignificant differences. NS = non-significant (P>.05).

^cSix pigs were sampled; however, one sample was lost prior to analysis.

TABLE XXXVI

INCIDENCE OF ATROPHIC RHINITIS (AR) AND VISUAL SCORES FOR SKELETAL CORRECTNESS,
 NASAL TURBINATES AND HAIR AND SKIN CONDITION OF 91 KG. PIGS FED DIFFERENT
 LEVELS OF CALCIUM (Ca) AND PHOSPHORUS (P) FROM THREE TO NINE WEEKS
 OF AGE, THEN FED A COMMON DIET TO SLAUGHTER.

Dietary Ca, %	0.368	0.569	0.776	0.952	
Dietary P, %	0.279	0.443	0.610	0.729	SD ^a
Ration Designation	A	B	C	D	
Number of pigs ^b	3	2	2	2	
Pig condition scores					
Skeletal correctness ^c	2.3	2.5	2.3	2.4	NS
Hair and skin ^d	2.0	2.2	1.8	1.8	NS
Nasal turbinate atrophy ^e	2.2	1.8	1.5	1.8	NS
Number with AR ^f	0	0	0	0	NS
Incidence of AR, %	0	0	0	0	NS

^aSignificant differences. NS = non-significant (P>.05).

^bNumber slaughtered at approximately 91 kg. live body wt.

^c1=normal, 2=slightly crooked, 3=moderately crooked, 4=very crooked and 5=severe crookedness.

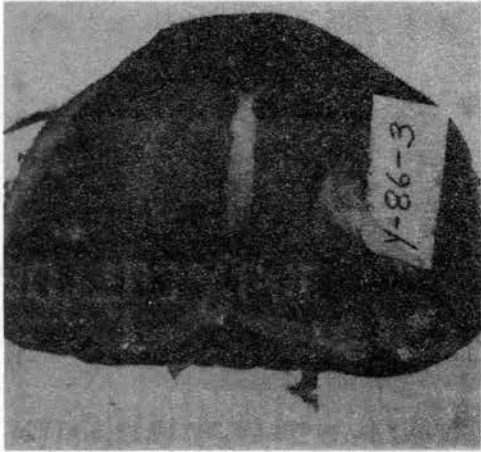
^d1=normal, 2=slightly dry and rough, 3=moderately dry and rough, 4=very dry and rough and 5=severely dry and rough.

^e1=normal, 2=slight, 3=moderate, 4=moderately severe and 5=severe.

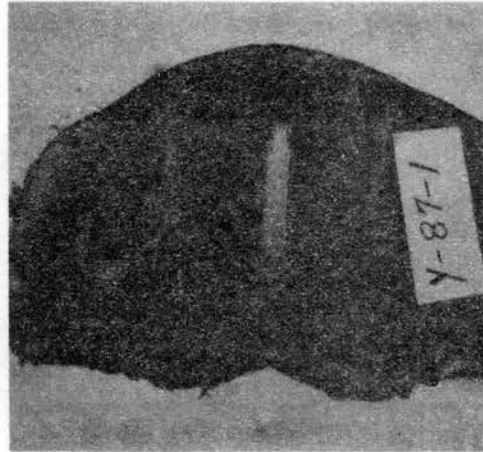
^fRepresents only those with gross evidence of nasal turbinate atrophy; microscopic and cultural examinations were not made. Gross examinations were performed by the Oklahoma State University Veterinary Pathology Department.

phic rhinitis. Nasal turbinates from each of the calcium-phosphorus treatment groups are pictured in Figure 12.

This study afforded an opportunity to make sex response comparisons in all of the performance, hematological and slaughter variables studied. For the most part, sex differences were small and nonsignificant ($P > .10$). However, 17 variables were significantly influenced by sex in this study and these variables are presented in Table XXXVII. One should keep in mind that pig numbers were small for the slaughter data and due to the large number of sex comparisons made, several significant differences could have occurred by chance.



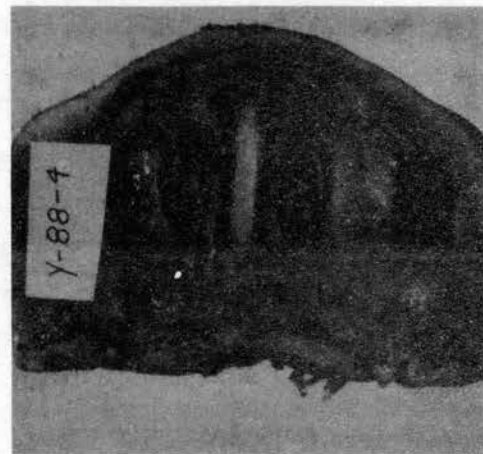
Ration A



Ration B



Ration C



Ration D

Figure 12. Cross-section of the Snout through the First Maxillary Premolar Teeth of Representative Pigs from Four Groups Fed Different Levels of Calcium and Phosphorus from Three to Nine Weeks of Age, then Provided a Common Ration to Slaughter at Approximately 91 kg.

TABLE XXXVII

VARIABLES SIGNIFICANTLY INFLUENCED BY SEX OF PIGS FED DIFFERENT
LEVELS OF CALCIUM AND PHOSPHORUS FROM
THREE TO NINE WEEKS OF AGE

Variable	Sex		SD ^a
	Males(M)	Females(F)	
Red blood cells, 3 wk., millions/mm. ³	4.95(38) ^b	5.61(41) ^b	F>M*
Mean corpuscular volume, 3 wk., cubic microns	67.59(38)	57.95(41)	M>F**
Mean corpuscular hemoglobin, 3 wk., micro-mcg.	22.44(38)	19.29(41)	M>F****
Clotting time, 3 wk., minutes	1.37(38)	1.73(41)	F>M***
Body wt. gain, 6 to 7 wk., kg.	2.78(36)	3.12(39)	F>M*
Feed intake, 6 to 7 wk., kg.	5.25(36)	5.94(39)	F>M*
Feed efficiency ^c , 8 to 9 wk., kg.	2.09(37)	1.69(39)	M>F*
White blood cells, 9 wk., thousands/mm. ³	10.61(37)	8.59(39)	M>F**
Relative stomach wt., gm./kg. live wt.	7.3(16)	6.2(9)	M>F**
Relative lung wt., gm./kg. live wt.	8.2(16)	7.2(9)	M>F**
Relative humerus length, mm./kg. live wt.	5.52(16)	4.72(9)	M>F**
Relative femur length, mm./kg. live wt.	6.13(16)	5.34(9)	M>F**
Relative ulna-radius length, mm./kg. live wt.	6.47(16)	5.72(9)	M>F**
Relative ulna-radius diameter, mm./kg. live wt.	1.40(16)	1.22(9)	M>F**
Femur ash, %	62.56(16)	64.58(9)	F>M*
Heart dry matter, %	21.92(16)	22.76(9)	F>M*
Heart ash, %	5.00(16)	4.72(9)	M>F**

^aSignificant differences. *P<.10, **P<.05, ***P<.025, ****P<.001.

^bNumber of animals included in the mean.

^cKg. of feed solids per kg. of body weight gain.

GENERAL SUMMARY AND CONCLUSIONS

One trial, involving a total of 79 purebred Yorkshire and Hampshire baby pigs of approximately equal sexes, was conducted at the Swine Rearing Laboratory located in the south wing of the Veterinary Medicine Building. This trial was initiated to study (a) the calcium and phosphorus requirements of 3 to 9-week old pigs fed purified diets and maintained under carefully controlled environmental conditions, and (b) the relationship between dietary calcium and phosphorus levels and development and condition of nasal turbinates. The baby pigs were obtained by collecting them into sterile bags as sows farrowed naturally. Each pig received pasteurized sow colostrum at birth and was fed a pasteurized, fortified cow's milk liquid diet for the first 2 weeks. On a dry matter basis, this diet contained 1.32 percent calcium and 0.98 percent phosphorus, by analysis. These pigs were reared in individual, isolated, disposable incubator boxes until they were 2 weeks of age. From 2 to 9-weeks of age, the pigs were housed in individual, open-topped, metal pens. During this period the pigs received one of the following four purified diets: Ration A, 0.37 percent calcium and 0.28 percent phosphorus; Ration B, 0.57 percent calcium and 0.44 percent phosphorus; Ration C, 0.78 percent calcium and 0.61 percent phosphorus; and Ration D, 0.95

percent calcium and 0.73 percent phosphorus. Data collected included body weight gain, feed consumption, feed efficiency, determination of various blood components and slaughter information at the end of the treatment period (9 weeks of age).

Ration A appeared adequate to effect normal feed efficiency, serum calcium and inorganic phosphorus levels, blood clotting time, hematocrit, hemoglobin, red and white blood cell counts, mean corpuscular volume, mean corpuscular hemoglobin and mean corpuscular hemoglobin concentration. Maximal skeletal development, as measured by fresh bone weight, specific gravity, ash content, calcium and phosphorus content, femur diameter and 8th rib and ulna-radius length, occurred in pigs consuming Ration D. Ration D also provided maximum survival and growth rate and optimal feed efficiency during the 6 weeks treatment period. Serum alkaline phosphatase levels were inversely related to dietary calcium and phosphorus levels. Calcium and phosphorus contents of the left kidney were positively related to the ration intake. Heart and liver calcium and phosphorus concentrations did not appear to bear this close relationship to dietary intake, although, total organ calcium and phosphorus appeared to bear a direct relationship. With the exceptions of the thyroid and the lungs, whose absolute weights were depressed by Rations A and B, no significant differences were observed in absolute and relative weights of various glands and organs. There was no incidence of atrophic rhinitis in any of the

treatment groups. With the calcium and phosphorus levels used in this study, it was concluded that the minimum calcium and phosphorus requirements of the 3 to 9-week old pig are at least 0.95 percent and 0.73 percent, respectively.

The calcium and phosphorus requirements as determined by the results of this study may be low when applied to practical rations. In practical rations, these nutrients are present in their natural state. In most cases, they are in different forms than when they are fed in purified diets. This indicates that there is likely to be a difference in their availability and thus some difference in requirements as determined with natural and purified rations. This does not mean that purified rations should not be used in studying nutrient requirements. Rather it indicates that some degree of reservation should be used when applying the data and that the nutrient should also be studied with natural rations.

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