

- I. RELATION OF NUTRITION AND AGE AT FIRST CALVING
TO LIFETIME PERFORMANCE OF BEEF COWS
- II. THE EFFECT OF LOW PHOSPHORUS RATIONS
UPON CAROTENE METABOLISM IN RUMINANTS

By

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Master of Science

State College of Washington

Pullman, Washington

1948

Submitted to the Faculty of the Graduate School of
the Oklahoma Agricultural and Mechanical College
in Partial Fulfillment of the Requirements

for the Degree of

DOCTOR OF PHILOSOPHY

1951

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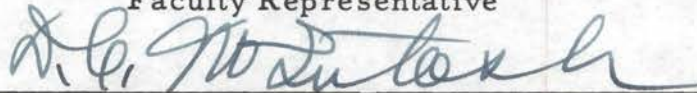
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ACKNOWLEDGEMENT

The author gratefully acknowledges the assistance of Dr. O. B. Ross, formerly of the Animal Husbandry Department, Oklahoma Agricultural and Mechanical College, for his valuable assistance in the planning and the initiation of these studies, and the assistance of Dr. C. K. Whitehair and Dr. A. E. Darlow, Animal Husbandry Department, Oklahoma Agricultural and Mechanical College, for their helpful suggestions during these studies.

Likewise, the author wishes to express his appreciation to Dr. R. W. MacVicar, Dr. W. D. Gallup, and Dr. V. G. Heller, Department of Agricultural Chemistry, Oklahoma Agricultural and Mechanical College, for technical advice and assistance in chemical determinations.

The assistance of W. D. Campbell and D. F. Stephens, who aided in collection of blood samples and weight data, is also appreciated.

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RELATION OF NUTRITION AND AGE AT FIRST CALVING TO LIFETIME PERFORMANCE OF BEEF COWS

INTRODUCTION

The importance of proper nutrition of cattle during growth, gestation, and lactation has been demonstrated by a number of investigators. A problem of major importance is the effect of different methods of management upon the lifetime usefulness of range beef cows. The various methods of management have not been observed under identical conditions, and a comparison of the relative merits of each has not been possible. In many cases, economics or available feed has dictated the system to be employed.

Reproductive ability is fundamental to economical beef production. In the production of replacement heifers, cattlemen are confronted with questions as to the effects of managerial, environmental, and nutritional factors upon growth and future usefulness of the heifers as mature cows. During the summer, most cattlemen follow the general practice of grazing their heifers on native grass; during the winter, however, methods of handling heifers may differ considerably, and it is during this season that nutritional deficiencies are most likely to occur. Levels of winter feeding, as measured by gain in weight, have been shown to affect growth and development of young cattle the ensuing summer grazing season, but there is insufficient information relative to the effect of level of wintering upon size of the mature cow, regularity of breeding, and size of the calf at weaning.

Although most commercial cattlemen breed their replacement heifers to calve as three-year-olds, some adhere to the practice of breeding the heifers

as yearlings to calve at two years of age. The breeding of commercial range heifers, wintered at medium or low levels, to calve as two-year-olds is a questionable practice. It is not uncommon to breed liberally fed heifers to calve under three years of age. This practice is prevalent in registered herds. Little information is available regarding the effect of age at time of calving and effect of level of wintering upon the subsequent performance of replacement heifers.

The investigation reported herein was undertaken to provide experimental data comparing the effect of level of wintering and the effect of age at breeding upon subsequent lifetime performance of heifers kept as breeding cows. In addition, this study was to provide information on the optimum amount of feed required and the cost of raising replacement heifers.

The advisability of feeding thyroprotein to beef cows for the stimulation of milk production was another part of this study. The measure of the effect of thyroprotein was the gain or loss in body weight of the mother and the size of the calf at weaning.

REVIEW OF LITERATURE

Extensive investigations concerning the wintering of beef cattle have been conducted for many years. These investigations were made in an effort to find suitable feeds for wintering cattle, as well as to study the most economical methods of wintering. Limited investigations has been completed in regard to the breeding of immature animals and the residual effect of this early breeding upon subsequent lifetime performance of the animals.

The effect of early breeding upon range cows was studied by McCampbell (1920). Eighty head of weanling heifers were divided into two groups, one-half being bred to calve as two-year-olds and the other half bred to calve as three-year-olds. One half of each of these two groups was fed a liberal amount of grain and roughage; the other half was fed only roughage. Heifers calving as two-year-olds produced smaller calves, regardless of the method of feeding, and the calf crop was reduced 30 per cent when the heifers were fed only roughage during the winter. The results of this experiment indicated that development of cows without grain and breeding them to drop their first calves at three years was the most practical method of management under range and semi-range conditions.

Withycombe, Potter, and Edwards (1930) conducted an experiment on age of breeding of beef cows. Their results indicated that the effects of early breeding were not changed by light or heavy winter feeding. At the age of six and one-half years, cows which had produced their first calves as two-year-olds had produced an average of 0.7 calf more for the entire time than cows which produced their first calves as three-year-olds. Early breeding did not

affect the size of the calves produced in subsequent years. Two-year-old heifers suckling calves weighed 200 pounds less than dry two-year-olds. This difference in weight was reduced to less than 100 pounds by the time the heifers were four years old. The effects of early breeding were not changed by light or heavy winter feeding. Cows producing their first calves as two-year-olds were more profitable than cows producing their first calves as three-year-olds.

Eckles and Swett (1918) stated that the most decided effect upon the size of mature dairy cows results from a combination of light rations during the growing period and early calving. They further stated that, next to hereditary factors which may determine the upward limit of growth, the combination of early calving and light rations during the growing period is the main cause, in numerous commercial herds, for a number of undersized cows. There is a tendency for animals to recover from retarded growth if conditions are favorable later. This may be accomplished by a more rapid rate of growth or by prolonging the period of growth. If the retardation, especially in skeletal growth, has gone too far, the animals will not, however, reach normal size. The amount of digestible nutrients consumed during the growing period has some effect upon rate of growth of the skeleton but has a greater effect on the weight of the animal. Reed, Fitch, and Cave (1924) stated that Holstein heifers bred to calve at 24 months of age did not develop as well as the animals given the same feed and bred to calve at 30 months; however, milk-producing ability was not affected by early calving.

Eckles (1919) made a study of the birth weight of calves of primarily the

Jersey and Holstein breeds and reported that the breed was the most important factor influencing the weight of calves at birth. The relatively small influence of the nutrition of the dam upon the development of the fetus may be explained by the dependence of the fetus for nourishment upon the blood stream of the dam and, therefore, only indirectly upon the food of the dam. It is a well-known physiological fact that there is a strong tendency for the composition of the blood to remain almost constant, even under adverse conditions of nutrition. It is a common observation by cattlemen that cows poor in flesh, to the point of emaciation, may bear calves of normal size for the breed. On the other hand, cows fattened to excess during gestation may have calves small for the breed and lacking in vigor as well. Apparently, only extreme cases of poor nutrition or the lack of some constituent in the ration for a long period of time may be expected to exert any marked influence upon the size of the calf.

Eckles and Palmer (1916) reported that underfeeding of cattle is manifested by a marked increase in the percentage of fat in the milk and, in some cases, by an appreciable decrease in the proportion of total protein, ash, and casein.

Eckles (1915) concluded that it is possible to influence the rate of growth, size of mature animal, and type, to some extent, by the liberality of the ration during the growing period and by the age at first calving.

Eckles (1916) stated that results from carefully controlled experiments lead to the conclusion that the amount of nutrients necessary to develop the bovine fetus is so small that it cannot be measured by ordinary methods of experimentation. Eckles stated that these results might be due to better use of feed during gestation, decreased maintenance during pregnancy, and the small amount of dry matter in the fetus.

Trowbridge, Moulton, and Haigh (1918) studied the effect of limited food supply on the growth of young beef animals and concluded that the condition of the animal appeared to have a decided effect upon the digestive ability. A very low plane of nutrition depressed the coefficient of digestion. These workers stated that the growth of the skeleton continued under very adverse feeding conditions and that this growth consisted of an increase of protein and of fat, as well as of mineral constituents.

Hart and Guilbert (1928) stated that the failure of beef cows to conceive is due, frequently, to a faulty plane of nutrition, resulting in lack of proper functioning of the ovary and no manifestation of heat periods. Cows that have weaned calves in the fall and have become pregnant again must gain during the early winter season if they are to be at normal weight by the next calving time.

Black, Quesenberry, and Baker (1938) wintered cows with and without a supplement of cottonseed cake and found that weight losses of cows that did not receive the supplement were significantly greater but that the increased weight of calves, at weaning time, from the supplement-fed cows did not compensate for the increased wintering feed costs. It was found that, for greatest economy, the use of cottonseed cake should be limited to seasons in which range conditions are severe. The experiments indicated that one pound of cottonseed cake fed on the range would replace approximately ten pounds of good quality hay in the feed lot. Lantow (1933) found that, for wintering beef cattle, one pound of cottonseed cake per head per day was more profitable than 2, 3, or 4 pounds and that the heavier feeding of cottonseed cake resulted in slower gains during the summer. Snapp (1939) emphasized

the fact that the plane of nutrition on which cows and heifers are wintered greatly affects their future usefulness. Ross, et al. (1947) reported a four-year study of two systems of cowherd management. They found that grazing cows year long and supplementing the cured grass with cottonseed cake was more economical than grazing cows during the summer and feeding them prairie hay and cottonseed cake, in a trap, during the winter. They reported that there was no difference in the condition of the cows at the end of the experiment or in the size of the calves at weaning.

The influence of winter gain or loss of steers upon summer gains made on grass pastures has been noted by a number of investigators. It is generally observed that steers making the greatest winter gain make the least summer gain. Kincaid (1939) reported a significant negative correlation between winter gain or loss and summer gain from pasture for both yearling and two-year-old steers. A gain of 100 pounds by yearling steers during the wintering period reduced pasture gains by 58 pounds. Dickson, et al. (1943) stated that, for economical production of feeder steers, it is necessary that steer calves be wintered in a thrifty and growing condition. Guilbert, et al. (1944) stated that efficient meat production and efficient use of range feed involves supplemental feeds. Thus, a plane of nutrition may be attained that will promote continuous growth and development--a consideration especially important in young animals at the time when growth rate is potentially greatest and when live-weight gains are most economical. Darlow, Taylor, and Campbell (1945), using weanling steer calves, studied the effect of level of wintering upon subsequent summer gains on grass. They found that steer calves wintered at a medium level were almost as heavy at the end of the summer

grazing season as steers wintered at a higher level. Black, Quesenberry, and Baker (1939) stated that significantly greater total range and feed-lot gains can be made by steers wintered on a high plane of nutrition but that significantly cheaper gains can be made by steers wintered on a low plane.

An extensive study on the effect of nutritional plane on the changes in proportions of steer carcasses was reported by Moulton, et al. (1921). In this work, the higher planes of nutrition proved to be the more efficient. Undernutrition resulted in a slow rate of gain. Hindquarter development was retarded more than the forequarter by undernutrition and was stimulated most by a high plane of nutrition.

According to Stephens, et al. (1948), yearling steers on bluestem pasture, whether wintered at a high, medium, or low level, will reach about the same weight by the end of the grazing season. A winter gain of approximately 3/4 pound per head daily seems the most desirable to produce the most profitable feeder yearlings; however, when yearling steers were full fed following early summer grazing, those on the high level of wintering proved most profitable.

The influence of the nutrition of the breeding ewe on growth and survival of progeny has long been recognized. Several recent investigations have shown that extreme differences in levels of feeding ewes during pregnancy affects the birth weights of lambs. Briggs (1936) summarized the results of a six-year trial studying the effects of breeding ewe lambs at about 9 months as compared to the more recommended practice of breeding ewes at about 21 months, or what is commonly termed the yearling age. Briggs stated that it took the early-bred lambs about ten months longer to reach their mature weight than it did those bred as yearlings. The immature,

bred ewes maintained their weight during the trial as did the ewes first bred as yearlings. The early-bred ewes produced an average of .69 lamb, or 30.96 pounds more of live lamb than the late-bred ewes. The most noticeable difference observed in the two groups of ewes studied was the fact that the teeth of the early-bred ewes showed wear at an earlier age than those of the late-bred group. Bowstead (1930) conducted an experiment on the effect of breeding immature ewes and concluded that lamb breeding did not cause a decrease in mature weight. Ewe lambs bred at 9 months of age produced more lambs, heavier and thriftier lambs at two and three years of age than the yearling-bred ewes produced at the same age. Wether lambs from lamb-bred mothers made as good gains during the suckling period as wether lambs from yearling-bred mothers.

Wallace (1948) studied the effect of extreme levels of diet upon pregnant ewes during two periods--early pregnancy and late pregnancy. He concluded that the level of nutrition prevailing during the period over which the gravid uterus makes the maximum growth is more important than the state of body reserves of the mother at that time.

Williams, et al. (1950) reported a cooperative study of pregnant ewes at five experiment stations in Canada. They concluded that legume hay gave more satisfactory results than did nonlegume hay, as indicated by ewe body weight, lamb weight, and vigor of lambs at birth. They found that a "change-over" ration, in which nonlegume hay was fed during the early pregnancy period and legume hay during the late pregnancy period, gave results comparable to those obtained when legume hay was fed for the entire pregnancy period.

Ewes are sometimes fed concentrates before and during the breeding period.

This practice of "flushing" is generally considered beneficial for ewes in thin condition and results in high fertility. However, Darlow and Hawkins (1932) and McKenzie and Phillips (1933) suggested that this practice also allowed the ewes to be bred earlier. Darroch, Nordskog, and Van Horn (1950) compared range ewes--supplemented with beet pulp pellets during the prebreeding, breeding, early pregnancy, and late pregnancy periods--with ewes which received no supplementation. They concluded that no important effects on birth or weaning weights were observed from the feed treatments; however, flock fertility was increased about 10 per cent by feeding supplement in the prebreeding and breeding periods. Ewes in good condition produced 11 per cent more lambs at birth and weaning than the thin ewes.

Neumann, Patton, and Gifford (1950) reported that lambs nursing ewes which were fed 1 gram of thyroprotein per 100 pounds of body weight daily significantly outgained a control group of lambs whose dams received a similar but unsupplemented ration.

Thyroprotein, a substance possessing thyrotropic activity, has been shown by many workers to stimulate milk production of dairy cows if the thyroprotein is fed during the stage of lactation when milk production is declining. Blaxter (1945) has suggested that, for best results, thyroprotein should be fed at a level that will raise milk yields about 20 per cent and that for this increase the feed intake should be increased 20 per cent. This practice largely eliminated the loss in body weight when thyroprotein was fed at a 15-gram level.

Moore and Sykes (1947) stated that a 20 per cent increase in the feed intake of a normally fed cow not receiving thyroprotein will increase milk production about 13 per cent. If, by feeding thyroprotein, 20 per cent more milk could

be produced with 20 per cent more feed, an increase of 7 per cent in milk production would be left to pay for the the thyroprotein and the extra trouble in feeding. Thomas, Moore, and Sykes (1949) advocated the addition of extra feed (25 per cent over the requirement) when thyroprotein was fed. They observed that poor-producing cows responded less than good producers and that it is not possible to make a good producer out of a poor producer by feeding thyroprotein. Blaxter (1945) stated that poor-producing dairy cows in milk for 10 weeks and yielding 20 pounds daily showed an average increase in milk production of 3.8 pounds after receiving thyroprotein. The milk production of good cows in milk 30 weeks and yielding 30 pounds daily increased by 9.1 pounds , on the average.

Hibbs and Krauss (1947) fed thyroprotein to cows at a rate of 1 gram to 53 pounds of body weight. They reported an increase in milk production that varied greatly from cow to cow. They also noted an increase in pulse and respiratory rates, as well as a decrease in body weight.

Reece (1944) stated that the feeding of 10 grams of Protamone daily for 3 weeks to a group of 5 dairy cows increased the butterfat content of milk from 3.62 per cent to 4.11 per cent. In a later paper, Reece (1947) observed that the daily feeding of 10 grams of thyroprotein to 9 cows resulted in an initial increase of 7.6 per cent in milk production; the milk production of five cows receiving 15 grams of thyroprotein daily increased 19.7 per cent. The average daily milk production in the second week after the withdrawal of thyroprotein was 19.5 per cent below that of the last week of thyroprotein feeding in 5 cows receiving 10 grams of thyroprotein; in three cows receiving 17

grams of thyroprotein, the decrease was 34.2 per cent.

Blaxter (1946) showed that iodinated casein could be placed into cubes; no loss of potency occurred either during the moderately high temperature (not over 130°) or during subsequent storage under farm conditions.

EXPERIMENTAL

Objectives

1. To study the effect of age at first calving upon subsequent performance of heifers kept as replacement breeding cows.
2. To study the effect of wintering at different levels of supplementation upon the nutritional status of the animal and upon subsequent performance of those heifers.
3. To evaluate these factors in terms of economy of production of replacement cows in the herd.
4. To determine the effect of supplemental summer feeding of two-year-old cows suckling calves.
5. To determine the effect of feeding thyroprotein upon the cow and upon the suckling calf.
6. To determine the effect of different levels of wintering upon certain blood constituents indicative of the nutritional status of the animal.

Procedure

One hundred twenty choice, weanling, Hereford heifers were selected for this study. One hundred five of the heifers were purchased from the Moon Ranch at Mill Creek, Oklahoma, and fifteen of the heifers were produced in the Experiment Station herd. The heifers were divided into eight lots of fifteen head each, in accordance with accepted experimental procedure, and were started on the experimental rations October 28, 1948, at the Lake Carl Blackwell Experimental Range. This range is located approximately 13 miles west of Stillwater, Oklahoma, on the north side of Lake Carl Blackwell. The

supplemental feeding during the winter months was adjusted to provide for three rates of gain--designated as low, medium, and high levels--the gain in weight being the measure of the level of wintering. The level of feeding designated lots was selected at random.

Lots 1 and 2, selected to be wintered at a low level, were fed to gain 0.00 to 0.10 pound per head per day. Lots 3, 4, 7, and 8, to be wintered at a medium level, were fed to gain 0.40 to 0.60 pound per head per day. Lots 5 and 6, to be wintered at a high level, were fed to gain 1.10 to 1.30 pounds per head per day.

During the 1948-1949 winter phase (October 28, 1948 to April 18, 1949), each group of heifers to be fed the designated levels of winter feeding--low level (Lots 1 and 2), medium level (Lots 3 and 4), high level (Lots 5 and 6), and medium level (Lots 7 and 8)--was placed in comparable 200-acre native-grass pastures to facilitate winter feeding. These pastures are a part of the experimental range area north of Lake Carl Blackwell. The groups of heifers were rotated among the four pastures, each group being grazed the same number of days in each pasture, to reduce possible effects of pasture variation; the pastures were originally fenced to be as uniform as possible. A statistical analysis indicated variation in the different pastures did not affect rate of winter gain. In addition to the dry, cured grass, the heifers were fed the following supplemental feeds: Lots 1 and 2--0.93 pound of cottonseed cake per head daily (considered to be a low level of wintering); Lots 3, 4, 7, and 8--1.97 pounds of cottonseed cake per head daily (medium level of wintering); and Lots 5 and 6--1.98 pounds of cottonseed cake and 2.69 pounds of oats per head daily (high level of wintering). Until June of 1949, the heifers grazed

the same native-grass pastures they grazed during the winter. Throughout the winter and summer periods, all lots had access to a mineral mixture of 2 parts ground rock salt and 1 part steamed bone meal.

Unfortunately, some heifers were found to be pregnant after the initiation of the experiment and were removed from the experiment the first winter. One heifer from each of Lots 1 and 5, two heifers from each of Lots 6 and 8, and three heifers from each of Lots 4 and 7 were removed from the experiment. These heifers were replaced with heifers from the Experiment Station herd which were of the same age and weight and which had comparable winter feeding.

The heifers were pasture bred to registered Hereford bulls. The bulls were placed with the heifers of odd-numbered lots (1, 3, 5, and 7) on May 1, 1949, and were removed on September 1, 1949. In June 1949, the heifers of all lots were removed to the Fort Reno Experiment Station, El Reno, Oklahoma, and were grazed in comparable native-grass pastures. The principal grasses in these pastures were Big and Little Bluestem and Indian grass.

During the summer of 1949, several heifers developed foot rot. One Lot 3 heifer was taken from the experiment because foot rot developed to the extent that one toe had to be removed. A heifer of comparable age and breeding which had received about the same amount of winter feed was placed in Lot 3.

During the 1949-1950 winter phase (October 25, 1949 to April 25, 1950), the dry, native grass was supplemented as follows: Lots 1 and 2--0.93 pound of cottonseed cake per head daily; Lots 3, 4, 7, and 8--2.32 pounds of cottonseed cake per head daily; and Lots 5 and 6--2.32 pounds of cottonseed cake and 2.79 pounds of oats per head daily. The heifers of all lots had access to

a mineral mixture of 2 parts ground rock salt and 1 part steamed bone meal.

The heifers of Lots 1, 3, 5, and 7 started calving, as two-year-olds, in February 1950. When the winter phase was terminated, the heifers of all lots except Lot 7 were placed in one large, native-grass pasture. The heifers of Lot 7 were grazed in a pasture providing forage and acreage per head comparable to that grazed by other lots but were fed 1.5 pounds of cottonseed cake and 3.5 pounds of oats from July 1 to October 9, 1950, at which time the calves were weaned. This ration was fed to determine the effect of supplemental feeding during part of the lactation period upon the ultimate size of the heifers at maturity, upon breeding efficiency, and upon size of the calf at weaning. All calves were dehorned at the age of approximately 4 months. The bull calves were castrated, and all calves were vaccinated for blackleg in June 1950.

Bulls were placed with the heifers of all lots on May 1, 1950, and removed on September 1, 1950. The calves were weaned on October 9, 1950; the summer grazing period ended October 31, 1950.

During the 1950-1951 winter phase (October 31, 1950 to April 23, 1951), the dry, cured grass was supplemented as follows: Lots 1 and 2--0.97 pound cottonseed cake per head daily; Lots 3, 4, 7, and 8--2.46 pounds cottonseed cake per head daily; and lots 5 and 6--2.47 pounds cottonseed cake and 2.93 pounds of oats per head daily. The heifers of all lots had access to a mineral mixture of 2 parts ground rock salt and 1 part steamed bone meal.

All heifers started calving in February 1951. At the termination of the winter phase, April 23, 1951, all lots were placed in one large, native-grass pasture and grazed together until July 2, 1951, at which time the heifers and

calves of Lots 7 and 8 were removed and placed in two different pastures provided forage and acreage comparable to the pasture grazed by the other six lots. From July 2, 1951, until the calves are weaned in October, the cows of Lot 7 were fed the same supplemental ration they received the previous summer--1.5 pounds cottonseed cake and 3.5 pounds of oats per head daily. The supplemental ration fed to the heifers of Lot 8 consisted of 1.5 pounds cottonseed cake, 3.25 pounds oats, 0.25 pound molasses, and 15 grams thyroprotein. In order to insure the daily consumption of 15 grams of thyroprotein by each cow, the feeds were thoroughly mixed and pelleted. Thyroprotein was fed to determine whether or not it would cause an increase in milk production in the heifers. The weaning weights of the calves are to serve as an indication of the efficacy of the thyroprotein on milk production; gain or loss in weight of the cows should give some information on the advisability or danger of feeding thyroprotein to beef cows during the summer months.

Bull calves were castrated April 9, 1951, and on May 28, 1951, all calves were dehorned with "bell-irons"; all calves were vaccinated for blackleg.

Bulls were placed with all heifers on May 1, 1951; the bulls to be removed September 1, 1951. Calves are to be weaned about the middle of October.

In order to evaluate the nutritional status of the animals, eight heifers from each of the first six lots were bled from the jugular vein, at intervals of approximately one month. The blood was collected in oxalated tubes and was kept under refrigeration until aliquots were taken for the various chemical determinations. The chemical determinations made and the methods

employed were plasma-carotene, Kimble (1939); plasma-inorganic phosphorus, Youngburg and Youngburg (1930); red blood cell count, indirect turbidimetric procedure using the Evelyn Colorimeter, turbidity readings standardized in terms of red blood cell by direct counting procedures; hemoglobin, an acid hematin method standardized by the method of Wong (1928); plasma-proteins, copper-sulfate specific gravity method of Phillips, et al. (1945).

The average plasma values of each lot are presented in Table 4.

Throughout this experiment, the following records were maintained:

1. Feed consumption and yearly feed costs.
2. Weights of heifers each 28-day period.
3. Birth and weaning weights of calves.
4. Breeding efficiency.
5. Percentage calf crop.
6. Calving dates.
7. Calving difficulty, if any.
8. Blood composition.

All supplemental feeds were analyzed chemically by A.O.A.C. methods (1945). In addition, grass samples were collected periodically and analyzed chemically. The samples--Big and Little Bluestem and Indian grasses--were collected from approximately the same area in each pasture; grass was cut one to two inches above the ground and collected in paper bags. Chemical compositions of feeds and grasses used in this experiment are given in Table 5.

The data in this experiment was analyzed statistically by the methods of Snedecor (1946).

RESULTS AND DISCUSSION

1948-1949

The results of the wintering and summer grazing of the first year are shown in Table I.

Heifers of Lots 1 and 2, wintered together at the low level on the range and fed 0.93 pound of cottonseed cake daily, gained only 28 and 17 pounds, respectively, for the 172 days of the winter period. The heifers in these two lots were thinner than heifers of other lots but, nevertheless, were in a strong and thrifty condition. The total cost of the winter feed was \$10.65 per head. Lots 3, 4, 7, and 8, wintered alike at a medium level--1.98 pounds of cottonseed cake per head daily, gained an average of 53.5 pounds per head. This gain was 31 pounds greater than the average gain of Lots 1 and 2. The average total winter feed cost, however, was \$18.40 per head.

The feeding of 1.98 pounds of cottonseed cake and 2.69 pounds of oats daily to Lots 5 and 6 (high level of wintering) resulted in an average gain of 88 pounds. This gain was 34.5 pounds greater than the average winter gain of Lots 3, 4, 7, and 8, but little difference in thrift was observed among the six lots. The average winter feed cost of Lots 5 and 6 was \$31.84 per head. This wintering cost was \$21.19 greater than the average cost per head of Lots 1 and 2; however, the gain of the heifers of Lots 5 and 6 was only 65.5 pounds greater per heifer than the gain of heifers of Lots 1 and 2.

During the summer grazing season, the total average summer gains for all heifers wintered on low, medium, and high levels were 289.5, 271.0 and 257.5 pounds, respectively, whereas the average winter gains were 22.5, 53.5, and 88.0 pounds. Statistical analysis indicated that the difference in

TABLE 1.

GAINS OF BEEF HEIFERS WINTERED AT DIFFERENT LEVELS (1948-49)

Lot number	1	3	5	7	2	4	6	8
Level of wintering	Low	Med	High	Med	Low	Med	High	Med
Heifers per lot ¹	15	15	15	15	15	15	15	15
Winter Phase (172 days)								
Average Weights								
Initial weight 10/28/48	473	471	476	481	473	461	470	478
Final weight 4/18/49	501	529	561	536	493	520	561	520
Total winter gain	28	58	85	55	17	59	91	42
Daily gain	.17	.33	.49	.32	.10	.34	.53	.25
Average Daily Ration (lbs.) ²								
Cottonseed cake	.93	1.96	1.98	1.98	.93	1.96	1.98	1.98
Oats	----	----	2.69	----	----	----	2.69	----
Range	Ad lib.	Ad lib.	Ad lib.	Ad lib.	Ad lib.	Ad lib.	Ad lib.	Ad lib.
Mineral	.07	.07	.07	.07	.07	.07	.07	.07
Cost of winter feed per heifer (dollars)	10.65	18.32	31.84	18.49	10.65	18.32	31.84	18.49
Summer Phase (190 days)								
Average weights								
Initial weight 4/18/49	501	529	561	536	493	520	561	520
Final weight 10/25/49	802	810	836	836	771	759	801	784
Total summer gain	301	281	275	300	278	239	240	264
Daily gain	1.58	1.48	1.45	1.58	1.46	1.26	1.26	1.39
Average Daily Ration								
Range	Ad lib.	Ad lib.	Ad lib.	Ad lib.	Ad lib.	Ad lib.	Ad lib.	Ad lib.
Mineral	.02	.02	.02	.02	.02	.02	.02	.02

TABLE 1. (Continued)

Lot number	1	3	5	7	2	4	6	8
Cost of summer feed per heifer (dollars)	14.07	14.07	14.07	14.07	14.07	14.07	14.07	14.07
Cost per cwt. gain (summer)	4.67	5.01	5.12	4.69	5.06	5.89	5.86	5.33
Total gain from 10/28/48 to 10/25/49	329	339	360	355	295	298	331	306
Total cost of summer and winter feed (dollars)	24.72	32.39	45.91	32.56	24.72	32.39	45.91	32.56
Cost per cwt. gain (entire year)	7.51	9.55	12.75	9.17	8.38	10.87	13.87	10.64
Initial value per head 10/28/48 (28.00 per cwt.) (dollars)	132.44	131.88	133.28	134.68	133.28	129.08	131.60	133.84

¹ Unfortunately, some of the heifers were bred when purchased. Because of pregnancy, one heifer each from Lots 1 and 5, two heifers each from Lots 6 and 8, and three heifers each from Lots 4 and 7 were removed during the winter phase. Data reported herein are only on those heifers that were open at the start of the experiment. During the summer of 1949, one heifer in Lot 3 was removed from the experiment because foot rot developed to such extent that one toe had to be removed. This heifer was replaced with one from the Experiment Station Herd of comparable breeding and which had received comparable feed.

² During inclement weather, 2000 pounds of hay were fed to each lot. The cost and amount of hay fed is not included in the above table.

Feed Prices

Cottonseed cake	\$86.50 per ton
Prairie Hay	13.00 per ton
Oats	0.925 per bu.
Range, winter phase	3.50 per head
summer phase	14.00 per head
Mineral mixture	1.97 per cwt.

summer gains was highly significant. Data of summer gains of beef heifers wintered at different levels are similar to those reported by Darlow, et al. (1945), Ross et al. (1947), and Stephens, et al. (1948). These workers studied the summer gains of steers wintered at different levels and found that steers which gained the least during the winter gained the most during the summer. Steers wintered at low and medium levels were almost as heavy at the end of the summer grazing season as those wintered at the high level.

The heifers of Lots 1, 3, 5, and 7, which were exposed to bulls during the summer grazing season, gained approximately 34 pounds more than the open heifers; these differences in gain were highly significant.

The total gain (winter and summer) for the low, medium, and high levels of wintering were 312.0, 324.5, and 345.5 pounds respectively.

The year's total feed costs for the low, medium, and high level lots were \$24.72, \$32.39, and \$45.91 per head. Although, for the year, the medium-lot heifers made a total gain of 12.5 more than heifers wintered at the low level, this additional gain cost \$7.67. The yearly gain of heifers wintered at the high level was 33.5 pounds greater than the gain of heifers wintered at the low level; this additional gain cost \$21.19.

At the end of the first year, there was very little difference observed among the lots in terms of total weight gain; however, heifers of Lots 1 and 2 (low level) did not appear to be quite as "growthy" as those wintered at the medium and high levels.

Data on cost of feed per hundred pounds gain are summarized in Table 1.

Gains made by heifers of Lot 1 (low level of wintering) during the first year were approximately twice as economical as those made by Lot 6 (high level of wintering).

1949-1950.

Heifers Calving as Two-Year-Olds (Lots 1, 3, 5, and 7)

Production data and weight gains during the second year are given in Table 2.

At the start of the winter feeding period, the average weights of Lots 1, 3, 5, and 7 were 802, 811, 836, and 816 pounds, respectively. All heifers lost weight from the onset of the winter-feeding period until calving and showed a loss in weight for the entire winter. The average winter loss in weight for the heifers of Lot 1 was 87 pounds. This loss was 48 pounds greater than that of heifers of Lot 3, 59 pounds greater than that of heifers of Lot 5, and 14 pounds greater than the loss of Lot 7. Some Lot 1 heifers were noticeably thin at the end of the winter-feeding period. During the 1950 summer grazing season, the heifers of Lot 1 made greater gains than the heifers of Lots 3 and 5, the summer gains for these lots being 207, 181, and 193 pounds, respectively. Lot 7, which received additional feed during the summer, showed a gain of 244 pounds. The average weights of the heifers of Lots 1, 3, 5, and 7, on October 31, 1950, after the calves were weaned, were 922, 952, 1001, and 995 pounds, respectively. At this time, the heifers of Lot 1 (low level) did not appear to be materially smaller or less thrifty in appearance than heifers of the other lots.

The heifers of Lots 3 and 7 were managed alike each winter, but those of Lot 7 were fed a supplement during the summer, from July 1 until the calves were weaned October 9, 1950. The heifers of Lot 7 gained 63 pounds more, during the summer, than the heifers of Lot 3 and at the end of the summer grazing period weighed 43 pounds more; however, at weaning,

WINTERING HEIFERS AT DIFFERENT LEVELS (1949-50)

Lot number	Exposed to bull summer 1949				Open			
	1	3	5	7	2	4	6	8
Level of wintering	Low	Med	High	Med	Low	Med	High	Med
Heifers per lot	15	15	15	15	15	15	15	15
Winter Phase (182 days)								
Average Weights								
Initial weight 10/25/49 ¹	802	811	836	816	771	758	804	779
Weight prior to calving 2/2/50	786	798	831	800	---	---	---	---
Gain up to calving	-16	-13	-5	-16	---	---	---	---
Final weight 4/25/50	715	771	808	751	782	788	825	799
Total winter gain	-87	-39	-28	-73	11	30	21	20
Daily gain	-.48	-.21	-.15	-.40	.06	.17	.11	.11
Average Daily Ration (lbs.)								
Cottonseed cake	.93	2.32	2.32	2.32	.93	2.32	2.32	2.32
Oats	----	----	2.79	----	----	----	2.79	----
Range	Ad lib.	Ad lib.	Ad lib.	Ad lib.	Ad lib.	Ad lib.	Ad lib.	Ad lib.
Mineral	.07	.07	.07	.07	.07	.07	.07	.07
Cost of winter feed per heifer (dollars)	10.91	19.39	31.91	19.39	10.91	19.39	31.91	19.39
Summer Phase (189 days)								
Average Weights								
Initial weight 4/25/50	715	771	808	751	782	788	825	799
Final weight 10/31/50	922	952	1001	995	1103	1066	1104	1082
Total summer gain	207	181	193	244	321	278	279	283
Daily gain	1.10	.96	1.02	1.29	1.70	1.47	1.48	1.50
Average Daily Ration (lbs.)								
Range	Ad lib.	Ad lib.	Ad lib.	Ad lib. ²	Ad lib.	Ad lib.	Ad lib.	Ad lib.
Mineral	.02	.02	.02	.02	.02	.02	.02	.02

TABLE 2. (Continued)

Lot number	Exposed to bull summer 1949				Open			
	1	3	5	7	2	4	6	8
Number of calves born	14	15 ₃	13	14 ₄	---	---	---	---
Number lost at time of birth	1	1	2	2 ₄	---	---	---	---
Number of heifers helped	6	8	4	10	---	---	---	---
Average calving date, March	23	15	17	5	---	---	---	---
Average birth weight	69	65	70	66	---	---	---	---
Per cent calf crop	87	93	73	80	---	---	---	---
Average weaning weight of calves 10/9/50	406	404	391	416	---	---	---	---
Total feed cost per heifer 10/28/48 to 10/31/50 (dollars)	50.70	66.85	92.89	81.48	50.70	66.85	92.89	67.02

- ¹ Differences in final weights for the first year of the trial, October 25, 1949, and the initial weight for the beginning of the 1949-50 wintering phase of the experiment were due to the substitution of additional heifers for those removed from the experiment because of pregnancy the first year. In general, the heifers substituted had been managed in the same manner as those heifers of the lot in which they were placed.
- ² From July 1, 1950 until October 9, 1950, when the calves were weaned, the heifers of Lot 7 received a supplemental ration of 1.5 pounds cottonseed cake and 3.5 pounds oats per head per day.
- ³ One cow in Lot 3 died during calving (April 25, 1950).
- ⁴ One cow in Lot 7 was injured during calving. Calf born dead March 14, 1950. Cow subsequently failed to conceive and was marketed June 8, 1950.

Feed Prices

Cottonseed cake	\$67.00 per ton
Oats	0.79 per bu.
Range, winter phase	4.00 per head
summer phase	15.00 per head
Mineral mixture	1.91 per cwt.

calves of Lot 7 were only 12 pounds heavier than those of Lot 3. The additional weight gain of the Lot 7 cows and their calves was obtained at an additional cost of \$14.46.

The average weaning weights of the calves from the heifers of Lots 1, 3, 5, and 7 were 406, 404, 391, and 416 pounds, respectively. The level of wintering of the heifers appeared to have little influence upon the weaning weight of the calves. The percentage calf crop weaned from the heifers of Lots 1, 3, 5, and 7 was 87, 93, 73, and 80, respectively. Again, the low level of wintering did not adversely affect the number of calves weaned. The heifers of Lot 5 (high level) had the lowest percentage calf crop.

The average calving dates for Lots 1, 3, 5, and 7 were March 23, 15, 17, and 5, respectively. The respective average birth weights of the lots were 69, 65, 70, and 66 pounds. As shown in Table 2, a total of six calves from all lots was lost at the time of birth; from four to ten heifers in each lot were given assistance at the time of calving. One cow in Lot 3 died during calving; one cow in Lot 7 was injured in calving and was later marketed. One heifer each in Lots 1 and 7 and two heifers in Lot 5 failed to breed.

The total feed costs for the two-year period for Lots 1, 3, 5, and 7 were \$50.70, \$66.85, \$92.89, and \$81.48, respectively.

On the basis of the results of one year, it would appear that heifers bred to calve at two years of age and wintered at a low level can produce satisfactorily.

On October 31, 1950, the two-year-old heifers which had been suckling calves weighed from 100 to 175 pounds less than comparable dry two-year-olds. These results are in general agreement with data reported by Withycombe, et al. (1930).

The average calving dates for Lots 1, 3, 5, and 7 were March 23, 15, 17, and 5, respectively. The respective average birth weights of the lots were 69, 65, 70, and 66 pounds. As shown in Table 2, a total of six calves from all lots was lost at the time of birth; from four to ten heifers in each lot were given assistance at the time of calving. One cow in Lot 3 died during calving; one cow in Lot 7 was injured in calving and was later marketed. One heifer each in Lots 1 and 7 and two heifers in Lot 5 failed to breed.

Heifers Exposed to Bulls in Summer of 1950 only (Lots 2, 4, 6, and 8)

All heifers made a slight gain during the winter, the heifers of Lot 2 (low level) making the least. During the summer grazing season, the greatest gain was made by Lot 2 heifers. The average weights of heifers of Lots 2, 4, 6, and 8, on October 31, 1950 (the end of the summer period), were 1103, 1066, 1104, and 1082 pounds, respectively.

The total feed costs per heifer for the two years of the experiment for Lots 2, 4, 6, and 8 were \$50.70, \$66.85, \$92.89, and \$67.02, respectively. After a consideration of the total gain and total cost of feed, it would appear that the most economical method of producing replacement heifers bred to calve at three years of age is to winter them at a low level (1 pound of cottonseed cake per head per day plus dry grass). At the end of two years, the heifers of Lot 2 (low level of wintering) weighed approximately the same as heifers of Lot 6 (high level of wintering). The additional feed for heifers of Lot 6, above that fed the heifers of Lot 2, was provided at the cost of \$42.10 per heifer for the two-year period.

1950-1951

Heifers Calving as Two-Year-Olds (Lots 1, 3, 5, and 7)

Data on weight gains, feed costs, and calf production during the third year are shown on Table 3.

The heifers of all lots lost weight from the onset of the winter period until calving; however, heifers of Lot 5 (high level) lost about one-fourth the weight lost by heifers of other lots. The total losses in weight during the winter period for heifers of Lots 1, 3, 5, and 7 were 209, 194, 144, and 244 pounds, respectively.

TABLE 3.

WEIGHT GAINS, FEED COSTS AND PRODUCTION DATA OF HEIFERS WINTERED AT DIFFERENT LEVELS (1950-51)

Lot number	Exposed to bull summers 1949 and 1950				Exposed to bull summer 1950 only			
	1	3	5	7	2	4	6	8
Level of wintering	Low	Med	High	Med	Low	Med	High	Med
Heifers per lot	15	14	15	14	15	15	15	15
Winter Phase (174 days)								
Average Weights								
Initial weight 10/31/50	922	952	1001	995	1103	1066	1104	1082
Weight prior to calving 2/29/51	864	901	988	941	1026	1025	1097	1024
Gain up to calving	-58	-51	-13	-54	-77	-41	-7	-58
Final weight 4/23/51	713	758	857	751	795	830	905	821
Total winter gain	-209	-194	-144	-244	-308	-236	-199	-261
Daily gain	-1.20	-1.12	-.83	-1.40	-1.77	-1.35	-1.14	-1.50
Average Daily Ration (lbs.)								
Cottonseed cake	.97	2.46	2.47	2.46	.97	2.46	2.47	2.46
Oats	----	----	2.93	----	----	----	2.93	----
Range	Ad lib.	Ad lib.	Ad lib.	Ad lib.	Ad lib.	Ad lib.	Ad lib.	Ad lib.
Mineral	.03	.03	.03	.03	.03	.03	.03	.03
Cost of winter feed per heifer (dollars)	11.61	21.70	36.60	21.70	11.61	21.70	36.60	21.70
Summer Phase (96 days)								
Average Weights								
Initial weight 4/23/51	713	758	857	751	795	830	905	821
Weight 7/2/51	897	941	1013	933	1006	1012	1085	1020
Total summer gain to 7/2/51	184	183	156	182	211	182	180	199
Daily gain to 7/2/51	2.65	2.61	2.23	2.60	3.01	2.60	2.56	2.84
Weight 7/28/51	911	967	1026	950	1026	1024	1097	968
Gain from 7/2/51 to 7/28/51	14	26	13	17	20	12	12	-52
Daily gain to 7/28/51	2.06	2.18	1.76	2.07	2.40	2.02	2.00	1.55

TABLE 3. (Continued)

Lot number	Exposed to bull summers 1949 and 1950				Exposed to bull summer 1950 only			
	1	3	5	7	2	4	6	8
Average Daily Ration (lbs)								
Range	Ad lib.	Ad lib.	Ad lib.	Ad lib. ²	Ad lib.	Ad lib.	Ad lib.	Ad lib. ³
Mineral	Ad lib.	Ad lib.	Ad lib.	Ad lib.	Ad lib.	Ad lib.	Ad lib.	Ad lib.
Number of calves born	14	14	13	14	15	14	14	15
Number lost at time of birth	0	1	0	0	0	0	2	1
Number of heifers helped	0	0	0	0	0	0	1	0
Average calving date	3-13	3-14	3-7	3-5	3-5	2-26	2-27	2-26
Average birth weight	67	69	72	71	69	65	70	64
Per cent calf crop	93	93	87	100	100	93	80	93
Average weight of calves								
7/2/51	245	244	275	270	262	265	286	258
7/28/51	294	273	326	329	313	307	339	308
Gain in weight of calves								
from 7/2/51 to 7/28/51	49	29	51	59	51	42	53	50
Total feed cost per heifer								
10/28/48 to 4/23/51								
(dollars)	62.31	88.55	129.49	103.18	62.31	88.55	129.49	88.72

¹ One heifer in Lot 8 died of undetermined causes on July 7, 1951.

² From July 2, until the calves are weaned in October 1951, heifers of Lot 7 will receive a daily supplement of 1.5 pounds cottonseed cake and 3.5 pounds oats.

³ From July 2, until the calves are weaned in October 1951, heifers of Lot 8 will receive a daily supplement of 1.5 pounds cottonseed cake, 3.25 pounds oats, 0.25 pound molasses and 15 grams thyroprotein.

Feed Prices

Cottonseed cake	\$77.50 per ton
Oats	0.93 per bu.
Range, winter phase	5.00 per head
summer phase	15.00 per head
Mineral mixture	1.79 per cwt.

Lot 7 heifers, which had received additional feed the previous summer, lost the most weight. The average weights in April 1950 (the end of the winter-feeding period) were 715, 771, 808, and 751 pounds, respectively, for the heifers of Lots 1, 3, 5, and 7. The respective weights in April 1951 were 713, 758, 857, and 751. Lot 3 heifers had a loss of 13 pounds, Lot 5 a gain of 49 pounds, whereas the heifers of Lots 1 and 7 maintained, approximately, the same average weights.

The percentage calf crops from the heifers of Lots 1, 3, 5, and 7 were 93, 93, 87, and 100. Lot 1 heifers, wintered at a low level, had a higher percentage calf crop both years than heifers of Lot 5, wintered at the high level. The heifers of Lot 7, which received additional feed during the summer, produced a 100 per cent calf crop.

The average calving dates of the heifers of Lots 1, 3, 5, and 7 were March 13, 14, 7, and 5. The low level of wintering did not adversely affect the breeding efficiency of the Lot 1 cows.

Birth weights of calves for Lots 1, 3, 5, and 7 were 67, 69, 72, and 71 pounds, respectively. No help was required for heifers at the time of calving; only one calf (lot 3) was lost at the time of birth.

The average weights of the calves on July 2, 1951 were 245, 244, 275, and 270 pounds for Lots 1, 3, 5, and 7. The weights of these calves on July 28 were 294, 273, 326, and 329 pounds, respectively. The calves of Lot 7, whose dams were receiving additional feed from July 2 until weaning time, made a greater gain in weight during this 26-day period than did calves of any other lot.

Heifers Calving as Three-Year-Olds (Lots 2, 4, 6, and 8)

The heifers of Lot 2 (low level) lost average of 71 pounds from the start of

From April 23, 1951 until July 28, 1951, the summer gains of all heifers were about the same, except for the high-level heifers of Lot 5 whose gain was approximately 30 pounds less than the gain of heifers of other lots.

The total feed costs, from October 28, 1948 to April 23, 1951, for the heifers of Lots 1, 3, 5, and 7 were \$62.31, \$88.55, \$129.49, and \$103.18.

At the present time, on the basis of two years' data, it would appear that replacement heifers bred to calve at two years of age can be wintered at a low level and still produce satisfactorily. Unless subsequent performance of heifers wintered at a low level is very poor, it would appear that the practice of wintering replacement heifers bred to calve at two years of age at a high level of nutrition is less economical than wintering them at a low level.

One heifer in Lot 1 and two heifers in Lot 5 failed to produce calves in 1950 and 1951. In 1950, one heifer in Lot 7 weaned a calf weighing 295 pounds. This weight was 121 pounds less than the lot average. The calf produced in 1951, by this same heifer, weighed 225 pounds on July 28. This weight was 104 pounds less than the lot average. The birth date of the calves for both years was earlier than the average of the lot.

Heifers Calving as Three-Year-Olds (Lots 2, 4, 6, and 8)

The heifers of Lot 2 (low level) lost average of 77 pounds from the start of the winter period up to calving, whereas the heifers of Lots 4, 6, and 8 lost 41, 7, and 58 pounds, respectively, for the same period. The average total losses for the heifers of Lots 2, 4, 6, and 8 during the winter period (including loss at calving) were 308, 236, 199, and 261 pounds, respectively. Weight losses were from 50 to 100 pounds greater than losses of heifers of Lots 1, 3, 5, and 7,

all calving for the second time. The average weights at the start of the winter period for the heifers of Lots 2, 4, 6, and 8 were 1103, 1066, 1104, and 1082 pounds, respectively, whereas at the end of the winter period, the respective weights were 795, 830, 905, and 821 pounds. During the winter the heifers on the low level of wintering (Lot 2) lost the most weight.

To date, the heifers of Lot 2 have made the greatest gains during the summer grazing season, and, as of July 28, 1951, the average weights were 1026, 1024, 1097, and 968 pounds, respectively, for heifers of Lots 2, 4, 6, and 8.

The percentage calf crops for the heifers calving for the first time as three-year-olds were 100 for Lot 2 and 80 for Lot 6; the heifers of Lots 4 and 8 averaged 93. The heifers of Lot 6, wintered at a high level, had the lowest percentage calf crop. This was true of the Lot 5 heifers wintered at a high level and calved as two-year-olds.

One heifer (Lot 6) required help at the time of calving. Two of the three calves lost at the time of birth were in Lot 6.

The heifers calving as three-year-olds for the first time calved from one to two weeks earlier than three-year-old heifers which were calving for the second time. The heifers of Lot 2 calved an average of one week later than heifers of Lots 4, 6, and 8.

The average birth weights of calves of Lots 2, 4, 6, and 8 were 69, 65, 70, and 64 pounds, respectively. The average weights of the calves on July 2, 1951 were 262, 265, 286, and 258 pounds, respectively, for Lots 2, 4, 6, and 8. On July 28, the average weights were 313, 307, 339, and 308 pounds. There was little difference in weight gain of the calves during this twenty-six day period; however, Lot 8 calves gained an average of 8 pounds more than calves of Lot 4.

Lot 8 heifers given a supplemental ration composed of 1.5 pounds cottonseed cake, 3.25 pounds oats, and 0.25 pound molasses, and 15 grams of thyroprotein from July 2, 1951 until the calves are weaned on October, lost an average of 52 pounds per head for the first twenty-six feeding period. This loss in weight of cows receiving thyroprotein is in agreement with work reported by Blaxter (1945) and Seath (1945).

After a consideration of total weight gain and total cost of feed, it would appear that the most economical method of producing replacement heifers bred to calve at three years of age is to winter them at a low level. Under conditions of this study, heifers fed 2.5 pounds cottonseed cake or 2.5 pounds cottonseed cake plus 3 pounds oats, per head per day, during the winter were not superior in size or appearance to heifers which received only 1 pound of cottonseed cake.

McC Campbell (1920) stated that the development of heifers without grain and bred to drop their first calves at three years of age is the most practical method under range and semi-range conditions.

Blood Composition

The lowest blood plasma-carotene levels were found to occur during the months of January, February, and March. This low level was probably due to the fact that the carotene content of the dry, cured grass was at a minimum during this period. The heifers of Lots 1, 3, and 5, exposed to bulls during the summer of 1949, tended to have slightly higher plasma-carotene levels than the respective open heifers wintered at the same level. This difference was noted particularly from October 1949 to April 1950. During the first

TABLE 4.

AVERAGE BLOOD CONSTITUENTS OF THE HEIFERS
(Units per 100 ml plasma)

Lot No.	1948		1949						1950	
	10/28	12/9	1/7	2/17	4/1	5/26	9/15	10/25	12/6	1/3
	<u>Carotene (mcg)</u>									
1	214	114	96	60	434	521	401	811	412	288
2	274	124	96	61	492	628	517	446	376	235
3	276	81	72	46	434	509	462	729	352	260
4	224	72	74	43	408	485	535	469	291	224
5	239	99	52	28	522	586	483	748	526	260
6	227	79	46	23	389	474	536	456	441	223
	<u>Hemoglobin (gms)</u>									
1	13.1	10.8	13.2	13.8	13.1	13.1	13.1	12.2	12.9	13.0
2	13.9	12.8	13.6	13.8	12.9	12.3	13.3	12.6	12.5	12.8
3	13.7	12.7	14.2	14.1	13.0	13.2	13.1	12.8	12.1	13.4
4	14.1	12.9	14.2	14.4	13.0	12.6	12.8	12.6	12.0	13.4
5	13.3	12.7	13.7	14.2	12.8	14.0	12.7	12.6	12.7	13.7
6	13.4	11.1	14.1	13.0	12.8	12.2	12.6	12.1	12.4	13.8
	<u>R B C (millions)</u>									
1	6.60	4.90	6.63	5.98	5.84	5.62	5.77	5.92	5.63	6.22
2	7.02	4.93	6.68	5.80	5.69	5.50	5.74	6.30	5.63	6.16
3	7.04	5.29	6.41	5.13	5.76	5.84	5.76	6.40	5.41	6.30
4	6.87	5.42	6.66	5.08	5.56	5.19	5.73	6.34	5.52	6.49
5	6.66	5.97	6.84	5.89	5.87	5.74	5.73	6.25	5.63	6.42
6	7.02	5.47	7.12	6.05	5.77	5.27	5.75	6.20	5.52	6.76
	<u>Plasma Protein (gms)</u>									
1	7.70	8.06	7.73	7.43	7.09	7.91	7.65	7.95	7.88	7.65
2	7.89	7.60	7.64	7.31	7.32	7.79	7.56	7.77	7.96	7.77
3	7.98	7.30	7.45	7.17	7.10	7.80	7.59	7.88	7.49	8.03
4	8.00	7.74	7.67	7.29	7.23	7.59	7.61	7.76	7.63	8.31
5	7.97	7.93	7.51	7.36	7.16	8.06	7.45	7.46	7.72	8.05
6	7.83	7.82	7.38	7.68	7.20	7.74	7.56	7.57	7.83	8.20
	<u>Phosphorus (gms)</u>									
1	3.4	8.5	6.7	6.7	5.4	6.0	5.2	5.0	5.9	5.1
2	3.6	7.1	5.9	6.6	5.6	4.7	6.0	5.3	6.0	5.2
3	4.2	7.6	5.2	6.9	5.7	6.5	5.4	5.0	6.0	5.8
4	3.7	7.4	5.4	7.2	5.4	5.3	5.6	5.3	6.7	6.2
5	3.4	8.3	5.8	6.9	6.2	5.7	4.8	5.1	6.2	5.7
6	3.5	8.1	6.3	7.2	5.9	5.3	4.8	5.0	6.1	5.9

TABLE 4. (Continued)

Lot No.	1950									
	2/2	2/28	3/28	4/25	5/31	6/28	7/24	8/31	10/9	10/31
	<u>Carotene (mcg)</u>									
1	304	388	751	1124	1116	1050	1058	899	907	699
2	267	358	986	1127	994	1082	1014	876	799	651
3	208	451	853	1251	1196	958	1003	944	991	718
4	172	407	1039	1139	1035	1084	1003	848	769	620
5	198	401	842	1403	1037	1014	1044	900	863	630
6	198	399	909	1151	962	996	941	778	721	634
	<u>Hemoglobin (gms)</u>									
1	13.0	12.0	12.5	12.2	12.0	12.4	13.4	12.6	12.1	12.3
2	13.2	12.9	12.6	13.2	12.6	12.8	14.2	13.4	12.9	12.8
3	13.1	13.5	14.0	13.4	12.6	13.2	13.9	12.7	12.5	12.3
4	12.4	13.2	13.7	13.7	13.3	12.9	13.9	13.4	12.5	12.8
5	13.2	12.7	13.0	13.0	12.7	12.4	13.7	12.5	12.0	12.2
6	13.6	13.1	12.9	13.5	12.9	12.9	13.8	12.9	12.3	12.5
	<u>R B C (millions)</u>									
1	6.12	5.35	6.38	6.09	5.44	7.50	6.36	5.88	5.49	5.51
2	6.28	5.74	6.14	6.52	5.73	7.64	6.57	6.11	5.69	5.75
3	6.06	5.73	6.52	6.38	5.68	7.43	6.63	5.98	5.52	5.50
4	5.86	5.72	6.51	6.44	6.03	7.48	6.70	5.93	5.38	5.62
5	6.38	5.47	6.26	6.97	5.51	7.36	6.53	5.99	5.84	5.59
6	6.68	5.59	6.24	6.44	5.60	7.37	6.50	6.11	6.04	5.49
	<u>Plasma Protein (gms)</u>									
1	7.97	7.68	7.88	8.11	8.14	8.29	7.96	7.94	8.11	8.23
2	8.16	8.01	7.76	8.20	8.33	7.74	7.79	8.00	8.00	7.85
3	8.08	8.19	8.08	8.41	8.14	7.70	7.70	7.84	7.94	8.01
4	8.12	8.20	8.23	8.24	8.34	7.94	8.06	7.82	7.74	7.52
5	8.52	8.23	7.79	7.86	8.12	7.70	7.80	7.63	7.68	7.52
6	8.66	8.54	7.95	8.24	8.44	7.92	8.23	8.08	7.96	7.72
	<u>Phosphorus (gms)</u>									
1	5.4	4.6	5.1	4.4	5.0	5.5	5.1	5.7	4.9	5.9
2	5.4	5.6	6.3	6.0	6.2	5.8	5.8	5.8	5.0	6.2
3	5.7	4.7	5.2	6.5	5.7	5.3	5.4	5.2	4.7	6.0
4	5.9	5.7	6.5	6.3	6.3	6.1	5.8	5.6	5.5	6.4
5	5.8	5.8	5.7	5.4	5.7	5.2	5.7	4.9	4.6	5.8
6	6.4	6.1	6.4	5.7	6.0	5.9	6.2	5.0	5.1	5.9

TABLE 4. (Continued)

Lot No.	1950			1951					
	12/4	1/4	1/29	2/26	3/26	4/23	5/28	7/2	7/28
	<u>Carotene (mcg)</u>								
1	233	83	49	45	193	757	1418	1063	1009
2	245	79	46	40	215	843	1378	979	838
3	232	97	53	62	294	942	1273	883	863
4	183	75	47	57	251	967	1262	885	881
5	160	57	41	53	321	978	1134	767	656
6	159	62	45	41	296	1104	1217	718	818
	<u>Hemoglobin (gms)</u>								
1	11.8	11.8	11.7	11.3	11.6	11.3	11.0	11.5	13.3
2	12.6	12.7	12.4	12.5	12.5	11.7	11.9	12.2	13.7
3	12.5	12.4	12.6	12.7	13.0	11.9	11.8	12.5	14.1
4	12.3	12.5	13.0	12.9	12.5	12.5	12.2	12.8	13.9
5	11.8	11.9	13.1	12.2	12.3	11.9	11.5	12.2	13.2
6	12.3	12.5	13.2	12.1	13.1	12.0	10.7	12.6	13.7
	<u>R. B. C (millions)</u>								
1	5.08	6.13	5.20	5.04	5.64	4.95	5.07	7.16	6.04
2	5.13	6.37	5.47	6.00	5.49	5.10	5.49	7.80	6.23
3	5.29	6.08	5.35	5.48	5.88	4.93	5.45	7.62	6.30
4	5.12	6.10	5.43	5.51	5.55	5.00	5.57	7.79	6.38
5	5.00	6.10	5.56	5.34	5.73	4.76	5.16	8.02	6.08
6	5.16	6.32	5.61	4.51	5.72	4.76	5.44	7.88	6.22
	<u>Plasma Protein (gms)</u>								
1	7.88	7.71	8.11	6.86	7.25	7.53	7.80	8.13	8.68
2	7.44	7.34	7.73	7.21	7.42	7.67	8.05	8.13	8.46
3	8.19	7.85	7.95	7.89	7.64	7.89	7.95	8.11	8.59
4	7.78	7.90	8.02	7.85	7.71	8.10	8.05	7.95	8.78
5	8.06	7.94	8.53	8.16	8.30	8.31	8.25	7.96	7.71
6	7.84	8.00	8.40	7.52	8.33	8.24	8.30	7.93	9.01
	<u>Phosphorus (gms)</u>								
1	6.9	5.3	6.0	5.9	5.7	4.2	5.6	4.0	4.0
2	6.5	4.9	5.7	5.8	6.0	4.0	5.2	4.2	3.6
3	5.6	5.3	7.9	7.3	6.1	5.1	5.3	4.3	3.8
4	6.8	5.8	6.8	6.8	6.8	4.6	5.4	4.3	3.8
5	6.8	6.1	6.6	6.6	6.3	4.7	5.3	4.4	3.8
6	6.5	5.7	6.2	5.4	5.5	5.2	5.5	4.2	3.9

winter of the experiment, the heifers in the lots wintered at the low level (Lots 1 and 2) had higher carotene levels than those of the other lots. This fact suggested the possibility that the heifers of Lots 1 and 2 were grazing more or at least eating greater amounts of green winter grasses found growing in limited amounts in pastures. During January and February 1951, the average carotene values of the lots ranged from 40 to 97 micrograms. No vitamin A deficiency symptoms were observed in any of the heifers. Payne and Kingman (1947) reported that aged Hereford cows in Colorado have normal health and reproduction with as little as 83 micrograms of carotene per 100 milliliters of plasma.

The lowest inorganic blood plasma-phosphorus levels of the heifers were observed at the initial bleeding, October 28, 1948, and at the final bleeding on July 28, 1951. Consistent differences among the lots were not observed until the fall of 1949. From October 25, 1949 to May 31, 1950, at almost every bleeding date, the heifers of Lots 1, 3, and 5 had lower phosphorus levels than the respective open heifers of the lots wintered at the same level. This lower level of blood phosphorus may have been due to the greater demand for phosphorus entailed by the growth of the fetus and by subsequent lactation.

During the winter and early spring months, the heifers wintered at the low level (Lots 1 and 2) tended to have lower plasma-protein levels than heifers of other lots. In general, the heifers of the odd-numbered lots suckling calves had lower plasma-protein levels during the spring months of 1950 than open heifers of Lots 2, 4, and 6. No consistent differences in plasma-protein levels were observed during 1951.

Consistent differences in number of red blood cells among the lots were not observed.

The heifers of Lot 1 tended to have slightly lower hemoglobin levels at nearly every bleeding date than heifers of other lots; however, very little difference was noted among the other lots.

TABLE 5.

AVERAGE CHEMICAL ANALYSES OF FEEDS AND GRASSES; AVERAGE OF TWO YEARS

	Percent dry Matter	Per cent composition of dry matter						
		Ash	Protein	Fat	Fiber	N.F.E.	Ca.	P.
Cottonseed cake	92.95	6.52	43.76	7.34	8.08	34.30	.24	1.10
Oats	88.60	3.88	14.66	2.92	10.68	67.86	.12	.37
Cured Grass ¹								
May		9.18	9.45	3.13	29.19	49.05	.30	.217
June		8.34	7.20	3.11	32.86	48.49	.284	.183
July		7.09	7.45	3.09	33.44	48.93	.356	.183
October		6.44	3.48	2.19	37.86	50.05	.242	.072
November		6.63	2.30	1.68	40.58	48.81	.253	.073

¹ These grass samples, taken at the Fort Reno Experiment Station in 1950, represent combined average analyses of the following species: Little Bluestem, Big Bluestem, and Indian Grass.

SUMMARY

One hundred and twenty, choice, weanling, Hereford heifers of comparable breeding were divided into eight lots of 15 head to determine the effect of supplements fed during the winter and the effect of age of breeding upon their subsequent performance as herd cows. Three levels of wintering were included in this study. Two lots of heifers were wintered at a low level, four lots at a medium level, and two lots at a high level. One-half of the heifers wintered at each level were bred to calve for the first time as two-year-olds, whereas the other half of the heifers were bred to calve as three-year-olds.

As yearlings, heifers wintered at a low level made the least gain during the winter, but made the most gain on grass the following grazing season.

There was little difference observed in the birth weights of calves produced by heifers calving for the first time as two-year-olds and those produced by these same heifers as three-year-olds. Calves produced by heifers for the first time as three-year-olds were about the same weight at birth as those produced by heifers calving as two-year-olds.

More assistance was required at time of calving for heifers bred to calve at two years of age than for those calving for the first time at three years of age.

Heifers wintered at a low level and calving for the first time at either two or three years of age have produced more calves than those heifers wintered at a high level.

Two-year-old heifers, suckling calves, weighed approximately 120 pounds less than dry two-year-olds wintered at the same level.

At the present time, on the basis of data of three winters, it would appear that the practice of wintering heifers bred to calve at two years of age at a high level of nutrition is not as economical as wintering at a low level. Heifers wintered at a low level for three winters have successfully produced two calf crops; however, it appears, on the basis of comparative weights, that the low-level heifers will be slower in attaining their mature weights than heifers which have been wintered at a high level.

The supplemental summer feeding given to heifers wintered at a medium level and which calved as two-year-olds resulted in gain in weight of the heifers, but only a slight gain in weaning weight of the calves as compared to the weaning weights of calves whose dams received no supplemental feed. Although it is too early to evaluate the practicality of summer feeding followed in this experiment, it appears that any advantage would be more than offset by the cost of such feed.

At the end of two years, heifers bred to calve at three years of age and wintered at a low level weighed approximately the same as heifers wintered at a high level. Under conditions of this study, it would appear that the most economical method of producing replacement heifers bred to calve at three years of age is to winter them at a low level (1 pound of cottonseed cake per head per day plus dry grass).

The summer feeding of 15 grams of thyroprotein, in addition to 5 pounds of supplemental feed, per head per day to the heifers wintered at a medium level and which calved as three-year-olds resulted in a loss of 52 pounds per head for the first twenty-six days of the feeding period. Weaning weights of the calves, and the heifers' gain or loss in weight will be observed to give

some indication as to the advisability or danger of feeding thyroprotein to beef cows during the summer months.

Seasonal variation in plasma-carotene levels were observed during the years of this study. Heifers exposed to bulls in the summer of 1949 had slightly higher carotene levels during the following winter than respective open heifers.

Heifers calving at two years of age had lower inorganic phosphorus levels than open heifers wintered at the same level.

The plasma-protein level tended to be lower for heifers wintered at a low level than for heifers of the other lots. No consistent differences between lots were observed in red cell count or hemoglobin level.

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THE EFFECT OF LOW PHOSPHORUS RATIONS UPON CAROTENE METABOLISM IN RUMINANTS

INTRODUCTION

This part of the thesis was a study on the effect of low phosphorus rations upon carotene metabolism in ruminants. Preliminary results on the influence of phosphorus on carotene metabolism were recently reported by Ross and Gallup (1949). They found an inverse relationship between the carotene and inorganic phosphorus content of the blood plasma of beef cows. Cows with abnormally low plasma-phosphorus levels were observed to have higher plasma-carotene levels than cows with normal or above normal levels.

Experiments to determine the effect of low phosphorus rations upon carotene metabolism were conducted with lambs, yearling Hereford steers, and aged Hereford cows. The research has been divided into three experiments: Experiment 1 was a study of low phosphorus rations upon carotene metabolism in lambs; experiment 2 was a study of the effect of low phosphorus rations upon the carotene metabolism and storage of vitamin A in the liver of beef steers; and experiment 3 was a study of the effect of low phosphorus rations upon metabolism of carotene, storage of vitamin A in the liver, concentration of carotene and vitamin A in the colostrum and milk of beef cows and upon phosphorus, carotene, and vitamin A of the blood of their calves, upon liver storage of carotene and vitamin A of the calves, and upon their growth.

REVIEW OF LITERATURE

Although numerous investigators have reported on the symptoms of phosphorus and vitamin A deficiencies in ruminants and on the requirements for each, very little work has been reported on the direct effect of low phosphorus rations upon carotene metabolism. Ross and Gallup (1949) first reported an inverse relationship between inorganic phosphorus and carotene in the blood of cows maintained on different levels of phosphorus. The data from which they observed this relationship were obtained from a study on the phosphorus requirement of beef cows grazed in two different areas of Oklahoma (Ross, et al., 1948, 1949, and Robertson, 1948). Each area was stocked with heifers which were fed low, medium, or high levels of phosphorus. The phosphorus intake of the low level group at each area was restricted to forage and protein supplement during the winter; the medium level group received an intake of 1.5 grams phosphorus per 100 pounds body weight; the high level group received an intake of 2.5 grams phosphorus per 100 pounds body weight. Blood samples were taken at periodic intervals, and inorganic phosphorus and carotene determinations were made on the blood plasma. The blood-phosphorus value of the low phosphorus group at the Wilburton area was abnormally low during the winter and early spring months; however, their carotene values tended to be slightly higher than values of cows receiving a high level of phosphorus, even though the high level cows consumed more prairie hay. The cows receiving the high level of phosphorus had a greater intake of carotene but less carotene in the blood than cows receiving the low level; it appeared that the high level cows may have been more efficient than the low level cows in the conversion of carotene to vitamin A. The phosphorus values

of the high level cows rose sharply during the winter when those cows ate the amount of phosphorus provided. The plasma-carotene and inorganic phosphorus values of cows receiving the medium level of phosphorus ranged between values of low and high level cows. Analyses of blood of cows at the Stillwater range revealed practically the same picture.

Van Arsdell, et al. (1949) fed a low phosphorus-high calcium rachitic diet to weanling rats to determine its effect upon utilization of carotene. Twenty-six rats were divided into two groups: one group was fed the rachitic diet supplemented with calcium carbonate, and the other group was fed the rachitic diet supplemented with monocalcium phosphate. Both groups were supplemented with 0.75 mg. carotene plus 0.5 mg. alpha-tocopherol per day. The rachitic diet contained 1.22 per cent calcium and 0.234 per cent phosphorus; the non-rachitic diet contained 0.74 per cent calcium and 0.417 per cent phosphorus. A paired-feeding technique was used. Three rats in each group were killed after one week and vitamin A determinations were made on the blood and liver and phosphorus determinations on the blood plasma. At the end of a month, all rats were killed, and analyses were determined on the blood plasma and liver. There was very little difference in vitamin A content of the blood and liver between the two group; despite this fact, the phosphorus level of the blood of the rats of the nonrachitic diet was approximately twice as high as that of the rats on the rachitic diet.

Phosphorus

The requirements of the animal body for phosphorus and calcium, for upkeep and normal development, have been the subject of research for many years. Conclusive evidence that phosphorus, in combination with calcium, constitutes

the greatest portion of the mineral matter of the animal body has been furnished by numerous determinations.

In addition to the important role of phosphorus in normal skeletal development, it is known to be one of the most important elements in the physiological process of the body. Gortner (1929) stated that phosphorus is concerned in the metabolism of nearly all body tissues. It is present in the nuclei of all cells and is vitally concerned with the complete role in the chain of events occurring in muscular contraction and in the transfer of energy. Phosphorus is considered to be indispensable in carbohydrate, fat, protein, and lipid metabolism, and as a constituent of certain enzymes. The maintenance of the reaction of the blood, as well as the maintenance of calcium equilibria of the blood, is a function attributed to phosphorus.

Numerous studies have been conducted relative to the phosphorus requirements of livestock. A report of the committee on Animal Nutrition of the National Research Council (1945) advised that cattle should be allowed free access to a phosphorus-rich mineral mixture if the forage is apt to fall much below 0.15 per cent phosphorus on a dry-matter basis. This committee recommends a range of 12 grams of phosphorus per head daily for wintering weanling calves and yearlings to 24 grams for cows nursing calves.

Theiler (24) reported that the first symptom of a phosphorus deficiency was usually depraved appetite as evidenced by the chewing of bones, sticks, hair, and the excessive ingestion of salt and dirt in an effect to secure the needed phosphorus. This condition was accompanied by a considerable loss of weight which greatly impaired the development of the body. The advanced stages of this deficiency caused marked skeletal changes, and Theiler (1931) concluded

that rickets and osteomalacia were definitely caused by a lack of phosphorus. Schmidt (1926) observed that growing animals frequently developed a peculiar stiff, "creep" gait as though it were painful to walk. Madsen (1942) stated that osteomalacia, a disease of mature animals, is characterized by a softening and replacement of bone with osteoid tissue resembling uncalcified bone.

In the classical studies of Theiler, Green, and Du Toit (1924), it was found that a lack of adequate available phosphorus in the diet was manifested by a lack of appetite and failure to utilize food economically. Riddell, Hughes, and Fitch (1927), (1934), and Eckles and Gullickson (1927) disclosed that a lack of phosphorus did not lessen the digestibility of the ration but it rearranged the metabolism so that the digestible nutrients were utilized inefficiently. The latter investigators concluded that cows on a low phosphorus diet required 20 per cent more digestible nutrients to maintain live weight. Kleiber, Goss, and Guilbert (1936) also noted that a phosphorus deficiency is a limiting factor in the economical utilization of feed.

It has been conclusively demonstrated that the utilization of phosphorus in the ration is modified by many factors relating to the ration or to the environment. The ratio of calcium to phosphorus in the diet has been found to have an important influence upon the metabolism of both elements. If either is present in inadequate amounts, the other is not utilized properly, even though it is present in normal quantities.

Early recommendations on the ratio of these two elements were based on the fact that they exist in rations of about 2:1 and 1.25:1 in the bone and milk, respectively. McCollum and associates (1921) observed in their early studies that a ratio of calcium to phosphorus in the diet was perhaps of greater

significance to the welfare of the animal than the absolute amounts of each. Meigs, et al. (1926), using four lactating cows, suggested that an excess of calcium in the ration may interfere with the assimilation of phosphorus.

Haag, Jones, and Brandt (1932) indicated that a calcium-phosphorus ratio of 10.5:1 was no more detrimental than one of 7.6:1. Bethke, Kick, and Wilder (1932) pointed out that, for optimum growth and bone formation, a calcium-phosphorus ratio of 2:1 gave best results. Increasing the ratio of these two elements from 1:5 to 5:1 causes a progressive decrease in growth, bone ash, and the inorganic content of the blood. When the ratio was changed to 1:0.25, growth was markedly depressed. Maynard (1947) stated that the desired ratio of these two elements is between 2:1 and 1:2 but that adequate nutrition is possible outside these limits. With adequate vitamin D in the ration, the ratio becomes of less importance.

Significance of Blood-Phosphorus Determinations

The concentration of inorganic phosphates in the blood plasma of cattle as an aid in clinical diagnosis of a phosphorus deficiency even before physical symptoms became apparent was first reported by Palmer and Eckles (1927).

Thieller, et al. (1927) reported that the phosphorus content of the blood of cattle receiving adequate phosphorus varied from 4 to 9 mg. per 100 ml. of whole blood, with an average value of 5.2 mg. MacVicar (1947) found that blood plasma values were higher than whole blood values by approximately 32 per cent.

Black, et al. (1942) observed symptoms of aphosphorosis in cattle grazing a phosphorus-deficient range when the blood-phosphorus content fell below

4 mg. per 100 ml. of whole blood. In contrast to this work, Knox, Benner, and Watkins (1941) found that cows with blood plasma levels of from 2.0 to 3.0 mg. per cent phosphorus for short periods in winter and spring and from 3.00 to 4.5 mg. per cent in the summer were in excellent health. Huffman and others (1933) stated that blood plasma-phosphorus values lower than 4.0 mg. per cent are to be questioned, especially if the animal is less than one year of age. Greaves, Maynard, and Reeder (1934) gave 5.0 mg. per cent as the borderline value.

Van Landingham, Henderson, and Bowling (1935), studying the effect of age on the inorganic phosphate of the whole blood, found that values tended to rise, reaching a peak at seven months of age and decreasing progressively to 24 months. Malan (1928) and Green and Macaskill (1928) found that the blood of newborn calves contained twice as much inorganic phosphorus as that of their dam. This finding has been confirmed by Eckles, Gullickson, and Palmer (1932).

Small-percentage calf crops, breeding difficulties, and difficulty at time of calving have been associated with a lack of available phosphorus in the feed by Theiler (1924), Eckles, et al. (1926), Schmidt (1926), Black, et al. (1943), Lantow (1933), Knox, et al. (1941), Ross, et al. (1948), (1949), and Hart and Guilbert (1928).

Vitamin A

The first evidence that vitamin A was needed for reproduction was reported by Hart, et al. (1911), (1924). In 1926, Jones, Eckles, and Palmer showed vitamin A to be an indispensable factor in the diet of calves.

Bechdel, et al. (1928) studied the effect of feeding five heifers, one to two and one-half years of age, a ration deficient in vitamin A. They observed that

the vitamin A deficiency caused edema in the front legs, declining appetite, and increased respiratory rate. One animal became blind. In two cases of pregnancy, the calves were born dead, one being born more than two months premature.

Guilbert and Hart (1935) found that night blindness was the first detectable clinical symptom of vitamin A deficiency and that it was a delicate index upon which minimum requirements could be based. They stated that the daily minimum carotene requirement of cattle is 26 to 33 micrograms per kilogram live weight. Carotene intake at the level of about 29 micrograms per kilogram live weight prevented or cured clinical symptoms of deficiency. Vitamin A deficiency that had progressed to the point of night blindness and convulsions did not inhibit the occurrence of oestrus. Guilbert and Hart advanced the hypothesis that the vitamin A requirement is related to body weight rather than to energy requirement. Reports of their later experiments (1937) substantiated this hypothesis.

Watkins and Knox (1950) determined the carotene content of blood plasma of Hereford breeding cows grazing on a southern New Mexico range. These values seemed to indicate that a vitamin A deficiency might occur only in case of prolonged drought or extremely abnormal conditions. Hart and Guilbert (1933) stated that in the bovine, as in other species, the concentration of vitamin A in the liver varies with carotene concentration of feed. The reserve supply that has accumulated in the body during the green feed season is sufficient to carry animals through the dry season under average conditions.

Guilbert and Hart (1934), Riggs (1940), Fry and Jensen (1946), and Eden (1948) have stated that the ability of cattle to store vitamin A in times of

abundance against periods of privation is extremely important under range conditions. During the period of active growth, there is a greater requirement for vitamin A and, consequently, less vitamin A is stored than when adult size is attained.

Davis and Madsen (1941) reported that an inadequate carotene intake in cattle could be determined by blood analysis. The carotene and vitamin A content of blood plasma at any particular time was dependent on the current intake and previous storage of these factors. When cattle were depleted of carotene reserves, the blood-carotene level was dependent on intake alone. Vitamin A values tended to reach a stable level and did not increase proportionally with intake as did the carotene level. Braun (1945) and Hibbs (1946) stated that the plasma content of carotene and vitamin A does not give an absolute measure of the storage of these substances in the animal's body. Although the plasma content of carotene and vitamin A may not be an absolute measure of the storage in an animal's body, these values do reflect the intake of the factors.

It is generally accepted that the amount of vitamin A or carotene needed for normal reproduction in cows is greater than the amount needed for protection against night blindness, a well-known symptom of vitamin A deficiency. Kuhlman and Gallup (1941), (1942), have set forth minimum requirements for carotene intake in Jersey cattle for normal conception, reproduction and lactation, as follows: conception, 20-29 mcg. per pound of body weight; reproduction, 40-45 mcg. per pound of body weight; and lactation, 40-45 mcg. per pound of body weight. In a later report, Kuhlman and Gallup (1942) stated that, when blood plasma values in 18 and 24-month old Jersey heifers dropped below 150 mcg. per 100 ml. plasma, difficulties were often encountered at parturition.

Moore, et al. (1947) stated that cows at the Beltsville Experiment Station which received 80 to 100 milligrams of carotene per day gave birth to normal calves. Guilbert and Rochford (1940) reported that a 1000-pound beef cow required about 75 milligrams of carotene during the last months of gestation; this amount is five times that needed for maintenance. Payne and Kingman (1947) report that the carotene-blood plasma level of ~~frange~~ Hereford heifers calving for the first time must be at least 118 and that for aged cows must be at least 83 micrograms per 100 millileters, for adequate health and reproduction.

Sutton and Soldner (1943) reported that blood plasma-carotene and vitamin A values of dairy cows remained nearly constant up to a week before calving. Just prior to calving, a decline of both plasma-carotene and vitamin A occurred, and a further drop was observed immediately after parturition. Sutton, et al. (1945) found that the maximum decrease in blood plasma-carotene values of lactating cows occurred one week following parturition. The maximum decrease in blood plasma-vitamin A was reached three days after parturition.

Thomas, et al. (1946) found that the concentration and total output of vitamin A and carotene in the colostrum of the cow is influenced by the ration fed during the two months immediately prior to parturition. Speilman, et al. (1946) stated the the prepartum diet of the normal bovine may influence markedly the vitamin A and carotene reserves of the newborn calf. However, Hart and Guilbert (1933) found the the livers of newborn animals tend to be comparatively low in vitamin A, irrespective of the diet of the dam during gestation.

Keener, et al. (1942) observed the effects of vitamin A deficiency in calves and found effects similar to those previously reported by other workers, including exophthalmia, roughness of coat, slowness of growth, muscular incoordination, diarrhea, pneumonia, blindness, and death. The gross and microscopic pathology observed at autopsy included a cystic condition of the pituitary, a constriction of the optic foramen, and various degrees of histopathological changes in the tissues of the intestines, liver, kidney, and testicle.

Hansen, et al. (1946) found that vitamin A of colostrum from barn-fed heifers in the first lactation was more than twice the vitamin A content of the colostrum from the same cows in the second lactation. Seven-fold variation in the colostrum vitamin A potency occurred in these cows, even though the cows were fed identical rations and were maintained under uniform conditions during two lactating periods. An increase in the blood plasma-vitamin A concentration of the newborn calf was observed following the ingestion of colostrum, and the percentage increase tended to reflect the concentration of vitamin A present in the colostrum of their dams.

Sutton, Warner, and Kaeser (1947) found a rapid decline of carotene and vitamin A in colostrum and milk with each successive milking. Colostrum was approximately ten times as potent in carotene and six times as potent in vitamin A as normal milk. Levels closely approaching normal milk were reached at the end of the third day.

Henry, et al. (1949) studied the relationship between the maternal reserves of vitamin A, the vitamin A content of the milk, and the stores of vitamin A present in the young at birth and amassed during suckling; they reported that the vitamin A content of the milk and the livers of the young

rats was influenced by the amount of vitamin A taken by the mother during lactation more than by her liver reserves.

Factors Affecting Carotene Metabolism and Storage of Vitamin A in the Liver

Thorbjarnarson and Drummond (1938) stated that the storage of vitamin A in the liver is facilitated by the presence of fat in the diet and its subsequent deposition in the liver. Considerable storage of vitamin A takes place when the animals are given an excess, but the reserves are depleted when an excess is no longer given. They further stated that fat leaving the liver may take with it vitamin A.

Bentley and Morgan (1946) found that when carotene was fed there was a definite depression of vitamin A storage on the high-fat as compared with the low-fat diets.

Moore (1940) stated that the vitamin A reserves of rats kept for prolonged periods on a diet deficient in vitamin E but a diet to which vitamin A was supplied as halibut oil were always much lower than those of control animals receiving supplements of vitamin E. Kachmar, et al. (1950) stated that the feeding of a low vitamin E ration did not impair the utilization of carotene and vitamin A. Harris, Swanson, and Hickman (1947) reported that the efficiency of transfer of ingested vitamin A and carotene into vitamin A and carotene in the milk was not increased by supplementation with vitamin E alone.

Gallup (1941) reported that the daily administration of cod liver oil to calves on winter rations caused about a 50 per cent decrease in plasma-carotene. Deuel, et al. (1942) observed that the decrease in milk carotene,

which followed administration of 2,100,000 I. U. of shark liver oil, appeared to be caused by vitamin A per se. A rough proportionality existed between the levels of carotene in blood and in the milk.

Sexton, Mehl, and Deuel (1946) stated that no increase in vitamin A could be demonstrated after the parenteral injection of carotene. The localization of the intestine as the site of transformation of beta-carotene to vitamin A in the rat has been demonstrated by Mattson, Mehl, and Deuel (1947), Goodwin and Gregory (1948), and Glover, Goodwin, and Morton (1948).

Since Wooley (1947) stated there was an antagonistic relationship between a number of nutritional factors and their structural analogs, a number of investigators have reported antagonistic relationships between certain precursors of vitamin A and vitamin A per se. Kemmerer, Fraps, and Demottier (1947) found that a mixture of xanthophylls in a concentrate from spinach decreased the storage of vitamin A in the liver of vitamin-A deficient rats supplemented with carotene. Johnson and Baumann (1948) stated that cryptoxanthine was equal to beta-carotene in producing stores of vitamin A and that its relative activity for storage was twice that for growth. Kelley and Day (1950) stated that large amounts of xanthophyll administered with beta-carotene or vitamin A decrease the utilization of of these substances for tissue storage of vitamin A in rats. They had no effect on the rate of disappearance of vitamin A already present in the tissues, indicating that inhibitory action may occur in the alimentary tract. Kelley and Day further stated that the effect of the xanthophyll is not due to a specific impairment of the enzymatic mechanism for the conversion of carotene to vitamin A.

Esh and Sutton (1948) stated that the feeding of lecithin with a vitamin A

or carotene supplement to rats resulted in greater gains in weight and increased liver storage of vitamin A. When choline was fed with these supplements, the liver storage of vitamin A was no different from that of the groups which received the vitamin supplement alone. These workers concluded that lecithin enhanced both the absorption and utilization of vitamin A and carotene and that the choline fraction of the lecithin molecule was not responsible for this effect. Slanetz and Scharf (1945) reported that commercial soybean lecithin markedly influenced storage and blood levels of vitamin A in the rat. Shaw, Moore, and Sykes (1951) found that a marked depression in plasma and liver vitamin A produced by feeding ground raw soybeans to calves on an adequate, but low, intake of carotene.

Heimer, Maslow, and Sobel (1949) divided litters of wistar rats into three groups--control, thyroidectomized, and thyroxin treated--and killed them after 45 days of feeding. It was found that vitamin A storage in the livers of rats on a vitamin A-free diet was highest in the thyroidectomized rats. Baumann and Moore (1939) found that when groups of normal and hyperthyroid rats containing uniform stores of vitamin A were fed a diet low in vitamin A the amounts of this factor remaining in the liver were usually at least as high in the hyperthyroid group as in the control group. Johnson and Baumann (1947) reported that, in some trials, the administration of thyroxine actually appeared to decrease the rate of utilization of vitamin A. Hyperthyroid rats fed moderate amounts of fish liver oil stored approximately as much vitamin A as control rats.

Johnson and Baumann (1948) studied the relative significance of growth and metabolic rate upon the utilization of vitamin A by the rat and concluded

that rats whose growth was restricted by diets inadequate in calories, thiamine, or tryptophane retained more hepatic vitamin A than control rats that grew normally. Comparisons between rats of similar size suggested that metabolic rate also influenced vitamin A depletion; desiccated thyroid hastened the depletion of vitamin A reserves somewhat, whereas thiourea and thiouracil delayed it.

The time taken to exhaust the reserves of vitamin A in the liver is dependent not only on the initial size of the reserve but also on the rate of utilization of the vitamin by the body. This rate is increased in growing animals and also in animals where the basal metabolic rate has been raised as in hyperthyroidism (Johnson and Baumann, 1948). Hickman (1946) suggested that the rate of depletion is also dependent on the actual size of the initial reserve. This hypothesis has been confirmed by Frey and Jensen (1946 and 1947), who showed that, for steers, the rate of depletion decreased as the amount of vitamin A in the liver decreased.

Davies and Moore (1935) gave adult rats, already possessing high liver reserves of vitamin A, massive doses of vitamin A concentrate. The mean liver levels rose to 18,000 B.U./gram, an amount which was enough, theoretically, to last a century if used up at a rate corresponding to physiological requirements. The rats were then restricted to a diet free of vitamin A, and a rapid elimination of vitamin A from the liver took place, the mean level falling to 400 B.U./gram after twelve weeks. At this level, a condition of stable storage of the vitamin appeared to have been attained, as no further fall occurred after twelve additional weeks on the vitamin A-free diet.

The fate of excess vitamin A stores during depletion was studied by

Popper and Brenner (1942). They used fluorescent microscopy for the location of vitamin A in the liver cells and suggested that the destruction of excessive vitamin A probably takes place in the Kupffer cells. Normally, vitamin A is stored in the other liver cells; however, when massive doses are given, the Kupffer cells also show intensive fluorescence, characteristic of vitamin A. During depletion, as much as can be determined by fluorescence, the Kupffer cells are the first to give up vitamin A. Decrease of fluorescence is not accompanied by an increased concentration of vitamin A in the blood; hence, the evidence suggests that the disappearance of vitamin A from the Kupffer cells does not represent a release of the vitamin into the blood but that it is actually being destroyed in these cells.

Krause (1949) reported an examination made of the relationship between serum level and liver content of vitamin A in normal rats and those under the influence of varying degrees of vitamin A depletion. The following points were observed: (1) An inverse relationship was observed between blood and liver levels of vitamin A in normal male and female rats and those depleted female rats whose liver content ranged as low as 600 I.U. per total liver. (2) When the total liver content fell below 600 I.U. in the male rats, there was a parallelism between blood and liver levels.

Vavich and Kemmerer (1950) stated that the size of the rats used as experimental animals markedly influenced the utilization of carotene for liver storage of vitamin A.

THE EFFECT OF LOW PHOSPHORUS RATIONS UPON CAROTENE METABOLISM IN LAMBS

EXPERIMENTAL PROCEDURE

Lambs were fed a ration low in phosphorus for a period of 268 days, in order to deplete them of phosphorus. They were of different ages and averaged 55 pounds when placed on experiment August 2, 1949. At the start of the experiment, they were treated with nicotine sulphate and copper sulphate. During and following the depletion period, the lambs were kept inside without access to green material of any kind.

The lambs were fed a ration composed of 1/4 pound corn gluten meal, 1 1/4 pounds cottonseed hulls and approximately 5 grams salt per head per day. This ration provided an intake of 1.1 grams of phosphorus and 1.4 grams calcium per 100 pounds live weight and had a calcium-phosphorus ratio of 1.3:1. Because of its relatively low phosphorus content, corn gluten meal was selected as the protein supplement. Cottonseed hulls were used because they are low in phosphorus and devoid of carotene. The chemical composition of the feeds is given in Table 1.

Blood samples were taken at the beginning of the experiment and at intervals of approximately one month during the depletion period. The blood was collected in citrated tubes and analyzed for vitamin A by the Kimble procedure (1939) and for inorganic phosphorus by the Fiske and Subbarow method (1925).

One hundred ninety-two days after the lambs were placed on the depletion ration, the average inorganic plasma-phosphorus level was 8.5 milligrams and the vitamin A-plasma level was 3.2 micrograms per 100 milliliters of

TABLE 1.

CHEMICAL COMPOSITION OF FEEDS USED IN EXPERIMENT 1

	Percent dry Matter	Per cent composition of dry matter						
		Ash	Protein	Fat	Fiber	N.F.E.	Ca.	P.
Corn Gluten Meal	92.45	1.85	45.16	2.53	3.28	47.18	0.09	0.343
Cottonseed Hulls	92.00	----	3.69	----	----	-----	0.12	0.037
Dicalcium Phosphate	96.52	----	-----	----	----	-----	25.10	18.74
Calcium Carbonate	99.14	----	-----	----	----	-----	38.30	-----

plasma. The average phosphorus values were higher than they were at the start of the experiment. Calcium determinations were made, by use of the Clark and Collip procedure (1925). The average calcium level was found to be 8.2 milligrams per 100 milliliters of plasma; this average was lower than the normal range of 9 to 12 milligrams of calcium in the blood plasma of sheep, as reported by Dukes (1947).

The ration was changed because of the low energy content of the ration, the high inorganic phosphorus content, and the low vitamin A and calcium content of the blood plasma. The ration fed the lambs for the remainder of the depletion period was composed of 0.15 pound corn gluten meal, 0.15 pound starch, 0.15 pound sugar, 1 pound cottonseed hulls, 2 grams of calcium carbonate, 0.45 gram Delsterol, which provided approximately five times the minimum vitamin D requirement (National Research Council 1945), and sufficient carotene concentrate to provide approximately nine times the minimum carotene requirement. This ration provided an intake of 0.67 gram of phosphorus and 2.27 grams of calcium per 100 pounds of live weight and a calcium-phosphorus ratio of 3.4:1.

The minimum carotene requirement was estimated from the work of Guilbert, et al. (1937, 1940); 13.6 micrograms per pound of body weight was used as the minimum. The carotene used was a crude carotene concentrate prepared from alfalfa which had a potency of 20,000 micrograms per gram. Seventy grams of the crude carotene concentrate, 400 ml. of corn oil, and 100 ml. of Skelly Solve "F" were placed in a Waring Blendor and mixed until an emulsion was formed. After cooling, the emulsion was stirred by an electric stirrer; and 5 cc. aliquot portions, containing 13,600 micrograms, were pipetted

into 1/4-ounce capsules. These capsules were kept under refrigeration, and one capsule was given to each lamb every other day.

Two of the nine lambs died of unknown causes during the depletion period. Phosphorus values of blood samples taken one day before death were above 6 milligrams per 100 milliliters of plasma. Bone sections from the femur of these two lambs showed the shaft to be small and the walls very thin.

After the lambs had been on the depletion ration for 268 days, they were divided into two lots according to their body weights, plasma-vitamin A and plasma-inorganic phosphorus levels. Blood samples were obtained, and the lambs were weighed at approximately ten-day intervals. The lambs were kept together in a dry lot and were fed individually, in stanchions, twice a day.

The three lambs in Lot 1 were continued on the low phosphorus ration; the ration of the four lambs in Lot 2 was supplemented with sufficient dicalcium phosphate to provide approximately three grams of phosphorus per 100 pounds of live weight. The composition of the ration fed to each lot is given in Table 2. All lambs continued to receive approximately nine times the minimum carotene requirement and approximately five times the minimum vitamin D requirement. The ration of the lambs in Lot 1 was supplemented with sufficient calcium carbonate to provide the same intake of calcium for both lots.

Thirty-two days after phosphorus supplementation was given to the lambs of Lot 2, all lambs were slaughtered. The livers were collected at this time; representative samples were wrapped in cellophane paper and kept frozen until they were analyzed for vitamin A. The method of analysis was that of Gallup and Hoefler (1946). A femur was taken from each carcass, and a cross-section of this was ashed, according to the A.O.A.C. method (1945).

TABLE 2.

RATIONS FED TO THE LAMBS OF EXPERIMENT 1¹

Lot No.	Number of Lambs	Corn Gluten Meal	Cottonseed Hulls	Sugar	Starch	Calcium Carbonate	Dicalcium Phosphate	Daily Intake gms/100 lbs. body weight		Ca:P Ratio
		lbs.	lbs.	lbs.	lbs.	gms.	gms.	Ca	P	
1	3	0.20	1.00	0.20	0.20	5	---	4.03	0.85	4.7:1
2	4	0.20	1.00	0.20	0.20	---	8	4.38	3.30	1.3:1

¹ Each lamb supplemented with 13,500 micrograms carotene every other day. All lambs received 0.45 Delsterol every day.

RESULTS AND DISCUSSION

The lambs maintained their weight throughout the experiment; however, growth was retarded, and the lambs appeared weak and emaciated and exhibited a depraved appetite; some were observed to eat wool. These symptoms are similar to those reported by Beeson, et al. (1937) for a phosphorus deficiency.

The inorganic phosphorus values in the blood plasma of the lambs are given in Table 3. In spite of the 268-day depletion period during which the lambs received from 0.67 to 1.1 grams of phosphorus per 100 pounds live weight, the lambs had approximately the same average inorganic phosphorus values as they did at the start of the depletion period. The average values for all lambs had only decreased from 5.6 to 5.0 milligrams phosphorus per 100 milliliters of plasma. At one bleeding, 192 days after the start of the experiment, the average phosphorus value for all lambs was 8.5 milligrams. The calcium value at this time was 8.2 milligrams. The low calcium level in the plasma may have been due to an inadequate intake of calcium or to a vitamin D deficiency, because the lambs were kept inside and no vitamin D was supplemented at that time. The low energy intake may have caused metabolic disturbances which were reflected in low calcium-plasma levels. The metabolism of calcium, phosphorus, and vitamin D is closely interrelated; rickets may tend to develop when these interrelations are not normal. In rickets, there is an increase of the phosphatase content of the blood and bones.

The average phosphorus values of the lambs in Lot 1, which received no phosphorus supplement, decreased from 4.99 milligrams at 268 days to 3.43

TABLE 3.

INORGANIC PHOSPHORUS CONTENT OF PLASMA OF LAMBS ON EXPERIMENT 1

AND PER CENT ASH OF BONES

Lot	Lamb No.	Plasma Inorganic Phosphorus (mg per 100 ml)					Per cent Ash Dry, Fat-free Bones	
		Aug. 2	Feb. 10	Apr. 27 ¹	May 10	May 19	May 29	May 29
Days on experiment		0	192	268	281	290	299	299
1	1208	4.4	8.88	4.88	5.04	3.72	4.60	67.60
	1475	4.2	5.32	5.48	1.16	1.32	1.20	65.83
	1500	4.6	9.20	<u>4.60</u>	<u>3.08</u>	<u>3.36</u>	<u>4.48</u>	<u>66.99</u>
Lot average				<u>4.99</u>	<u>3.09</u>	<u>2.80</u>	<u>3.43</u>	<u>66.81</u>
2	1471	6.4	8.80	2.84	8.40	9.64	8.92	67.41
	1474	6.3	8.60	5.64	5.20	5.16	5.84	67.57
	1478	7.2	11.04	5.72	6.32	6.12	6.20	67.51
	1485	5.8	6.48	<u>6.08</u>	<u>7.12</u>	<u>6.24</u>	<u>9.24</u>	<u>69.00</u>
Lot average				<u>5.07</u>	<u>6.76</u>	<u>6.79</u>	<u>7.55</u>	<u>67.87</u>
Average all lambs		5.5	8.3					

¹ Phosphorus supplementation was started at this date for lambs of lot 2.

milligrams at the termination of the experiment. Two lambs in Lot 1 maintained, apparently, normal values, whereas the phosphorus values of lamb 1475 dropped from 5.48 to 1.20 milligrams at the end of the experiment. The results of these phosphorus determinations on the lambs of Lot 1 substantiate the previous work of Beeson, et al. (1937) who reported that lambs receiving 1.31 grams of phosphorus daily showed signs of a phosphorus deficiency. These workers stated that, after 133 days of feeding on the low phosphorus ration, the blood-phosphorus levels were reduced from 5.00 milligrams to 3.87 milligrams per 100 milliliters of plasma. Several of these lambs had a lower value than that indicated by the average, and other lambs had a normal plasma-phosphorus level. In another lot which received 1.7 grams of phosphorus, the average plasma-phosphorus level was 5.25 milligrams, although, obviously, some of these lambs were definitely deficient in phosphorus.

In view of the fact that some of the lambs in this experiment exhibited symptoms of a phosphorus deficiency and yet maintained normal inorganic phosphorus levels in the blood plasma, it is apparent that the concentration of the inorganic phosphorus in the blood plasma might not be a true indication of the phosphorus metabolism of lambs which have been receiving a low intake of phosphorus for an extended period.

The average phosphorus values of the lambs in Lot 2, which received additional phosphorus, increased from 5.07 milligrams at 268 days to 7.55 milligrams at the end of the experiment. Beeson, et al. (1937) and Gallup and Briggs (1950) stated that lambs require about 2.5 grams of phosphorus daily per hundred pounds of live weight for efficient and rapid gains.

A section of the femur from each lamb was ashed; the per cent ash of the dry, fat-free bones is given in Table 3. There was very little difference between the ash of the bones of the lambs from the two lots; however, the walls of the bones of Lot 2 lambs appeared to be thicker than those from Lot 1. The average ash content of the bones from the lambs of Lot 1 was 66.81, whereas the Lot 2 average was 67.87. Lamb 1475, which had an inorganic phosphorus level in the plasma of 1.20 milligrams, the lowest of any lamb, had an ash value of 65.83, the lowest ash value for any lamb. These ash values are lower than those found by Nash (1950), who reported ash values averaging 69.5 per cent for ewes maintained on apparently inadequate intakes of phosphorus.

The vitamin A content of the blood plasma of the lambs is shown in Table 4. The average vitamin A values increased in all lambs following carotene supplementation given 192 days after the initiation of the experiment. The vitamin A values of the lambs on the 268th day varied from 9.8 to 19.7 micrograms. The average values were about 20 micrograms lower than that usually found in comparable lambs given fattening rations containing roughage of high carotene content (Gallup, et al., 1951). The lambs were nervous and excitable; these symptoms of vitamin A deficiency correspond to those reported by Klosterman, et al., (1949).

From the 268th day until the end of the experiment, the vitamin A values increased for lambs of both lots. The average plasma-vitamin A level of the lambs of Lot 1 was 14.3 at 268 days and 20.4 micrograms at the end of the experiment. For the lambs of Lot 2, the average vitamin A values increased from 15.1 to 18.2 micrograms. Although the vitamin A values

TABLE 4.

VITAMIN A CONTENT OF PLASMA AND LIVER OF LAMBS IN EXPERIMENT 1.

Lot	Lamb No.	Plasma Vitamin A (mcg per 100 ml)					Liver Vitamin A (mcg per gram)	
		Aug. 2	Feb. 10 ¹	Apr. 27 ²	May 10	May 19	May 29	May 29
Days on experiment		0	192	268	281	290	299	299
1	1208	31.2	3.6	13.6	20.3	20.3	20.3	15.7
	1475	21.5	2.8	17.7	27.5	28.7	20.8	12.0
	1500	20.0	1.9	<u>11.7</u>	<u>23.5</u>	<u>22.4</u>	<u>20.0</u>	<u>12.0</u>
Lot average				14.3	23.8	23.8	20.4	13.2
2	1471	28.7	2.8	19.2	24.9	25.2	19.2	10.8
	1474	42.7	3.2	11.7	15.6	18.2	15.1	15.2
	1478	16.8	None	9.8	9.8	15.1	15.3	10.3
	1485	33.2	7.0	<u>19.7</u>	<u>25.2</u>	<u>23.0</u>	<u>23.0</u>	<u>10.8</u>
Lot average				15.1	18.9	20.4	18.2	11.8
Average all lambs		27.8	3.1					

¹ All lambs supplemented with nine times carotene requirement starting this date.

² Lambs of Lot 2 supplemented with additional phosphorus this date.

increased more for the lambs in Lot 1 than for those in Lot 2; the increases made by both lots were small. Values for both lots were considered low, indicating that vitamin A storage in the liver was at a minimum.

Liver analyses for vitamin A indicated that there was very little difference between the two lots. As shown in Table 4, the average vitamin A content of the livers of the lambs in Lot 1 was 13.2 micrograms of vitamin A per gram, whereas the livers from the lambs of Lot 2 contained 11.8 micrograms per gram. Although all lambs received approximately nine times the minimum requirements of carotene for a period of 108 days, it appears that the storage of vitamin A in the liver was at a minimum at the start of carotene supplementation and that very little storage occurred after supplementation with nine times the minimum requirement.

SUMMARY

Seven lambs were fed a low phosphorus ration for a period of 268 days, after which they were divided into two lots following a consideration of the inorganic phosphorus and vitamin A content of the plasma and body weights. The three lambs in Lot 1 were continued on the low phosphorus ration, whereas the four lambs in Lot 2 were supplemented with sufficient dicalcium phosphate to approximate a phosphorus intake of 3 grams per hundred pounds body weight.

The lambs were bled at intervals of approximately ten days, and analyses were made for inorganic phosphorus and vitamin A. Although the additional phosphorus fed to the lambs of Lot 2 caused an increase in plasma-phosphorus values, this increased phosphorus level in the plasma did not influence the vitamin A content of the plasma.

Analyses of livers from lambs slaughtered at the end of the experiment indicated that there was very little difference in the vitamin A content between the two lots.

Under conditions of this experiment, a low phosphorus ration did not appear to affect the metabolism of carotene.

THE EFFECT OF LOW PHOSPHORUS RATIONS
UPON CAROTENE METABOLISM AND UPON STORAGE
OF VITAMIN A IN THE LIVER OF BEEF STEERS

EXPERIMENTAL PROCEDURE

Nine head of yearling Hereford steers were used in this study. They weighed approximately 680 pounds at the start of the experiment, August 2, 1949. These steers had received different feeds the previous winter but had been grazed together in the same pasture during the summer at the Lake Carl Blackwell Range Experimental Area, west of Stillwater, Oklahoma. At the start of the experiment, the steers were placed in a dry lot at the Experimental Steer Feeding Barn near the campus and were fed a ration composed of 2 pounds corn gluten meal and prairie hay, free choice, in order to deplete the steers of phosphorus. Corn gluten meal was used as the protein supplement because of its relatively low phosphorus content. This ration furnished an intake of 1.04 grams of phosphorus per 100 pounds live weight. After the steers had been on this ration for a period of 117 days, the amount of corn gluten meal fed was reduced from 2 to 1 pound per day. This change was made because the previous intake of 1.04 grams of phosphorus had not lowered the inorganic phosphorus level of the blood plasma. The phosphorus intake per 100 pounds live weight for the remainder of the 309-day depletion period was 0.5 gram.

Blood samples from the jugular vein were collected at the start of the experiment and at intervals of approximately one month during the depletion period. The blood was collected in citrated tubes and analyzed for carotene and vitamin A by the Kimble (1939) procedure and for inorganic phosphorus

by the Fiske and Subbarow method (1925). After the steers had been on the depletion ration for a period of 270 days, liver samples were obtained and were analyzed for carotene and vitamin A by the method of Gallup and Hoefler (1946).

The procedure used to obtain the liver samples from the steers on this experiment was one developed by Whitehair, Van Arsdell, and Thomas, of the Oklahoma A. & M. Animal Husbandry Department (1950). This procedure is partially illustrated by Figures 1 and 2. As shown in Figure 1, the steer was tied to a fence, little restraint being necessary for this operation; an area 4 by 5 inches was shaved on the steer, the center of the shaved area being 12 to 14 inches from the backbone, between the 12th and 13th ribs on the right side of the animal. The shaved area was cleaned with tincture of Merthiolate and alcohol. A local anesthesia, composed of 50 cc. of 3 per cent Procaine Hydrochloride, was injected subcutaneously and intramuscularly near the two ribs, using 20-gauge needles, $3/4$ inch and $1\ 1/2$ inches long. After about five to ten minutes, a 3-inch incision was made midway and parallel with the ribs. A 6-inch, blunt-ended trocar and canula with a 1-inch diameter was then inserted through the muscle fibers and into the abdominal cavity. The trocar was withdrawn, and a flashlight with an extension cable was inserted to determine the exact location of the liver. When the liver was located, the canula was pressed against the liver, and the instrument designed to obtain these samples, shown in Figure 2, was inserted in the canula, pressed against the liver, and, after two complete turns, was withdrawn. The operative area was treated and cared for as in accepted practices. The entire procedure, as outlined by Whitehair, et al. (1950) normally required about

TABLE 1.

RATIONS FED TO STEERS IN EXPERIMENT 2¹

Lot	Number of Steers	Ration		Calcium Carbonate	Dicalcium Phosphate	Average Daily Intake		Ca:P Ratio
		Corn Gluten Meal	Prairie Hay			gms per 100 lbs. body weight		
		lbs.	lbs.	gms.	gms.	Calcium	Phosphorus	
1	5	1	10	62	----	6.45	0.50	12.8:1
2	4	1	10	----	100	6.48	3.03	2.1:1

¹ After one month on this ration, the daily ration of each steer was supplemented with 136,000 micrograms of carotene.



Figure 1. Local anethesia being given to steer in area of operation.

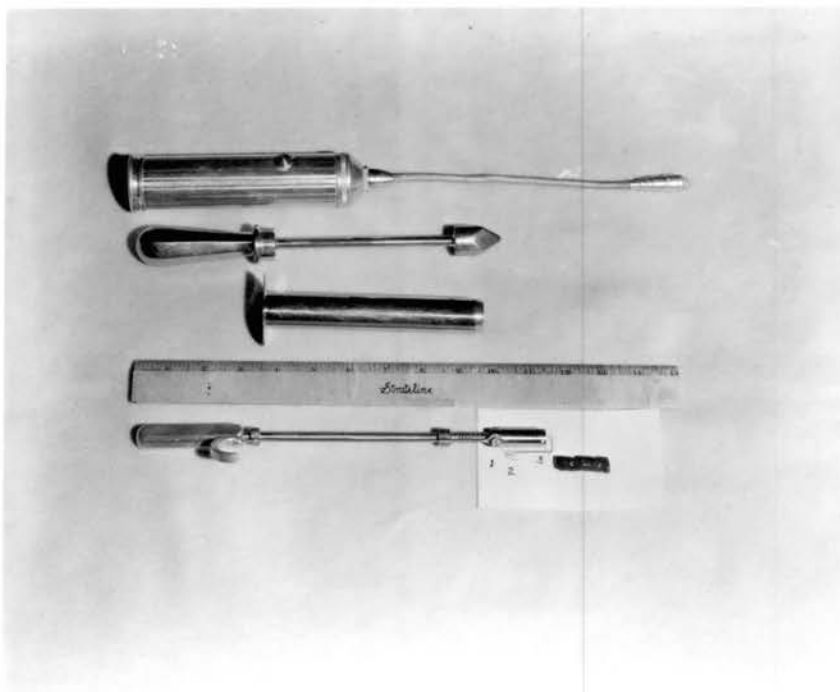


Figure 2. Instruments used in obtaining liver samples--from bottom to top--instrument designed to obtain liver samples and a 2.2 gram liver sample, ruler, canula, trocar, flashlight with extension cable.

20 to 30 minutes. The 0.5 to 3.0 gram samples obtained from the steers in this manner were wrapped in cellophane, placed in labeled jars, and immediately frozen on dry ice. These samples were kept frozen until they were analyzed for carotene and vitamin A.

After 309 days in the phosphorus depletion ration, the steers were divided into two lots according to their live weight, blood and liver levels of carotene and vitamin A, and plasma level of inorganic phosphorus. The steers were placed in stalls in the Animal Husbandry Area and fed individually, in order that the intake of phosphorus and carotene might be controlled. Lot 1 steers were continued on the low phosphorus ration, whereas the ration of Lot 2 steers was supplemented with sufficient dicalcium phosphate to provide a total phosphorus intake of approximately 3 grams per 100 pounds of live weight. The ration fed each lot is shown in Table I.

Blood samples were obtained at ten-day intervals and liver samples at monthly intervals.

In order to determine whether or not the phosphorus intake of the steers would influence carotene metabolism when the carotene intake was high, the ration of all steers was supplemented with sufficient carotene to provide a total intake of approximately 15 times the minimum requirement. This supplementation was begun thirty days after the steers were allotted. The minimum requirement of carotene was estimated from the work of Guilbert, et al. (1937, 1940); 13.6 micrograms per pound body weight per day was used as a minimum. The carotene used was a crude carotene concentrate prepared from alfalfa (Valley Vitamins, Inc.) which had a potency of 20,000 micrograms per gram. One hundred and forty grams of the crude carotene concentrate,

400 ml. of corn oil, and 100 ml. of Skelly Solve "F" were placed in a Waring Blender and mixed until an emulsion was formed.

This carotene emulsion was placed in glass jars and kept in the refrigerator. At each feeding period, the jars were thoroughly shaken and 25 cc. of the carotene concentrate emulsion were measured in a graduated flask and poured on the feed of each steer. This amount provided 136,000 micrograms carotene per steer per day in addition to that received from the prairie hay.

The steers were slaughtered on September 11, 1950, 405 days after the initiation of the experiment and 96 days after the steers of Lot 2 received additional phosphorus. At this time, the livers were collected, and weighed; representative samples wrapped in cellophane were kept frozen until they were analyzed for vitamin A. The method used for this analysis was that of Gallup and Hoefler (1946). A foreshank of each steer was also obtained at the time of slaughter, and a cross section of the foreshank was later analyzed for bone ash by the method described in the Official and Tentative Methods of Analysis of the Association of Official Agriculture Chemists (1945).

The chemical composition of feeds used in Experiment 2 are given in Table 2.

TABLE 2

CHEMICAL COMPOSITION OF FEEDS USED IN EXPERIMENT 2

Item	Date	Dry Matter	Per cent composition of dry matter							Carotene p.p.m.	
			Ash	Protein	Fat	Fiber	N.F.E.	Ca	P		
Corn Gluten Meal	11/22/49	-----	----	41.60	----	----	----	----	0.41	0.47	----
" "	12/21/49	93.36	1.30	42.37	2.59	3.30	50.53	0.03	0.275	----	
" "	1/2/50	94.46	----	42.22	----	----	----	0.03	0.314	----	
" "	4/24/50	92.40	2.21	51.68	----	----	----	----	0.392	----	
" "	6/22/50	93.00	----	----	----	----	----	----	0.232	----	
" "	7/10/50	93.00	----	----	----	----	----	----	0.261	----	
" "	9/11/50	93.00	----	----	----	----	----	----	0.791	----	
Prairie Hay	11/22/49	-----	----	4.00	----	----	----	----	----	19.1 ¹	
" "	12/15/49	91.94	7.69	4.22	2.57	32.77	52.75	0.49	0.050	8.8	
" "	1/12/50	94.22	----	----	----	----	----	----	----	14.1	
" "	2/9/50	94.43	7.03	4.34	2.18	32.64	53.81	0.45	0.048	17.1	
" "	3/15/50	93.45	7.96	4.28	2.48	32.00	53.38	0.45	0.056	17.6	
" "	6/27/50	93.00	----	----	----	----	----	----	----	11.4	
" "	7/10/50	93.00	----	----	----	----	----	----	----	2.9	
" "	9/10/50	93.00	----	----	----	----	----	----	----	3.1	
Dicalcium Phosphate		-----	----	----	----	----	----	25.00	18.00	----	
Calcium Carbonate		-----	----	----	----	----	----	40.00	----	----	

¹ Crude carotene--all other carotene values reported as true carotene.

RESULTS AND DISCUSSION

All steers gained weight during the early part of the phosphorus depletion period. The average weights of the steers at the beginning of the experiment was 679 pounds, and after 117 days of feeding, the average was 759 pounds. At this time, the amount of corn gluten meal fed was reduced, and the steers gradually lost weight until the end of the 309-day depletion period, at which time the average weight was 672 pounds. The steers exhibited some outward symptoms of a phosphorus deficiency such as poor condition, rough hair coat, and a depraved appetite. Although prairie hay was fed, free choice, the amount consumed by the steers decreased from 15 pounds during the first 117 days of the experiment to 10 pounds at the end of the depletion period. The steers of both lots gained weight during the period of phosphorus supplementation. Lot 1 steers averaged 657 pounds at 309 days and 717 pounds at 405 days, the end of the experiment. Lot 2 steers weighed 691 and 785 pounds at the two respective periods.

The plasma-inorganic phosphorus level of the steers at the start of the experiment averaged 6.0 milligrams per 100 milliliters of plasma. After the steers had been on a low phosphorus ration for 309 days--a ration which provided an intake of 1.04 grams of phosphorus per 100 pounds live weight or 7.7 grams phosphorus per steer per day for the first 117 days and an intake of 0.50 grams phosphorus per 100 pounds live weight or 3.4 grams phosphorus per steer per day for the remaining 192 days of the depletion period, the average inorganic phosphorus level of the plasma had only dropped to 4.0 milligrams. These results are similar to those obtained by

Beeson, et al. (1937), who reported a definite phosphorus deficiency in steers which had been receiving a daily phosphorus intake of 8.23 grams for 100 days. The blood-phosphorus level in these steers dropped from 6.71 to 4.40 milligrams. Beeson further stated that the phosphorus deficient steers required 25 per cent more feed to make 100 pounds gain and that the beef steer apparently requires about 2 grams of phosphorus daily per 100 pounds of live weight for normal growth and fattening.

The analyses of the blood plasma for phosphorus during the experimental period are given in Table 3 and Figure 3. Both lots had the same average, 4.0 milligrams of phosphorus per 100 milliliters of plasma, at the start of the experimental period. After one week of supplemental phosphorus feeding, the blood-phosphorus level of Lot 2 steers had increased to 12.3, and the level for Lot 1 steers had increased to 5.6 milligrams. These values gradually decreased during the experimental period, as is graphically illustrated in Figure 3. At the end of the experimental period, Lot 2 had an average of 7.2; Lot 1 had an average of 4.3 milligrams. This difference was highly significant (Snedecor, 1946). Steer number 93 in Lot 1 and steer number 92 in Lot 2 had consistently lower values than those of the other steers.

Carotene

The average carotene level of the steers at the start of the experiment was 364 micrograms per 100 milliliters plasma; this value decreased to 105 micrograms at the end of the 309-day depletion period. The Lot 1 steers had an average carotene level of 98 at the start of the experimental period, whereas the level of carotene for Lot 2 steers was 115 micrograms.

TABLE 3

INORGANIC PHOSPHORUS CONTENT OF PLASMA OF STEERS IN EXPERIMENT 2

		Plasma Inorganic Phosphorus (mg per 100 ml.)										
Lot	Steer Number	8/2/49	5/9/50	6/17 ¹	6/27	7/7	7/20	7/31	8/9	8/21	8/30	9/11
Days		0	280	318	329	339	353	363	372	384	393	405
1	93	6.2	2.4	3.5	3.2	2.7	2.5	2.4	2.3	2.6	2.5	2.1
	94	5.6	4.2	6.6	6.0	6.4	5.0	5.2	5.4	6.8	5.6	5.2
	95	5.9	4.9	6.5	5.2	6.2	5.4	6.0	5.6	6.2	5.0	4.9
	99	7.2	4.5	5.6	5.6	5.7	4.6	4.2	4.8	5.9	4.9	4.1
	101	5.5	<u>3.9</u>	<u>6.0</u>	<u>5.1</u>	<u>5.2</u>	<u>5.2</u>	<u>6.4</u>	<u>5.0</u>	<u>6.2</u>	<u>4.5</u>	<u>5.2</u>
Lot average			<u>4.0</u>	<u>5.6</u>	<u>5.0</u>	<u>5.2</u>	<u>4.5</u>	<u>4.8</u>	<u>4.6</u>	<u>5.5</u>	<u>4.5</u>	<u>4.3</u>
2	92	5.6	3.6	11.8	8.4	9.0	8.4	7.6	7.5	6.8	7.3	6.2
	97	5.5	3.4	12.1	10.1	11.4	10.2	10.4	8.9	7.8	6.8	7.0
	98	6.0	4.8	12.5	9.8	11.2	9.6	9.8	8.8	8.8	7.3	8.0
	100	6.5	<u>4.1</u>	<u>12.6</u>	<u>9.1</u>	<u>9.9</u>	<u>8.9</u>	<u>9.4</u>	<u>8.4</u>	<u>8.1</u>	<u>9.2</u>	<u>7.5</u>
	Lot average			<u>4.0</u>	<u>12.3</u>	<u>9.3</u>	<u>10.3</u>	<u>9.3</u>	<u>9.3</u>	<u>8.4</u>	<u>7.9</u>	<u>7.7</u>
Initial average all steers		6.0										

¹ First bleeding date after the ration of Lot 2 steers had been supplemented with additional phosphorus.

**Significant at the 1% level.

AVERAGE PLASMA VALUES

PHOSPHORUS

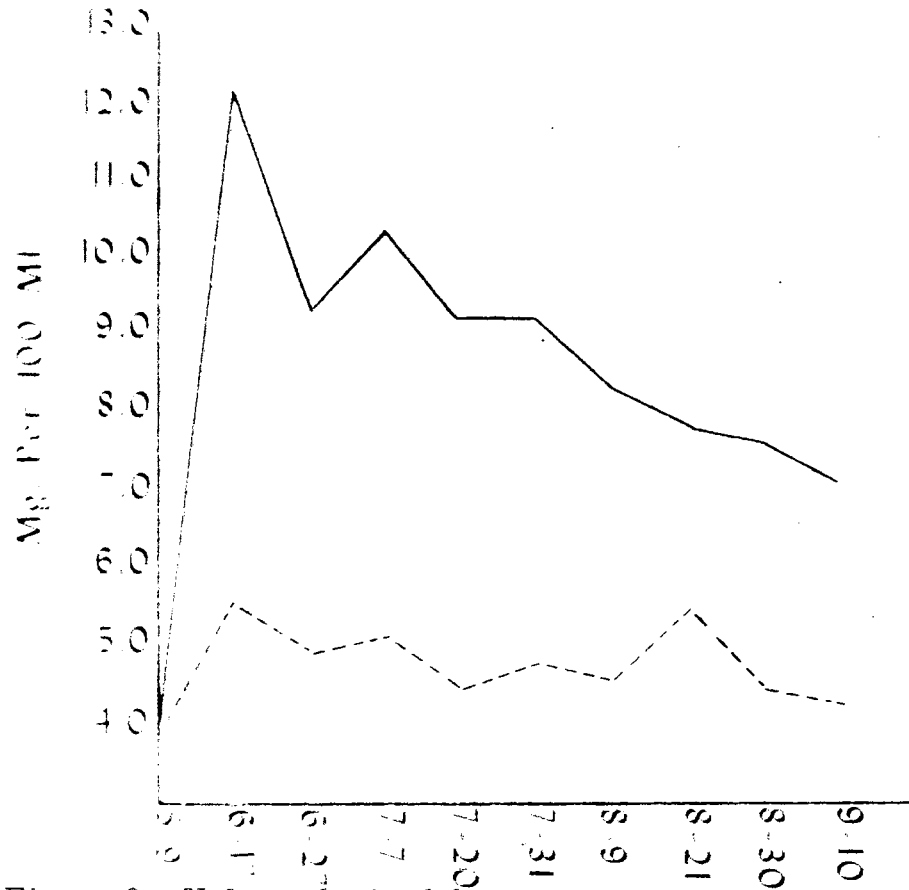


Figure 3. Values obtained from steers in Experiment 2.

- 1 - Low phosphorus
- 2 - High phosphorus

CAROTENE

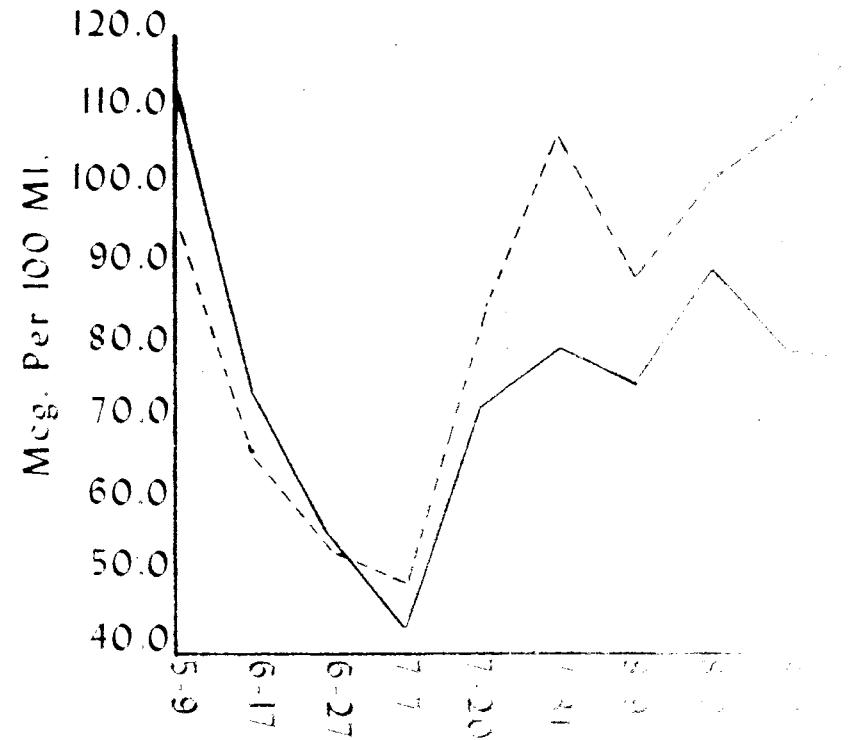


Figure 4. Values obtained from steers in Experiment 2. All steers received additional carotene from July 7 until end of experiment.

The average carotene levels dropped considerably during the following month, as is shown in Table 4 and Figure 4, at which time the average was 49 for Lot 1 steers and 43 micrograms for Lot 2 steers.

The carotene values increased in both lots after carotene supplementation; however, the increase was greater for the steers in Lot 1 than for the steers in Lot 2. After 66 days of supplemental feeding with carotene, the average carotene values were 119 for Lot 1 and 76 for Lot 2. This difference was not significant, the lack of significance probably due to the consistent high level of carotene in steer number 97 in Lot 2.

These results indicate that steers receiving a low intake of phosphorus tend to have higher carotene values than steers receiving a high intake of phosphorus. These data substantiate, to some extent, the work of Gallup and Ross (1948), who reported that pregnant beef cows which received a low intake of phosphorus had higher carotene values than those which received a high intake of phosphorus.

Vitamin A

At the start of the depletion period, the average vitamin A level in the plasma of all steers was 29 micrograms per 100 milliliters. This average value gradually decreased to 18 micrograms at the end of 309 days. When the steers were allotted, the Lot 1 steers had an average of 16 and Lot 2 an average of 20 micrograms. One month later, when carotene supplementation was begun, both lots had an average of 13 micrograms. As shown in Table 5 and Figure 5, the level of vitamin A in the plasma of both lots increased after the steers were given carotene; however, the increase was

TABLE 4

CAROTENE CONTENT OF PLASMA OF STEERS IN EXPERIMENT 2

Lot	Steer Number	Plasma Carotene (mcg per 100 ml)										
		8/2/49	5/9/50	6/17	6/27	7/7 ¹	7/20	7/31	8/9	8/21	8/30	9/11
Days		0	280	318	329	339	353	363	372	384	393	405
1	93	467	125	56	47	44	67	103	82	105	115	104
	94	329	71	40	40	34	86	107	70	94	91	109
	95	456	110	78	58	50	98	110	103	104	113	126
	99	319	87	69	54	46	80	96	83	81	82	101
	101	312	<u>98</u>	<u>89</u>	<u>71</u>	<u>73</u>	<u>87</u>	<u>124</u>	<u>110</u>	<u>124</u>	<u>138</u>	<u>157</u>
Lot average		98	66	54	49	84	108	90	102	108	119	
2	92	309	96	65	46	35	66	78	70	73	66	69
	97	434	182	125	99	73	125	135	137	151	146	133
	98	303	77	52	40	34	50	50	48	81	62	65
	100	351	<u>103</u>	<u>56</u>	<u>35</u>	<u>28</u>	<u>48</u>	<u>56</u>	<u>47</u>	<u>56</u>	<u>44</u>	<u>43</u>
Lot average		115	75	55	43	72	80	76	90	80	76	
Initial average all steers		364										

¹ Starting this date, the daily ration of each steer was supplemented with 136,000 micrograms of carotene.

TABLE 5

VITAMIN A CONTENT OF PLASMA OF STEERS IN EXPERIMENT 2

Lot	Steer Number	Plasma Vitamin A (mcg per 100 ml)										
		<u>8/2/49</u>	<u>5/9/50</u>	<u>6/17</u>	<u>6/27</u>	<u>7/7¹</u>	<u>7/20</u>	<u>7/31</u>	<u>8/9</u>	<u>8/21</u>	<u>8/30</u>	<u>9/11</u>
Days		0	280	318	329	339	353	363	372	384	393	405
1	93	27	20	16	15	13	13	13	12	14	14	15
	94	29	17	16	16	14	13	19	8	11	16	14
	95	21	15	15	16	12	13	13	13	7	20	17
	99	24	16	16	18	13	11	14	11	15	15	21
	101	39	<u>11</u>	<u>19</u>	<u>18</u>	<u>11</u>	<u>15</u>	<u>17</u>	<u>15</u>	<u>11</u>	<u>22</u>	<u>17</u>
Lot average		16	16	17	13	13	15	12	12	17	17	
2	92	20	13	14	13	9	15	13	14	17	24	19
	97	40	27	23	22	18	18	21	23	23	34	25
	98	36	22	11	15	12	11	11	18	22	16	22
	100	24	<u>19</u>	<u>18</u>	<u>15</u>	<u>13</u>	<u>12</u>	<u>18</u>	<u>15</u>	<u>17</u>	<u>22</u>	<u>20</u>
Lot average		20	17	16	13	14	16	18	20	23	22*	
Initial average all steers		29										

¹ Starting this date, the daily ration of each steer was supplemented with 136,000 micrograms of carotene.

*Significant at the 5% level.

AVERAGE VITAMIN A VALUES

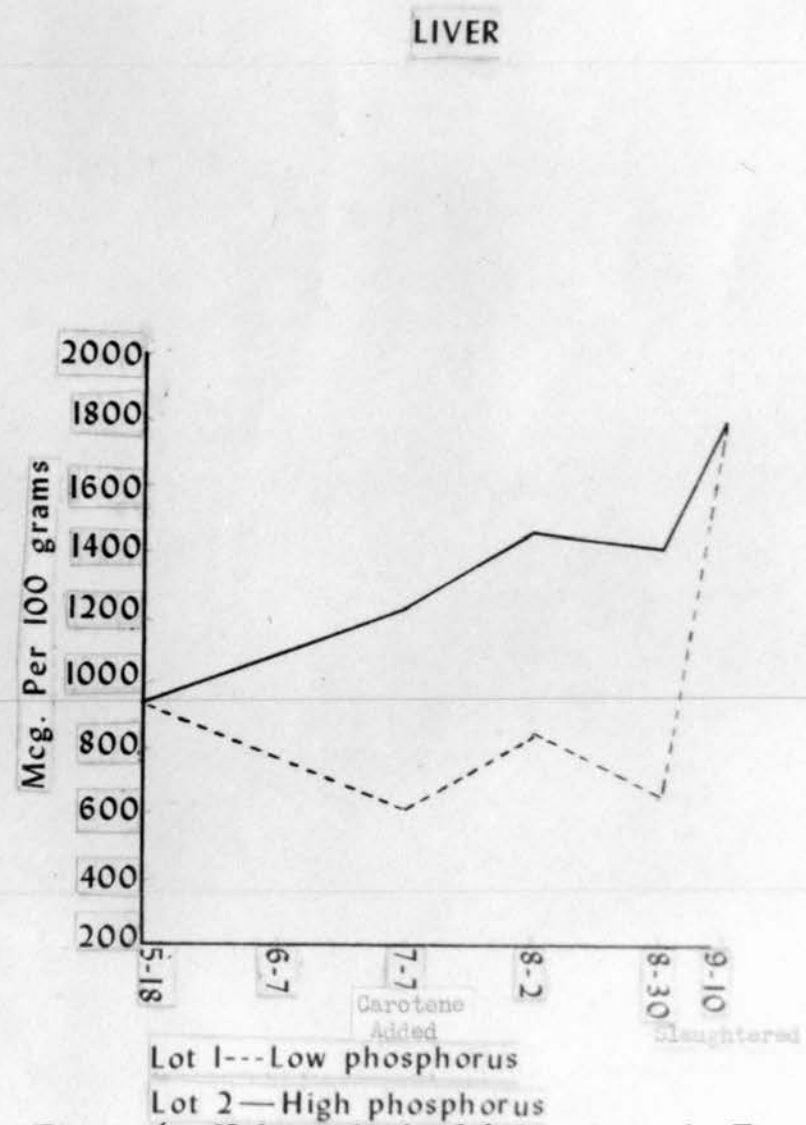
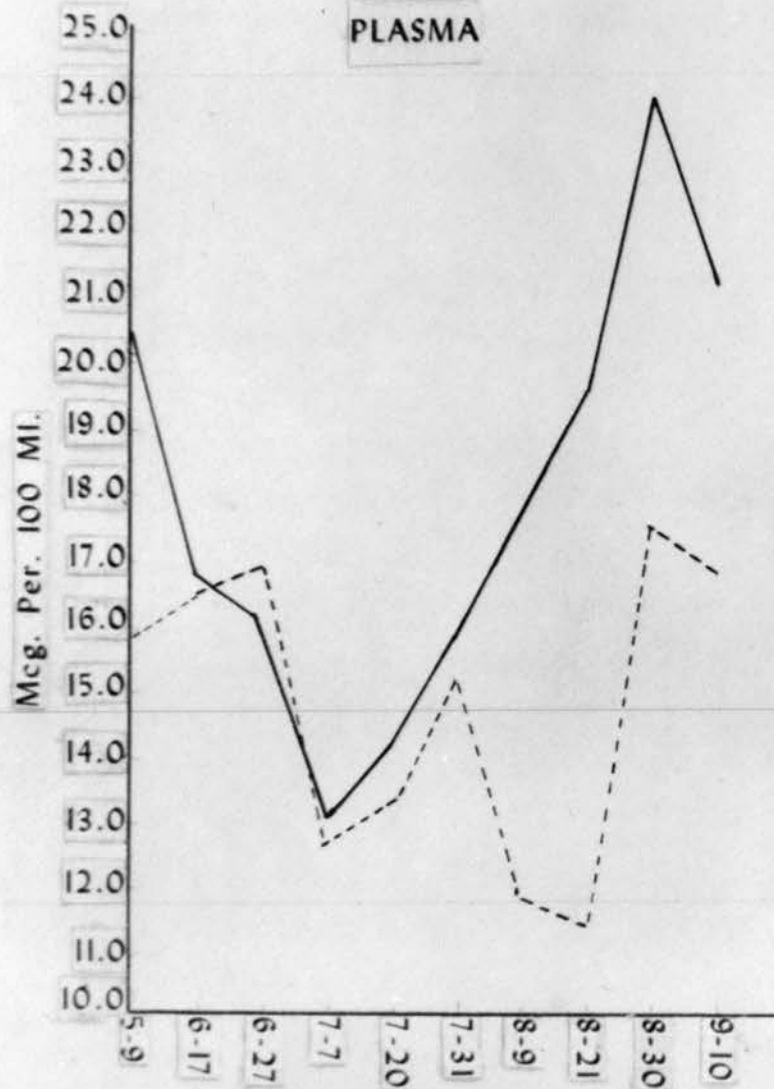


Figure 5. Values obtained from steers in Experiment 2. All steers received additional carotene from July 7 until end of experiment.

Figure 6. Values obtained from steers in Experiment 2.

greater for the steers of Lot 2 than for the steers of Lot 1. At the end of the experiment, the average values were 17 for Lot 1 and 22 micrograms for Lot 2. This difference was significant (Snedecor, 1946) although there was very little difference in the values between the two lots.

The location of the intestine as the site of transformation of beta-carotene to vitamin A in the rat has been demonstrated by Mattson, et al. (1947), Sexton, et al. (1946), Goodwin and Gregory (1948), and Glover, Goodwin, and Morton (1948). Although there can be no doubt that beta-carotene is the precursor of vitamin A in the animal body, little is known of the mechanism of the transformation of carotene into Vitamin A. The results of this study indicate a trend for steers having low plasma-phosphorus values to have higher carotene and lower vitamin A values than steers having high plasma-phosphorus values. It is suggested that the level of phosphorus in the blood plasma may play some role in the transformation of beta-carotene into vitamin A, under certain physiological conditions.

Liver

The results of the analyses of liver samples, obtained by biopsy and at the time of slaughter, for carotene and vitamin A are shown in Table 6 and Figure 6. Liver samples obtained 280 days after the initiation of the experiment showed that the livers of the Lot 1 steers contained 225 and Lot 2 steers 158 micrograms per 100 grams of liver. At the time of slaughter, the values for Lot 1 and Lot 2 were 201 and 192 micrograms, respectively. The values of carotene and vitamin A for steer 99 at 280 days and for steer 101 at 339 days were extremely low in comparison with later values and were not

TABLE 6

CAROTENE AND VITAMIN A CONTENT OF STEER LIVER
(Obtained by biopsy and slaughter)
(mcg per 100 gm liver)

Steer Number	5/9	7/7 ¹	8/2	8/30	9/11 ²	Weight Liver lbs.	Total mcg
Days	280	339	365	393	405		
<u>Carotene</u>							
Lot 1, Low Phosphorus							
93	214	113	205	174	214	6.2	6,024
94	139	126	189	182	261	5.6	6,636
95	264	129	213	202	251	5.0	5,697
99	(18) ³	116	135	95	126	6.1	3,489
101	284	(None) ³	237	173	251	6.7	7,635
Lot average	225	121	196	165	201	5.9	5,896
Lot 2, High Phosphorus							
92	149	100	189	125	170	6.2	4,785
97	224	204	212	251	251	6.0	6,837
98	72	(None) ³	177	145	225	6.4	6,538
100	186	(None) ³	137	163	122	6.3	3,489
Lot average	158	152	179	171	192	6.2	5,412
<u>Vitamin A</u>							
Lot 1, Low Phosphorus							
93	961	379	864	557	1833		51,595
94	1041	776	861	490	2474		62,899
95	1086	811	965	1037	1781		40,429
99	(8) ³	495	738	560	1570		40,340
101	665	(87) ³	770	549	1270		38,631
Lot average	938	615	840	639	1786		46,778
Lot 2, High Phosphorus							
92	127	453	730	483	1125		31,667
97	1022	1066	1697	2179	2467		65,147
98	477	1044	1309	1023	944		27,429
100	2173	2424	2143	1998	2683		76,739
Lot average	950	1237	1470	1421	1804		50,746

¹ Starting this date, the daily ration of each steer was supplemented with 136,000 micrograms of carotene.

² Values obtained at time of slaughter.

³ These values were considered low in comparison with later values and were not averaged with values obtained from the other steers.

averaged with the values obtained for the other steers.

The amount of vitamin A in the livers of the steers of Lot 2 was greater at 280 days than for the steers of Lot 1, the respective values being 950 and 938 micrograms per 100 grams of liver. The amount of vitamin A increased in the livers of Lot 2 steers, whereas for Lot 1 steers there was a decrease; however, analyses of liver samples taken at the time of slaughter showed practically the same value for both lots. Although the vitamin A values obtained from liver samples taken by biopsy were reasonably consistent, these values were much lower, as a rule, than values obtained from samples taken at the time of slaughter; this difference was especially noted in values obtained for Lot 1.

The variation in results of analyses of liver samples taken by biopsy and those taken at the time of slaughter may have been due to the fact that most of the liver samples taken by biopsy contained some scar tissue and were engorged with blood, whereas the samples taken at the time of slaughter contained no scar tissue and little blood. Some error could result from the weighing and handling of the relatively small (0.5 to 2.5 grams) samples obtained by biopsy.

Bone Ash

The per cent ash of dry, fat-free bones is given in Table 7. The average for both lots was about the same; however, there was considerable variation in the Lot 1 steers. Steer 99 had an ash of 73.2 per cent, whereas steer 101 had an ash of 53.2 per cent. The average per cent bone ash was 65.1 for Lot 1 and 65.5 for Lot 2 steers.

TABLE 7

PER CENT ASH OF DRY, FAT-FREE BONES FROM STEERS IN EXPERIMENT 2

Lot 1, Low Phosphorus		Lot 2, High Phosphorus	
Steer Number	Per cent Ash	Steer	Per cent Ash
93	63.34	92	65.23
94	65.40	97	65.23
95	65.26	98	65.72
99	73.22	100	<u>65.74</u>
101	<u>53.22</u>		
Average	65.09	Average	65.48

SUMMARY

Nine steers were fed a low phosphorus ration for a period of 309 days, after which time they were divided into two lots, on the basis of the inorganic phosphorus level of the plasma; carotene and vitamin A content of the plasma and liver; and body weights.

The five steers in Lot 1 were continued on the low phosphorus ration, from which they received approximately 0.5 gram of phosphorus per 100 pounds live weight, whereas the ration of the four steers in Lot 2 was supplemented with sufficient dicalcium phosphate so that the daily intake of phosphorus per steer was approximately 3 grams per 100 pounds live weight.

Blood samples obtained at intervals of about 10 days were analyzed for inorganic phosphorus, carotene, and vitamin A. Data from these analyses indicate a trend for the Lot 1 steers, which had low inorganic phosphorus levels in the plasma, to have higher carotene levels and lower vitamin A levels than the steers of Lot 2, which had higher inorganic phosphorus levels. It is suggested that a certain level of phosphorus in the blood of steers may be necessary in some manner for the conversion of carotene into vitamin A in the intestinal wall.

Liver samples obtained by biopsy and taken at the time of slaughter of the steers, 96 days after experimental feeding was begun, were analyzed for carotene and vitamin A. The carotene values of liver samples taken by biopsy from Lot 1 steers tended to be higher and the vitamin A values lower than values obtained from Lot 2 steers; however, the values of carotene and vitamin A were about the same for both lots at the time of slaughter.

THE EFFECT OF LOW PHOSPHORUS RATIONS
UPON CAROTENE METABOLISM AND UPON STORAGE
OF VITAMIN A IN THE LIVER OF MATURE BEEF COWS

EXPERIMENTAL

Objectives

1. To determine the effect of low phosphorus rations upon the blood plasma-inorganic phosphorus, carotene, and vitamin A levels of beef cows.
2. To determine the effect of low phosphorus rations on liver storage of carotene and vitamin A in beef cows.
3. To determine the effect of low phosphorus rations upon carotene and vitamin A content of the colostrum and milk.
4. To determine what effect the feeding of low phosphorus rations to beef cows will have upon the blood and liver values of their calves and upon the subsequent storage of carotene and vitamin A in the liver.

Procedure

Nine grade Hereford cows, averaging 11 years of age, were placed on a low phosphorus ration November 16, 1951. These cows had been on an experiment in which they received different feeds, but the phosphorus intake of each cow in preceding years had been approximately the same. The previous summer, the cows had grazed in pastures of comparable forage and acreage at the Lake Carl Blackwell Experimental Range west of Stillwater, Oklahoma. All cows had been pasture bred, and, after examination, were thought to be safely in calf. The cows were placed in a dry lot at the Experimental Steer Feeding Barn near the Oklahoma A. and M. College campus and were fed a

low phosphorus ration composed of 1 1/2 pounds of corn gluten meal and prairie hay, free choice. Corn gluten meal was used as the protein supplement because of its relatively low phosphorus content. The ration provided a daily intake of about 0.5 gram phosphorus per 100 pounds live weight.

Blood samples were taken from the jugular vein at the start of the experiment and thereafter at intervals of approximately one month until calving. The blood was collected in citrated tubes and analyzed for carotene and vitamin A by the Kimble procedure (1939) and for inorganic phosphorus by the Fiske and Subbarow method (1925).

After the cows had been on the depletion ration for 71 days--a time estimated to be about four weeks before parturition, liver samples were obtained by biopsy technique (Whitehair, et al, 1950) and were analyzed for carotene and vitamin A by the method of Gallup and Hoefler (1946). Analyses of blood and liver samples taken at this date were used in dividing the cows into two lots. From this time until the cows calved, the four cows in Lot 1 were fed the phosphorus depletion ration; the five cows in Lot 2 were fed the same ration supplemented with sufficient bonemeal to provide an intake of approximately 3.0 grams phosphorus per 100 pounds live weight. Up to this time, the cows consumed similar amounts of hay which furnished approximately 80 milligrams of carotene per cow per day.

At the time of calving, blood, liver, and milk samples were obtained from the cows, and blood and liver samples were obtained from the calves; birth weights were also recorded. The cows were inspected frequently, and immediately following parturition, calves were separated from the cows without being allowed to nurse until blood, liver, and milk samples had been procured.

In a few cases, calves may have nursed before samples were taken. When blood and liver samples had been obtained, the cows and their calves were placed in stalls in the Animal Husbandry Arena and were fed individually, in order to control phosphorus and carotene intake. The ration fed each lot after calving is given in Table 1; the composition of the feeds used in the experiment is given in Table 2.

At weekly intervals for one month and then at the sixth and eighth week, blood samples were taken from each cow and her calf and milk samples from each cow. Milk samples were frozen until they were analyzed for carotene and vitamin A by the method of Boyer, et al. (1944). Calves were weighed at the time of each bleeding.

Results of these blood determinations and those of vitamin A in the liver indicated that the cows were receiving sufficient carotene to maintain liver storage of vitamin A. Consequently, the carotene content of the ration was reduced by feeding cottonseed hulls or beet pulp instead of prairie hay.

Cows 33 and 46 in Lot 1 and cows 19 and 50 in Lot 2 calved earlier than the other cows and received cottonseed hulls as a replacement of part of the prairie hay during the second month. Because cottonseed hulls proved to be unpalatable, when comprising two-thirds of the roughage of the ration, the hulls were replaced with beet pulp at the end of the second month.

Cow 41 in Lot 1 and cows 90 and 98 in Lot 2 received the same total intake of prairie hay during the second month. In addition to prairie hay, cow 41 received beet pulp, whereas cows 90 and 98 were fed cottonseed hulls the first two weeks and beet pulp the second two weeks of the second month of the experiment.

TABLE 1

AVERAGE DAILY RATIONS FED TO COWS IN EXPERIMENT 3

Lot	Cow No.	Corn Gluten Meal	Prairie Hay	Cotton- seed Hulls	Beet Pulp	Calcium Carbonate	Bonemeal	Average Daily Intake		Ca:P Ratio	Average ¹ Daily Carotene Intake
								gms/100 Ca	lb. body wt. P		
		lbs.	lbs.	lbs.	lbs.	gm.	gm.	gm.	gm.		mg.
<u>End of depletion period until parturition</u>											
1	All	1.5	10	---	---	125	---	6.91	0.58	11.9:1	81
2	All	1.5	10	---	---	---	150	7.14	3.00	2.4:1	81
<u>From parturition until end of fourth week</u>											
1	All	1.5	15	---	---	125	---	8.43	0.71	11.9:1	116
2	All	1.5	15	---	---	---	150	9.11	3.38	2.7:1	116
<u>From four weeks to five weeks after parturition</u>											
1	33, 46	1.5	10	5	---	100	---	6.09	0.70	9.6:1	81
2	19, 50										
	90, 98	1.5	10	5	---	---	125	7.78	3.20	2.4:1	81
1	41 ₃	1.5	5 ²	---	10	100	---	12.12	1.33	9:1:1	45
1	43 ₃	1.5	--	---	10	100	---	8.78	0.66	13.3:1	11
2	29 ₃	1.5	--	---	14	---	125	10.93	3.32	3.3:1	11
<u>From five weeks to eight weeks after parturition</u>											
1	33, 46	1.5	5	10	---	100	---	5.49	0.72	7.6:1	45
2	19, 50										
	90, 98	1.5	5 ⁴	10	---	---	125	7.05	3.23	2.2:1	45
<u>From eight weeks to twelve weeks after parturition⁵</u>											
1	33, 46	1.5	5	---	10	100	---	9.12	1.00	9.1:1	40
2	19, 50	1.5	5	---	10	---	125	11.80	3.77	3.1:1	40
<u>From twelve weeks to twenty weeks after parturition⁶</u>											
1	33, 46	1.5	---	---	14	100	---	10.20	0.73	14:0:1	11
2	19, 50	1.5	---	---	14	---	125	12.35	3.72	3.3:1	11

TABLE 1 (Continued)

-
- 1 This value includes 11 milligrams of carotene which was present in 1.5 pounds of corn gluten meal.
 - 2 Cow 41 received this ration until the end of the seventh week; the five pounds of prairie hay were not fed during the eighth week in order that the total intake of prairie hay for the month would be equal to that consumed by cows 90 and 98.
 - 3 Cows 43 and 29 received this ration from the fourth to the end of the eighth week.
 - 4 Cows 90 and 98 were fed this ration only to the end of the sixth week; from the sixth to the eighth week they were fed 1.5 pounds corn gluten meal and 10 pounds beet pulp.
 - 5 The feeding of 5 pounds of prairie hay was discontinued at the end of the eleventh week; two pounds of beet pulp were added to the ration during the eleventh to twelfth week.
 - 6 From the twelfth to the thirteenth week, the cows received only 12 pounds of beet pulp.

TABLE 2

CHEMICAL COMPOSITION OF FEEDS USED IN EXPERIMENT 3

Item	Date	Dry Matter	Per cent composition of dry matter							Carotene p.p.m.	
			Ash	Protein	Fat	Fiber	N.F.E.	Ca	P		
Corn Gluten Meal	2/24/51	-----	-----	-----	-----	-----	-----	-----	0.095	0.518	15.5 ¹
Prairie Hay	2/24/51	-----	-----	-----	-----	-----	-----	-----	0.410	0.037	15.5 ²
Corn Gluten Meal	4/30/51	92.62	3.00	45.51	1.65	4.70	45.14	0.11	0.57		
Molasses Beet Pulp	4/30/51	91.63	7.19	7.88	0.47	14.85	69.61	0.80	0.08		
Cottonseed Hulls	4/30/51	90.62	2.86	4.04	0.70	46.50	45.90	0.11	0.05		
Bonemeal	4/30/51	97.30	90.57	3.73	-----	-----	5.70	33.14	15.11		
Calcium Carbonate	4/30/51	100.00	99.54	-----	-----	-----	-----	38.00	-----		
Prairie Hay	4/30/51	-----	-----	-----	-----	-----	-----	-----	-----		12.8 ²
Bonemeal	6/12/51	98.27	87.62	7.18	0.12	-----	5.20	31.04	15.57		
Beet Pulp	6/12/51	91.30	2.98	8.93	0.47	22.72	64.90	0.81	0.04		

¹ This amount of carotene present in corn gluten meal was included in calculation of daily intake of the cows.

² Air-dry basis.

Cow 43 in Lot 1 and cow 29 in Lot 2 were fed only beet pulp as roughage during the second month.

Cows 33 and 46 in Lot 1 and cows 19 and 50 in Lot 2, because they calved earlier than the other cows, were continued on the experiment until twenty weeks after calving; the experiment was terminated for all other cows at the end of the eighth week. From the twelfth to the twentieth week, the two cows in Lot 1 and the two cows in Lot 2 were fed a carotene-deficient ration; however, a small amount of carotene was found to be present in corn gluten meal.

Cow number 46 in Lot 1 lost her calf at the time of calving; in order to keep her in the experiment on a basis comparable to that of the other cows, a Holstein calf was obtained for the cow to suckle. No data on this calf were included in the experiment.

At the completion of the experiment, the cows were turned to grass.

RESULTS AND DISCUSSION

The cows in the high phosphorus lot required no assistance at the time of calving, and all calves appeared to be strong. One cow in the low phosphorus lot required assistance at calving, and one calf died shortly after birth; all calves in the low phosphorus lot were weak at birth.

The results of blood plasma analyses are given in Table 3. The average plasma inorganic phosphorus level of both lots was 2.8 milligrams per 100 milliliters plasma. At parturition, the average for the low phosphorus lot was 4.0, whereas the the average for the high phosphorus lot was 7.9 mgs. Eight weeks after parturition, the average for Lot 1 was 2.0; the average for Lot 2 was 5.6 mgs.

At the initial bleeding, the average carotene values were about the same for both lots. From the first bleeding until parturition, the average carotene values decreased from 194 to 180 micrograms per 100 milliliters plasma for the low phosphorus cows and from 190 to 165 micrograms for the cows of Lot 2. The carotene values for both lots were lower one week after calving than at calving. Gallup and Kuhlman (1941), Sutton and Soldner (1943), Kuhlman and Gallup (1944), and Sutton, et al. (1945) observed that in dairy cattle there is a drop in plasma-carotene levels a few days before parturition and a further drop immediately following parturition. The carotene values for the low phosphorus cows increased at the second week, whereas values for the high phosphorus cows decreased. The average carotene value of the low phosphorus cows was higher at each date of bleeding for the first eight weeks than those of the high phosphorus cows. These results are similar to

TABLE 3

BLOOD PLASMA ANALYSES OF COWS IN EXPERIMENT 3

End of ¹ Depletion Period	Partu- rition	Weeks Postpartal									
		1	2	3	4	6	8	12	16	20	
<u>Carotene (mcg per 100 ml)</u>											
Lot 1, Low Phosphorus											
33	160	138	138	160	155	174	172	186	54	32	24
46	252	214	114	180	190	161	219	186	44	39	40
41	208	209	206	204	178	155	95	78			
43	<u>156</u>	<u>157</u>	<u>128</u>	<u>121</u>	<u>129</u>	<u>130</u>	<u>58</u>	<u>38</u>			
Av.	194	180	147	166	163	155	136	122	49	36	32
Lot 2, High Phosphorus											
19	188	149	122	117	115	113	149	168	60	29	4
50	145	151	117	133	116	157	171	171	104	50	24
29	128	147	115	108	115	96	54	52			
90	257	215	190	182	198	192	139	77			
98	<u>234</u>	<u>162</u>	<u>161</u>	<u>135</u>	<u>137</u>	<u>151</u>	<u>165</u>	<u>101</u>			
Av.	190	165	141	135	136	142	132 ²	120 ²	82	40	14
<u>Vitamin A (mcg per 100 ml)</u>											
Lot 1, Low Phosphorus											
33	5.3	8.7	7.9	8.7	7.0	8.7	10.5	12.8	11.0	27.0	28.7
46	8.9	7.5	11.0	5.8	10.5	5.8	10.1	13.7	21.0	23.4	21.9
41	8.2	10.5	4.0	8.3	7.9	8.7	20.0	15.0			
43	<u>13.2</u>	<u>10.1</u>	<u>4.9</u>	<u>7.5</u>	<u>7.0</u>	<u>6.2</u>	<u>7.0</u>	<u>12.3</u>			
Av.	8.9	9.2	7.0	7.6	8.1	7.4	11.9	13.5	16.0	25.2	25.3
Lot 2, High Phosphorus											
19	11.0	9.2	8.7	11.0	9.2	7.0	11.0	8.3	21.5	30.7	4.9
50	13.9	12.8	5.3	10.5	6.6	12.8	13.7	13.7	23.4	32.2	17.7
29	7.4	5.8	3.6	0.7	0.7	6.6	9.2	22.9			
90	8.9	11.0	5.8	9.6	6.6	11.4	11.0	13.7			
98	<u>8.2</u>	<u>7.9</u>	<u>12.3</u>	<u>5.3</u>	<u>7.0</u>	<u>7.0</u>	<u>6.2</u>	<u>17.3</u>			
Av.	9.9	9.3	7.1	7.4	6.0	9.0	10.6 ²	15.1 ²	22.5	31.2	11.3
<u>Phosphorus (mg per 100 ml)</u>											
Lot 1, Low Phosphorus											
33	2.9	1.3	2.0	1.5	2.1	2.1	3.0	2.7	4.0	2.7	-2.6
46	3.2	1.7	3.2	3.7	3.5	4.2	5.7	2.9	3.5	3.6	2.3
41	2.4	6.2	2.9	2.6	3.0	3.5	2.2	1.4			
43	<u>2.8</u>	<u>6.6</u>	<u>5.8</u>	<u>3.3</u>	<u>1.7</u>	<u>2.9</u>	<u>1.7</u>	<u>1.0</u>			
Av.	2.8	4.0	3.5	2.8	2.6	3.2	3.2	2.0	3.8	3.2	2.5
Lot 2, High Phosphorus											
19	1.8	5.1	5.1	6.2	5.7	6.7	3.8	4.3	2.9	3.8	3.6
50	2.8	7.6	11.7	11.0	4.5	4.4	6.7	7.8	6.0	3.3	6.2
29	3.0	8.2	3.9	6.0	7.7	6.5	5.5	4.6			
90	3.4	9.5	8.0	7.2	6.6	5.5	5.6	5.8			
98	<u>3.2</u>	<u>9.2</u>	<u>7.2</u>	<u>4.8</u>	<u>7.0</u>	<u>7.1</u>	<u>6.2</u>	<u>7.0</u>			
Av.	2.8	7.9	7.2	7.0	6.3	6.0	5.5 ²	5.8 ²	4.5	3.6	4.6

¹ Number of days from this date (January 26, 1951) until date of calving was 31 for cow 33, 23 for cow 46, 113 for cow 41, 83 for cow 43, 21 for cow 19, 27 for cow 50, 65 for cow 29, 55 for cow 90, and 52 for cow 98.

² Weighted averages.

those reported by Ross and Gallup (1949), who observed an inverse relationship between plasma-inorganic phosphorus and plasma-carotene in cows receiving a limited intake of phosphorus. The carotene values of cows 33 and 46 in Lot 1 (low phosphorus) and of cows 19 and 50 and Lot 2 (high phosphorus)-- which received little or no carotene from the eighth to the twentieth week after parturition--decreased quite rapidly.

At the end of the depletion period, the average vitamin A level in the blood plasma of Lot 1 cows was 8.9 micrograms per 100 milliliters of plasma, whereas for Lot 2 the average value was 9.9 micrograms. At the time of parturition, the average values were 9.2 and 9.3 micrograms, respectively. From the first to the fourth week after parturition, no consistent difference was noted in the vitamin A values between lots. The vitamin A values increased for most cows from the end of the fourth week to the eighth week when the carotene intake was decreasing. Two cows in Lot 1 and two cows in Lot 2 were observed to have much higher values at the twelfth and sixteenth week than at the fourth week, and the carotene intake was only one-fifth as much. The cows of Lot 2 had higher vitamin A values at twelve and sixteen weeks than the cows of Lot 1. Cow 19 in Lot 2 had a value of 4.9 micrograms vitamin A per 100 milliliters plasma at the end of twenty weeks. This cow was excitable and showed some signs of night blindness and muscular incoordination. These symptoms of a vitamin A deficiency are similar to those reported by Moore (1940). Davis and Madsen (1941) reported that an inadequate carotene intake in cattle could be determined by blood analyses and that carotene and vitamin A content of blood plasma at any particular time was dependent on the current intake and previous storage of these

factors. When cattle were depleted of carotene reserves, the blood carotene level was dependent on intake alone. Davis and Madsen further stated that the critical level of carotene in the blood plasma was 25 micrograms and that for vitamin A was 16 micrograms per 100 milliliters of plasma.

Liver Analyses

The carotene and vitamin A values of liver samples obtained from cows by biopsy are shown in Table 4.

Analyses of liver samples taken before calving showed that the average content of carotene was slightly higher for cows in Lot 1 than for cows in Lot 2, the respective values being 1,192 and 1,114 micrograms per 100 grams of liver. The average carotene content of the livers of cows from both lots was lower at each subsequent bleeding. Carotene values of Lot 1 were higher than those of Lot 2 at each date of sampling, except at the eighth week. Cow 19 (Lot 2) had a high level of carotene in the liver at this date, a value not consistent with previous values obtained. This high value might have been due to an error in sampling or in liver analyses. From the eighth to the twentieth week, when cows 33 and 46 in Lot 1 and cows 19 and 50 in Lot 2 received little or no carotene, the carotene level in the livers dropped sharply. At the end of the twentieth week, the average carotene value for these two cows in Lot 1 was 306 micrograms per 100 grams of liver and 157 for the two cows in Lot 2.

Analyses of liver samples, obtained prior to parturition, showed an average of 27,314 micrograms of vitamin A for Lot 1 and 19,739 micrograms per 100 grams liver for Lot 2. Values for both lots gradually decreased until the eighth week. From the eighth to the twentieth week, there was a

TABLE 4

CAROTENE AND VITAMIN A CONTENT OF COW LIVER
(Samples obtained by biopsy)

	End of Depletion Period	Partu- rition	Weeks Postpartal			
			4	8	16	20
<u>Carotene (mcg per 100 gm)</u>						
Lot 1, Low Phosphorus						
33	901	693	868	589	359	260
46	1,391	1,240	790	794	420	352
41	1,136	1,031	1,023	480		
43	<u>1,340</u>	<u>915</u>	<u>1,075</u>	<u>667</u>		
Average	1,192	970	939	633	394	306
Lot 2, High Phosphorus						
19	824	753	688	1,017	254	197
50	534	525	502	326	199	117
29	1,251	908	852	681		
90	1,146	982	951	725		
98	<u>1,316</u>	<u>1,083</u>	<u>740</u>	<u>703</u>		
Average	1,014	850	747	685 ¹	227	157
<u>Vitamin A (mcg per 100 gm)</u>						
Lot 1, Low Phosphorus						
33	25,194	21,405	23,042	22,555	8,545	7,932
46	22,248	22,142	21,872	13,713	6,957	6,404
41	27,244	32,655	33,779	13,085		
43	<u>34,570</u>	<u>28,110</u>	<u>31,826</u>	<u>20,009</u>		
Average	27,314	26,078	27,630	17,341	7,751	7,168
Lot 2, High Phosphorus						
19	15,982	13,933	14,465	17,436	2,312	2,889
50	9,991	11,082	11,292	9,307	2,068	1,646
29	18,980	15,565	14,876	10,561		
90	32,466	26,211	25,602	21,642		
98	<u>21,276</u>	<u>13,885</u>	<u>11,418</u>	<u>9,916</u>		
Average	19,739	16,135	15,531	13,271 ¹	2,190	2,268

¹ Weighted averages.

sharp drop in vitamin A levels for the two cows in Lot 1 and the two cows in Lot 2 which received little or no carotene from the ration during this period. The Lot 1 averages at the sixteenth and twentieth week were 7,751 and 7,168 micrograms, respectively, whereas for Lot 2 the average values for the two dates were 2,190 and 2,268 micrograms per 100 grams of liver. This decrease in liver reserves of vitamin A from the eighth to the twentieth week is in agreement with results reported by Frey and Jensen (1946). They showed that, for steers, an increasing loss of hepatic reserves of vitamin A occurred with decreasing carotene intake. The rate of depletion of large reserves of vitamin A was studied by Popper and Brenner (1942). They stated that the rate of depletion is greater than can be accounted for by the needs of the body. Glover, Goodwin, and Morton (1947) reported that the plasma-vitamin A levels of rats were maintained near normal even when liver stores approached exhaustion

Colostrum and Milk Analyses

The analyses of colostrum and milk for vitamin A and carotene are shown in Table 5.

Considerable variation in concentration of carotene and vitamin A of the colostrum and milk was observed. The carotene content of colostrum from cows of Lot 1 ranged from 61.6 to 172.6 micrograms, with an average value of 126.7 micrograms per 100 milliliters. The average value of cows of Lot 2 was 169.1, these values ranging from 69.4 to 486.0 micrograms. One week after parturition, the average value for Lot 1 was 5.6; for Lot 2 the average was 6.2 micrograms carotene. From the second to the eighth

TABLE 5

CAROTENE AND VITAMIN A CONTENT OF COLOSTRUM AND MILK

Colostrum	Weeks Postpartum						
	1	2	3	4	6	8	
<u>Carotene (mcg per 100 ml)</u>							
Lot 1, Low Phosphorus							
33	61.6	2.9	9.7	1.7	None	8.1 ¹	1.7
46	172.8	12.0	14.6	4.7	6.6	4.4	0.2
41	152.6	5.3	4.4	3.5	5.6	2.6	2.0
43	<u>119.8</u>	<u>2.0</u>	<u>2.9</u>	<u>1.1</u>	<u>2.0</u>	<u>4.4</u>	<u>3.5</u>
Average	126.7	5.6	7.9	2.8	3.5	4.9	1.9
Lot 2, High Phosphorus							
19	100.8	10.7	3.5	2.0	1.7	0.8	0.2
50	69.4	2.0	4.4	0.8	3.5	3.2	0.2
29	92.8	4.4	5.3	2.6	None	2.9	0.2
90	96.4	4.1	3.8	2.0	1.7	2.3	0.8
98	<u>486.0</u>	<u>9.7</u>	<u>3.2</u>	<u>2.6</u>	<u>None</u>	<u>0.5</u>	<u>4.4</u>
Average	169.1	6.2	4.0	2.5	1.4	2.3 ²	1.7 ²
<u>Vitamin A (mcg per 100 ml)</u>							
Lot 1, Low Phosphorus							
33	87.1	3.8 ¹	15.1	3.4	1.1	1.5	3.8
46	156.3	12.1	11.4	4.6	6.4	1.2	2.5
41	242.5	5.5	6.9	6.7	6.5	0.6	1.2
43	<u>294.5</u>	<u>3.7</u> ¹	<u>8.6</u>	<u>2.2</u>	<u>3.5</u>	<u>3.8</u>	<u>9.6</u> ¹
Average	195.1	5.0	10.5	4.2	4.4	1.8	4.3
Lot 2, High Phosphorus							
19	234.0	24.0	7.0	5.5	10.6	6.1	5.3
50	206.9	9.6	10.9	6.2	7.6	5.0	5.9
29	148.3	8.3	11.9	5.5	3.5	7.0	6.4
90	269.0	4.1	2.1	5.5	1.2	4.0	3.1 ¹
98	<u>521.6</u>	<u>15.7</u>	<u>1.9</u>	<u>5.0</u>	<u>2.3</u>	<u>1.5</u>	<u>6.2</u> ¹
Average	276.0	12.3	6.8	5.5	5.0	5.2 ²	5.6 ²

¹Analyses repeated, results confirmed.²Weighted averages.

week, the carotene values of milk were higher for cows of Lot 1 than for cows of Lot 2. It was observed that the cows of Lot 1 had higher carotene levels in both blood plasma and milk than cows of Lot 2 during most of the experiment. Deuel, et al. (1942) reported, in an experiment with dairy cattle, a rough proportionality between levels of carotene in the blood and milk.

The average content of vitamin A in the colostrum was 195.1 for Lot 1 and 276 micrograms per 100 milliliters colostrum for cows of Lot 2. At the end of one week, these values had decreased to 5.0 and 12.3 micrograms. The amount of vitamin A in the colostrum was 10 to 100 times that found in milk obtained one week after parturition. Dann (1933) and Henry, et al. (1940) determined the vitamin A content of Shorthorn colostrum, found a marked variation from cow to cow, and found that, in some samples, the concentration of vitamin A was as much as 100 times that of later milk. Van Arsdell, Ross, and MacVicar (1950) reported the following average vitamin A values found in colostrum in three lots of Hereford cows: 114.2, 37.3, and 96.8 micrograms per 100 milliliters. These values decreased to 10.7, 9.4, and 9.8 micrograms at the end of ten days.

Calves

The average birth weight of the three calves produced in Lot 1 was 68 pounds, whereas the average for Lot 2 calves was 61 pounds. When calves were four weeks of age, the weights were 100 and 92 pounds respectively; at the end of eight weeks, 129 and 120 pounds.

The blood plasma analyses of the calves are given in Table 6. Blood samples taken at birth showed an average plasma inorganic phosphorus level

TABLE 6
BLOOD PLASMA ANALYSES OF CALVES IN EXPERIMENT 3

Birth	Weeks of age									
	1	2	3	4	6	8	12	16	20	
<u>Phosphorus (mg per 100 ml)</u>										
Lot 1, Low Phosphorus										
33	4.2	9.3	8.9	9.1	7.5	7.7	7.8	6.0	5.3	6.8
41	7.4	7.6	7.3	7.1	6.5	7.4	7.6			
43	8.8	7.1	8.4	7.3	7.8	7.4	7.5			
Average	6.8	8.0	8.2	7.8	7.3	7.5	7.6	6.0	5.3	6.8
Lot 2, High Phosphorus										
19	7.3	9.6	9.8	8.4	7.4	8.2	8.0	7.7	8.3	6.6
50	7.2	9.5	8.2	9.2	8.2	7.3	9.4	8.2	6.4	8.5
29	6.7	8.5	8.9	8.5	8.6	8.5	6.7			
90	6.6	8.4	7.3	7.4	7.6	7.3	7.6			
98	7.0	8.6	8.5	7.7	8.5	8.2	8.2			
Average	7.0	8.9	8.5	8.2	8.1	7.9	8.0	8.0	7.4	7.6
<u>Carotene (mcg per 100 ml)</u>										
Lot 1, Low Phosphorus										
33	None	6	Trace	11	8	8	40	24	40	17
41	None	22	22	29	24	6	19			
43	2	18	23	12	31	21	6			
Average	1	15	15	17	21	12	22	24	40	17
Lot 2, High Phosphorus										
19	None	13	8	None	None	6	46	44	38	20
50	10	None	None	None	7	8	40	71	64	33
29	1	20	14	17	23	28	8			
90	2	10	6	None	13	36	23			
98	1	13	13	None	10	13	13			
Average	3	11	8	4	11	18	26	58	51	27
<u>Vitamin A (mcg per 100 ml)</u>										
Lot 1, Low Phosphorus										
33	None	14.1	15.9	15.0	6.6	4.0	7.9	7.9	12.3	2.3
41	1	10.1	1.1	14.6	6.2	5.8	3.2			
43	10.1	11.9	15.5	5.3	7.7	8.3	9.2			
Average	3.6	12.0	10.6	11.6	6.8	6.0	6.7	7.9	12.3	2.3
Lot 2, High Phosphorus										
19	None	15.9	5.8	9.2	6.6	Trace	3.2	10.5	12.3	None
50	4.0	15.9	16.4	12.3	14.6	7.0	7.0	9.2	15.9	None
29	Trace	4.9	9.2	8.3	5.3	8.6	8.9			
90	4.4	6.6	8.3	1.5	7.0	5.8	4.0			
98	2.8	14.6	11.0	10.5	1.9	8.7	8.7			
Average	2.2	11.6	10.1	8.4	7.1	6.0	6.4	13.9	14.1	None

of 6.8 milligrams per 100 milliliters plasma for Lot 1 calves and 7.0 for calves of Lot 2. The phosphorus value of the calves of Lot 1 was nearly twice as high as the phosphorus value of their dams. Green and Macashill (1928), Eckles, Gullickson, and Palmer (1932), and Godden and Allcroft (1932) have reported findings of greater concentrations of inorganic phosphorus in newborn calves than in their dams. There was an increase in phosphorus values for calves of both lots from birth to one week of age. At each bleeding date, phosphorus values of calves from Lot 2 cows (high phosphorus) were slightly higher than values of Lot 1 calves.

Only a small amount of carotene was found in the blood plasma of five calves at the initial bleeding; no carotene was present in blood plasma of two of the calves. From the first until the fourth week after birth, calves of Lot 1 had higher carotene values than those of Lot 2.

At the time of birth, the average vitamin A value of Lot 1 was 3.6 micrograms, whereas the average value of Lot 2 was 2.2 micrograms per 100 milliliters plasma. After one week, the average levels of vitamin A were 12.0 and 11.6 micrograms, respectively. Consistent differences in vitamin A were not observed between the two lots during the remainder of the trial. Van Arsdell, Ross, and MacVicar reported average vitamin A values of three lots of Hereford calves. Values at birth were 4.2, 4.3, and 2.3 micrograms; one week later, the average values of the calves were 9.5, 13.5, and 9.1 micrograms per 100 milliliters plasma.

Carotene and Vitamin A Content of Liver of Calves

A measurable amount of carotene was found in the liver of only one calf at the time of birth. After eight weeks, carotene was present in the liver of

only two calves. The results of analyses of liver for carotene and vitamin A are shown in Table 7.

A greater quantity of vitamin A was present in the liver of Lot 1 calves at the time of birth than in the liver of Lot 2 calves, the values being 429 and 164 micrograms per 100 grams of liver, respectively. Calves whose dams had the greatest concentration of vitamin A in the liver at parturition had the greatest reserves of vitamin A at birth. A great increase in storage of vitamin A in the liver of calves in both lots was noted during the first four weeks. At four and eight weeks, the average values for Lot 1 calves were 3,317 and 2,021 micrograms; for calves of Lot 2, at these dates, the concentration of vitamin A was 1,491 and 1,747 micrograms per 100 grams of liver, respectively. Hart and Guilbert (1933) found that the livers of newborn animals tend to be comparatively low in vitamin A, irrespective of the diet of the dam during gestation. Speilman, et al. (1946) stated that the prepartum diet of the normal bovine may influence markedly the vitamin A and carotene reserves of the newborn calf. Henry, et al. (1949) conducted vitamin A studies with rats and concluded that the vitamin A content of the milk of the mothers and of the livers of the young was influenced more by the amount of vitamin A taken by the mother during lactation than by her liver reserves.

TABLE 7

CAROTENE AND VITAMIN A CONTENT OF CALF LIVER
(Samples obtained by biopsy)

Birth		Age in weeks				
		4		8		
Carotene	Vitamin A	Carotene	Vitamin A	Carotene	Vitamin A	
<u>(mcg per 100 gm)</u>						
Lot 1, Low Phosphorus						
33	None	176	None	1,496	41	2,067
41	30	628	8	4,183	None	3,691
43	<u>None</u>	<u>482</u>	<u>4</u>	<u>4,273</u>	<u>None</u>	<u>305</u>
Average	10	429	4	3,317	14	2,021
Lot 2, High Phosphorus						
19	None	162	None	1,674	19	1,515
50	None	58	None	910	None	1,995
29	None	113	None	3,071	None	1,396
90	None	329	None	1,341	----- ¹	----- ¹
98	<u>None</u>	<u>158</u>	<u>None</u>	<u>458</u>	<u>None</u>	<u>2,083</u>
Average	None	164	None	1,491	5	1,747

¹ A liver sample was not obtained from calf 90 at this date.

SUMMARY

Nine mature, pregnant, Hereford cows were fed a low phosphorus ration for a period of 71 days. At the end of this period--a time estimated to be approximately four weeks prior to parturition, the cows were divided into two lots on the basis of phosphorus level of plasma, carotene and vitamin A content of the plasma and liver, and body weights.

Four cows were fed the low phosphorus ration, which consisted of 1 1/2 pounds corn gluten meal and 15 pounds prairie hay. This ration provided an intake of approximately 0.7 gram of phosphorus per 100 pounds body weight. Five cows received the same ration plus sufficient bonemeal so that the daily intake of phosphorus per cow was approximately 3 grams per 100 pounds body weight.

Blood and milk samples were taken from the cows, and blood samples taken from the calves at parturition and weekly for four weeks and at the sixth and eighth week. Liver samples, obtained by biopsy, were obtained from the cows and calves at parturition and at four and eight weeks. Blood samples were analyzed for inorganic phosphorus, carotene, and vitamin A. Colostrum, milk, and liver samples were analyzed for carotene and vitamin A.

Data from these analyses indicated a trend for cows receiving the low phosphorus ration to have higher carotene levels in the blood plasma than cows receiving a high phosphorus ration. No consistent differences in vitamin A levels were observed between the two lots.

Considerable variation was observed in the concentration of the carotene and vitamin A in the colostrum. Cows fed the low phosphorus ration tended

to have higher carotene levels in the milk than cows fed the high phosphorus ration.

Concentration of carotene and vitamin A in the liver of both lots of cows decreased as the carotene intake of the cows was reduced.

Eight weeks after parturition, the low phosphorus lot had lost 36.5 per cent of its original reserves of vitamin A in the liver, whereas the high phosphorus lot had lost 32.8 per cent.

Approximately twice as much inorganic phosphorus was present in the plasma of newborn calves in the low phosphorus lot as was present in the blood plasma of their dams.

Only a slight amount of carotene and vitamin A was present in the blood plasma of the calves at birth. At the end of one week, these values had increased 6 to 22 per cent.

A measurable amount of carotene was found in the liver of only one calf at the time of birth. After eight weeks, carotene was present in the liver of only two calves.

Calves whose dams had the greatest concentration of vitamin A in the liver at parturition had the greatest reserves of vitamin A at birth.

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THESIS TITLE: I. Relation of Nutrition and Age at First Calving
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II. The Effect of Low Phosphorus Rations
Upon Carotene Metabolism in Ruminants

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