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THE RELATIVE VALUE OF RAW ROCK PHOSPHATE AND BONE LEAL AS SOURCE OF CALCIUM AND PHOSPHORUS FOR LACTATING

DAIRY COWS

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DAIRY COWS .

Thesis

Respectfully submitted to the faculty of Michigen State College in partial fulfillment of the requirements for the degree of Master of Science.

> by Wright B. Jones

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ACKNOWLEDGMENTS

The author wishes to acknowledge his indebtedness to C. F. Huffman, Research Assistant in Dairying, for his aid in planning and conducting the experiment and for his kindly criticism of the manuscript.

He also wishes to express his gratitude to Professor O. E. Reed, Head of the Department of Dairy Husbandry, for timely advice and assistance in the investigation.

Likewise the writer wishes to acknowledge his appreciation of the help given him by Doctor A. J. Patten, Head of the Chemistry Experiment Station and Doctor C. S. Robinson, Research Assciate in Chemistry, both of whose suggestions and aid made the chemical analyses possible.

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INTRODUCTION

The ...dern dairy cow has become such an efficient milling machine that the ordinary feeds can no longer supply calcium and phosphorus in quantities large enough to keep her up to full production. Both elements are needed in relatively large amounts by the animal body. This need is intensified by increase in growth, by lactation, and by pregnancy. About three-fourths of the ash of the body is calcium photphate, and as the animal increases in size the need for calcium and phosphorus in the feed is increased. A heavily lactating cow supplies a large anount of calcium and phosphorus in the allk as milk contains both of these elements. The work of most investigators points to the fact that during heavy milk production there is a loss of calcium and photohorus from the body. Also the body of a new born calf weighing eighty pounds contains according to Eckles and Trowbridge, about 2.8 pounds of mineral matter of which three-fourths is calcium and phosphorus. These elements cust be furnished by the mother and adds to the amount rejuired in the feed.

Most of the roughages and especially the letunes contain calcium in abundance. Cottonseed meal, linseed oil meal, wheat bran and in fact nearly all concentrates have relatively large amounts of phosphorus. The ordinary animal on a well balanced ration of grains and legume hay probably has plenty of calcium and phosphorus in her diet. It is different, however, with a heavily lactating animal. The supply in the common feeds is not enough to meet her requirements. There is a need for supplements which have a high percentage of both calcium and phosphorus.

Bone meel and raw rock phosphate are two minerals which contain an abuldance of these elements. Bone meel is a socelled organic source. Bone meel is a by product of the meat industry and can be secured from packing houses only in limited quantities. This makes bone meel a relatively expendive sup leadnt. Raw rock phosphate is an inrogenic source of celcium and phosphorus. It is simply the natural rock very finely ground. The supply of raw rock phosphate is practically unlimited and is therefore inexpensive. If there was not difference in the feeding value, raw rock phosphate would be a much chesper supplement than bone meel.

No work with dairy cows had been done directly comparing bone meal and raw rock phosphate and this experiment was undertaken in order to determine the relative availability of the calcium and phosphorus of bone meal and raw rock phosphate when fed to lactating dairy cous.

REVIEW OF LITERATURE

Calcium and phosphorus are considered the limiting mineral elements in animal nutrition, since they are present in the body in much larger quantities than any of the other elements. Larger amounts of both are lost from the body during the normal life processes; and probably the main use of mineral supplements such as raw rock phosphate and bone meal is to replace the calcium and phosphorus that is used up in this way.

Normal Functions of Calcium and Phosphorus

Calcium and Phosphorus of the Skeleton

About three-fourths of all the ash of the body is calcium phosphate. (21) About five percent of the live weight or twelve percent of the dry weight of the carcass of the ox is calcium phosphate. (86) Lawes and Gilbert have made the following analysis: (60)

	Half fat ox	Fat ox	Calf
Calcium	1.508%	1.281%	1.177%
Phosphoru s	0.803%	0.677%	0.670%

By far the greatest percentage of both calcium and phosphorus is found in the skeleton. Probably 99% of all the calcium and 85% of the phosphorus of the body are found in the bones. The two elements make up about 90% of the ash of bone. (42) They are largely combined, seven-eighths of the bone ash being phosphate of lime. (21) Beaunis found the following composition of bone: Lime 37.58%

Phosphoric Acid 53.31%

Hart, McCollum and Humphrey of the Wisconsin Station (34) have shown that the animal skeleton acts as a reserve storehouse for mineral matter supplying calcium and phosphorus when the amount in the feed is below requirements, in order that the metabolic processes of the body may continue. Under such conditions the calcium of the flesh and other soft parts of the body remain as high as in animals liberally fed with these elements. These investigators found that a cow fed a ration deficient in calcium during three and one half months gave off 5.5 pounds more calcium in milk and excrement than was in the feed. This was fully 25% of all the calcium in her body, in cluding the skeleton, at the beginning of the trial.

Forbes and associates (23) showed that liberal milk production on common practical rations, fed in sufficient quantities to maintain live weight and cause regular nitrogen and sulphur storage, caused consistent losses of calcium, magnesium and phorphorus from the cow's skeleton. Losses occurred in spite of liberal supplies of these elements in the feeds. (24) With a ration of common feeds chosen to provide maximum supply of mineral nutrients all calcium, magnesium and phosphorus balances were negative. With increase in these elements by increasing the food consumed and by addition of calcium carbonate and bone meal all calcium end all but one magnesium balance remained negative, the phosphorus balance, however, became positive.

These experiments indicate that minerals are normally

removed from the skeleton during liberal milk production.

Later studies at the Ohio Station (25) gave no evidence that the limited utilization of calcium compounds is due to the difficult solubility of calcium compounds in the ration. Calcium lactate, calcium chloride, and precipitated bone phosphate, all soluble compounds of calcium, were added to the rations without making the calcium balance positive. During this experiment the phosphorus balances were negative.

All of these experiments show that it is a normal function of the skeleton to give off calcium and phosphorus during liberal lactation.

This loss of minerals seems to take place until the impulse to secrete milk has largely spent itself after which a retention begins to take place. Forbes and associates (27) showed in a series of studies that in 49 negative balances of calcium during liberal milk production, without one exception, the animal began to store calcium as soon as the milk production had decreased to such an extent that the calcium outgo did not exceed their capacities to assimilate calcium.

Meigs and Woodward (81) found that milk cows store large amounts of calcium and phosphorus during a dry period, if they are properly fed.

It appears from the results of all these investigators that the skeleton is made up largely of calcium and phosphorus; that both of these elements are stored in the skeleton during growth and periods of rest from lactation; that during heavy milk production the animal uses up part of the calcium and phosphorus of the skeleton.

Calcium and Phosphorus in Other Tissues

Both calcium and phosphorus are found in other tissues of the body. Phosphorus is found in relatively large quantities in muscle, brain, nerves, liver and lungs. Gilbert and Posternack (32) estimate the total phosphorus of the human body as 1600 grams P_2O_5 at middle life. This 1600 grams is made up of about 1400 grams in the skeleton, 130 grams in muscles, 12 grams in brain, 8 grams in nerves, 10 grams in liver, 6 grams in lungs, and about 4 grams in blood. Calcium is also found in these tissues but in much smaller quantities. (28)

Calcium and Phosphorus in Milk

Very large quantities of both calcium and phosphorus are contained in milk. This accounts for the enormous drain on the calcium and phosphorus of the skeleton during liberal milk production. The phosphorus occurs mostly as phosphates of calcium, magnesium, and aluminum. (40) Calcium occurs as phosphate, citrate and caseinate.

According to various authors the content of lime and phosphoric acid in cows milk is as follows:

Author	% C _a O	% P ₂ 0 ₅
Barge (1874)	0.1599	0.1974
Schrodt & Hansen (188	5) 21.45 % of ash	24.11% of ash
Trunz (1903)	0.1524	9.1821
	0.2841	0.3284
Jensen (1904)	0.2831	0.1983
Babcock	0.1400	0.1700
Katagama (1908)	0.1670	0.2160 (Average composition of milk of improved breeds)
	0.1780	0.225 (Average composition of milk of native breat
	0.5400	0 914

A 3 4 G 4

The percentage of calcium and phosphorus in milk is subject to but little variation. Some authors note (11) success in increasing the calcium content of milk by addition of calcium salts to the ration. An increase in the amount of calcium and phosphorus in milk, however, is hardly the purpose of the feeder in adding mineral supplements to the ration. The value of these feeds lies in their ability to supply enough calcium and phosphorus to meet the normal demand and thus prevent the loss of these elements from the skeleton.

Calcium and Phosphorus in Blood

Calcium and phosphorus are also important constituents of the blood. Phosphorus is present as potassium and calcium phosphate and inorganic combination as phosphatides and neucleoproteins or nucleins and according to Panella as phosphocarnic acid. (29) Lecithin was found by Gobley in 1851. Calcium occurs in blood serum as $C_a(HCO_3)_2$. (63) Other forms of calcium are present but in smaller quantities.

The analysis of blood as found by several investigators is as follows:

Ox blood Bull blood Corpuscles Serum Corpuscles Serum (1) Parts per 1000 of blood P205 0.2392 0.1646 0.2360 0.1560 1000 " 17 C_aO 0.0805 0.0730 (80) 9.6 - 9.8 milligrems calcium per 100 grams blood plasma (67) Oxen 6.43 milligrams celcium per 100 c.c. whole blood (8) Ox blood 7.82 milligrams calcium per 100 c.c. whole blood

In human beings the average calcium in the blood is between 9 and 11 milligrams per 100 c.c. whole blood. (39), (50), (10) In rats 9.5 to 10.5 milligrams per 100 c.c. and that of phosphorus 7 to 8.5 milligrams. (57) The amount of phosphorus in both the plasms and the corpuscles is extremely variable and depends to a large extent upon the amount available in the feed. Calcium, however, is quite constant. (53)

Calcium and phosphorus may both be used for specific purposes in the blood. For instance, calcium is necessary for clothing of the blood, and alkali compounds of phosphates are of great importance in maintaining almost constant reaction of body fluids. It is doubtful, however, if the feeding of minerals containing calcium and phosphorus can effect these functions. It is the amount of calcium and phosphorus that is being carried by the blood to replace that lost from the tissues through the normal life processes that may be affected by a diet rich in calcium and phosphorus.

<u>or Phosphorus Metabolism</u>

An insufficient supply of either calcium or phosphorus or a disturbed metabolism of these elements may lead to a number of pathological changes.

Among these osseous cachexia, rickets, and teteny are the most important.

Osseous Cachexia or Osteomalacia (In Human Beings)

Osteomalacia of domestic animals differs from human osteomalacia. In humans the disease is of an entirely unknown cause (30) One of the prominent symptoms is an absorption of selts from the bone and their replacement by tissues not having the normal content of bone salts. There is usually a marked · · ·

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loss of mineral matter generally, of calcium, a less marked loss of phosphorus, and a decided increase in magnesium. (83) A moderately severe case studied by McCrudden (79) showed that the relative emount of inorganic matter of the dried bone was 28% compared with 48-1/2% in normal bone. The calcium was decreased to nearly one-helf its normal value, while the relative amount of other mineral constituents was increased. The P_2O_5 was two-thirds normal, although the ratio P_2O_5 : C_AO was greater than normal. Magnesium increased fourfold and sulphur increased nearly fourfold. The relative increase of inorganic constituents other than calcium was greater than could be accounted for by simple decalcification. McCrudden assumes that this is a manifestation of the organism to supply other material to replace C_aHO_4 .

The method by which these changes take place, as stated by Pommer (91), in 1885 is through a continuous building of new lime-free bone tissue, a cessation of the deposition of lime, and a local removal of lime from the parts containing it.

Forbes states (31) "While the cause of the deposition of the salts in bone is not definitely known we must admit: (1) That there resides in the osteogenous tissue some properly which acts like an affinity for the bone salts. (2) That this property tends to maintain a constant proportion between the salts deposited. (3) That variations in the composition or reaction of the blood, as affected by disease, may modify the character of this affinity, as indicated by the composition of the bones, in definite and consistent ways, even if not to unlimited degrees."

It is apparent that human osteomalacia is not due primarily to the lack of phosphorus or calcium compounds in the food, nor to too repid growth, pregnancy, lactation, or senility, though naturally these may be contributory or accentuating conditions, but is due rather to a disturbance of the normal functions of the bone cells.

Osseous Cachexia or Osteomalacia (In Domestic Animals)

Osteomalacia of domestic animals, however, is due principally to the deficiency of the food in bone forming material, although it may be due to a lack of correct proportion between the mineral elements, bone-forming or otherwise, contained in the food. (30) The condition is easily curable. The principal predisposing factors are pregnancy, growth, lactation, starvation, and unhygenic condition. Bone meal and raw rock phosphate may be of great value as preventative or cure in regions where the disease is prevalent.

<u>Symptoms</u> The symptoms of the disease in domestic animals can be divided into four phases:

Initial phase. (84), (90), (105), (96).

Irregularity, diminution and sometimes perversion of appetite. Loss of spirits. Some interference with movement. Tendency to remain lying in the stable. This phase of the disease is seldom noticed except in a region where the disease is prevalent. The second phase, however, is more noticeable.

Second phase

Difficulty in rising. When standing still patient may seem to be in pain. The least muscular effort when lying down may cause it to moan. Weakness becomes marked and appetite very irregular. Swellings appear, due to arthritis of extremities. Secretion of milk diminishes or ceases, and abortion is not uncommon.

Third phase

Characterized by fractures. Even the slightest muscular effort or violence may cause fracture. These fractures are seldom accompanied by excessive bleedings, and they show little tendency to repair.

Fourth phase

Softening of the bones. This phase is very rarely seen in cattle because the accident so often accompanying the preceding stage necessitates slaughter. The bones become elastic, soft, and depressible. Bones of the head are usually the first to suffer becoming greatly enlarged. Osseous tissue, properly so-called, slowly disappears.

The development of the disease is slow, lasting one to two months as a rule.

Good milking cows seem to be most frequently attacked, probably because of great losses of nutritive material which occur through milk. According to J. M. Parker (90), pregnant cows seem to be the most susceptible. In some localities of Texas 50% of such cows are affected.

Occurrence. The disease has been reported among domestic animals in several countries and regions. D. Hutcheon (49) reports disease among cattle, sheep, goats and horses over a large area of Cape Colony, most prevalent on the easter. dosst. In Hawaii the disease occurs on soils lacking in lime. (17) Thieneux (61) reports the disease in cattle, horses, sheep and goats. In lucensland osteomalacia is a condition affecting meinly the bones of cows and heifers chiefly of dairy stock and high producers. Creeping sickness, as the disease is sometimes called. occurs along the coastal plain region during the dry season. (19) and the first symptoms of the disease may often be noted in sections of Michigan. The disease is somewhat prevalent emong cattle in sections of Montana. J. A. Reid and B. C. Aston (96) report the disease in sheep on soils lacking in lime. A Scheumut, A. Schattke and E. Loetsch (101) fed hay and oats to horses and osteomalacia appeared. The hay was somewhat lower in calcium than ordinary hay. No other marked differences were found.

<u>Cause.</u> As has been stated above the chief cause of osseous cachexia in domestic animals is a deficiency of the food in bone forming constituents. It may, however, be due to a lack of correct proportion between the mineral elements contained in the food. (30) W. Dibbelt fed certain salts and found that it is possible to continuously withdraw calcium salts (excretion in feces and urine) from the organism of a full grown animal. If animals so treated became pregnant, the fetus developed normally even as regards skeleton formation. The loss of calcium salts from the organism of the mother, however, was intensified and led to osteomalacia and atrophy of osseous tissue. Etuani (16)

shows that administration of C_aCL_2 in considerable amounts over long periods causes an inital retention of calcium followed by increased elimination. The loss is derived chiefly from the bones and may reach 15%. Deformations results which resemble osteomalacia. Both of these cases tend to show that the case may be an improper proportion of mineral elements in the feed.

Nearly all cases of osteomalacia in the domestic animal have occurred on soils lacking in lime and phosphorus or following the use of feeds deficient in these elements. This fact backs up the belief of Forbes that the disease is due primarily to feeds low in calcium and phosphorus.

<u>Treatment.</u> The treatment of the disease consists simply of supplying additional calcium and phosphorus to the ration in the correct proportions.

Moussu and Dollar (84) suggest as a cure the use of ground bone, tri basic or di basic calcium phosphate and feeds rich in calcium and phosphorus. They also recommend the use of cod liver oil, although it may be expensive. Forbes claims that the disease is readily curable by the use of calcium and phosphorus rich feeds. J. Hogen (45) says that lime and phosphate should be added to the ration. D. Hutcheon (49) claims bone meal or bone ash controls the disease.

All these results point to the use of mineral supplements as a preventitive or cure for osteomelacia of domestic animals.

Rickets

Rickets is essentially a disease of dietary orgin. It is due to a lack of deposition of calcium and phosphorus and in this way differs from osteomalacia which is caused by a withdrawal of them from the skeleton. According to many investigators a deficiency in the ration in calcium or phosphorus leads to rickets. It is therefore possible that young animals fed a ration low in one or both of these elements may develop the disease.

<u>Symptoms.</u> McCollum (70) says the disease varies in severity, and individual cases present pictures which differ in some detail but all of which have certain characterisites in common. Rickets leads to deformity, due to abnormal enlargements of the ends of bones, and to distortion due to bending because of the lack of resistance of the bones to body weight, muscular tension, and to atmospheric pressure. The latter factor is especially important in changing the form of the thorax. Bow legs, knock knees, enlarged joints, flat or deformed chests, and abnormal conformation of the skull, are results of the failure of bones to develop in the normal manner.

Rickets is essentially a disease of infancy and early childhood although it occurs in children of five or six years of age and even later ('rachitis tarda). The disease may be menifested by the second month of life. It is most frequent from the second month to the end of the year. It is usually accepted that the disease is rarely, if ever, present at birth, but since clinically recognized symptoms may occur very early in life, and since the development of the disease is slow, it must in meny cases have its beginning in the earliest days of extra uterine life.

<u>Occurrence</u>. Rickets occur chiefly in Europe and North America especially in large industrial centers where the people are living in conditions differing very widely from the natural state. Park (89) states that the disease is so common in the large cities of America and Europe that few children among the poorer classes are untouched by it.

Etiology. There are several factors that may be considered in the etiology of rickets. These are (1) the supply of calcium in the ration, (2) the supply of phosphorus in the ration, (3) calcium and phosphorus of the blood, (4) the proportion of these elements in the ration, (5) the presence of an organic substance (vitamin D) which aids in their assimilation, (6) and to these five we might add exercise and certain kinds of light.

Effect of rations low in calcium

It has been believed for a long time that a lack of sufficient cakium in the diet is of prime importance in the development of rickets. A survey of the history of the disease shows that rickets is most common in those parts of the world where milled cereal products, rubers, and muscle meats form the principal components of the dietary. This type of diet is essentially lacking in mineral constituents and vitamins, and has repeatedly been shown to be inadequate for the production of satisfactory milk by the lactating mother.

Recent experiments by several investigators have shown that rickets can be produced by feeding diets low in calcium. (87), (14), (74), (76), (75), (73)

Effect of rations low in phosphorus

Other experiments have illustrated the fact that rickets may also be developed by feeding rations low in phosphorus. (87), (103), (68)

Calcium and phosphorus of blood in rickets

McCollum and associates (77) state that there are two main kinds of rickets. One is characterized by a normal or nearly normal blood calcium and a low blood phosphorus (inorganic). The other by a normal or nearly normal phosphorus but a low blood calcium.

The above results of different investigators indicate that a lack of either, or perhaps both, of these elements in the diet may lead to rickets. It is, therefore, possible that an addition of mineral supplements containing calcium and phosphorus to the ration might be of value in the prevention of rickets.

The proportion of celcium and phosphorus in the dist

Another factor involving the metabolism of calcium and phosphorus may also be of importance in the development of the disease. McCollum (69) makes the following statement: "It would seem from the results of a larger number of experiments now available, that the physiological relation in the diet within certain limits between the two elements (calcium and phosphorus) is of much greater importance in insuring normal calcification, then the absolute amount of the salts themselves." He mentions the series of experiments carried on by his associates and himself and also those of Sherman and Pappenheimer (102) Hess, McCann and Pappenheimer (43) in support of this theory.

In discussing Sherman and Peppenheimer's work, McCollum says (72): "The basal ration employed by these investigators was deficient in celcium, sodium, cholorin, iron, and possibly potassium, in water-soluble B, in fat-soluble A, and in

the orgenic factor playing a role in the prevention of rickets. When calcium was added in the form of calcium lactate to the ration a marked disproportion in calcium-phosphate ratio was produced, the calcium being nearly optimel and the phosphorus very low. Fat soluble A was almost lacking and the condition was such under which we should expect severe rickets to develop. (The authors reported rickets on this diet) When neither calcium nor phosphorus was added, the ratio was more nearly optimum than after the calcium addition. Rickets did not develop. When potassium phosphate in addition to calcium lactate was added to the ration the calcium to the phosphate ratio was again more favorable and rickets was prevented."

In an experiment of McCollum and associates (71) a diet containing about twice the optimum amount of calcium and deficient in phosphorus and fat-soluble A was fed to rats. Very severe lesions developed. The disease depended in severity upon the deviation from the optimel calcium-phosphorus ratio. These investigators repeatedly observed that the addition of excessive amounts of calcium carbonate to diets which were deficient in phosphorus and fat-soluble A induced most pronounced disturbances in the growth of the bones.

The effect of feeding a ration containing calcium and phosphorum in a ratio opposite to that described in the above ratio was tried by Shipley, Park, Simmonds and McCollum. (104) A diet consisting of cereal grains and legumes did not induce any growth because it was too poor in calcium. It was likewise lacking in phosphorus although not to so marked a degree as in case of calcium low in fat soluble A, and the proteins were not

of the best quality. If this diet was supplemented with common selt, celcium and fat soluble A it supported good growth, although the animals were not normal. Without this supplement the animals were simply brought into a state of nutritional stability. They did not grow and did not develop rickets, although the bones were not entirely normal.

The addition of 10% casein, a phosphorized protein, enhanced the food value both with respect to phosphorus and amino acids, yet when this was done rickets promptly developed. This showed that the addition of phosphorus without exceeding a concentration which is about optimum if more calcium were available, may cause damage when it leads to an unfavorable ratio between the quantities of the two elements. When calcium salts were added with the casein the diet was greatly improved and the bones tended toward mormal structure.

These results show rather conclusively that the quantitative ratio between calcium and phosphorus in the diet is an important factor in the etiology of rickets.

The Mineral Retention Factor

The third factor concerned in the etiology of rickets is the so-called vitamin D. This substance is present in considerable quantities in cod-liver oil and to a very slight degree in butter-fat. Cod-liver oil as a folk remeay has been used for many years but no direct evidence of its curative action in rickets had been obtained until recently. McCollum and Simmonds (78) showed that cod-liver oil caused deposition of calcium selts in the bones of the rachitic rat. By means of the X-rey, Howland and Park (47) showed that the administration

of cod-liver oil to rachitic children was followed by the deposition of lime salt in the cartilage and bone after a period of 15 to 21 days.

In the treatment of rickets with codliver oil many investigators have obtained favorable results. J. A. Schabad (99) found that cod-liver bil increased retention of calcium in rachitis. Addition of phosphorus to the oil increased the favorable action. Both the plain oil and the phosphorized bil increased the phosphorus retention. The same author later found (100) in the use of phosphorized cod-liver oil in rachitis, the particular salt of calcium fed with it caused marked differences in calcium retention. Calcium acetate caused retention in large degree. Calcium citrate and calcium phosphate did not cause retention. Phosphorus retention corresponded to calcium retention.

Rucklin (98) obtained distinct improvement in a patient and an increase in calcium retention after administration of cod-liver oil with $Ca_{\Xi}(PO_4)_{2^{\bullet}}$

H. Chick, E. J. Dalyell, H. Hume, H. H. H. Machey, H. H. Smith and H. Wimberger (9) observed that rickets developed in children during winter and spring under excellent hygenic conditions when receiving a diet composed of milk to which sugar was added. The disease was prevented by addition of cod-liver oil, less carbohydrates and a somewhat larger amount of milk.

S. V. Telfer (106), however, found that the retention of calcium and phosphorus in an eight months old child was no better on a diet of skimplik and cod-liver oil

than on a diet of cow's milk or cow's milk to which batterfat had been added.

A series of experiments by McCollum and associates led to their making the following conclusions: (75) "Codliver oil contains in abundance some substance which is present in butterfat in but very slight amounts and which exerts a distinct influence on bone development and enables animals to develop with an inadequate supply of calcium much better than they could otherwise do. This substance is apparently distinct from fat soluble A which is essential for growth and which is associated with the prevention of ophthalmia."

This factor is also thought to be present in fresh, green forage and to be destroyed by curing in sunlight,

Hart, Steenback and Hoppert (36) experimenting with cows and gosts showed that milking snimels receiving grain and dry oats straw are brought into a decidedly negative calcium balance, 1.6 to 2.79 grams $C_{g}O$ daily. When the dry straw was replaced by an equivalent in dry matter of fresh, green material the negative balance was only 0.6 grams per day. Another gost showed a change from 1.5 to 2.5 grams $C_{g}O$ to 0.3 to 0.8 grams $C_{g}O$ per day. As the intake was only 8 to 9 grams $C_{g}O$ daily a positive balance was not expected. Apparently there was something in the fresh, green food which affected calcium assimilation.

The same investigators later showed that oat hey dried out of sublight but in a fairly well lighted room seemed to retain the properties of fresh, green oats. (37)

In enother series of experiments Hart, Steenback, Hoppert, Bethke and Humphrey (38) observed that cows

producing 20 to 45 pounds of milk deily fed a ration of grains, silage and timothy hay were in negative calcium and phosphorus balance. Substituting alfalfa hay for timothy hay, reduced the losses but did not make the balance positive. Positive balances with liberal milking cows was contrary to their former results. However, the alfalfa hay used in the first experiment had been cured under caps, while that used in the latter experiment was cured in windrows with exposure to air and sunlight. These differences in the effects of the two alfalfa hays may be attributed to a difference in the degree of destruction.of the vitamin assisting in assimilation due to the curring process.

It is evident from these results that the same factor which is present in cod-liver oil is also probably present in some green plants. Not all green plants, however, contain this substance.

The retention of calcium and phosphorus by dairy cows depends largely on the presence of a vitamin in the hay or grasses.

Relation of exercise and sunlight to rickets

The theory that leck of exercise is one of the main causes of rickets has not been upheld by investigators.

Park and Howland (89) and later Baldwin confined puppies in small cages for two or three months at a time without inducing rickets. Mellanby (82) showed that puppies on a properly constituted diet did not develop rickets when confined in such a way that they received very little exercise.

Light, however, has been found to exert direct influence in offering protection from rickets.

The seasonal variation indicates that sunlight

plays a very importent part in prevention of the disease. Schmorl (89) made a study of the occurrence of the disease and found that, although rickets may begin at any time, the highest percentage of early manifestations of the disease is between November and May when there is the least sunlight. Raezynski in 1912 (95) correlated the relationship which exists between the incidence of rickets and lack of sunlight. He pointed out that the curve representing the number of cases admitted to the hospital began to rise sharply in January, reached a maximum in May and fell rapidly in June. This shows practically the same thing as the work of Schmorl.

Hess, Unger, and Pappenheimer (44) fed rats a diet adequate in celcium but lacking in phosphorus. Rachitic lesion developed regularly. These could be prevented by exposure to direct sunlight.

Powers (92) placed 18 rats on a diet high in celcium but low in phosphorus and fat soluble A but in other respects well constituted. Twelve of the rats were exposed to 242 hours of sunlight over a period of 62 days. Six were kept under ordinary room light as controls. The controls were killed after 60 days and all showed rickets. The rats emposed to sunlight were without exception free from rickets confirmed by histological examination.

Raczynski (95) reported an experiment with puppies born of the same mother in May. One was kept in sunlight from morning to evening and the other was kept in total darkness. Both pups were nursed exclusively by the mother. At the end of six weeks the two were killed for examination. The one that had lived in light was normal whereas the one kept in darkness had

but poorly assimilated the mineral salts necessary for formation of a skeleton.

The use of light in caring of rickets began with Huldschinsky's work (89). He found that under the influence of the ultra violet ray in children suffering from rickets there was a deposition of calcium selts in the ends of the long bones which were observable in radiographs. Control children who were not treated with the rays showed no improvement.

Winkler, Putzig, Karger, Riedel, Sacks, Erlaeker, Mengert, and Hess have since corroborated this work and have proven beyond doubt the curative value of the ultra violet ray.

The effect of light on celcium assimilation seems to be the same as the effect of vitamin D. It must be noted, however, that neither can prevent rickets if the supply of calcium or phosphorus in the feed is too low. In rations deficient in these elements the supply of celcium and phosphorus may perhaps be kept up by the use of mineral supplements.

Tetany

A disease often associated with rickets is tetany. A low percentage of blood celcium and celcium of the tissues usually accompanies the disease. This fact suggests the possible use of celcium rich feeds as a preventative and cure for tetany.

It may be another case where raw rock phosphete or bone meal can be used to advantage.

Calcium in blood serum in teteny. The disease is usually accompanied by a lowering of the percent of calcium in the blood serum. The normal empoint as mentioned before is 10 - 11 milli-

grams per 100 c.c. serum. Howland, McKim and Marriott (46) found that calcium may fall as low as 3.5 milligrams. The average of 18 cases was 5.6 milligrams. Brown, McLachlan and Simpson (7) studied 8 cases of tetany and found the calcium to vary from 5.7 to 8 dilligrams. W. G. McCollum and Carl Voegtlin (65) showed that the blood of animals killed in tetany had about one-half normal concentration of calcium. F. F. Tisdell, B. Kramer, and J. Howland (107) and A. Orgler (93) mention the fact that calcium content of the blood is markedly low during tetany.

A reduction of 2.9 to 4 milligrams per 100 c.c. of serum was found by Paul Trendelenberg and w. Goebel. (108)

All of these results show that tetany is accompanied by low calcium content of blood serum.

<u>Celcium balance in tetany</u>. Accompanying the low blood celcium in tetany is very often, though not always, a negative celcium balance.

In four cases of tetany studied by Artzenias (3) three showed negative balance of calcium. The fourth patient showed symptoms of latent tetany but no disturbance of the calcium balance.

A. Orgler (93) found a negative balance of calcium in many cases of tetany.

Underhill, Tileston and Bogert (109) pointed out that a patient with tetany showed a greater tendency to store calcium on a calcium-rich diet and a greater tendency to lose calcium on a calcium-poor diet then normal individuals under the same condition. Tetany in infants largely depends on the diet (93). Potassium and phosphorus increase the symptoms. Calcium and magnesium havesgood influence. In many cases the calcium balance is negative. Parathyroid tetany and spontaneous tetany of infants all show the same type of calcium metabolism.

<u>Parathyroids and teteny.</u> That there is a relation between the parathyroid glands and calcium in the development of teteny is clearly shown by a large number of experiments. According to Robert H. Kunner (58) the appearance of tetany following the hypo-functioning of the parathyroids is due to the disturbance of the balance between calcium and the products of metabolism. The deficit of the calcium deprives the calls of their defence, facilitating the entrance of toxic products which give rise to tetany.

Paul Trendelenberg and W. Goebel (108) found in tetany resulting from parathyroidectomy there is a deionization of calcium and the total calcium of the serum is depressed. Avidence of calcium deficiency appears within six hours after the operation.

I. Ott and J. C. Scott (94) proved that removal of parathyroids alone caused tetany. This tetany was not due to a lack of calcium but to a poison in the blood.

Working with cets E. Farmer and R. Eline (20) found that small accessory parathyroids can always be found. These authors removed in forty cats the four principal parathyroids. Effect was not the same in all animals. Often acute and sub-acute tetany developed which ended fatally; other animals remained in good health after the operation. This was probably not due to the remaining accessory glands as there was no correspondence

between their existance and the symptoms.

The authors twice transplanted parathyroids and temporary improvement was seen.

Noel Patton has shown that guanidine or methyl guanidine produce symptoms of tetany. The authors consider tetany an intoxication with these compounds, the functions of the parathyroids being to destroy (probably oxidize) them. The good results following calcium treatment would be due to the fact that calcium salts give a precipitute with guardiane compounds. The intensity of the tetany would depend on the quantity of parathyroid tissue and quantifie compounds in the body.

This idea is also held by 2. Uhlenhulth (110) who states that the thynus glands secrete a tetany producing substance which is antagonized by the parathyroids. In parathyroidectomized animals this substance may reduce the colcium content of the organism. This also has a highly injurious effect on the central mervous system, resulting in permanent operandic muccular contraction and paralysis of almost the entire nervous system. The most important function of the parathyroids is to prevent tetany toxin from reaching the central nervous system by antagonizing it.

Use of calcium solts. Calcium solts have been found to be of great value in the cure of tetany.

Farmer and Klinge (20) in their experiment with parathyroidectomized cats had favorable results from feeding celcium salts.

Uhlenhuth (110) prevented the muscular contractions by introduction of colcium lactate into the body.

W. G. MacCallum and Carl Voegtlin (66) found that the

administration of celcium salts has an immediate and specific curative effect on parathyroid tetany. While much of the evidence pointed to a type of acid intoxication the removal of calcium must be one of the factors responsible for tetany.

Four grams deily of $C_{a}C_{2}$ or calcium lactate was found to decrease the symptoms of tetauy. (3)

Both calcium and magnesium have good influence in infantile tetany (93).

Howland, McKim and Marriott (46) showed celcium administration causes marked effect on the course of tetany. In a few hours the symptoms disappear. Calcium must be continued for a long time. C_aCL_2 administration per os causes an increase in calcium content of the serum coincident with dessation of symptoms although calcium of the serum does not usually return to normal.

Codliver oil and phosphorus was also fund to be of benefit in cases of tetany. Eight cases studied by Brown, MacLachland and Simpson were cured by administration of phosphorized cod-liver oil. The blood calcium was increased within a period of 10 to 17 days.

The results of neerly ell the investigators point to a distrubance of the calcium metabolism as a cause of tetany. The feeding of calcium salts seems to be of great benefit in telieving the animals from the symptoms, and suggests that raw rock phosphate and bone meel may be important in the ration as a preventative for tetany.

Ion antagonism and tetany. Several investigators have advanced the theory that tetany is due to an irritant effect

of certain ions. Loeb (62) experimenting with frags, found that when certain ions came in contact with herves a tetanous condition resulted, while other ions, if present, tended to counteract this effect. Sodium fluoride, disodium phosphate and phosphate, sodium oxelste, so ium citrate, sodium tartrate, and acid sodium carbonate, or the compounds of sodium whose enious form insoluble celeium compounds. The celeium ions have a soothing effect and counteract the sodium ions. When anions of salts of sodium mentioned above come in contact with celcium ions incoluble celcium compounds are formed and the sodium ions are left free to cause irritability. OH and H ions have a catelytic action and speed up the contraction of muscles due to sodium ions, and the tetanus of muscles in presecue of sodiums salts may be due to presence of OH in the solution. This action is counteracted by the constant production of $H_2CO_{\mathbf{x}}$ by the muscles which increase the solubility of the calcium selts. If OH ions are added this effect will be counteracted again. Alkalinity seems to aid sodium ions in producing irritability. The addition of CaCLy to a sodium citrate solution stopped the contact reaction.

A. Orgler (93) found that calcium and megnesium have a good influence on the symptoms of tetany. Potassium and phosphorus seem to increase the irritation.

Parhow and Dumitresco (08) found that celeium seems to exercise a sedative action on nerves.

Joseph and Maltzer (52) using caldium chloride were able to completely inhibit all irritability of nerves and

muscle; this inhibition was promptly reversable by subsequent use of sodium chloride.

Several authors believe that tetany is a condition in which the normal balance between calcium and magnesium on one hand and sodium and potascium on the other hand is disturbed so that the amount of calcium and magnesium becomes relatively smaller in proportion to the amount of sodium and potassium, which causes an increase in the irritability of the nerves system. (15), (34), (85)

Tisdele, Kramer and Howland found that the sodium and magnesium content of the serum of children suffering from active infantile tetany falls within limits of normal. The K content is somewhat elevated. The calcium markedly diminished below normal. The ratio (Na,K) : (Ca,Ng) is normal infants is 27.6 : 1, while in cases of active tetany it is 44.5 : 1. However, if calcium were to remain the same the ratio would be 27.8 The change in ratio is, therefore, due almost wholly to decrease in calcium concentration.

The results of these investigators point to the fact thar calcium and magnesium ions have a spothing effect on the nervous system while sodium and potassium ions produce irritability. Tetany would seem to be due to the presence of more sodium and potassium than calcium and magnesium in the body tissues. The feeding of mineral supplements high in calcium may correct this condition.

Calcium and Phosphorus Requirements

The literature concerning the ash requirements of dairy

cows is quite fragmentary and unsatisfactory. This makes it difficult to get any trustworthy estimates of the smaunt or kind of calcium and phosphorus compounds required in the feed of the dary mnimals.

The influence of function is so great that it is necessary to discuss this subject from a functional point of view.

Requirements for maintainence

Several factors may cause a change in the amount of ash necessary in the feed for meintaining the ash in the body. Armsby (4) states that "The effort of the body to maintain the osmotic pressure of its fluids by removing a surplus of some one ingredient may bring about an impoverishment as regards other elements and so create a need for a supply of the latter in the feed. The action of the kidneys in eliminating surplus salts and so preventing an increase in the osmotic pressure is not confined to the particular salt in excess but extends to others also." He adds, "Small amounts of some acids tend to escape oxidation in the body and to be excreted in the urine carrying a corresponding amount of base with them. Oxelic acid and its salts are oxidized with difficulty and tend to impoverish the body in calcium by the formation of insoluble calcium oxelete. This acid is liable to be especially injurious to young ruminants while in mature ruminants it seems to be largely destroyed in the first stomach". "Long continued maintainence on abnormal feed or under conditions favoring acid production in the body may result in extracting from the body comparatively large empunts of mineral matter even to the extent

apparently of bringing about pathological changes. These fluctuations of bone ash effect the ash as a vhole and the percent composition of the ash remains ab ut constant." "With rations containing a larger proportion of roughage there is no reason to fear losses either specifically of fixed bases or in general of total ash. Such would shoost always be the case with ordinary maintainance rations of cattle. As regards maintainance it seems clear that the ash requirement is a qualitative rather than a quantitative one. It is the proportions far more than the total amounts of ash that are important."

Effect of Lactation

The supply of calcium and phosphorus in milk must come either from the feed or from the body of the animal. If the skeleton is supplying these materials to the milk, the animal must be able to replace these elements from the ash of the food, otherwise pathological changes will take place and the animal may die. It is necessary, therefore, to have sufficient calcium and phosphorus in the feed to cover the supply excreted in the milk.

Effect of Pregnancy

According to Eckles and Trowbridge (42) the body of a new born calf weighing 80 pounds contains 2.8 pounds of mineral matter. Additional calcium and phosphorus should be furnished in the feed of the pregnant animal in order to meet the demands of the developing fetus. This ash material would necessarily have to come from the pregnant animal and should be supplied in the feed.

Amount of Calcium and chosphorus needed in feed.

It is hard to arrive at any definite figures as to the

amount of coldium and phosphorus necessory in the feed. The work of several authors throws some light on the subject but as yet there is nothing definite. The main difficulty arises from the fact that we do not know what percent of the ash in the feed is digestible.

Anger (2) showed that storage of phosphorus and caldium took place when large anounts were fed, d0 grams $C_{\rm E}O$ and 90 to 100 grams P_2O_5 being sufficient. The cows he worked with did not give over 15.48 Kg milk (about 32 pounds), daily.

Hart, McCollum, Humphrey (35) found that cows fed 190.5 grams of phosphorus daily had positive balances at different metabolism periods during three months time. Cows receiving only 46.7 grams per doy had negative balances.

Nordon, Hart and Patton (51) found a cow storing phosphorus when fed only 37 grams deily. This cow was giving 16.72 kilos deily (about 34 pounds) and weighed 966 pounds.

From work of Khuen (55) Forbes computed that in addition to the empunt of phosphorus in the milk a cow must receive more than .07 milligrams P_2O_5 per kilogram live weight. From Khuen's work it can also be figured that .007 grams C_gO per kilogram live weight above the amount in milk is needed.

A. R. Rose (97) concludes that the phosphorus requirements of a milk cow is the amount of phosphorus eliminated in the milk plus .026 grams of phosphorus per kilo of live weight or .06 grams $P_{\rm g}O_{\rm f}$ per kilo live weight.

Henneberg (41) determined the maintainence requirement of a steer for calcium and phosphorus per 100 kilogram live weight as 100 grams lime and 50 grams P_2O_5 .

Kellner (56) in computing the requirements of the milk cow adds to these figures three times the smoult of lime and phosphoric acid content of 20 kilograms of milk produced per 1000 kilograms live weight by milk cows and then obtained a total of 200 grams of lime and 140 grams of phosphoric acid as the requirements of milk cows per 1000 kilograms live weight.

It is Kellner's table that was used for figuring the celcium and phosphorus requirements of the cowe used in this experiment. It is as follows:

Calcium grams Phosphorus grams

For	maintainence 1000 pounds	32	10
For	production 20 pounds mill	x %9	15

This table gives only an approximation of the requirement, and is based on the assumption that a cow can assimilate only one third of the calcium and phosphorus in the feed. The truth of this assumption remains to be proven.

Sources of Calcium and Phosphorus for Dairy Cows

Celcium end phosphorus in neturel feeds

Many of our common feeds contain relatively large quantities of both calcium and phosphorus. The use of such feeds in the ration will largely eliminate the abnormal condition that very often occurs when a ration that is low in calcium or phosphorus or low in both is fed.

The following table (48) shows the calcium analysis of several of the common feed stuffs: (Table computed by Huffman from different analysis)

Feeds high in calcium -

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1	Tankage	3.342,5	64 pounds	in	8	ton
2	Cow pea hay	2.029,5	40 ⁿ	TT	11	17
3	Soy been hey	1.378%	27 "	17	11	17
4	Skim milk	1.376%	27 "	17	11	17
5	Clover hay	1.236,5	25 "	11	77	17
6	Alfalfa	1.120,5	23 "	11	11	11
7	Beet pulp	•729,5	15 "	11	11	TT
Feeds low in cal	lcium -					
l	Peal hominy	.005%	.10 "	11	11	17
2	Rice	•009 <i>,</i> 5	.18 "	11	17	11
3	Corn	•014%	•≈8 ^{II}	11	17	17
4	Wheat	•056,5	.112 "	11	17	n
5	Oat s	.112,5	2.24 "	Ħ	Ħ	11
6	Timothy	.192%	3.84 "	T	17	Ħ
7	Wheat straw	•2175	4.34 "	11	11	11

Wehave no roughages that are high in phosphorus. It is necessary to feed some concentrate if the ration is to be high in phosphorus content. The table (48) shows the phos phorus analysis of some common feeds. (Table by Huffman) Feeds high in phosphorus

1	Tankage 1.	4 79]5	29.6	pounds	per	ton
2	Cottonseed mesl	1.789%	35.6	17	11	17
3	Wheat bran	1.233%	24.0	17	17	11
4	Skim milk	.979%	18.6	17	17	17
5	Linseed oil meel	•789%	15.8	17	17	17
6	Soy beens	•64 9 %	13.0	Ħ	17	17
7	Cow pees	•532 ⁷ /2	10.6	11	17	17

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Feeds low in phosphorus -

1	Wheat staw	•009%	1.40	20 mae	per	ton
2	Beet pulp	078,5	•8	11	11	17
3	Corn stover	•102%	2.04	17	Ħ	17
4	Rice	•104%	2.1	17	11	17
5	Timothy	•1:23,0	2.5	n	17	17
6	Clover	.183/5	7.7	11	TT	17
7	Alfelfa	•238%	4 .7	17	17	17

Such feeds as corn, oats, wheat and gluten feed may be classed as medium in phosphorus.

As a rule if feeds high in the two elements are used in the rations, additional calcium and phosphorus in the form of mineral supplements is not needed. For high producing cows, however, additional minerals are a necessity.

Mineral supplements

Some of the most common sources of calcium are Raw rock phosphate which also contains phosphorus Acid phosphate which also contains phosphorus Bone meal which also contains phosphorus Lime stone rock, marl, chalk and wood ashes.

Raw rock phosphate, acid phosphat., limestone rock, and marl are so-called inorganic sources while bone meal is an organic source. Bone meal and raw rock phosphate are the two minerals that were compared in this experiment.

Bone meal and raw rock phosphate are made up to a large extent of the same chemical compound, tri-calcium phosphate $Ca_3(PO_4)_2$. However, there is a difference in the

physical structure of the two materials. Bone meal is classed as an organic substance while raw rock phosphate is classed as an inorganic substance.

The phosphate rock used in this experiment came from Tennessee fields. These deposits are the remains of very ancient micro-organic sea life. They are classed in the ordovician period. Forms of life were crustacea, and the phosphatic beas of rock are the leached residues of limestone beas formed at locations where these types of life were most abundent and which constituted a high percentage of the limestone beds. Later most of the calcium carbonate was leached out leaving the phosphatic residue.

The organisms making up these deposits became coated with silicions layers of a coral like structure, and it is to these encasements that we owe the existance of the calcium phosphate contents today; because if these encasements had not been impervious to acid waters, such contents would have leached away ages ago.

However, these same silicious walls today present a problem in the preparation of raw rock phosphate for both soil application and animal feeding. They must be broken down before the calcium phosphate contents are available. This silica and other impurities may cause a difference in the availability of the calcium and phosphorus of raw rock phosphate and of bone meal, as the calcium phosphate of bone meal is not encased in these silica walls.

The rock phosphate used in this experiment was a very finely ground rock from the Telmessee beds. It was the commerical raw rock phosphate "floats" and had been specially

prepared by the Thomson Phosphate Company of Chicago, Illinois.

The bone meal was obtained from Darling and Company of Chicago and was made from raw bones which were steamed to remove flesh and fats and to destroy any disease garms and then finely ground.

The raw rock phosphete contained 38.92% C_g0 and 26.7% P_2O_5 . Bone meal analysis was about 48% C_g0 and 35% P_2O_5 .

The supply of raw rock phosphate is practically unlimited. Bone meal, however, depends on the supply that can be obtained from the packing industry. The price of bone meal is just about twice that of raw rock phosphate. If the feeding values of the two were found to be nearly equal it would be much cheaper to use the raw rock phosphate.

Feeding Experiment with Bone Heal and Raw Rock Phos hate

No work has been done with dairy cattle comparing bone meal and raw rock phosphate as a source of calcium and phosphorus. However, experiments with other animals gives a general idea of their relative value.

Köhler (59) found precipitated calcium phosphate, bone ash, and steamed bone much all assimilable by lambs, the first preparation four times as completely as the last two. Hart, McCollum and Fuller (33) studied the role of inorganic phosphorus in nutrition of swine by feeding, slaughter, and balance experiments. They made the following conclusions:

l Animals fed a low protein, supplemented with inorganic phosphates, made as vigorous developments as others receiving their phosphorus supply wholly in organic form.

2 Precipitated calcium phosphete, a mixture of ai and tri calcium phosphates, gave no better results than did floats, a crude tri calcium phosphate.

Forbes and associates at the Ohio Experiment Station in a series of experiments with pgis compared different compounds of phosphorus and calcium. Theyfound (21) that bone meal when added to a ration which was low in calcium and phosphorus greatly increased the ash and the strength of bones, but did not increase the percentage of protein in the growth produced. This would indicate that bone meal cannot serve all the bodily needs for phosphorus. Some other compounds of this element are needed for muscle and tissue. Phytin from wheat bran caused a marked increase in muscle development.

A cereal ration (26) was fed to growing swine. This ration was supplemented by pulverized limestone, <u>bone four</u>, <u>raw rock phosphate floats</u>, and calcium carbonate, and the membolism with the different supplements studied. On the basal ration of corn, oil meal, wheat middlings and M_aCl there was a loss of celcium, a subnormal retention of magnesium and phosphorus, and a high acidity and NH₃ in the urine. The feeding of lime materially caused marked increase in calcium, magnesium, and phosphorus retention. The carbonate lowered urinary acidity and NH₃, while precipitated bone flour increased these products. Precipitated calcium carbonate and steemed bone produced relatively very dense and strong bones, while the raw rock phosphate produced bones that were only slightly denser and not quite as strong as when no supplement was fed. Precipitated bone was intermediste. Rock phosphete

produced no greater strength of skeleton in proportion to live weight than did a ration without supplement. Steemed cone, precipitated bone flour, pulverized limestone and $C_a CO_3$ caused marked increase in the strength of bone.

Forbes also determined the palatibility of mineral salts preparations to cattle (26). He found that steamed bone is more palatable to cows then is precipitated C_aCO_3 , precipitated bone phosphete, or rock phosphete. This whe preparation is also more palatable to heifers than was marl, pulverized limestone, or precipitated C_ECO_Z. Ordinary packer's steamed bone meel was more pelatable to cattle than special steamed bone meal, although the latter was taken freely. Hart, Steenback, Hoppart, Bethke and Humphrey (38) working with dairy cattle producing 20 to 40 pounds of milk found that the animals were in a negative balance on a ration composed of grains and their by-products, corn silege and timothy hey. Substituting alfelfa for timothy reduced the losses but did not make the balance positive. This was contrary to their former work, but in the earlier attempt the alfalfa had been cured under caps while the alfalfa used in this experiment had been cured in windrows with exposure to air and sunlight for four days. Addition of bone meel to the timothy hay ration did not result in positive balances or even equilibrium although the losses were reduced to some extent as compared with unsupplemented hay.

B. F. Kaupp (54) raised chicks from hatching, to eight weeks of age on a scratch ration of cracked corn, cracked wheat, and pinhead oats 50:33:17 and a basel mash of fish meal, wheat mids, pulverized oats, and corn meal 15:20:20:40. To every 95 pounds of mash fed to a part of the lots of chicks 4.5 pounds of

bone meal and 0.5 pound salt were added. In lots receiving minerals the bones were larger, birds stronger, average weight somewhat greater than those without mineral. Bone meal was evidently assimilable by the chicks, although the salt may have had something to do with the difference in the lots.

It would seem from the limited amount of vork that has been done that bone weal is superior to raw rock phosphate. This is true at least as far as growth is concerned. Perhaps, however, raw rock thosphate is equal to bone meal for milk production and may make just as good a supplement to the rations of milking cows.

Discussion of Review of Literature

Under normal conditions large quantities of calcium and phosphorus are needed in the feed of lactating dairy cows. Hart, McCollum and Hamphrey showed that a heavily lactating animal will draw these elements from the skeleton if the supply in the feed is below requirements. Forbes showed that the removal of calcium and phosphorus took place in spite of liberal amounts in the feed. Later studies at the Wisconsin Station indicated that the amount drawn from the skeleton depends to a large extent on the quality of roughage used. Forbes and associates and Heiss and Woodward found that cows store large quantities of both calcium and phosphorus during a period of low milk production or during the dry period.

These results suggest the use of a mineral suggement in the feed in order to supply sufficient calcium and phosphorus in the ration during heavy lactation and during periods in which the animal may store these elements in the skeleton.

If there is not sufficient supply of calcium and phosphorus in the ration a number of pathological changes may take place.

Ossebus cachexia or betenuelacia of domestic animels is essentially a disease of dictory orgin and is due principally to a deficiency of the food in bone forming material. The predisposing factors of the disease are prognamely, lactation, growth and starvation. Osteomalacia is a disease of the coues and is due to a removal of salts from the bone and their replacement by tiscue not having the normal content of bone salts. It is easily curable. Treatment consists of supplying additional colorum and phosphorus to the ration and suggests the use of a mineral sup lowent containing these elements.

Richets is a disease of young animals and is due to a lack of deposition of calcium and phosphorus. Several authors have shown that rickets may be developed by feeding rations low in calcium and others have shown that rickets can be produced by feeding rations low in phosphorus. McCollum points out the fact that there are two kinds of rickets, one characterized by normal phosphorus and low calcium of the blood and the other by normal calcium and low phosphorus. McCollum has also shown that the quantitative ratio between calcium and phosphorus in the diet is an important factor in the etiology of rickets.

Recentwork has pointed to an absence of a factor which has been called vitamin D and which aids in the assimilation of calcium as a cause for rickets. This factor is present in large quantities in cod-liver bil and to a small extent in

butterfat. It is also present in certain green plants. Treatment of rachitic petients with cod-liver oil has produced good results.

Sunlight and ultra violet ray neve the same effect on calcium assimilation and rickets as the antirachitic vitamin.

Rickets does not occur smong dairy cattle. Conditions similar to rickets, however, exist among dairy calved. Most authors believe that rickets is not present in the unborn fetus, but since clinically recognized symptoms appear very early in life, the first stage must occur in the earliest days of extra uterine life. The diet of the mother may have some effect on the development of rickets in the offspring and the ration of the pregnant milk dow should contain sufficient calcium and phosphorus.

Teteny is another disease that may be due to a disturbed calcium metabolism. Low calcium of the blood and usually negative calcium balances accompany tetany. A review of the literature shows that tetany can be cured by feeding or injecting calcium salts. The work of several investigators show that tetany is due to an increased proportion of initating ions in the tissue over the ions that suppress irritability. Sodium and potassium ions stimulate, while calcium and magneaium ions repress or sooth. Feeding minaral supplements high in calcium would have a curative effect on tetany.

There is very little to be found in the literature regarding the calcium and phosphorus requirements of dairy cows. Small amounts of each are needed for maintainence because these elements are withdrawn with others during the ordinary

life processes. The large quantities of both in milk and in the fatus make the requirment much greater during lactation and pregnancy. No definite conclusion can be drawn as to the exact requirements.

Many of our common feeds contain large quantities of both calcium and phosphorus. In general, calcium is found in largest quantities in roughages and especially legumes. Most of the common g ains are low in calcium. Phosphorusis found in larger quantities in the concentrates especially cottonseed meal, wheat bran, skimmilk, and linseed oil meal. All of the roughages are low in phosphorus.

Among the common mineral supplements the following contain both celcium and phosphorus, raw rock phosphate, acid phosphate, and bone meal. Those containing celcium alone are limestone rock, marl, chalk, and wood ashes. Bone meal is a so-celled organic source while the others are inorganic sources.

Most of the work comparing mineral supplements has been done with animals other than dairy cows. The review of literature would indicate that bone meal is more assimilable and as far as growth is concerned superior to raw rock phosphate. It is at least more palatable.

No work has been done with dairy cattle directly comparing bone meel and raw rock phosphate and this experiment was started in order to determine the relative availability of the calcium and phosphorus of tone meal and raw rock phosphate when fed to lactating dairy cows.

OBJECT OF THE ALDERIGENT

The object of the experiment is to determine the relative value of raw rock phosphate and one meal as sources of calcium and phosphorus when used as supplements to the ration of dairy cattle.

The availability of the calcium and phosphorus from bone meal and from raw rock phosphate will be compared through balance experiments on lactating cows. At the same time the palatability of the two supplements will be noted.

In addition, the effect of adding calcium and phosphorus to the feed in the form of bone meal and raw rock phosphate will be noted on the amount of the two elements found in the blood, on the percentage fat in mill, and on the hydrogen ion concentration of the faces.

PIAN OF THE LEPERTIES

It is planned to use three lactating cows for the experiment. The animals selected will be high producers and will show dairy temperament. If possible, animals that have recently freshoned and are giving more than forty bounds of milk a day will be used.

The experiment will be divided into four periods of twenty days each. A basal ration as low in calcium and phosphorus as practical will be fed during the first period. Raw rock phosphate will be added to the ration to the extent of three per cent by weight of the grain ration in the second period. During the third period the animals will again be placed on the basal ration without any mineral supplement. While in the last period three per cent by weight of the grain ration will consist of bone meal.

During the last five days of each period balance experiments will be conducted on the animals. The feeds and water and the urine, milk and feees will be analyzed for calcium, phosphorus, and mitrogen and the balances of these elements for all of the five day period will be determined. The first and third puriods where no mineral supplement is fed will be check periods. The retention of the three elements by the animals during the bone meal period will be compared with the retention during the raw rock phosphate period and the retention during these periods will be com**pared** with that during the two check periods. From these belance tests the relative availability of the calcium and phosphorus of bone meal and raw rock phos hate will be determined. In addition the effect of these supplements on the retention of nitrogen will be noted.

The palatability of raw rock phosphate and bone neal will be apparent from the ray the animals eat the grain mixtures containing these two compounds.

The animals will be bled at the end of each metabolism period and the blood analyzed for calcium and phosphorus so as to determine the effect of adding mineral supplements upon the percent of these elements in the blood.

Composite samples of milk will be taken deily throughout the experiment and will be tested for butterfat by the Babcock method. This will show the effect, if any, of bone meal and raw rock phosphate on the percents of butterfat in milk.

During each metalolism period, portions of fresh faces will be used for the hydrogen ion determination in order to determine the effect of feeding bone meal and raw rock phosphate on the hydrogen ion concentration of the faces.

The appearance and health of the animals will be carefully noted each day during the experiment. They will be weighed daily at four A. M. because the digestive tract is probably more nearly empty at this time than at any other time during the day and the daily weights will therefore be more uniform.

All records will be kept in permanent record books.

ILLIND OF LARLERILLICATION

Peediug

Three lactating covs were selected for the experiment which was divided into four periods.

During the first period of 28 days the animals were fed the basel ration which consisted of timothy hey, corn silage, and grain. This ration was low in both phosphorus and calcium. Collection of urine, faces, and milk were made during the last five days of this period and these, together with the feeds, were analyzed for nitrogen, calcium, and phosphorus to determine the amounts of these elements utilized.

Raw-rock phosphate was saded to the basal ration during the second period which covered 23 days. Balances were determined during the last five days of this period.

The basal ration alone was fed the following period of 15 days. This period had two purposes. One was to check up on the first period, and the other was to give the animals a chance to recover from the effect of the raw rock phosphate feeding. A five day balance was taken as usual.

Bone meal was added to the basal ration for the fourth period which was continued for 20 days. The nitrogen, phosphorus, and calcium balances were determined for the last five days.

From the balances for these different periods the relative availability of raw rock phosphete and bone meal was determined.

Selection of Aminels

The animals chosen were as follows: Cows No. 33, a pure-bred Holstein-Priesian, five years of age, weighing 1,539 pounds at the beginning of the experiment. At the time this enimel was placed in the metabolism stall, she was giving about 65 munds of z.E. milk deily and had been fresh two months. This cow had a seven day record at four years, twenty-two days of 498.8 nounds milk, 26.28 butter. She had freshened normally and scened to be in extremely good condition. Cow No. 71, a pare-bred Jersey, senior three year old, weighed 809 pounds and was giving 45 pounds of 5, milk daily at the beginning of the experiment and had been fresh one month. She had a two year old Class AA record of 526 pounds fat. This dow had freshened normally and was in good condition. She showed excellent milking qualities. Cow No. 70 was a pure-bred Jersey, four and onehalf years of age, weighing 1,079 pounds when placed on experiment. She was giving about 23 pounds of 5% milk daily at the beginning of the experiment and had been fresh a little over one month. The was fat and rounded in sonegrance. showing a tendency towards beefiness. Her yearly record was only 220 pounds fat as a three year old. The first two cows showed an inherent tendency towards high production. No. 70, however, was clearly a low producing enimel.

<u>Equipment</u>

The metabolism stalls used were a modification of

these described by Forbes on page 18, bulletin 763, Obs Experiment Station. The cows were kept in structions, but individual mangers were specially built to correspond to those shown on pages 12 - 18 of the above Bulletin. The mangers were ab at 4-1/2 feet high. The frants of the mangers were removable as as to allow feeting and vatering. Hinged gates, made to fit chose to the animal's means, could be swing back from the side of the magers while the animals were eating the thus prevent them from throwing any of the feed out of the manger. These gates were swing forward out of the animal's way then they were not feeding. The animals were bedded with shavings except for a few days before and a during the metabolism periods, at which time couves matrices were used.

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An accurate gran belance expable of weighing up to 29,000 grams was used for weighing feeds, urine and feeds. The milk was weighed on a spring balance, (graduated so as to weigh accurately to a tenth of a pound).

The faces was collected on large scoop shouls and immediately transferred to galvanized bushel baskets which were kept covered at all times. The wrine was collected in pails to which long upoden handles were attached. The wrine was intediately transferred to covered milk cans. Deparate pails, should, cans and baskets were used for the individual cows. The ordinary covered top milk pail, weighed, was used to collect the milk.

Care, Feeding and Watering

Cow No. 33 was milked four times a dey throughout the

experiment. The other two were milked three times a day. Grain was fed at the time of milking. Silage was fed to all three animals twice a day, at 4:70 A.H. and 3:70 P.H. Hay was also fed twice a day, at 6A. H. and 5 P. H.. The cows were watered from pails inhediately after they had eaten their grain.

The cows were kept in metabolism stalls during the entire experiment but every day about 10 o'clock were allowed in a dry lot to exercise except during the metabolism experiment.

Ground timothy hey was fed. The grinding was done mainly to insure a better sample for analysis. It was found, however, that grinding made the hey more palatable.

The corn silage fed had been held over from the previous year and was strongly acid.

Cow No. 33 received the following grain mixture from February nineteenth to March twenty-fifth:

1 00	o orang a	homing feed		
100	17	Distiller's	grains	(corn)
100	17	gluten mesl		
100	**	oil meel		
		l, selt		

About the twenty-first of Earch Ko. 33 refused to est the grain mixture and on the trenty-fifth it was found necessary to change her ration so as to make it more palatable. The new ration was used from the twenty-fifth of Earch until the end of the experiment. It was made up as follows:

200	Domig a	gluten feed
612	17	hominy feed
200	11	bran
ಜರಿರಿ	11	oats
412	17	Diamona glaten
462	17	oil meal
512	11	Distiller's grains
		l% selt

After the change to the above ration there was no further trouble as to palatability.

Cows Nos. 70 and 71 received the following ration:

200 pounds hominy feed

- 100 " Distiller's grains
- 100 "gluten meal
- 100 " oil meal

There was no change in their ration throughout the experiment. The feeds were all mixed ten times to insure uniformity and to eliminate as far as possible error in sampling.

Collection of Experimental Data

Weighing

The animals were weighed daily at four A. M. They were weighed at this time so as to have the digestive tract as empty as possible and therefore the weights more uniform. It was found that the weights varied very little from day to day. The daily weights were placed in a permanent record book immediately.

Records of Feed

Records of the feeds given to the animals were kept in

a permanent record book and also on cards at the barn where the feeds were weighed out. These cards were kept in sight at all times so as to prevent any error in weighing out the feeds.

Sampling and enelysis of feeds

The feeds for each period were all prepared in advance. The timothy hay was ground and placed in burlap sacks in a room separate from other feeds. Samples of the hay were taken from the top, center, and bottom of each suck and placed in air-tight jars for analysis. The silage was sampled at each feeding and the samples placed in a large air-tight jar for analysis. The grain was all mixed before each feeding period and stored in burlap sacks in a separate feed room. A grain sampler was used to take several samples from each bag. These samples were also kept in air-tight jars.

Before ta king samples for finel analysis from the composites collected, the feeds were all finely gound, (ground until they would go through a 30 mesh sieve) the silage having been first dried. The large samples were then very thoroughly mixed and quartered. Two quarters were discarded. The remainder was mixed again and quartered. This process was continued until the quantity desired was reached. In this way it was possible to get a very representative sample of the lot.

The method used for analysis was as follows:

From the sample of feed ten grams were weighed out in duplicate. The ten gram sample was then ashed, treated with hydrochloric acid and filtered. The filtrate was made up to 250 cc. volume. 59 cc. was then taken for analysis of calcium and 25 cc. for analysis of phosphorus. The calcium was precipitated as oxelate and titrated with permangarate solution. The phosphorus was precipitated with annohium molybdate solution. This was dissolved in sodium hydroxide and titrated back with nitric acid.

The nitrogen of the feeds was determined from a weighed portion by the Hjeldel method.

Samples of water were taken at each watering pariod. The water was analyzed for calcium and pho-phorus. The water was evaporated and the ash treated as in the preceding analysis. Collection and Analysis of Urine and Paces

During the metabolism period, one attendent was present at all times. The wrine was collected by means of long handled pails and immediately placed in covered milk cans. The urine was weighed daily and thoroughly mixed by pouring back and forth. A sample was then taken. The samples were taken in duplicate and kept in glass stoppered bottles. Formaldehyde (3 drops) was used as a presevative. All the metabolism experiment started at four A. M. and samples were taken about 4:30 A. M. each day. The samples were analyzed for nitrogen during the same morning so as to guard against any loss.

The feces were collected in large scoop shouls and i undistely duced in gelvenized weighed bushel bashets which were kept covered. The faces were weighed every morning and the mixed. A sample of 2,005 grass was taken. This sample was analyzed dealy for mitrogen and then placed in drying racks. After drying, the faces were finally ground and representative sample obtained by the quartering as thos used in case of feed.

In the determination of nitrogen of urine 3 cc of the sample was taken. The Kheldahl method was used.

For calcium 50 cc of urine was used. Three cc. of nitric acid was added and the solut on evaporated. The residue was ashed, treated with hydrochloric acid and filtered. It was then precipitated with exalate and titrated with permangenate solution.

Twenty-five cc of urine was taken for the phosphorus analysis. Three cc of magnesium nitrate was added and the solution evaporated. The residue was treated with nitric acid and amnonium molhyodate solution. The precipitate was dissolved in sodium hydroxide and titrated back with nitric acid. The feces were analyzed in the same manner as the feeds.

Collection and Analysis of Milk

Milk was collected in weighed milk pails. The milk was weighed on spring scales whichwere graduated to twentieth of a pound. The weight in grams was figured from this weight. The milk was poured back and forth seve al times to make certain of an even mixture. Composite samples were taken in triplicate form. One sample was used for the determination of butter fat present, and the other two were used for analysis of nitrogen, calcium, and phosphorus. The samples were kept in glass stoppered bottles and formaldehyde was used as a preservative.

The Babcock method was used to determine the percentage of butterfat in the milk.

Nitrogen was determined by the Kheldahl method. Five cc of milk was taken for this purpose.

For calcium determination 25 cc of milk was used. The method was the same as that used for the determination of

celcium in urine.

For the phosphorus determination 10 cc of mild, and 2 cc of magnesium nitrate solution was used. Otherwise the method was the same as described for wrine.

Blood Dicture

The effect of the feeding of bone meal and raw rock phosphate on the percent of calcium and inorganic phosphorus in the blood was determined. The cows were all bled at the end of the first and at the end of the second period. They were bled about the hiddle and at the end of the third and fourth periods. In order to get further data on the effect of the raw rock phosphate on the percent of calcium antiphosphorus in the blood, the basal ration was fed for a fifth period of twenty-three days. They were bled three times during this period. Then raw rock phosphate was added to the ration and after seven days they were bled again.

The following method was used for blood analysis: Inorganic phosphate, Briggs modification of Bell-Doisy method. Calcium, precipitated as calcium oxalate and titrated with permanganate colution.

Hydrogen Ion Concentration of the Feces

During each five day metabolism period samples of feces were taken and the hydrogen ion concentration determined.

The electrometric method was used for this analysis.

EADLAILANIAL DATA

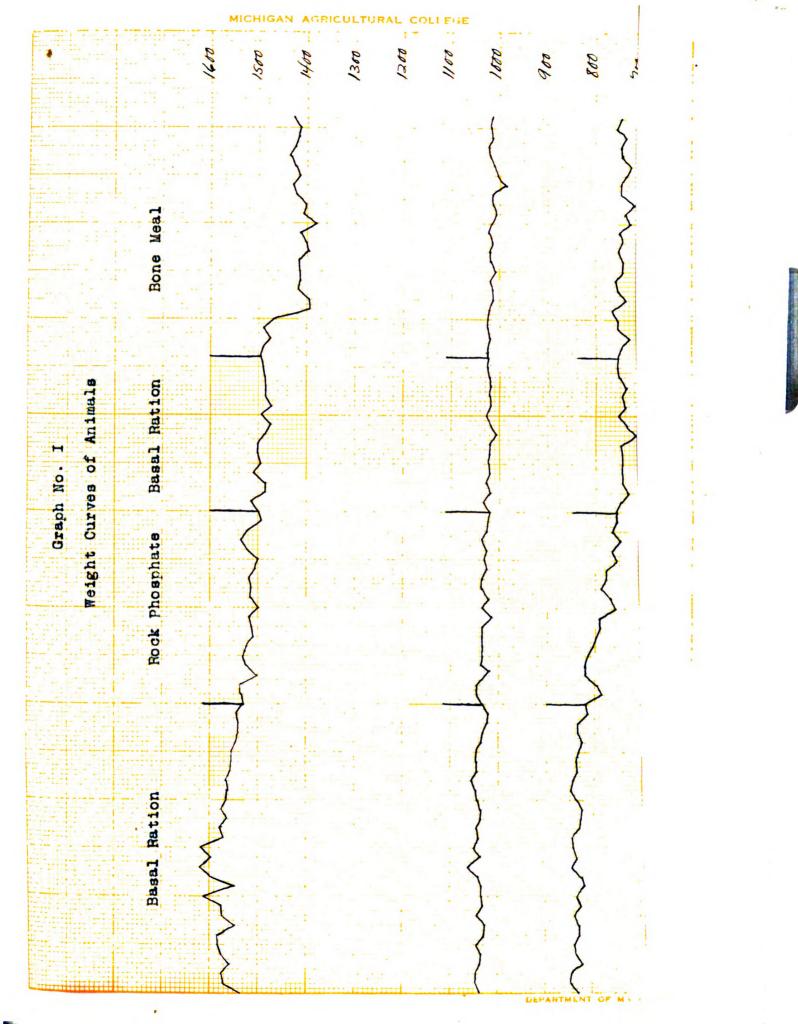
Palatability of Raw Rock Phosphate and Bone Meal

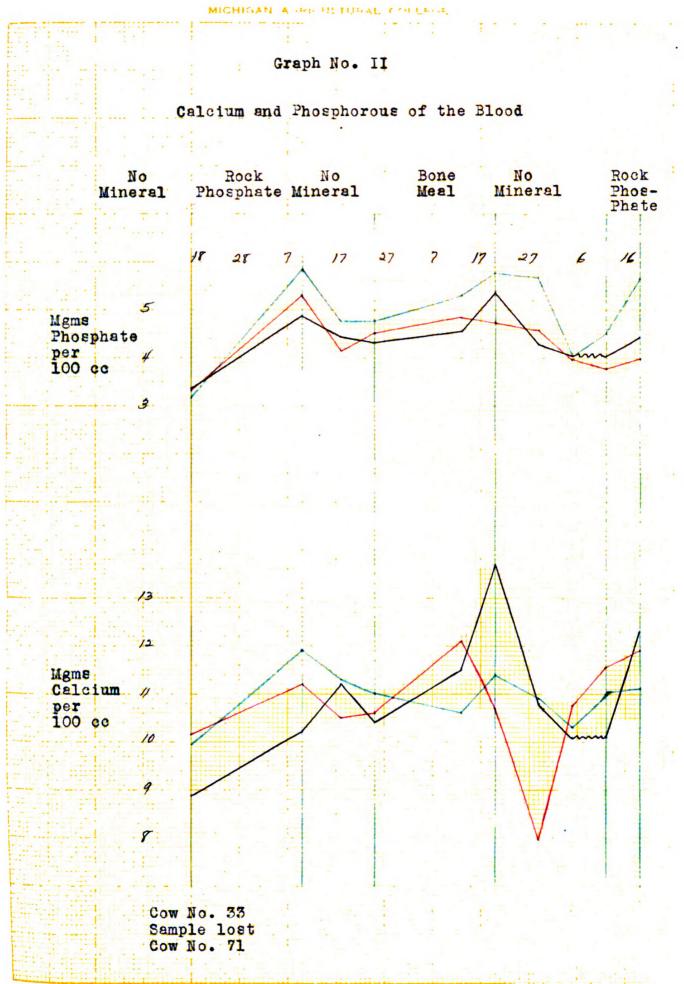
Both No. 33 and 71 refused to eat raw rock phosphate when it was mixed with grain, and ate only sparingly when it was fed on silage. No. 70 ate grain containing raw rock phosphate, but her appetite was poor and she played withher food for a long time before eating.

When bone meal was fed all three snimals ate heartily.

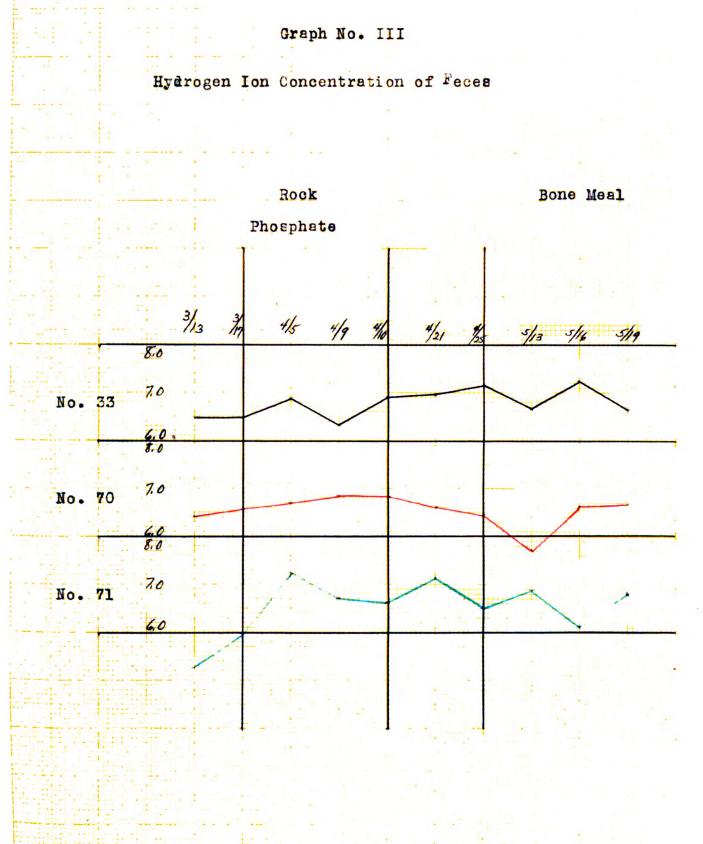
Appearance of Aminals During Experiment

At the beginning of the experiment all three animals were in excellent condition. They had every appearance of good health, were well fleshed, the hair was smooth and glossy, and their eyes were bright. At the end of the experiment there was a marked change in No. 37 and No. 71. No. 70, except for a little loss of flesh, looked as good as at the beginning. Both 37 and 71 had lost a large amount of flesh, they appeared geunt, the hair was coarse and lusterless, and they were very irritable.





DEPARTMENT OF MATHEMAT



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Hydrogen Ion Concentration of Feces

	Cow No. 23	Cow No. 70	Cow No. 71
Dat e	$\overline{0}$ H	рH	<u>nH</u>
Besal Ration Elone			· · · · · · · · · · · · · · · · · · ·
March 13, 1934	û•48	6.42	5.27
March 17, 1924	ů ₀ 49	6 .54	5.98
B asal ration and raw	rock phosphate	3	
April 5, 1924	ů € 89	ΰ .7 0	7.31
April 9, 1924	6.29	6.84	∂.71
April 10, 1924	ċ.91	6.82	6.65
Basal ration alone			
April 21, 1924	6.98	ü•60	7.12
April 25, 1924	7.18	û .45	6 .53
Basal ration and bon	e me al		
Ma 13, 1924	6.70	5.76	6 .88
May 16, 1924	7.27	6.6 1	6.18
May 19, 1924	6.68	6.68	6.82

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Talle No. II

	Cow N	o. 33
	jo Calcium	[*] ∕ ₽205
First Period	.1 239	.190
Second Period	.1 3⋶7	•20 6
Third Period	• 1 066	.157
Fourth Period	.1974	. 2403
	Cow N	o . 70
	, Celcium	% ≥ ₂ 0 ₅
First Period	•>046	.2576
Second Period	•20 1	•2488
Third Period	.199	.2370
Fourth Period	.179	. 228
	Cow N	o. 71
	5 Caleium	% P205
First Period	.193	•26 7
Second Period	•184	. 2398
Third Period	.176	•223
Fourth Period	.126	.186

Average Percent Calcium and P205 in Milk

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Table No. III

Blood Data

	Date	Aniasl Number	Phosphorus nys. per 100 c c whole blood	Celcium Myr. per 100 c.c. Thile blood
Basal ration clone	5/18	33	7.5	83.8
Basal ration and Raw rock phosphate	4/10		4.88	10.2
Basel ration clone	4/18		4.43	11.2
	4/25		4.71	10.4
Basel ration and Bone Meal	5/13		4.56	11.5
	5/20		5.78	17.7
Basal ration alone	5/29		4.28	10.77
	6/5		4.03	10.10
	6/12		sample lost	
Basal ration and Raw rock phosphete	6/19		4.43	13.3
Basal ration clone	7/18	70	₹.00	10.15
Basel ration and Raw rock phosphate	4/10		5.70	11.2
	4/18		4.10	10.5
	4/25		4.50	10.6
Basal ration and bone meal	5,⁄13		4 • 8 4	12.1
	5/20)	4.73	10.7
Basal ration clone	5/2 9		4 . 5 9	7.95
	∂∕5		3• 98	10.75
Bescl ration and raw Rock phosphate	6/12 0/19		3.79 3.98	11.55 11.90

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Table	1 3	T T T	4.551	4 -
Tarta	_ ∎∎ ∪ ●		90.a.	· •

	Date	Animel Number	Phosphurus most for 100 c c vhole blood	1038. 66 r 103 c c
Basal ration alone	7/18	71	≈.1 4	9.95
Basal ration and Raw Roch Phosphate	4/10		5.84	11.9
	4 / 18		4.77	11.3
	4/25		4.77	11.0
Basal ration and Bone Meal	5/13		5.30	10.6
	5/20		5 . 79	11.4
Basal ration clone	5/9		5.08	10.9
	c/5		4.03	10.3
	0/12		4.00	11.02
Basal ration and Raw Rock Phos Late	J /19		5.03	11.1

Table No. IV

Effect of food of Bone Meal & Rev Rock

PhonyAute on Parcent fat.

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		: Tat : : Pormas:	ý . :	Average Daily	:	Average Daily	:
No. 33	•		:	Lilk Pound e	:	Fat Dounds	•
	•	• •	•		•		<u> </u>
1st. Period	1024.4	:40.3777	2.855:	5e .01	:	1.0502	:
2nd Period	879 . 9	:26.6958	5.084:	₹8 . 20	:	1.1607	:
3rd Period	452.5	:12.7057	2.608:	20 .17	:	.8470	:
4th Poriod	40⊕.0	:12.1441	L.C17:	18.56	:	. 48 58	:
No. 70				angegen og en en de sen de			
lst. Period	597.4	:32.1572	5.385:	21.34	:	1.1485	:
2nd Period	435.8	:22.0759	5.00 5:	18.95	:	•9ë99	:
3rd Period	278 . 3	:13.5758	4.804:	18.55	:	. 90∷4	:
4th Period	454.2	:22.1705	4.370:	18,188	:	.8855	:
No. 71	******	****				8-12-19- <u> 2-</u>	
lst. Period	1125.2	:59.6372	5.299:	40.19	:	2.1294	:
2nd Period	528.2	:29.3065	5.548:	22.96	:	1.2742	:
5rd Period	783.1	:19.6893	5.139:	25.54	:	1.71:26	:
4th Period	580.8	:30.5489	5.200:	23.272	•:	1.2220	:

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TABLE NO. V. CaO Grums

No. 33		Intake	k6		Gruns		0	Outgo		
Date	Silage	Grain	Нау	Tater	Tctal	Milk	Urine	F - C 0 B -	Total	Bulance
3/13/24	21.17	14.06	30.12	20.2	74.42	31.24	61.4	77.44	80 • 50	- 0.03
3/14	21.12	14.06	30.12	9.36	74.71	33•01	たった	42.10	25.35	-10.64
3/15	21.12	14.06	30.12	C . 5	74.60	59.50	50°3	45.08	10.13	- 6.41
3/16	21.13	1 4.06	30.12	10.25	75.60	11. 6	4.68	16.75	83.11	- 7.51
3/17	21.12	14.06	30.12	10.25	75.60	32.15	5.25	42.73	22.22	- 4.14
	105.85	70.50	150.50	48.18	374.93	157.98	25.29	226.43	02.204	-74.77
Raw Roc	Rock Phosphate	ate							Алетаее -	- 6.55
4/2/5H	93 . 84	19.61	22.53	5.65	140.63	24.13	07 . 4	54.72	127.55	17.08
1/6	98.60	15.40	06.45	6.95	145.85	23.94	5.66	91.62	121.28	23.45
4/7	97.25	13.36	21.21	7.24	139.69	21.58	4•65	73.64	99.67	40.02
4/8	96.12	13.93	22.24	10.78	143.12	52.35	4.31	109.59	137.25	5.87
6/†	105.23	15.61	52.25	8.13	151.13	23.11	5.85	116.85	145.81	5.32
	70.194	73.96	113.64	41.75	720.42	115.91	25.17	486.48	627.56	92•86

Average 18.57

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TAPLE NO. V

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Balance ັງ**.** ຮູຮູ 2.28 2.68 29.31 8.00 00 1.47 27.00 1.16 30.61 129.49 34.71 36.41 66.56 AVETEGE 5.86 93.66 88.45 87.75 316.35 65.01 60.92 05.004 62 • 78 61.31 Tctal 513.42 27.SIt 107.52 76.51 82.01 73.26 4C • 79 40.03 40.00 13.54 40·77 70.17 Feces Outgo и. 50 1. 1. 20 1. 1. 20 1. 1. 20 1. 1. 20 1. 1. 20 1. 6.10 25.37 6 H 6 6 H 6 1.58 12.53 Urine 1.10 15.63 15.7*2* 16.42 77.79 15.97 14.05 **M11k** 12.57 12.17 12.47 13.61 13.58 65 . 60 69.50 12.63 345.63 68.66 68.84 69.48 125.04 124.16 123.87 127.16 124.16 620.39 Total Grams CaO 7.54 5.89 7.07 7.71 7.71 7.83 7.83 25.79 5.5 Teter 1. . 24 24.10 32.08 32.03 32.03 24.10 24.10 120.50 32.98 24.10 24.10 32.98 164.90 НЕУ 11. 58 11.58 11.58 11.58 57.90 74.07 74.07 74.07 11.58 74.07 35.35 Intake 7:+.07 Grain 11.87 11.57 11.87 26.09 26.09 26.03 130.45 26.09 26.09 11.87 59.35 Silage 11.37 Bone Meal 5/15/24 42/02/4 No. 33 ħ2/ħ 5/16 4/23 5/18 5/19 Date 12/1 4/22 5/17

Average 25.90

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No. 70		Intake		Grana Grana	0		Ó	Outgo			
Date	Silage	Grain	Y.	Water	Total	12118	Urine	Fecea	Total	Balance	1
3/13/24	14.11	9°7.9	17.57	3.36	14. ta	20.19	1 .58	60.00	42.36	2.00	
3/14	1,4.11	9. T	17.57	01 61 01	8+++	13.53	1.53	11.13	42.20	63 63 63	
3/15	14.11	хо М С	17.57	2.77	43.83	17.13	1.20	23.93	42.31	1.52	
7/16	14.11	9.38	17.57	3.12	44.13	27.73	1.07	23.67	0. 10 1	- 2.34	
7/17	14.11	9.73	17.57	11 •=	43.53	19.25	1.72	22.85	43.43	0.7	
	70.55	145.30	87.85	15.44	220 • 74	97.93	6.70	112.19	216.82	3.52	
Raw Rock	<u>р</u> .	Ð						Average	3e •73		
4/5/54	77.11		13.88	3.71	77.23	16.76	1.26	ло. 60	17.47	0 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1	
t;/6	77.11	1+7 • E7	15.53	3.12	78.44	16.61	1.08	14.95	77.10	1.34	
۲/۲	77.11	47.87	15.68	94 10	73.26	16.94	1.11	64.71	82.76	0ú• tr	
4/8	77.11	47.87	14.36	3.24	12 · 5H	16.70	1.92	53.09	77.61	37	
5/1;	22.11	47.87	13.09	3.59	76.32	01.71	2.18	59.81	29.09	- 2.77	
	58.85	239•35	69.22	16.60	327.49	£4.11	7.55	299.61	391.27	- 3.78	02-1
								Average	se756		J

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Balance - .07 - 65 .18 3.53 2.78 5.77 11.78 10.40 6.80 6.41 13.14 48.53 36.18 39.52 41.60 42.24 41.53 68.52 69.84 72.97 74.13 65.63 352.09 203.07 Total verage 1.15 23.00 44.70r 19.16 21.06 48.83 258.30 22.70 21.52 51 • 29 54 • 71 55 • 48 42.94 Fe ces Outgo 1.96 2.12 21.15 2.41 2.25 2.41 2.31 2.25 2.46 2.17 12.40 Urine 7.21 16.35 17.13 17.60 17.60 17.06 16.34 17.33 16.50 16.09 16.19 15.48 S 1.39 MIIK 77.67 41.53 41.59 41.71 42.30 80.30 80.24 Tctal 208.84 400.62 80.54 17.91 Grama CAO 3.18 3.24 3.36 3.36 3.95 17.09 3.59 3.53 3.06 Tater 3.83 3.06 17.07 14.46 14.46 14.46 14.46 14.46 72.30 19.75 19.79 19.79 19.79 19.79 98.95 Hay Grain 6.49 6.49 6.49 6.49 32.45 45.05 45.05 45.05 45.05 225.25 Intake 87.00 17.440 04.71 04.71 04.71 17.440 Silage 11.87 11.87 11.87 11.87 59.35 11.37 Bone Meal 5/15/24 4/20/5h No. 70 5/16 /23 ₩2/t 5/18 -/22 5/19 Date 4/21 5/17

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Average

Balance

- 5.35

- 7.07

- .39 - 2.14

- 9.95

- 4.05 1.40

6.96

16.5-

28.14

85.29 57.33 55.21 56.63 64.72 296.65 64 • 51 62.76 - 1.99 66.93 85.78 Total 73.02 375-53 Average 19.61 Outgo 63.48 46.45 443.53 65.80 52.62 92.711 28.64 20.62 22.18 271.83 26.54 Feces 1.36 4.60 1.50 . 000 •79 6 Urine .61 24. . 23 2.75 2. **6**6**•** 53 35.62 35.47 36.24 34.04 33.09 174.46 21.42 20.25 20.28 19.08 100.90 **M11k** 19.87 57.65 56.54 57.35 286.70 81.24 68.35 52.35 57.41 92.95 Total 47.56 405.39 11.02 Grams g 2.5**9** 5.24 5.95 5.65 5.65 3.89 2.77 3.30 2.77 28.20 Tater 5.71 15.32 22.59 22.59 22.59 15.68 15.68 22.59 112.95 15.68 22.59 15.31 15.68 78.03 Huy Intake 15.00 15.CO 15.00 15.00 15.00 6.40 5.70 6.33 00.27 Crain 4.54 30.67 Raw Rock Phosphate S114g9 14.11 14.11 14.11 14.11 56.57 42.08 70.55 67.80 14.11 67.80 47.12 281.37 3/16 3/13/24 4/2/5A No. 71 **+**τ/ź 3/15 3/17 Date 4/6 t /2 **1**/3 6/+

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29.86

Average 5.97

CBO

Balance - 4.07 - 1.85 1.02 1.02 - 3.46 11.46 9.15 -20.26 2.65 17.15 2.77 70.43 98.72 75.40 61.67 75.15 44.10 40.99 38.89 39.71 38.72 202.48 Average - .59 52.32 70.443 20.50 92.72 55.58 75.40 47.13 61.67 Tctal 381.38 Out go 19.32 <u>17.42</u> 96.79 54.34 11.91 19.04 289.67 21.90 Feces • 56 3.05 • 65 ·¹⁵ 57 000 40. - [‡] -5.17 .76 ର ଅନ୍ତି <u>с</u> Uri ne 102.64 21.32 21.23 19.40 19.89 20.50 17.55 17.58 19.02 14.13 MITK 20.06 63.34 79.55 78.45 73.05 78.82 40.03 39.91 39.91 40.03 199.02 Total 77.93 392.84 Grams 15.97 2. tr 2. tr 3. tr 4. tr 4. tr 5. 53 7. tr 5. tr 7. н. 06 1.94 1.33 2.41 15.24 Water 14.46 14.46 14.46 14.46 14.46 14.46 14.46 27.91 27.91 27.91 27.91 93.95 19.79 Hay Intake Grain 32.45 45.05 45.05 45.05 45.05 45.05 225.25 73.30 Bone Meal 15.66 15.66 15.66 15.66 15.66 10.68 10.68 10.68 10.63 10.58 53.40 Silage 5/15/24 120/24 No. 71 4/2H 2/1/5 5/16 5/18 12/1 t/22 +/23 Date 5/19

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Average

P205

10. 33		Inteke		Grane Grane	τ μ		Outgo	ට හු	
Date	Ω.	Gráin	Нау	Totel	XL1X	Urine	т Ресев	Tctal	Balance
3/13/24	11.71	62.06	19.19	127.09	47.76	1.89	72.29	122.54	4.55
3/14		62 • 06	19.19	127 . 09	64.64	±• 65	84•33	158.47	-11.38
3/15	11.71	62.06	19.19	127.09	14.96	6.18	92.15	1,13.29	-16.20
3/16	17.11	62.06	19.19	127.09	0t,•2t	3.21	16.46	146.52	-19.43
3/17	17.11	90.79	19.19	127.09	45.38	2.93	27.92	141.23	-14.14
	65.55	453.95	95.55	635.45	235.99	18.18	436.20	692.05	-56.60
Raw Rock	k Phosphate	9					7	Average -	22.II
4/5/54	65.45	81.72	12.51	159.98	34.67	23.17	46.79	155.78	4-20
4/6	65.45	80.6 1	14.15	160.21	36.71	20.33	88.75	145.79	24.42
L/1	65•45	69 .91	12.40	147.76	. 35.36	23.39	60.49	123.84	23.92
;t/8	65.45	73.17	12.64	151.26	35.28	21.11	100.02	156.41	- 5.15
6/1	65.45	81.72	12.60	159.77	35.83	25.59	104.41	166.83	- 7.06
	327.25	387.13	64.60	778.98	178.85 1	114.59	455.21	748.65	30.33
								Average 6	6.066

P2 05 Grame

No. 33

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((•0N		Intake	Ø		Grume		Outgo		
Date	Silage	Grain	Hay	Total	71 I K	Urine	Feces	Total	Ealance
4/20/2H	19.38	65.23	18.12	102.73	46.13	7.81	61.04	90.79	11.94
1 2/†	19.38	65.23	13.12	102.73	23.01	ó. ĉ5	63.50	93.36	72.6
4/22	19.38	65.23	18.12	102.73	24 . 33	С. С.	69.57	24.02	3.71
t/23	19.38	65.23	18.12	102.73	23.59	5.20	66. 38	95.17	7.56
4/54	19.33	65.23	18.12	102.73	19.50	3.16	67.04	60.05	12.73
	96.90	326.15	30 • 6 <mark>0</mark>	513.65	112.67	28.54	327-53	468.74	15.14
Bone Meal	i.						Avera	୍ତ ୪.୨୪	
5/15/24	12.54	102.38	18.12	133.04	15.74	3.26	80. 80	102.36	30.18
5/16	12.54	102.38	18.12	133.04	16.38	5. 5 5	107.45	128.52	4.12
5/17	12.54	102.38	16.12	133.04	18.97	3.09	79.95	102.01	31.03
5/18	12.54	102.38	18.12	133.04	20.42	2•15	50.72	113.29	19.75
5 /1 9	12.54	102.38	18.12	133.04	CI • 02	2.98	76.08	99.16	н 1 20 20 20
	62.70	511.30	09 · 06	655 .20	11.76	1:4.07	435.06	546.24	113.96

Average 23.79

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Nc. 70		Intske		Grans			Outgo		
Date	Silage	Grain	Hay	Total	K11k	Urine	Feces	Total	Balance
3/13/24	11.41	52.55	11.20	75.16	25.19	• 59	41.17	66.95	8.21
3/14	11.41	52.55	11.20	75.16	22.74	• •	73.86	66.93	8.18
3/15	11.41	52.55	11.20	75.16	22.41	47.	56.05	79.20	+0•+ -
3/16	11.41	52 • 55	11.20	75.16	27 • 89	- 1 - 1	47.40	72.07	
21/2	τη・ττ	52.55	11.20	75.16	24.88	.93	50.40	76.21	- 1.05
	52.05	262.75	56.00	375.80	123.11	3.42	234.88	361.41	14.39
Raw Rock	k Phosphate	t) D					Average 2	2. 3. 3. 3. 3. 3. 3. 3. 3. 3. 3. 3. 3. 3.	
4/2/5h	10.39	65.23	2.89	£3.51	20.72	6.02	64.18	90.92	- 7. ⁴ 1
9/t	10.39	65.23	8.92	84.54	20.53	5.78	63.89	90.20	- 5.66
2/۲٫	10.39	65.23	8.92	84.54	22.12	4.71	52.44	95.27	-10.73
80/=t	10.39	65.23	6.1 6	87.78	41.02	5.13	60.81	86.08	- 2.70
4/9	10.39	65.23	7.44	87.06	20.20	0:1-1	63.86	53.46	- Л.40
	51.95	3:6.15	41.33	419.43	τζ.ξοι	26.04	321.18	450.93	-31.50
							AVETUGE	0 • • 4 •	

P205 Grame

No. 70

•	· · · ·	Intake		Grans	8		Outgo			
Date	Silago	Grain	Нау	Total	Milk	Urine	Feces	Tctal	Belance	
4/20/24	12.92	35.52	10.87	59.31	19.40	• 35	ुह •05	57.80	1.51	
1 2/t	12.92	35.52	13.01	59.31	20.27	• 63	10.97	ó1.ĉ7		
t1/25	12.92	35.52	10.37	59.31	21.18	0 1 0	39.68	61.36	- - - - -	
4/25	12.92	35.52	10.87	59-31	20.23	• 69	32.98	53.90	G.41	
tr/5t	12.92	35.52	10.87	59.31	12.54	. 85	35.65	57.04	2.27	
	64 • 60	177.60	54.35	296.55	100.62	3.02	163.33	26.165	4.58	
Bone Meul	ч						Aversge	0 0 •		
5/15/24	12.54	67.04	10.87	90 . 45	50.29	9†•	60.51	8 1. 26	9.19	
5/16	12.54	67.04	73.CI	90 • 45	19.03	5 ⁴ .	62 • 55	11.020	3.74	
5/17	12.54	40-29	72.CI	90.45	20.02	ະ ເງ	61.31	82.16	6°.3	
5/18	12.54	67.04	72.87	90.45	20.11	•00	63.93	84.77	10 • • •	
5/19	12.54	67. CH	73.CI	90.45	10.61	ot.	53.76	75.27	14.68	
	62 • 70	335.20	54.35	452.25	70.69	2.78	304.22	40.504	45.12	
							Average	42.9		

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-19.51 -19.51 -19.51 -19.51 -19.52 -15.65 -1.79 -1.79 -1.79 -1.79 Ealance

 Feces
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 61.93
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 107.62

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 555.77

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 56.44

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 83.56

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 0ut go Urtre 2.53 4.82 5.59 5.12 5.69 4.82 70.47 70.47 11.87 13.60 13.60 13.60 13.60 мллк 47.31 45.38 49.07 46.91 46.91 46.91 46.91 46.92 26.39 26.39 26.39 26.39 26.39 26.39 31.64 Total 109.88 109.88 109.88 109.88 109.88 549.40 71.04 71.04 91.18 91.18 53.66 Grams 502d 14 .39 14 .39 14 .39 71 .95 8 .92 8 .92 8 .92 8 .92 14 .60 Hay

 Silage
 Grain

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 11.42
 37.90

 12.51
 24.29

 13.55
 24.29

 148.36
 37.90

 13.55
 24.29

</tabular 64.19 Intake 60.16 Raw Rock · / 5/ 17 113/5# 3/14 3/25 3/16 3/17 4/5/24 4/6 4/7 10. 71 late

Average - 1.47

P20 5

- 4.12 - .25 1.84 1.84 .70 .70 6.58 -22.84 -11.57 17.10 1.33 - 9.40 Balarce 1.83 58.27 56.18 57.32 55.49 289.40 Average 22.61 112.03 100.76 Average 62.14 72.09 Tctal Outgo Гесев 31.25 28.17 29.21 29.53 29.53 29.53 58.17 58.17 58.17 57.07 72.70 50.33 50.33 327.67 3.71 2.04 2.04 2.07 2.07 2.07 2.07 2.07 2.12 3.12 3.57 2.68 Urine 3.31 27.18 25.56 24.93 25.33 25.33 130.82 21.65 21.84 24.49 19.03 MILK 25.15 112.21 Grams 53.02 53.02 58.02 58.02 290.10 89**.19** 89.19 89.19 89.19 Tctal 445.95 89.19 10.87 10.87 10.87 10.87 10.87 10.87 10.87 10.87 10.87 54.35 Hay Intake 67.04 67.04 67.04 35.52 35.52 35.52 35.52 35.52 177.60 Grain 335.20 11.63 11.63 11.63 11.63 11.63 11.26 11.26 11.28 Silage **11.28** 56.40 4/23 Bone Meal 1/20/5/1 **5/1**5/24 i4/24 71. 71 5/16 1/22 1*2*/1 21/5 Date 5/18 5/19

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Nitrogen Grams

Intake

No. 33

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Outgo

							D		
Date	Silage	Grain	Нау	Total	ALIM	Urine	Feces	Total	Balance
3/13/24	H6 .1 6	348.80	50 .01	26.444	109.23	165.18	147.66	422.07	22.90
3/14	46.16	348.80	50.01	26-444	116.96	160.13	160.97	438 . 06	6.91
3/15	46.16	348.80	50.01	26• 111 1	102.64	178.56	165.63	446.83	- 1.86
3/16	46.16	348.80	50.01	26.4414	111.02	160.18	157.09	428.29	16.68
3/17	46.16	348.80	50.01	72.444	109.69	168.24	149.00	426.93	18.04
And mag	230.80 Dhoanhata	1744.00	250.05	2224.85	549.54	832.29	780.35	2162.19	62.67
4/5/24		257.12	33.97	317.28	81.5 0	155.17	106.02	Average 342.69 -	age 12.53 - 25.41
4/6	26.31	253.64	37.54	317.49	83.18	135.14	66•#6	313.31	4.18
۲/4	25.84	219.97	32.89	278.70	76.93	131.52	76.07	284.52	- 5.82
tt /8	27.33	230.33	33.54	291.20	82.86	44°611	109.59	311.89	- 20.69
6/†	30.90	257.12	33.41	321.43	84.57	161.14	101.98	347.69	- 26.26
	136.57	1218,18	171.35	1526.10	40°604	702.41	488.65	1600.10	- 74.00
	•							Avera -	Average - 14.80
									.4

				·					
No. 33			TA	TABLE NO. VII, Nitrogen	, con'd. n				
		Intake		Grama		Ou	Outge		
Date	Silage	Grain	Hay	Tctal	LIIK	Urine	Fece s	Total	Balance
tt/20/5/tt	46.97	213.63	38.96	299•56	66.10	125.57	93.51	285.18	14.38
H/21	46.97	213.63	38.96	299.56	67.86	122•51	94.58	28 ⁴ •95	14.61
4/22	46.97	213.63	38.96	299•56	71.93	46.211	98.82	283.69	15.87
4/23	46.97	213 . 63	38.96	299•56	67.25	123.95	98.67	289.87	9•69
4/2H	46.97	213.63	38.96	299.56	60.62	117.22	100.46	278.30	21.26
23 Bone Meal	23 4.85 al	1068.15	194.60	1497.80	333.76	602.19	486.04 Av	1421.99 Average 15.16	16 ^{75.81}
5/15/24	26 . 81	167.61	47.57	241.99	47.26	100.63	78.43	226.32	15.67
5/16	26.81	167.61	47.57	241 . 99	48.63	97•31	92.10	238.04	3.95
5/17	26 .81	167.61	47.57	241 . 99	46.58	101.91	69.98	218.47	23.52
5/18	26.81	167.61	47.57	24 1. 99	50.66	71.75	82•65	205.06	36.93
5/19	26.81	167.61	47.57	241.99	46.51	109.59	69.06	225.16	16.83
·	134.05	838.05	237.85	1209.95	239.64	481.19	392.22	1113.05	0é•96
							Α	Average 19.38	38

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Nitrogen

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No. 70		Intake		Gran a	1 8 8	Ou'	Outgo		
Date	Silage	Grain	Нау	Total	MILK	Urine	Feces	Total	Balance
3/13/24	30.77	180.29	29.18	240.24	54.81	102.56	65.46	222 • 83 -	17.41
3/14	30.77	160.29	29.18	240.24	57-55	96.72	64.67	218.94	21.30
3/15	30.77	180.29	29 .18	240.24	13.64	77.43	74.12	201•36	36.88
3/16	30.77	160.29	29 •16	42.042	63.80	93.30	59•5 0	216.60	23.64
3/17	30.77	160.29	29.18	240.24	55.40	24.32	72.12	223.99	16.25
	153.85	901.45	145.90	1201.20	281.37	466.45	335.87	1083.72	117.48
Raw Roc	Raw Rock Phosphate	te					A	Average 23.50	20
4/5/54	20 .60	139.89	20.93	181.42	47.88	96.93	65.34	210.15	- 23.73
4/6	20.6 0	139.89	23•65	184.14	th7。74	93.57	54.62	195.93	- 11.79
۲/ ۲	20.60	139.89	23•65	184°181	48.32	94.51	60.63	203.46	- 19.32
4/8	20.60	139.89	21.65	182.14	48.17	89.22	59 . 42	196.81	- 14.67
4/9	20.60	139.89	19.73	18 0.22	48.32	88.71	58.52	195.55	- 15.33
	103.00	699.45	109.61	912.06	240.43	462.94	298.53	1001.90	- 89.84

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Average - 17.97

TABLE.NO. VII Nitrogen Grums

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No. 70	Int	Intek•		6 E E E E E E E E E E E E E E E E E E E	- 10 0 1 9 10 1		Outgo			
Date	Silage	Grain	Нау	Total	MILK	Urine	₩ 60 3	Total	Balance	ł
4/20/2H	31.31	19.041	23.37	195.29	42°54	66.33	62.06	196.79	- 1.50	ţ
4/2 1	31.31	140.61	23.37	195•29	47.50	86.68	63.19	197.37	- 2.06	
4/22	31.31	140.61	23.37	195.29	52•52	82•35	66.98	201-85	- 6.56	
4/23	31.31	140.61	23.37	195.29	47.92	56•77	54.98	180.83	·14 • 46	
#724	31.31	140.61	23.37	195.29	46.23	79.61	64.36	190.20	5.09	
	156•55	703.05	116.85	976.45	239.91	415.56	311.57	40.76	9.41	
Bone Meal	331						Ател	Average 1.88		
5/15/24	26 .81	140.61	28.54	195.96	48.95	93.29	61.98	204.22	- 8.26	
5/16	26.81	140.61	28.54	195•96	45.12	79.29	59.19	183.60	12.36	
2/17	26.61	140.6 1	28.54	195.96	66 • ††	63.99	64.19	198.17	- 2.21	
5/18	26 .61	140.61	28.54	195.96	46.07	83.68	65•99	200.74	- 4.78	
5/19	26.81	140.61	28.54	195.96	43.65	83.84	58.46	185.95	10.01	
	134.05	703.05	142.70	02.672	226.78	434.09	309.81	972.68	7.12	

Average 1.42

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Table NO. VII Nitrogen

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No. 71

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+		Intake		Grams	8		Outgo		
Dat e	Silage	Grain	Нау	Total	WIIK	Uri ne	Feces	Total	Balance
3/13/24	30.77	288.47	37.51	356.75	103.89	101.54	114 .20	319.63	37.12
3/14	30.77	256.47	37.51	356.75	101.86	107.67	136.08	345.62	11.13
3/15	30.77	288.47	37.51	356.75	104.09	89.83	57.101	295.67	61.08
3/16	30.77	288.47	37.51	356.75	102.13	150-55	98.76	351.44	5.31
3/17	30.77	288.47	37.51	356.75	57.72	106.63	116.84	321.25	35.50
	153.85	1442.35	187.55	1783.75	509.70	556.27	567.64	1633.61	150.14
Raw Roci	Raw Rock Phosphats	0)					Ачегаде	se 30.03	
4/5/5 4	9.89	132.13	23•65	165.67	58.58	78.06	65.77	202.41	- 36.74
4/6	6.58	138.44	23.65	168.67	54.15	62.00	46 . 24	162.39	6.28
۲/۲	16.4 8	138.44	23.65	178.57	1 6•65	97.30	20.7 4	204.31	- 25.74
4/8	16.48	130.60	23.65	170.93	56.92	89•36	63.16	111.002	- 38.51
6/1	8.00	93.74	23.65	125.39	58.73	70.85	49.55	179.43	- 54.04
	57.43	633•55	118.25	809.23	268.32	397.57	272.09	957.98	-148.75

Average - 29.75

TABLE NO. VII Nitrogen

No. 71		Int ak •		G	5 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1		Outgo	0	
Date	Silage	Grain	Нау	Totel	MIL	Urine	Feces	Total	Balance
4/20/24	28.18	140.61	23.37	192.16	62.87	97.57	64.84	225.28	- 33.12
1 2/4	26,16	140.61	23.37	192.16	62.03	14.17	59.12	192•56	9 1 .
4/22	26.15	140.61	23.37	192.16	57.44	63.64	1 4.19	182.49	9-67
4/23	26.18	140.61	23.37	192.16	58.16	60.73	65•73	191.00	1.16
42/4	26.16	140.61	23.37	192.16	60.61	69.41	62.52	192.54	- 38
	140.90	703.05	116.85	960.50	301.13	369.12	313.62	983.87	- 23.07
Bone Leal	Ч						Average	- 4.61	
5/15/24	24.12	140.61	28 • 5 4	193.27	14.84	69.58	62.27	180.32	12.95
5/16	24.12	140.61	28.54	193.27	48°79	71.82	64.89	185.50	7.77
5/17	24.12	140.61	28.54	193.27	55.42	77.56	62•58	195.56	- 2.29
5/18	24.12	140.61	28.54	193.27	41.25	69.57	56 . 44	167.26	26. 0 1
5/19	24.12	140.61	28.54	193.27	60.93	86.57	62.80	210.30	- 17.03
	120.60	703.05	02•24I	966.35	254.86	254.86 375.10	308.93	938.94	27 . 41

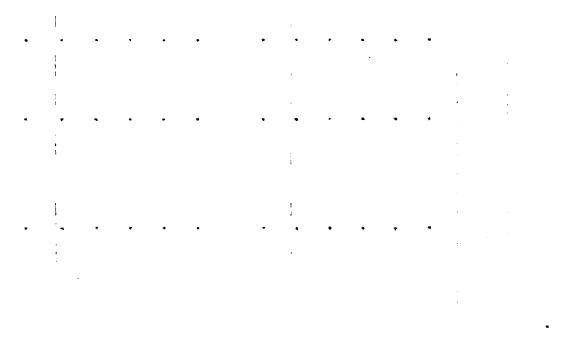
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Average 5.48

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DIBUUSCI A OF MADULIS

Palatability

The results plainly show that have med is much more palatable than raw rock phosphate.

Ap carance of the Minels

The change in the appearance of the animals during the experiment was probably not due entirely to a lack of nutrients in the feed, but to other factors. Cow No. 33, the Holetein, was giving large quantities of milk at the beginning of the experiment and after fifteen days on the basal ration began to lose weight as shown by graph No. I. This loss of weight would be only natural for a heavily lactating animal. When rock phosphate was added to her basel ration she refused to eat. It was necessary to practically starve her for a few days. Molesses was added to the ration and helped a lit le. The rock phosphete was then mixed with the silage instead of the grain. She ate the silage very slowly but still refused prain and it was necessary at last to change her grain ration. A grain mixture was fond that had practically the same analysis as the first ration but was more selatable. This ration was used and the animal ate better, altho she did not eat well as long as the rock phosphete was being fed. During this period she continued to lose weight.

After the rock phosphate was removed the animal atevith more relish.

About the twenty-eighth of March one querter of the udder of cow No. 33 became infected. The quarter swelled up and gave thick, watery milk and later bloody milk was produced. The blood disappeared just before the second metabolism period but the milk was still thick. This quarter remained infected until almost the end of the experiment.

Just after bone meal vas added to the basel ration the animal became sick, probably because of toxins produced in the infected quarter. This sickness accounts for the rapid loss of weight just after bone meal was added. For several days the animal refused food and water. About the fifth of Hay she slowly began to recover. The infection had gone from her udder and the milk was clear again. The recovered rapidly after this and increased considerably in her milk flow and gained in weight. The was entirely recovered before the fourth matabolism period.

No. 71, a Jersey, was the other animal that showed a striking difference in appearance at the beginning and at the end of the test. This animal showed constant weight during the basal ration period. When raw rock phosphate was added to her ration she immediately wont off feed and on the third day bloated. She lost weight rapidly for a few days. She was never in normal condition while raw rock phosphate was being fed, although during the five day metabolism period she had a fair appetite and ate quite regularly.

As soon as the rock phosphete was taken out of her ration she began to eat better and so n seemed entirely recovered

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except that see appeared rather irritable. Her weight remained fairly constant until the close of the experiment.

The third Lainel, No. 70, showed very little veristion in weight, ate beartily during the ontire experiment and se med perfectly nor all at all times.

The deleipm and Inergania Phosphorps of the Plood

Graph Lo. I shows a very definite relative between the amount of phosphorus in the feed and the inorganic phosphorus of the blood. All three minals were bled at the end of the first period, (March eighteenth) in which the basel retion slone was fed. This ration was low in both calcium and phosphorus. The amount of phosphorus in the blood was very low. On the tenth of April the animals were bled again. This was at the end of the period in which rock phosphate had been edded to the ration. All three animals showed a definite rise in the emount of inorgenic phosphorus in the blood. Luring the third period the cows were fed the basal ration alone. They were bled twice during this period, on April eighteenth and April twenty-fifth. Here, as in the first period when the feed as low in phosphorus, the inorganic phosphorus of the blood was low. On May thirteenth and nineteenth, they were bled again. This was during and at the ending of the period in which bone meal was fed along with the basal ration. All three aminals showed a higher inorganic hosphorus content of the blood. The basal ration was fed again from the nineteenth of May until the first of June. Blood samples were taken on May twenty-ninth, June fifth and June twolfth. The amount of inorganic phosphorus in the blood of all three animals showed a definite drop. Raw Rock phosphate was then added to the ration and the cows were bled again on the nineteenth of June. The three samples showed a rise in the phosphorus content over the previous samples. The evidence shows that the amount of inorganic phosphorus in the blood varies with the amount of phosphorus in the food.

It is very doubtful if the same is true of calcium. The results obtained in this experiment, however, show that there may be a tendency for the amount of calcium in the blood to depend on the amount of calcium in the ration. At the end of the first period when the tasal ration, low in calcium had been fed, all three enimals showed a low calcium content of the blood. After raw rock choschate had been added to the basal ration during the second period all three animals had a decided increase in the blood colcium. This sample was taken April tenth. In the next period on the basel ration both No. 70 and 71 showed a lowering in the colcium content of the blood, on both the eighteenth and twenty-fifth of April. No. 32, however, showed an increase on the eighteenth and then a decrease by the twenty-fifth. The amount of celcium in the blood on the twentyfifth, though, was higher than on the tenth. Bone meal was then added to the ration. The animals were bled on the thirteenth and twentieth of Lay. On the thirteenth cows 70 and 33 showed an increase in blood calcium; No. 71, however, showed On the twentieth all three animals were higher a decrease. than on the twenty-fifth of April and Nos. 71 and 73 showed a decided increase over the calcium content of the blood on the thirteenth of Abril. No. 70 was very much lower. During the period from May twentieth to June twelfth the basal ration was

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again fed. The animals were bled three times during this period and on all three processions showed lower calcium content of the blood then they did on the twentieth of May when bone meal was fed for the last time. (They did, however, show that the blood calcium was increasing towards the end of the period.) Raw rock phosphate was again added to the ration and the animals were bled on the hineteenth of June. All three showed a slight increase in blood calcium over the previous period. These results indicate that there may be a tendency for the amount of calcium in the blood to correspond to the amount of calcium in the feed.

Hydrogen Ion Joncentration of Feces

Table No. I giving the hydrogen ion concentration of the feces shows no relation between the amount of calcium and phosphorus in the food and the hydrogen ion concentration of the feces.

Percent of Celeiam and Phosphorus in the Milk

Table No. II showing the average percent of calcium and phosphorus in the milk during the different metabolism periods shows very little variation and it is probably safe to assume that the feeding of bone meal or of raw rock phosphote does not effect the percent of calcium and phosphorus in the milk.

Percent of Fat in Lilk

Table No. III shows that the percent of fat in milk is not increased by the additionof more phosphorus or calcium to the ration in the form of bone usal or raw rock phosphate.

Availability of Caldium and Phosphorus

The measurement of the availability of the coldium and phosphorus in Jone deal and rew rock doephate was the amount of these elements retained by the animal body. This was not an accurate measure because the milk production of an eminal declines somewhat as the lactation wrind advances, and the cows were, therefore, giving less milk at the time bone weal was fed than when raw rock phosphete was fed. Naturally, they would be more likely to lose caldium and phosphorus when rew rock chosplate was fed than when bone meal was fed because of the greater drain of higher milk production. This difference in milk production, however, the not great. No. 53 during the metabolism on raw rock phosphate gave 28.2 pounds of milk daily and on bone meal 27.6 pounds. No. 71 averaged 34.2 pounds while fed raw rock whosphate and 21.7 pounds while on bone most. No. 70 avoraged 18.4 powhas of milk during the raw rock phosphote period and 18.2 counds during the metabolism when bone meal was used.

The Calcium Balances

A study of table No. V giving the calcium balances shows that bone meal is superior to raw rock thosphate as a source of calcium.

On the basal ration when the first balance test was conducted dow No. 73 was giving 55 pounds of milk and had an average negative balance of - 0.95 grams of calcium daily. As explained before, when ray such prophete was added to the ration considerable trouble was experienced in getting the think 1 to consume her neuch chows of feed. Before the began to cat regularly

her milk production had decreased to 78 younds doily. She continaed this production during the metabolism trial with raw nock phosphite and aboved a positive caldium balance of + 13.57 grans daily. The great difference between this balance and the first balance when no raw rock phosphete was fed was probably due to a large extent to the greater milk production during the first period. because in thethird period when the basel ration alone was again fed allo had a prattive balance of + 5.00 grees caldium daily and vas giving 35 pounds of milk. The last period when bone meal wer fod she was giving only 23 pounds of mild, but she had a much greater coldium belance then during suy of the other periods. Her balance at this time was 25.00 group daily. No. 33 showed a definite increase in the amount of coldium retained when fed both raw rock phosphate and wone meal over the periods when the basal ration was fed slone. The positive balance was much stronger, however, when bone meal was fed than when raw rock phosphete was used.

No. 71 giving 40 pounds of milk during the first period had a slightly negative balance - 1.09 grans calcium deily. During the next period while on raw rock phoephate she was taken sick and went off feed. After she began to est regularly again she had dropped to 24 pounds of milk daily. Her calcium balance increased to a positive + 5.97 grams daily. This storing of calcium may have been due to her recovering from the sickness during the beginning of the period. The third period on basel ration alone the balance was very slightly negative, - 0.69 grams calcium daily. The fourth period with bone meal the balance was positive + 0.09 grams of calcium daily. There was little change in the milk flow after the animal had dropped to 24 pounds of milk per day. As in the case of No. 33, there was an increase in the amount of calcium retained when fed both raw rock phosphate and bone meal over the periods in which the basal ration alone was fed. The balance was less, however, on bone meal than on raw rock phosphate. This animal was never in normal condition after raw rock phosphate was added to the ration. The was very irregular in her feeding and appeared to be in poor health. It is doubtful if the results obtained with this animal should be used in making conclusions.

Cow No. 70 seemed to be normal throughout the experiment and the results with her are probably much more reliable than with the other two cows. The colcium balance for the first three periods with this dow were practically the same. The first period on basel ration alone the belance was positive + 0.78 calcium deily. The second period with raw rock phosphate added to the ration the balance was very slightly negative being -0.756 grams. The third period on basel ration alone the balance was positive + 1.11 grans daily. There was no apprecicble difference in the balances for these geriods. The fourth period, however, with bone meal in the ration the balance became very strongly positive + 9.71 grams calcium daily. The milk flow for this animal was practically the same throughout the entire experiment. The results with No. 70 yould indicate that the calcium of bone meal is more available than the calcium of raw rock phosphate.

The Phosphorus Balance (2205)

Table No. VI shows the phorphorus balances for all three animals during the metabolich periods.

Cows Nos. 33 and 70 showed a greater retention of phos-

phorus during the bone meal period than when fed raw rook phosphate. They also retained more phosphorus when fed bone meal than on the basal ration alone. No. 71 showed very little difference in the amount of phosphorus retained during any of the periods.

On the basal ration alone, during the first period No. 33 had anegative phosphorus balance of - 11.52 graps daily. For the next period raw rock phosphote and the third period basal ration alone the retention was about the same being + 6.066 grams phosphorus daily in the second and + 8.98 grams in the third. During these two periods and the last one the milk flow was much less than during the first period. The large amount of milk probably is the reason for the loss of phosphorus during the first period. When bone meal was fed the balance became very strongly positive. It was + 25.79 grams phosphorus daily.

Cow No. 70 showed a positive balance of # 2.88 grams phosphorus daily during the first metabolism period on basal ration alone. The balance became negative - 6.70 grams phosphorus daily after raw rock phosphate was added to the ration. The third period on basal ration alone the balance became slightly positive again, + 0.92 grams. Bone meal added to the ration for the fourth period brought the balance up to positive 9.24 grams phosphorus daily, showing the superiority of bone meal over raw rock phosphate as a source of phosphorus.

No. 71 showed no significent difference in the pholophorus balance throughout the experiment. As she was not in normal condition the results obtained with her are probably of fittle value.

The Nitrogen Balance

A study of Table No. VII showing the nitrogen balance indicates that raw rock phosphate causes a loss of nitrogen from the body. The first period on basal ration slone all three animals showed a very positive nitrogen balance, No. 33, - 12.53 grems of nitrogen daily, No. 70, - 23.50 grems, and No. 71, - 20.03 grams. During the second period after raw rock phosphate had been added to the ration the balances all became stronaly negative, being - 14.82 grams nitrogen daily for No. 33. - 18.97 grams for No. 70. and - 29.86 grams for No. 71. The basel ration was again fed alone during the third period and for all but No. 71 the balances became positive. For 71 the balance was still slightly negative but was much nearer positive than during the raw rock phosphete period. The balances for the third period were - 15.18 grams nitrogen daily for No. 33. - 1.88 grams for 70, and - 4.01 grams for No. 71. Bone meal when fed during the fourth period did not have the same effect as raw rock phosphete. The balance for No. 33 was - 19.38 grams nitrogen daily. for No. 70 - 1.42 grams and for No. 71 - 5.48 grams.

All three animals lost weight during the first metabolism period as shown by Graph No. I. Since they were on positive nitrogen balance, this loss in weight must have been due to loss of compounds other than those containing nitrogen. The second period on raw rock phosphete, No. 33 and 71 were losing weight rapidly, probably due to loss of nitrogenous compounds from the body. No. 70, however, although losing nitrogen, maintained a constant weight. During the third period the weights for all three animals varied only slightly. The fourth period No. 33 lost weight repially at first because of sickness. She recovered, however, before the metabolism period, and during the metabolism period the weights for all three animals were practically constant.

It is hard to explain thy each sninel lost nitrogen during the second feeding period. -vidently there is something in raw rock phosphate that hinders the retention of ritrogen.

This experiment may be critised for the shortness of the metabolism periods. Five days is hardly long enough from which to draw very definite conclusions. However, since all three animals show the same trend the results offer an indication of the effects of the minerals fed. Further work with longer metabolism periods is needed before definite conclusions can be drawn.

Phosphorus in the Urine

A close study of the analysis of $P_2 O_5$ in the wrine shown in Table No. VI brings out an interesting fact. During the raw rock chosplate period the urine of all three animals was extremely high in phosphorus.

No. 33 had on average of about 4 graps of P_2O_5 daily in the urine while on basal ration during the first period, about 22 graps of P_2O_5 in the urine during the raw rock period; this dropped again to about 5.5 graps in the third period and 1.8 graps in the fourth period. The urine of No. 70 contained 0.7 grams of P_2O_5 daily during the first period 5.0 grams while on raw rock phosphate, 0.55 grams the third period, and 0.5 grams the fourth period.

No. 71 had an average of 3.5 grams of P_2O_5 in her unite daily while on the basel ration the first period. This increased to 15.0 grams P_2O_5 daily while new rock hosphate was being fed. Then the basel ration was fed during the third period the amount of P_2O_5 in the unite dropped to 5.0 grams daily and remained the same during the fourth period when bone meal was fed.

Evidently raw rock photohete is digested by the animal and the phosphorus at least assimilated. Something, however, prevents its retention.

CONCERNING.

Bone meal is superior to raw rock phosphate as a source of calcium and phosphorus for lactating dairy cove.

The phosphoras of raw rock phosphate is digested and assimilated but is not retained by the animal organism as indicated by the increased amount of phosphorus in the urine during the period when raw rock phosphate was fed.

The inorgenic phosphorus content of the blond follows the phosphorus content of the feed.

Bone meel is much more galatable than rew rock thosphate.

The hydrogen ion concentration of the faces is not affected by the addition of either bone meal or raw rock phoschate to the ration.

The percentage of calcium and phosphorus in mill is not affected by the addition of more calcium and phosphorus to the feed in the form of raw rock phosphate or bone meal.

The percent of fat in the milk is not effected by adding raw rock choophate or bone meal to the ration.

Raw rock dosphate hinders the retention of hitrogen.

1. Abderhalden, 1398

Cited by Forbes " Keith, Technical Bulletin 5, Ohio Agricultural Experiment Station, page 152.

- 2. Anger, 1898
 Cited by Forbes & Keith, Technical Bulletin 5, Ohio
 Agricultural experiment Station, pages 420 to 421.
- Artzemias, G. K.
 Endocrinology 3,560-1 Dissertation 1919.
- 4. Armsby, H. P. Nutrition of Farm Animals, pages 335, 336, 338.
- 5. Armsby, H. P. Nutrition of Farm Animals, page 414.
- 6. Beaunis

Cited by Forbes & Keith, Technical Bulletin 5, Ohio Agricultural Axperiment Station, page 104.

- 7. Brown, Mac Lacklin, Simpson
 American Journal Diseases Children 19, pages 413 to 428.
- 8. Crown, D. M., Calhoun, H. A. Journal Biology Chemistry 37, pages 505 to 509.
- 9. Chick, H., Dalzell, A. J., Hume, N., Mackey, H.M.M., Smith, H. H., and Lancet, H. Winberger II, pages 7 to 11.
- 10. Dennis, W., Talbot, P. B. American Journal Diseases Children 21, pages 29 to 37.
- 11. Dibbelt W., 1908, Bahrhudt & Edelstein, 1910 Technical Bulletin 5, Ohio Agricultural Experiment Station, pages 571.

- 12. Dibbelt, W.
 Arb. Geb. path. Anat. Bakteriol Tubinger 7, pages 55 to 59,
 C. A. 6, 2761.
- Eckles & Trowbridge
 Feeds & Feeding, Henry & Morrison, page 82.
- 14. Elliot, Chrichton, & On British Journal Expt. Pathol 7, pages 10 to 19.
- 15. E merick & Loew Experiment Station Record 29. page 565.
- 16. Etienne,

Journal Physiol. Pathol. Gen 14, page 108.

- 17. Experiment Station Record 18, page 173.
- 18. Experiment Station Accord 22, page 783.
- 19. Experiment Station Record 42, page 777.
- 20. Farmer, E., Klinge, R. Endocrinology 5, pages 375 to 376.
- 21. Forbes, 2. B. Bulletin 201, Ohio Agricultural Experiment Station, page 146 and page 149.
- 22. Forbes, B. Bulletin 201, Ohio Agricultural Experiment Station, page 141.
- 23. Forbes & Associates Bulletin 295, Ohio Agricultural Experiment Station.
- 24. Forbes & Associates Bulletin 308, Ohio Agricultural Experiment Station.
- 25. Forbes & Associates Bulletin 330, Ohio Agricultural Experiment Station.

- 26. Forbes & Associates Bulletin 347. Ohio Agricultural Experiment Station.
- 27. Forbes & Associates Bulletin 363, Ohio Agricultural Experiment Station.
- 28. Forbes, E.B., Keith, H. Helen Technical Bulletin 5, Ohio Agricultural Experiment Station, page 104.
- 29. Forbes, E. B., Keith, M. Helen Technical Bulletin 5, Ohio Agricultural Experiment Station, page 149.
- 30. Forbes, 2. B., Keith, M. Helen Technical Bulletin 5, Ohio Agricultural Experiment Station, page 552.
- 31. Forbes, -. B., Keith, H. Helen Technical Bulletin 5, Ohio Agricultural Experiment Station, page 556.
- 32. Gilbert & Posternack Cited by Forbes & Keith, Technical Bulletin 5, Ohio Agricultural Experiment Station, page 104.
- 33. Hart, McCollum & Fuller Cited by Forbes & Keith, Technical Bulletin 5, Ohio Agricultural Experiment Station, page 327.
- Hart, McCollum & Fuller & Humbhrey
 American Journal Phys. 1909, Research Bulletin 5, Wisconsin.
- 35. Hart, McCollum & Humphrey Cited by Forbes & Keith, Technical Bulletin 5, Ohio Agricultural Experiment Station, page 420.

- Z6. Hart, Steenback & Hoppert Science 50, page 318.
- 37. Hart, Steenback " Hoppert Journal Biology Chemistry 48, pages 33 to 50.
- 38. Hart, Steenback & Hoppert, Dethie & Humphrey Journal Biology Chemistry 54, pages 75 to 89.
- 29. Haverson, J. O., Hohler, H. K., Bergheim, O. Proc. Soc. Biol. Med. 15, pages 11 to 12.
- 40. Heineman Milk, page 101.
- 41. Henneberg
 Cited by Forbes ~ Keith, Technical Bulletin 5, Ohio
 Agricultural ~xperiment Station, page 421.
- 42. Henry & MorrisonFeeds & Feeding, page 64 and page 82.
- 43. Hess, McCann, & PappenheimerJournal Biology Chemistry 47, page 105.
- 44. Hess, Unger, & Pappenheimer Journal Biology Chemistry 50, pages 77 to 81.
- 45. Högen, J.

Experiment Station Record 10, page 920.

- 46. Howland, McKim, Marriott, Quarterly Journal Medicine 11, pages 289 to 317.
- 47. Howland & Park Bulletin John Hopkins Hospital 32, page 341.
- 48. Huffman, 0. F. Unpublished tables compiled from work of several investigations
- 49. Hutcheon, D.

Agricultural Journal, Cape of Good Hope 24, No. 4, pages 479-43

- 50. Janison, M. H. Deutch Arch. klein med. 125 pages 167 to 187 From C. A. 12, 2757.
- 51. Jordan, mart & Patton Cited by Forbes & Keith, Technical Balletin 5, Ohio Agricultural Experiment Station, page 421.
- 52. Joseph & Heltzer American Journal Phys. 29, page 1.
- 53. Journal Biological Chemistry 37, page 1.
- 54. Kaup, B. F. Poultry Items 26, no2, pages 14 and 18.
- 55. Khuen, 1908

Cited by Forbes & Keith, Technical Bulletin 5, Ohio Agricultural Experiment Station, page 421.

56. Kellner

Cited by Forbes " Keith, Technical Bulletin 5, Ohio Agricultural Experiment Station, page 421.

- 57. Kremer, B., Howland, John. Bulletin John Hopkins Hospital 33, pages 313 to 517.
- 58. Kummer, Robert, Presse Medicole 28, pages 765 to 766. Chem. Abstracts 15, 557.
- 59. Köhler

Landw. Versuchsstationen Vol. LXI, 451, Vol. LXV, 349 Cited by Forbes, Bulletin 201, Ohio, page 149.

- 60. Lawes " Gilbert Bulletin 201, Ohio Agricultural Experiment Station, page 141.
- 61. Lienaux Am. Med. Vet. 56 no. 4, pages 192 to 200.

62. Loeb

American Journal Phys. 5, page 362.

- 63. Loew, O Munch Med. Wochsehe 61, pages 983 to 985, From Chem. Abstracts 8, 3307.
- 64. Lovett, J. Am. Med. Assoc. 78, page 3.
- 65. MacCallun, d.G., Voegtlin, Carl Trans. Associ. Am. Phys. 25, page 416.
- 66. MacCallum, W. G., Voegtlin, Carl Journal Exp. Med. 11, page 118.
- 67. Mazzocco, 2. Anales soc. quim. Argentine 9, pages 515 to 525 Chem. Abstracts 16, 3116.
- 68. McClendon, J. F. Am. Journal Phys. 61. pages 273 to 279.
- 69. McCollum,

Newer Kno ledge of "atrition, 1922 edition.

- 70. McCollum Newer Knowledge of Nutrition, 1922 edition, page 294
- 71. LeCollum

Newer Anowledge of Autrition, 1922 edition, page 312.

72. LeCollum

Newer Knowledge of Nutrition, 1922 edition, page 513.

- 73. McCollum, Shipley, Park & Simmonds Am. Journ. Hyg. I, pages 512 to 525.
- 74. McCollum, Simmonds, Park & Shipley Proc. Soc. Exp. Bi ol. Med. 18, page 275.

- 75. McCollum, Simmonds, Park & Shipley Journal Biol. Chem. 50, pages 5 and 30.
- 76. McCollum, Dimmonds, Park ω Shipley
 Journal Biol. Chem. 45, pages 333 to 341.
- 77. McCollum, Simmonds, Park & ShipleyAn. Journal Diseases Children 25, pages 91 to 100.
- 78. McCollum & Simmonds

Journal Biol. Chem. 50, page 5.

79. llcCrudden

Arch. Intern. Med. 5, pages 596 to 621.

- 80. Meigs, Blatherwich & Corey Journal Biol. Chem. Vol. 37, page 1.
- 81. Meigs & Woodward U.S.D.A. Bulletin 945.
- 82. Mellanby

Journel -hys. 52, page 11.

- 83. Mors & Muck, 1869 Cited by Forbes & Keith, Technic al Bulletin 5, Ohio Agricultural Experiment Station, page 555
- 84. Moussn & Dollar
 Diseases of Dairy Cattle 1920.
- 85. Nassan

Journal American Med. Association 76, 171d.

- 86. National Lime Association, Washington, D.C., Bulletin 176.
- 87. Pappenheimer, McCann & ZuckerJournal Exp. Med. 35, pages 421 to 426.

- Parkow & Dunitresco
 Chem. Abstracts 5, page 112
- 89. Park Physiological - Veviews 3 no. 1, page 106.
- 90. Parker

Exp. Station Aecord 16, 1924.

91. Ponner, 1885

Cited by Forbes & Keith, Pechnical Bulletin 5, Ohio Agricultural -xperiment Station.

92. Powers

Journal Am. Med. Association 78, pages 159 to 165.

93. Orgler, A.

Endocrinology 6, page 724.

94. Ott, I., Scott, J. C. New York. Med. Journal 90, page 359.

95. Roezyns_i

Newer -nowledge of Autrition, McCollum page 324.

96. Reid, H. A., Ashton, B. C., Journal Department Agricultural New Zealand, Nov. 15, 1910.

97. Rose, A. R. Cited by Forbes ∞ Keith, Technical Bulletin 5, Ohio Agricultural Experiment Station, page 421

98. Rucklin

Zentr Biochem Beophys 19, pages 701 to 702. Chem. Abstracts 14, 1137.

99. Schabad, J. A.
2 klein Med. 69, pages 435 to 474. Chem. Abstracts 5, 935.

100. Schabad, J. A.

Jahet Kinderheilk 72, pages 1 to 17. Chem. Abstracts 6, 565

- 101. Schuenert, G, Schottke, A, Loetsch, E. Biochem. Z 36, pages 240 to 244, Chem. Abstracts 6, 245
- 102. Sherman and Papjenheimer Proc. Soc. Exp. Biol. Med. 13, page 193
- 103. Shipley, Park, McCollum, Simmonds Bulletin John Hopkins Hospital 32, pages 160 to 166
- 104. Shipley, Park, Simmonds and McCollum An. Journal Hyg. I page 492.
- 105. Sourell, J. Experiment Station Record 17, page 1193
- 106. Telfer, S. V. Journal Pjysiol 54 CV
- 107. Tisdell, F.F., Kramer, 3., Howland, J. Proc. Soc. Exp. Med. 18, pages 252 to 253.
- 108. Trendelenberg, Psul, Goelel, W. Arch. Exp. Pathol. Pharm. 69, pages 171 to 199
- 109. Underhill, Tileston and Bogert Journal Biol. Chem. 50, page 39.
- 110. Uhlenhuth, E. Journel Gen. Phys. 1, pages 315 to 322

