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# INTERRELATIONSHIPS OF COPPER, MOLYBDENUM AND SULFATE SULFUR IN BOVINE NUTRITION

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# INTRODUCTION

For over one hundred years it has been known that certain pastures in restricted areas of England caused a disease in cattle known locally as "teart". This disease was characterized by severe diarrhea, loss of body condition, and discoloration of the hair. In 1938 Ferguson, et al., attributed the cause of "teart" to a high molybdenum content of these pastures. Proof for this theory was provided when these investigators were able to produce identical symptoms of the disease by either drenching experimental animals with a molybdenum salt or by increasing the content of molybdenum in forage by adding molybdenum to the soil as a top-dressing.

Attempts to reduce the high molybdenum content of the forage were under way when Brouwer, et al., reported that a similar condition in Holland responded to large doses of inorganic copper. Although the "teart" pastures were shown to have what was considered an adequate level of copper, it was found that copper sulfate fed at the rate of 2 grams per day for cows and 1 gram per day for calves, or much smaller doses injected intravenously, would correct the toxic condition caused by the high molybdenum intake. Later it was found that the condition in Holland was not caused by high molybdenum levels but by inadequate copper intake by the animals.

In Australia about this same time investigators studying an enzootic disease of cattle, haematuria vesicalis,

observed that pastures on which these animals grazed, and the tissues of the animals, were abnormally high in molybdenum. Further analysis of the tissues of these animals revealed them to be lower than normal in copper. From this work in Australia and later work in New Zealand with a condition known as "peat scours" it was found that molybdenum has a profound effect on the metabolism of copper in the ruminant. This finding was further substantiated by the successful use of molybdenum salts in treatment of chronic copper poisoning. In effect these workers suggested that molybdenum toxicity is complicated copper deficiency.

Since the time that it was first shown that high molybdenum content of pastures was the cause of "teart" in England, molybdenum toxicity has been reported in Australia, New Zealand, the United States, Canada, Sweden and Japan. It was soon realized, however, that the conditions causing the toxicity in various locations differed greatly. The molybdenum content of the forage in the "teart" pastures of England was much higher than that of forage which caused the toxicity reported in Australia and New Zealand. In addition, the feeding of molybdenum salts was not successful in developing the symptoms of molybdenum toxicity unless it was carried out at a rate far in excess of the rates observed in forages in toxic areas.

These inconsistencies indicated that molybdenum toxicity was complicated by other unidentified conditions. Dick, in Australia, showed that molybdenum could reduce the copper level in the tissues of sheep only when a high level of sul-

fate also was present in the diet. Davis, in Florida, has shown that low phosphorus levels also will influence molybdenum toxicity. Even these findings did not explain the occurrence of molybdenum toxicity in some areas of the United States, England, and New Zealand where relatively low levels of molybdenum in forage are found. Still unexplained, also, is the critical question of how molybdenum exerts its toxic effects on the animal. The mechanism by which molybdenum and other complicating factors affect copper metabolism still is not understood. Whether molybdenum has toxic effects on the body in addition to its effect on copper metabolism is not known.

Losses in animal production in areas where molybdenum toxicity exists can be heavy. Although deficiency symptoms in most areas can be corrected by treating the animal with copper, or top-dressing the land with copper, such treatments are both time consuming and expensive. It was important that more of the answers to this problem be sought through research.

The staff of the U. S. Plant, Soil and Nutrition Laboratory, Ithaca, New York, has been studying conditions which cause poor growth and production of cattle in some western states. The symptoms found in these animals strongly suggested that either copper deficiency or molybdenum toxicity was involved. Examination of the forage in some of these areas revealed an adequate copper content and an above-normal molybdenum level but not high enough to suggest molybdenum toxicity. The presence of some complicating factor being the

cause of this problem was suspected.

As a result of the experience of the New Hampshire Agricultural Experiment Station in mineral metabolism problems and the unusually low trace mineral content of forage grown in New Hampshire, the Agricultural Research Service of the United States Department of Agriculture requested the New Hampshire Station to undertake a study of the molybdenum-copper-sulfate problem on a contract basis. This thesis represents the results of that study.

# LITERATURE REVIEW

## THE ABSORPTION AND METABOLISM OF SULFUR

For many years the absorption and metabolism of sulfur was considered to be important only because of its presence in certain essential amino acids. It was believed that sulfur, which represents 0.15 per cent of the body, could be utilized by the body only as part of the sulfur containing amino acids. In recent years, with the development of the practice of using urea as a protein replacement for ruminants, there has been renewed interest in the metabolism of sulfur.

Harris and Loosli (79) demonstrated that methionine added to the ration of lambs increased the utilization of urea. Later Loosli et al.(101) revealed that 10 essential amino acids were synthesized in the rumen by micro-organisms. This information indicated that, at least in the ruminant, the importance of sulfur in the diet is not confined to the organic sulfur found in amino acids consumed in the diet.

With the use of urea as a source of nitrogen for ruminants, diets could be formulated with a very low sulfur content. Feeding such diets were shown to cause sulfur deficiency. The symptoms of sulfur deficiency in ruminants were described by Starks et al.(148) and Thomas et al.(150) as poor appetite, loss of weight, weakness, excessive lacrimation, profuse salivation, cloudy eyes, emaciation and death.

It was surprising to note that sheep would continue to produce wool with normal sulfur content until death. Inorganic forms of sulfur such as sodium sulfate and elemental sulfur, as well as organic forms, were able to relieve the symptoms of deficiency, according to Starks et al (146,147,148). Ruminants receiving inorganic forms of sulfur added to sulfur deficient diets were shown by Thomas et al. (150), Starks et al. (146,148), Garrigus et al (70) and Lassiter et al. (94) to gain weight normally; and in sheep wool production was shown to increase.

Diets low in sulfur have caused a negative balance for both nitrogen and sulfur. The value of sulfur in increasing nitrogen retention in a low sulfur diet has been well established (70,94,146,147, 148,150). Efforts by Albert et al.(2), Lofgreen et al.(99) and Starks et al. (146,148) to increase nitrogen utilization by the addition of sulfur to diets already containing more than 0.2% sulfur, however, were not successful with lambs. Similar results were obtained by Jones et al (88) with dairy cattle. These workers found no increase in milk production or feed utilization when 1% sodium sulfate or 0.5% methionine were added to a ration containing 0.1% sulfur. For lambs, optimum levels of sulfur in the form of methionine, sulfate, and elemental sulfur have been suggested by Albert et al. (2) as 0.640 $\pm$ 0.048%, 1.270 $\pm$ 0.44% and 0.471 $\pm$ 0.028%, respectively. Loosli et al .(100) and Starks et al (146) have proposed a diet nitrogen to sulfur ratio of 15 to 1 and 20 to 1, respectively, for good nitrogen uti-

lization.

The ability of the ruminant to utilize cellulose as a source of energy also is influenced by the level of sulfur in the diet. In vitro studies by Hubbert et al.(87) and Trenkle et al.(151) indicated that there was no digestion of cellulose by micro-organisms when the level of sulfur was much below 10 p.p.m. Optimum cellulose digestion was obtained with sulfate sulfur levels between 10 and 20 p.p.m. and only slight depression of cellulose digestion was found when it was supplied at a rate of 1000 p.p.m. Sulfite sulfur gave optimum cellulose digestion when added to the medium in the range of 10 to 15 p.p.m. Levels above 30 p.p.m., however, inhibited cellulose digestion drastically. Organic forms of sulfur were required in much higher levels than inorganic sulfur for optimum cellulose digestion than inorganic forms indicating other beneficial properties than sulfur content. Also, high levels of sulfur, organically combined, did not impair cellulose digestion as was observed with inorganic sources forms of sulfur.

Studies on the fate of inorganic sulfur in the rumen by Anderson (8) and Lewis (97) indicate that sulfate and sulfite ions are reduced rapidly to sulfide by enzymes produced by the micro-organisms found in the rumen. Concentrations up to 97 micrograms per milliliter of sulfide were obtained by Anderson (8) when sulfate was fed at the rate equivalent to 2250 micrograms sulfur per milliliter. He found that sulfide administered as sodium sulfide disappeared from the rumen

very rapidly. It was found that with an initial level of 113 micrograms sulfide per milliliter, one half had disappeared in 30 minutes and all had been removed from the rumen in 2-1/2 hours. It was further shown that sulfide concentrations of the rumen were not affected by the administration of organic sulfur. According to Anderson (8) removal of the sulfide from the rumen is accomplished by direct absorption through the epithelium, and incorporation into amino acids by the bacteria of the rumen. Block et al. (16,17) demonstrated the synthesis of sulfur-containing amino acids by feeding sulfur-35 as sodium sulfate to cattle and goats. Radioactive sulfur labelled cystine and methionine were recovered from the milk of these animals as soon as three hours after administration of the sulfate. Experiments at Michigan by Emery et al. (58, 59) indicated inorganic sulfur is incorporated into organic form faster when added to a diet high in concentrates. In these experiments, bacteria incorporated labelled sulfur twice as rapidly into cystine as they did into methionine. Large amounts of glutathione also were recovered. Attempts by Emery et al. (58) to identify organisms which could utilize inorganic sulfur for amino acid synthesis, revealed that this ability is found in only a few strains and that even these strains would use organic sulfur more readily.

Evidence is now emerging that inorganic forms of sulfur may be used by an animal directly without first being synthesized into amino acids by rumen micro-organisms. Data on the removal of inorganic sulfur from the rumen strongly

indicates that large amounts are absorbed into the body in the inorganic form (8,52,92,99,158). Machlin et al.(103) reported finding radioactive sulfur incorporated into cystine in hens eggs after sulfur-35 was included in the diet as sodium sulfate. These investigators found 1.2% and 0.1% of the radioactive sulfur administered in the diet incorporated into cystine in the egg albumin and egg yolk, respectively. It was interesting to note that no radioactive sulfur was found in the methionine of the egg. In other work Machlin et al (104) reported that 60% of sulfur-35 injected as sulfate into a chick embryo was found incorporated into taurine ($NH_2CH_2-CH_2-SO_3H$). These experiments indicate that higher animals have the ability to reduc inorganic forms of sulfur and incorporate them into organic form.

Recent advances in enzyme investigations reveal many new functions of sulfur in metabolism. Evidence that sulfhydryl groups are essential to the activity of many enzymes such as xanthine oxidase, homogentisate oxidase, 3 hydroxyanthanilic acid oxidase, p-hydroxyphenyl pyruvate oxidase and protocatechuic acid oxidase has been reported by Mason (114) and Harris (80). The existence of sulfhydryl groups in the metabolic regulators, glutathione and insulin; and the presence of sulfur in the vitamins, thiamine and biotin, have been known for some time.

The existence of inorganic sulfate in the blood has been regarded for many years only as the end product of sulfur

metabolism. As more sulfate bound compounds have been discovered in the body, the importance of this ion in body metabolism has been reevaluated. Many of these compounds such as aromatic sulfonates and steroid sulfates appear to be products of detoxification. Sulfate compounds like brain lipids and mucopolysacharides, however, have been shown to be essential to body metabolism. The oxidation of organic sulfur in animals has been reviewed by Fromageot (67). The metabolism of sulfate has been reviewed recently by Gregory and Robbins (74). According to Weir and Rendig (158) serum inorganic sulfate levels reflect the total sulfur intake in sheep. When an animal has been fasted for a period of time, sulfate levels of the blood become high due to the metabolism of proteins for energy. Normal ranges of serum sulfate appear to be 2.0 to 5.0 mg. per 100 ml. of serum for sheep.

Although the excretion of sulfur occurs mostly in the urine, some is found normally in the feces. Recent balance studies by Muller et al. (127) indicate that half of the sulfur excreted by the bovine and two-thirds of that by sheep is in the form of inorganic sulfate in the urine. Dick (52), using sheep, found that 95 per cent of sulfur-35 fed as inorganic sulfate was excreted within 72 hours and only 1 to 3 per cent was found in the feces. Kulwich et al. (92) reported recovering 34% of the sulfur-35 in the feces and 52% in the urine in the four days following its administration as sodium sulfate. There appears to be no explanation for these differences. Balance studies by Knappen et al. (90) in Germany indicate that

the urinary sulfur cannot be used to obtain information on sulfur balance.

Sulfur in the diet as either organic sulfur or sulfate sulfur is not known to be toxic, even at very high levels. According to Anderson (8) the liver has the ability to oxidize sulfides at an estimated rate of .006 grams per hour per gram of liver. At this rate the production of sulfide in the rumen from inorganic sulfate contained in the ration at a level of 2% would not be harmful. The work of Starks et al.(146) indicates that the palatability of both inorganic sulfate and elemental sulfur are such that animals cannot be made to consume levels higher than 1.78% and 0.6% respectively.

## COPPER ABSORPTION AND METABOLISM

The presence of copper in animal tissues has been known for over a hundred years, according to Underwood (152). Like many other elements, little attention was given to its metabolism because it appeared in very low concentrations and was thought only to be a biological contaminant. In 1928 Hart etal.(81) created the first real interest in copper metabolism in mammals when they found that copper was required in addition to iron for the formation of hemoglobin. Details of the progress in copper metabolism in animal research have been the subject of many recent reviews by Underwood (152,153), Mason (113), and Singer et al.(144) and should be consulted for extensive details.

Since copper was first shown to be essential for several metabolic processes in animals, deficiencies of this element have been reported to occur naturally in many parts of the world (4,21,31,39,152). Ruminants, particularly cattle and sheep, are more sensitive to copper deficiency than the non-ruminant species. The symptoms most commonly described in ruminants are anemia, depressed growth, achromotrichia, gastro-intestinal disturbances (diarrhea), poor bone formation, and ataxia. Alopecia has been described in cattle, and the wool in Merino sheep lacks the normal crimp.

The known functions of copper in the body have not explained satisfactorily the various symptoms which have been described for copper deficiency. Copper is now known to be

contained in the enzymes phenol oxidase, p-hydroxyphenylpyruvate oxidase, diphenylalanine (dopa) oxidase, butyrylcoenzyme A dehydrogenase, uricase, laccase, and cytochrome oxidase. Information on the properties of these enzymes has been reported by Mahler et al. (106,107,109), Singer et al. (144), and Waino (157). It is quite surprising that only the activity of cytochrome oxidase has been shown to be affected by experimentally induced copper deficiency. As early as 1939 it was noted by Schultze (140) that cytochrome oxidase activity was reduced during copper deficiency. Another enzyme, dopa oxidase, which is required to oxidize dihydroxyphenylalanine (dopa) and tyrosine to the pigment melanin is suspected of being reduced in activity during copper deficiency, thus explaining the loss of hair color.

It is interesting to note that the function of copper in hemoglobin formation, although it was the first evidence that copper was required in metabolism, still remains obscure. A recent review by Matrone (115) concerning the inter-relationship of iron and copper states that only in the formation of hemoglobin is there an apparent interaction. Work done with calves by Matrone at North Carolina (115) indicates that iron absorption is not impaired by copper deficiency in contrast to the lower absorption of iron in copper deficient swine as reported by Gubler et al. (75). The only consistent finding with respect to the addition of copper to copper deficient animals is an increase in the per cent of reticulocytes, as reported by Matrone (115). This increase in per cent of

reticulocytes was demonstrated in dogs, rats, rabbits and swine, but no similar work involving ruminants has been reported.

Copper has been reported by McCutcheon (116) to be an additional co-factor in the exchange of formate with inosenic acid. The exact function copper has in this reaction is unknown.

Bone formation abnormalities have been noted in copper deficient sheep, cattle, pigs, dogs, and chickens by Underwood (152). Studies with both dogs, by Follis (66), and swine, by Bush (24), indicate a similarity between the changes in bone formation during copper deficiency and scurvy. In both cases there is a normal growth of cartilage but the deposition of bone in the cartilage matrix has been stopped. There is as yet no indication as to how copper deficiency interferes with the functional activity of the osteoblasts. Deficiencies of other elements generally stop both the osteoblastic and chondroblastic activities together, thus resulting in shorter bones but with normal bone conformation. The bone growth in copper deficiencies is of normal length but the conformation is altered due to the softer nature of the bone.

The symptom which is most readily observed in copper deficiency of ruminants is the change in hair and wool. According to Hayashi (82) and Cunningham (31) achromotricia and alopecia are commonly reported in cattle. The texture of such hair has been described by Cunningham (31) as starring and rough. In Merino sheep the crimp normally found in wool is absent and the physical properties are inferior. Burley et

al. (22) have shown that such wool has a greater number of sulfhydryl groups and fewer disulfide groups than normal wool. More recent evidence by Burley et al. (22) indicates that copper deficiency interferes with the arrangement of polypeptide chains in keratin. These workers have shown that wool from copper deficient sheep contains more N-terminal glycine and alanine and sometimes more N-terminal serine and glutamic acid than normal wool. Cunningham (36) has reported that the copper content of hair was not changed by the level of this element in the diet but that the copper content of hair varied with hair color. Copper-containing proteins which have been shown to catalyze the aerobic oxidation of L (-) cysteine, L(-) tyrosine, and $\beta$ -3,4 dihydroxyphenyl alanine have been isolated from sheep's hide by Scaife (139).

Cardiac failure has been reported by Bennetts (14) in cattle grazing on pasture extremely low in copper in south-western Australia. The condition was claimed to be caused by atrophy of the myocardium with replacement fibrosis. A similar condition has now been reported in pigs by Gubler et al (75). These workers also reported cardiac hypertrophy to be common in copper deficient swine and suggested its cause to be an attempt to compensate for the loss of tissue respiration which occurs as a consequence of reduced cytochrome oxidase activity. Cardiac failure is thought to result from stress on a depleted myocardium.

The assimilation of dietary copper in the ruminant is influenced by many conditions and generally amounts to only a

few per cent of that consumed. Comar et al.(25,26), feeding copper-64 as the nitrate salt, found that 75 per cent of the activity was recovered in the feces and 3% was found in urine after five days. In these experiments about 10 per cent of the dose was found in the tissues. In spite of the very low amounts of copper-64 absorbed, 0.15 per cent of it was found in the blood only 19 minutes after being administered orally. It is surprising that only limited research has been carried out in determining the availability of different forms of copper to the animal, and even less research has been undertaken to learn the mechanism of its absorption. Schultze et al.(141,142), using rats as an experimental animal, showed that copper in the form of copper glycine amide biuret, alanine amide biuret, hemocyanin, caseinate, aspartate, citrate, nucleinate, pyrophosphate, citrate, and cysteine cuprous mercaptide could be used by animals fed a copper deficient diet, whereas, copper hematoporphyrin and sulfide were not utilized. These investigators could not establish any information on the form in which such copper was absorbed because the copper was shown to be released from many of these compounds in dilute acid media. Recently Mills (123) has investigated the forms in which copper is found in plants and the ability of the animal to assimilate plant copper. These investigations showed that only 8 to 22 per cent of the copper in the plant was in a form which was soluble in water or organic solvents. The remaining copper was shown to be released from its organic form only in a solution with a pH lower than 2. This pH is far

lower than is found in the digestive tract of animals. In studies with rats, however, this investigator showed that plant-contained copper was more readily absorbed than was inorganic copper of the same concentration. This would indicate that copper can be transported through the intestinal mucosa as an organic complex as well as the free ion. Further support for this theory can be found in the observation of Mitchell and Tosie (125) that copper ions are very rapidly absorbed by rumen micro-organisms and incorporated into an organic form. Mills (123) states that copper in an organic complex would pass the membranes more rapidly because it would not be free to combine with complexing agents found in the membrane. It is interesting to note that the quantity of free copper in plants showed seasonal variations. Davis (40) recently reported isolating from grasses a complexing agent which renders copper completely unavailable to animals. This worker suggests such a complexing agent is responsible for the occurrence of copper deficiency in areas where seemingly adequate copper is present.

The total amount of copper found in the tissues of an animal is quite small. According to Underwood (152) mammals at birth generally contain from 3 to 7 p.p.m. copper but the quantity goes down in the adult to 2.0 to 1.5 p.p.m. Data on the levels of copper for sheep and cattle are not available, but it is expected that the copper contents of these animals are higher than for other mammals because the analysis of many individual tissues shows that they contain more copper. Although there is relatively little copper found in the body,

the distribution is very wide and there is good evidence to believe that copper can be found in every cell of the body. The liver of most animals, particularly sheep and the bovine, have high concentrations of copper. Normal ranges of copper in these species are reported by Underwood (152), and Marston (112), to be 100 to 400 p.p.m. Copper levels in the blood of sheep and cattle also have been found to vary over a wide range. Underwood (152) and Marston (112) have summarized values from many investigators and set the normal range at 80 to 120 micrograms per 100 ml. of blood. Values as low as 50 and as high as 180 micrograms per 100 ml. of blood, however, are commonly found. The lever of copper in the blood has been shown to be affected greatly by many factors. Low levels of copper in the diet have been shown to cause decreased blood levels by Cunningham (31) and Bennetts et al. (14). In humans, pregnancy has lowered blood copper values, but no data on sheep or cattle are available. Heavy infestations of internal parasites have been shown by Bremner (19), Seekles (143) and Gibson (71) to lower both blood and liver concentrations of copper. The action of molybdenum in lowering blood copper levels will be discussed later.

Many balance studies have shown that the chief route of excretion of copper is through the intestinal tract. Comar et al.(25,27) reported that when copper-64 was injected intravenously into cattle only 3 per cent was excreted in the urine and an equal quantity in the feces. After reviewing the literature, Underwood (152) reported that 90 per cent of the dietary copper appears in the feces. Copper is excreted into

the intestinal tract by way of the bile.

The level of copper required in the diet for normal animal health is influenced greatly by many factors. In most areas of the world, levels of copper from 4 to 6 p.p.m. on a dry weight basis are adequate for normal health of ruminants, according to Cunningham (31), Marston (112), and Underwood (152). The form of copper in the diet has been discussed already in relation to assimilation and obviously is related to the level needed in the diet. The level of molybdenum in the diet, which will be discussed later, has a profound effect on the copper requirement. Recently, there has been growing evidence that zinc also has an effect on the copper requirement. High levels of zinc in the diet have been reported by Cox et al,(29) to require higher levels of copper. Zinc with lead has been shown by Gray and Ellis (73) to cause a lowering of hemoglobin values in rats but this could be overcome by copper supplementation. Earlier, Cunningham (31) reported zinc had no effect on copper metabolism. This investigator reported also that tungsten, manganese, vanadium, chromium, rhenium, uranium and tantalum have no effect on copper requirements.

Additional copper has been administered to both sheep and cattle successfully, both as copper salts added to the diet or as an injected cerate. Reports of copper being injected as a cerate indicate up to 90 per cent utilization in work reported by Branion (18) and Cunningham (31). Copper glycinate, given at the rateof 400 mg. for cattle and 150 mg. for sheep, has been used successfully for many years. A

recent report by Cunningham (35) on the use of cupric bis-8-hydroxyquinoline 5:7 disulfonic acid salt of tetradiethylamine in cerate injections showed copper in this form was slowly absorbed, as desired, and caused no harmful effects to the tissues, but copper citrate caused great tissue damage when used as a cerate. Branion (18) reported that copper administered as a cerate must be given three times per year to maintain animal health. Dent et al. (42) and Cunningham (30) reported some success in meeting the requirements of cattle for copper by offering a free choice of a mineral mixture containing copper. Top-dressing of 10 to 20 pounds of copper sulfate per acre on forage has met with only moderate results in raising the copper content of the forages according to Branion (18) and Cunningham (33). Such a practice has proved to be very uneconomical. The feeding of copper sulfate at the rate of 3 grams for cattle and 0.5 grams for sheep has been effective but such treatment is required 3 times per week.

Copper salts taken in large doses have been known to be toxic for many hundreds of years, but only recently has the recognition of chronic copper poisoning under natural conditions occurred. Copper poisoning has been described by Dick (55) and Underwood (152) both in sheep and cattle grazing on pastures having a high copper content. In many cases of such poisoning the levels of copper in the forage were only 15 p.p.m. Kidder (89) demonstrated that hemolytic jaundice could be caused by large doses of copper over a long period of time. Branion (18) reported that feeding 3 g to 5 g per

day for a period of 3 years, however, was not harmful. Sheep are much more susceptible to copper poisoning than are any other species. Frequent accidental poisoning with orchard sprays and copper salts used for parasitic treatment has been reported by Allcroft (7) and Underwood (152). According to Dick and Bull (55) copper poisoning is characterized by sudden hemoglobinemia and hemoglobinuria usually accompanied by the occurrence of icterus which increases rapidly.

Excess copper intake may also cause reduction in feed utilization by ruminants. Studies carried on in vitro by McNaught et al.(117) indicated that levels of copper above 10 p.p.m. greatly reduced protein digestion. In other in vitro studies, Hubbert et al.(87) showed a level of 1.5 p.p.m. copper as the sulfate reduced cellulose digestion and 2.5 p.p.m. stopped digestion of this substance completely.

## THE ABSORPTION AND METABOLISM OF MOLYBDENUM

The discovery that molybdenum is part of xanthine oxidase was made by two groups of investigators, de Renzo et al.(43) and Richert and Westerfeld (134), working independently. Mahler et al.(110) in 1954 reported that molybdenum is a part of liver aldehyde oxidase. The third molybdenum enzyme, hydrogenase, was reported by Shug et al.(144) also in 1954. Although these investigators were able to show increased enzyme activity with increased molybdenum levels in the diet, the element was not considered essential because growth and purine metabolism were not affected by changes in dietary levels. Tungsten, as the tungstate, has been shown by de Renzo (45) to reduce the molybdenum level in the tissues, presumably in the role of an antagonist. Higgins et al.(85), feeding tungstate to rats, showed that the xanthine oxidase activity could be reduced by three-quarters without changing the uric acid excretion. In this experiment the xanthine oxidase production of the intestines was completely stopped. These same investigators showed that feeding sodium tungstate to chicks caused a 25 per cent decrease in growth in five weeks. This treatment reduced the secretion of uric acid which was replaced by xanthine and hypoxanthine. Molybdate added to the diet overcame all these effects. More recent work with rats by Richert et al.(135) showed that both xanthine oxidase and aldehyde oxidase activity could be reduced by feed-

ing tungstate, to a point where normal metabolism did not occur.

Claims that molybdenum is an essential element have gained support in recent years. Ried et al.(132), in 1957, was able to increase the growth of turkey poults by adding molybdenum to the diet. Leach and Norris (96) were able to show a similar growth response with chicks but were unable to confirm a response in turkeys. Ellis et al.(57) reported a beneficial effect in sheep when 1 and 2 p.p.m. of molybdenum were added to a basal diet containing 0.36 p.p.m. molybdenum. These sheep made significantly faster gains with the added molybdenum than were made on the basal diet. The addition of the molybdenum to this diet significantly increased cellulose digestion without altering digestion of other components of the ration. It appears now that low molybdenum levels can interfere with normal purine metabolism in sheep. Askew (10) has demonstrated that low molybdenum levels in the diet contribute to xanthine calculi formation in the kidneys of sheep.

Molybdenum is absorbed readily as the molybdate ion. Comar et al.(27), using molybdenum-99, found that only 34 per cent was recovered in the feces after being fed as the molybdate ion. These workers found that 45 per cent of the molybdenum-99 was excreted in the urine. These results are in good agreement with those of Dick (52). This investigator has demonstrated that molybdenum levels in the blood and tissues of the animal reflect the animal's intake of the ele-

ment. Molybdenum will increase to a certain level in the body, depending on intake, and then remain constant. If the molybdenum level in the diet is decreased the tissue concentration will decrease. The bone, kidney, spleen, liver, and blood show the greatest concentration of this element according to Anderson (8) and Comar et al.(27). Molybdenum excreted in the milk appears also to reflect the intake. Comar et al.(27) found that about 2 per cent of a dose of molybdenum-99 was excreted in the milk of a cow.

The molybdenum content of the forage is dependent on the molybdenum content of the soil, soil pH, and species of plants. According to Barshad (11) high pH favors greater accumulation of molybdenum in plants. Legume plants growing in the same field as grasses will contain many times as much molybdenum (11,18,64,84). In many cases legume plants, particularly white clover, have been shown to contain 160-260 p.p.m. molybdenum. Normal contents of plants, however, are assumed to be in the range of 1 to 2 p.p.m. throughout the world (11, 18,64,84). Ferguson (64) reported that 70 to 80 per cent of the molybdenum in grass, 40% of that in hay and 10% of that in dead winter herbage was water soluble.

Molybdenum was first found to be important in animal nutrition because of its toxic effect. Ferguson et al.(62) in 1938 reported that molybdenum was responsible for the sickness of cattle and sheep pastured on the "teart" areas of England. Since that time molybdenum toxicity has been reported by Cunningham (31) in New Zealand, by Dick (51) in

Australia, by Hayashi (82) in Japan, Hallgren et al.(77) in Sweden, Cunningham et al.(30) in Canada, and by Britton and Gross (20), Robinson and Dever (136), and Davis (39) in the United States. The symptoms of this toxicity in cattle and sheep have been described as follows: achromotricia, ataxia, unthriftiness, debilitating spring scouring, anemia, increased susceptibility to internal parasites, proneness to fracture of the bones, loss of reproductive capacity, low serum copper and low liver copper (4,30,31,52,82,112, 118,149).

Hair and wool of ruminants suffering from molybdenum toxicity have shown many changes. In adult cattle black hair has changed to red or gray, and alopecia has occurred around the eyes, according to Cunningham (31), Ferguson (63,64) and Hayashi (82). Hayashi (82) has reported that individual hairs of these animals have been shown to have alternate clear and pigmented areas and the clear areas show less strength than other areas. Cunningham (31) and Underwood (152) have reported that black wool of sheep on toxic levels of molybdenum shows loss of pigmentation. Such wool lacks quality, and because the normal crimp is absent, it has a stringy appearance. Ataxia generally occurs in lambs between the ages of 3 and 4 months, but occasionally young cattle have been seen with the disorder, according to Cunningham (31). Allcroft (4) and Underwood (152) report that the condition is caused by diffuse symmetrical demyelination of the central nervous system.

The loss of reproductive capacity has been observed experimentally in bulls, by Hayashi (82,83), and Thomas and

Moss (149). In these experiments spermatogenesis stopped entirely and the individuals completely lacked libido. Histological examinations of the testes showed that there was damage to the seminiferous tubules and inflammation of the interstitial tissue. Copper administration has been shown to correct all symptoms of the disease with the exception of advanced ataxia, but copper has been shown to prevent this condition from occurring.

# THE INTERRELATIONSHIP OF MOLYBDENUM, COPPER AND SULFATE SULFUR

The original description of molybdenum toxicity by Ferguson (48) implied a relationship of this element with copper. Not only were many of the symptoms of this disorder the same as those described for copper deficiency, but the fact that copper sulfate was a successful treatment indicated a possible relationship. Dick and Bull (55), while studying copper metabolism, discovered that copper levels of sheep livers were below normal when a large amount of molybdenum was found in the diet. These workers used molybdenum to correct copper toxicity. Molybdenum toxicity, both in New Zealand (Cunningham, 31) and Florida (Davis, 39), was described as occurring with levels of molybdenum intake between 7 and 10 p.p.m. Davis (39) later reported that levels of molybdenum as low as 2 p.p.m. could cause molybdenum toxicity. It became apparent that the molybdenum toxicity in certain areas was caused by much lower levels of molybdenum than were originally described by Ferguson et al.(63). Furthermore, attempts to induce experimentally molybdenum toxicity required much higher levels than occurred naturally in toxicity areas.

According to Ferguson et al.(63) molybdenum toxicity in England was caused by very high levels of the element. Plants on the "teart" pastures contained from 20 to over 150 p.p.m. of molybdenum. Animals on these pastures are affected

by the high molybdenum content within a week, or two weeks, after the start of grazing. Most prominent symptoms include severe diarrhea, emaciation, poor production, and changes in hair color. Levels of molybdenum below the range indicated do not give these symptoms. Feeding molybdenum as molybdate at levels of 100 p.p.m. or more has been shown by Branion (18), Ferguson et al. (63), Hayashi (82), and Hallgren et al. (77) to cause the same symptoms.

Toxicity in New Zealand, Australia and Florida occurred on levels as low as 2 to 10 p.p.m. of molybdenum, according to Branion (18), Cunningham (31), Davis (39), Dick (52), Miller et al. (118), and Wynne and Clymont (159). The symptoms of the toxicity in these areas are not shown to occur quite as rapidly as those described in England but appear to be just as severe. Diarrhea, emaciation, anemia, and hair color changes are the first symptoms to be noticed. Analysis of the blood and liver for copper show levels to be below normal. Copper levels as low as 0.2 p.p.m. in the blood have been reported by Branion (18) and Cunningham (31). Molybdenum toxicity in these areas is most frequently associated with peat or muck soils. According to Cunningham (31), in New Zealand the problem was first encountered when a large program of reclaiming swamp land was carried out and cows and sheep were grazed on this new land.

Dick (49), while studying enzootic jaundice caused by high levels of copper, tried to reduce the copper in the animal with molybdenum and found that molybdenum was effective

in reducing copper with alfalfa but not with oat straw as forage. In later work this investigator indicated that the difference between the two diets which caused the effect was a high level of sulfate in the alfalfa hay. Dick (46,47,48, 49,50,51,52,53,55) further showed that supplements of neither sulfate nor molybdenum alone had any effect on copper metabolism separately, but together they caused all of the symptoms of molybdenum toxicity in sheep. Similar results with cattle have been reported by Cunningham (31) and Mylrea (128). It is interesting to note that studies by Kulwick et al.(93) using copper-64 in swine and rats, showed an increased copper deposition when molybdenum was added to the diet. These investigators suggest that copper may be unusable by the animal.

In studying the effects of sulfate on molybdenum metabolism, many investigators have become quite perplexed. In rats and other monogastric animals, sulfate was shown by Miller et al.(120,121), Reid et al.(131),and Van Reem (155, 156) to protect against the toxic effects of high molybdenum intakes in the diet. High levels of methionine were shown to have the same protective effect on molybdenum toxicity in rats, according to Gray and Daniel (72). Experiments with sheep by Dick (52) indicate that the molybdenum content of the blood and tissues can be reduced by the addition of inorganic sulfate to the diet. A single dose of sulfate was shown to increase greatly the level of molybdate excreted in the urine. Similar results have now been reported in cattle by Cunningham (37). Increases in the molybdenum content of both plasma and

blood cells were reported when only molybdenum was added to the diet but when sulfate was added with molybdenum the plasma alone showed an increase.

There is very little evidence to suggest that sulfate alone has any effect on copper metabolism. Mylrea (128) has reported that the liver copper of cattle was reduced from 144 p.p.m. to 63 p.p.m. by the addition of 1% gypsum to the diet. Similar experiments using other forms of sulfate have not shown any effect, according to Dick (52) and Cunningham (37). Singer et al. (144), in a review on copper enzymes, report that sulfate has an inhibitory effect on the enzyme laccase.

Indications of the phosphorus levels also adding to the severity of molybdenum toxicity have been reported by Cunningham (31) and Davis (39). In these situations normal phosphorus metabolism was shown to be altered, and bone fractures were frequent. In all cases, however, the phosphorus level of the diet was shown to be below what was considered an adequate level. Excess phosphorus has not been found to relieve molybdenum toxicity, according to Davis (39). Conflicting evidences to the effects of manganese in relieving toxicity caused by high molybdenum and sulfate intake has been found. Dick (51) has reported that manganese would counter the effects of high levels of sulfate and molybdenum. Mylrea (128) was unable to show similar results with cattle.

Despite the relations described above, certain areas

are known where animals show all the symptoms of molybdenum toxicity or copper deficiency, while in other areas identical conditions seem to exist without causing this malady. In England, Allcroft et al. (4) showed identical conditions of molybdenum, sulfate, and copper levels on two locations, but one had the symptoms of molybdenum toxicity in sheep and cattle and the other did not. In these areas copper was frequently as high as 24 p.p.m., molybdenum levels were about 7 p.p.m., and sulfate levels were as high as 1.1 per cent inorganic sulfate. Furthermore, even with high levels of sulfate, all of the symptoms of molybdenum toxicity could not be produced experimentally with levels of molybdenum below 50 to 100 p.p.m., as was shown by Branion (18) and Hayashi (82). A recent review of the conditions found in the Everglades area of Florida by Kretschmer and Beardsley (91) states that molybdenum has never been conclusively proven to be the causative agent for what was supposed to be molybdenum toxicity in this area. A response to copper supplementation, however, was well demonstrated. This author pointed out that many times the condition could be corrected by phenothiazine treatment, thus indicating that the problem was a copper deficiency brought on by internal parasites. Copper levels in the forage of this area are from 3 to 8 p.p.m. Such data as described here, indicate that there are still unidentified complicating factors involved in molybdenum toxicity.

That there exists a relationship among copper, molybdenum and sulfate has now been established by the work of Dick (52), Cunningham (37), and Mylrea (128) in the bovine

and sheep. The exact levels at which the intereaction affects animal health have not been established, nor has the exact mechanism by which this interrelationship operates been proven. Dick (52) has proposed that inorganic sulfate at high levels interferes with the transport of molybdenum across membranes. Such a membrane at which molybdenum is blocked would then interfere with the copper transport. Under this theory copper deficiency would occur due to reduced copper absorption from the alimentary tract. Further, this author suggests in the case of high levels of molybdenum, copper would be prevented from leaving the storage areas or the blood to enter the tissues where it is needed and thereby produce the symptoms of copper deficiency without lowering the copper levels of the blood or the liver. No details have been proposed by the proponent of this theory as to how sulfate stops molybdenum from passing through the membrane or how the latter in turn prevents copper from crossing a membrane. This theory, however, explains most of the observed conditions as to why blood and liver concentrations of copper vary so greatly when toxicity is caused by both high and low levels of molybdenum. Recent results of Cunningham (36) on the copper and molybdenum contents of hoof and hair of animals fed high levels of molybdenum and sulfate shows that molybdenum decreased the copper content of hair, wool and hoof, both with and without sulfate. The molybdenum content of the hair, wool and hoof was affected by the level of this element in the diet, but sulfate added to the diet decreased the level of molybdenum in these tissues.

This information would seem to contradict Dick's (52) theory because molybdenum alone seemed to have an effect on the movement of copper. It was interesting to note further that the copper level changes in the hair did not affect color, according to Cunningham (36).

A second theory on the copper, sulfate, and molybdenum interrelationship has been proposed by Mills et al.(124) and has been extended by Halverson et al.(78). Mills et al.(124) reported that high molybdenum levels depressed the sulfide oxidase levels in the liver of the rat by 56 per cent. They then proposed that high sulfide levels may be produced from reduction of sulfate in the rumen. This sulfide would be toxic to the animal because of a limited ability to oxidize this compound. The depression of sulfide oxidase activity was explained on the basis of the fact that both copper and hypoxanthine are required as co-factors. Halverson et al. (78) reported that cystine did not reduce the toxic effects of molybdenum as methionine and inorganic sulfate were shown to do. These investigators reported that high levels of cystine were not oxidized by the rat because of reduced sulfide oxidase activity, and that this cystine formed a complex with copper rendering the latter unavailable to the animal. Halverson et al.(78) suggested that small amounts of sulfide in the tissues, even though undetectable, could form insoluble copper sulfide and thus render the copper useless to the animal. It is interesting to note that Van Reem (155) found that feeding potassium thiomolybdate ($K_2MoS_4$) to

rats was extremely toxic. Singer et al.(144) have reported that hydrogen sulfide can be shown to inhibit the activity of copper enzymes by complexing with the copper.

The question of whether the interaction with copper is the only toxic effect molybdenum has on an animal has been raised several times, especially by Branion (18) and Miller et al.(118). This theory that molybdenum is toxic in some other system is strongly supported by the research of Branion (18) in which high levels of molybdenum caused the symptoms of toxicity to appear before any changes in copper levels of the blood and liver occurred. Further evidence that molybdenum per se is toxic is provided by the observation of Scaife (139) that the molybdate ion inhibited the action of three copper containing enzymes isolated from the skin of sheep. These enzymes, which were referred to earlier, catalyze the aerobic oxidation of L (-) cysteine, L (-) tyrosine, and dopa ( 3,4 dihydroxyphenylalanine).

## EXPERIMENTAL PROCEDURE

The experimental procedures used to study the interrelationship among copper, molybdenum,and sulfate sulfur will be described as four separate experiments for the sake of clarity. These will be designated as Experiments I, II, III, and IV.

### EXPERIMENT I

The purpose of Experiment I was to investigate the effects of various levels of molybdenum supplementation on milking first calf heifers fed a diet relatively low in copper, molybdenum,and sulfate. Fifteen springing Holstein heifers purchased in the last summer of 1957 from dairy farms were brought to the Dairy Nutrition Barn at the University of New Hampshire Agriculture Experiment Station for this experiment. The experiment was designed with three replications, each having five animals on different treatments. The treatments consisted of a control and levels of 5, 10, 20, and 50 p.p.m. of molybdenum added to the total ration. Table 1 illustrates the experimental design. Each animal was placed on experiment at parturition and was assigned to a treatment at random. All animals were kept on experiment for four consecutive periods of 75 days each.

The animals were fed a 17 per cent crude protein concentrate mixture, which was low in copper, molybdenum, and sulfur, according to the Morrison Feeding Table (126) with

TABLE 1

Design of Experiment I

| Number of animals | Molybdenum fed | Sulfate sulfur | Other ions fed | Days spent in Pole barn | Days spent in Stanchion barn | Days spent in Pasture |
|---|---|---|---|---|---|---|
| 3 | None | None | None | None | 300 | None |
| 3 | 5 p.p.m. | None | None | None | 300 | None |
| 3 | 10 p.p.m. | None | None | None | 300 | None |
| 3 | 20 p.p.m. | None | None | None | 300 | None |
| 3 | 50 p.p.m. | None | None | None | 300 | None |

the adjustment for first calf heifers. The composition of this mixture is found in Table 2 of the Appendix. Hay consisting predominantly of grasses grown under high fertilization was fed ad libitum. Concentrate and hay weights were recorded, and refuse was weighed back three times per week and recorded. Molybdenum in the form of sodium molybdate ($Na_2MoO_4 \cdot 2H_2O$) was used as the molybdenum supplement. The amount to be fed was calculated on a weekly basis using the previous week's total feed consumption as the basis for the calculation. The weekly allowance of the molybdate salt was weighed out on an anlytical balance, diluted with cane sugar, and then divided into seven portions. Each portion was placed in a quarter-ounce gelatin capsule and one of these was administered each day. A plain salt block was accessible to each animal and individual automatic drinking cups provided water.

The animals were exercised for a few hours each day in a small lot when weather permitted. Heat periods were recorded and each animal was serviced artificually during the first heat occurring after 60 days post-partum. Pregnancy examinations were made once per month until an animal was found to be with calf. The cows were milked twice a day by machine and the milk weights were recorded. At the end of 300 days on experiment the animals were slaughtered and a section of liver was saved for analysis.

## EXPERIMENT II

The purpose of Experiment II was to study the effects of various levels of molybdenum supplementation on milking first calf heifers fed a diet high in sulfate sulfur and relatively low in copper and molybdenum.. Fifteen springing first calf heifers were purchased for the experiment in the late summer of 1958 from dairy farms and brought to the Dairy Nutrition Barn. The experimental design of this experiment was planned to be identical with that of Experiment I except that 0.3% sulfate sulfur was added to the diet. As the experiment progressed, however, the following changes were made: At the end of 150 days on experiments two of the three animals receiving 5 p.p.m. of molybdenum showed positive reactions to a blood test for brucellosis and were removed from experiment and slaughtered to prevent further spread of the disease. The remaining 5 p.p.m. animal and the three 10 p.p.m. animals were carried on experiment for three consecutive periods of 75 days (275 days) and then divided into two new treatments with two animals receiving 100 p.p.m. and two getting 200 p.p.m. of supplemental molybdenum. These treatments were continued for one period of 75 days. At the end of four consecutive 75-day periods (300 days) on experiment, two animals from the control group and two animals from the 50 p.p.m. level were continued on treatment through the remainder of the first lactation, the ensuing dry period, and four consecutive 75-day periods during the second lactation. The

design of Experiment II is shown in Table 2.

The animals were placed on experiment and fed hay and concentrates of the same quality and in the manner as described for Experiment I except for the addition of potassium sulfate to the concentrate mixture. The feeding of sodium molybdate was carried out according to the same procedures as were used in Experiment I. The procedures followed with respect to milking, breeding, and housing were the same as for Experiment I with the exception of exercise. The animals in Experiment II were exercised for the first 150 days but this practice was discontinued in an attempt to prevent the spread of brucellosis.

## EXPERIMENT III

Experiment III was designed to investigate the effects of stress, pasture, and the addition of some ions which are found only in small amounts in New Hampshire grasses, on the development of toxicity in cattle fed moderately high levels of molybdenum and sulfate sulfur.

Fifteen first calf heifers were purchased in the late summer of 1959 and placed on experiment in the same way as in Experiments I and II. This experiment was designed with three replications of five different treatments. The treatments in each replication were as follows: 0.3 per cent sulfate sulfur - outdoors; 50 p.p.m. molybdenum - outdoors; 0.3% sulfate sulfur and 50 p.p.m. molybdenum - outdoors; 0.3% sulfate sulfur and 50 p.p.m. molybdenum - indoors; 50 p.p.m. molybdenum,

TABLE 2

Design of Experiment II

| Number of animals | Molybdenum fed | Sulfate sulfur | Other ions fed | Days spent in: Pole barn | Days spent in: Stanchion barn | Days spent in: Pasture |
|---|---|---|---|---|---|---|
| 3 | None | 0.3% | None | None | (1)*** 300<br>(2) 600 | None<br>75 days |
| 3 | 5 p.p.m. | 0.3% | None | None | 150 | None |
| 3 | 10 p.p.m. | 0.3% | None | None | 225 | None |
| 3 | 20 p.p.m. | 0.3% | None | None | 300 | None |
| 3 | 50 p.p.m. | 0.3% | None | None | (1) 300<br>(1) 675<br>(1) 600 | None<br>None<br>75 days |
| 2* | 100 p.p.m. | 0.3% | None | None | 75 | None |
| 2* | 200 p.p.m. | 0.3% | None | None | 75 | None |

* Animals were either 5 p.p.m. or 10 p.p.m. Mo. plus 0.3% Sulfate before this treatment.

*** Number in parenthesis indicates number of animals.

0.3% sulfate sulfur, 5 p.p.m. arsenic, 5 p.p.m. lead, and 100 p.p.m. rubidium — indoors. The designation "outdoors" and "indoors" refer to the conditions under which the animals were housed other than during the milking period. The "outdoor" animals were kept in a lot and had access to an open pole barn. The "indoor" animals were housed in a stanchion barn. After 225 days on experiment one animal from each treatment was selected at random and placed on pasture instead of receiving hay. With the exception of this change, all other conditions of the experiment remained the same for 300 days. The design of the experiment is shown in Table 3.

At the end of 300 days on Experiment III four animals which received 50 p.p.m. molybdenum and 0.3% sulfate sulfur and showed extensive hair color changes, were selected to receive additional copper. These animals were continued on their experimental diet but two received 10 p.p.m. copper and two received 20 p.p.m. copper added to the diet as copper sulfate. This treatment was then continued for 75 days at which time serum and liver samples were taken.

The animals received hay and concentrates of the same description as were used in Experiment I. The animals housed outdoors received hay in an open rack as a group and individual hay consumption data was not possible. Hay was fed to animals housed indoors in the same manner as described for Experiment I. All animals were milked and fed concentrates twice a day. The amount of sulfate required in the diet was supplied in the form of potassium sulfate mixed with the con-

TABLE 3

Design of Experiment III

| Number of animals | Molybdenum fed | Sulfate sulfur fed | Other ions fed | Days spent in Pole barn | Stanchion barn | Pasture |
|---|---|---|---|---|---|---|
| 3 | None | 0.3% | None | (2)* 300<br>(1) 225 | None<br>None | None<br>75 |
| 3 | 50 p.p.m. | None | None | (2) 300<br>(1) 225 | None<br>None | None<br>75 |
| 3 | 50 p.p.m. | 0.3% | None | (2) 300<br>(1) 225 | None<br>None | None<br>75 |
| 3 | 50 p.p.m. | 0.3% | None | None | (2) 300<br>(1) 225 | None<br>75 |
| 3 | 50 p.p.m. | 0.3% | 5 p.p.m. Pb<br>5 p.p.m. As<br>100 p.p.m. Rb | None | (2) 300<br>(1) 225 | None<br>75 |

* Numbers in parenthesis indicate number of animals.

centrates, as in Experiment II. Concentrate levels were determined by Morrison's Feeding Table modified for first calf heifers. The animals had free access to plain salt blocks and water.

Molybdenum as sodium molybdate, arsenic as sodium arsenate, lead as lead sulfate, and rubidium as rubidium chloride were used for administration to the animals. The amounts of each salt were calculated weekly using the previous week's feed consumption data as the basis for the calculations. For those animals which were fed in a group, estimates of hay consumption were made from data collected before the animals went on experiment. The weekly allowances of salts were weighed on an analytical balance, diluted with cane sugar and then portioned into seven gelatin capsules, one of which was administered daily.

Animals which were housed indoors received a few hours exercise each day in a small lot if weather permitted. Heat periods were recorded, and animals were bred during the first heat occurring after 60 days post-partum.

## COLLECTION OF DATA ON EXPERIMENTS I, II, AND III

Body weights of all animals in Experiment I, II, and III were recorded immediately following parturition and once each week thereafter while on experiment. At the time of weighing, each animal was given a body condition rating which indicated her state of flesh. The ratings used were as

follows:

| | | |
|---|---|---|
| Excellent | — | very heavily fleshed |
| Very good | — | well fleshed |
| Good plus | — | relatively good flesh |
| Good | — | average working condition |
| Fairly good | — | relatively poor flesh |
| Fair | — | thin condition |
| Poor | — | very thin condition (emaciated) |

The hair condition of each animal was examined weekly and a written description of the distribution, color, and texture was made. The bowel movements for all animals in the three experiments were observed and one of the following descriptions was recorded for each week:

Normal
Slightly loose
Loose
Very loose
Diarrhea

Bi-weekly hemoglobin determinations for all animals were made during the three experiments. The determinations were made by diluting 0.05 milliliters of heparinized blood to a volume of 10 milliliters with sodium bicarbonate and reading the optical density on a Klett-Summerson photo-electric colorimeter. A chart which was prepared with known standards was then used for converting the optical density readings into hemoglobin values.

Hematocrit values were determined on the blood samples drawn for hemoglobin determinations during the third and fourth periods of Experiment I, and bi-weekly during Experiments II and III. These values were obtained by a method described by Van Allen (154). Heparinized blood was pipetted into hematocrit tubes, and centrifuged in a 10-inch centrifuge

for 1-1/2 hours.

Serum protein percentages were determined three times during the last period of Experiment I and at the start of every 75 days thereafter during Experiments II and III. These values were obtained with the use of a hand protein refractometer. Inorganic phosphorus determinations were made each month on whole blood during the third and fourth periods of Experiments I and II and each month during Experiment III. These determinations were made by a method reported by Powers (130).

During the fourth period of Experiment III, sulfide levels of the rumen contents were determined. The sulfide determinations were carried out by a method reported by Fogo (65) with modifications to permit its use on rumen liquid and solid samples. Rumen samples were drawn by vacuum through a stomach tube into a clean 1000 milliliter flask. The samples then were centrifuged for 15 min. at 3000 r.p.m. to separate the liquid from the solid material. Samples of these fractions were diluted with distilled water and placed in a round bottom flask which had side arms connected through a stopcock. Glass tubing in a tight fitting rubber stopper permitted air to be bubbled through the sample and removed from the top of the flask. The gas removed from the flask in this manner was then bubbled through a basic solution of zinc acetate in a large test tube. A negative pressure, created by an aspirator, in the tube containing the zinc acetate, caused the flow of gases from the round bottom flask to the

receiving test tube. While the gas was flowing in this manner, concentrated hydrochloric acid was added to the sample of rumen material through the side arm of the flask. Any sulfide present in the sample was given off as hydrogen sulfide and collected in the zinc acetate solution. From this point the procedure used for determining hydrogen sulfide followed the method of Fogo (65).

After every seventy-five days on experiment a sample of liver obtained by biopsy, a blood serum sample, and a milk sample was taken from each animal and sent to the U. S. Plant, Soils and Nutrition Laboratory for analysis of copper and molybdenum. A set of samples also was taken at the beginning of Experiments II and III but not at the beginning of Experiment I.

The procedure for obtaining liver biopsies from the cattle was as follows:

1. The hair was clipped from an area over the two most posterior right ribs from the spinal column to a point 12 to 16 inches down the side of the animal.

2. The area was washed with soap and water and tincture of iodine was applied.

3. A small incision was made through the skin between the two most posterior ribs and about 6 inches from the spinal column.

4. An assembled trocar and cannula were forced through the muscle into the right lung cavity causing that lobe to collapse.

5. The trocar and cannula were then pushed through

the animal's diaphragm at which time the trocar was removed from the inside of the cannula.

6. The cannula, which was against the liver, was pushed into this organ cutting a small core.

7. The protruding end of the cannula was covered firmly with the thumb and removed from the animal.

8. A liver sample weighing from 0.1 to 0.8 grams was forced out of the cannula with the trocar onto filter paper.

9. The sample was blotted with filter paper to remove excess blood, placed in a small bottle, and dried in a vacuum over.

10. The trocar and cannula were washed in redistilled water after each sample was taken and placed in a bath of redistilled ethyl alcohol to await further use.

Blood samples were taken in 50 millliliter centrifuge tubes and permitted to clot. These samples were placed in a centrifuge and spun at 3000 r.p.m. for 15 minutes. The serum was poured off into glass test tubes and frozen. Premilk removed from the udder by hand was collected in a glass jar and stored in the frozen state.

All the bottles and test tubes used for the milk, liver, and serum samples were boiled in nitric acid and rinsed in redistilled water. Both the redistilled water and ethyl alcohol were distilled the second time in a glass still.

The calves born to the four animals from Experiment II which were carried through a second lactation, were sacri-

ficed at birth. The liver, spleen, and a sample of blood serum was saved from these calves and sent to the U. S. Plant, Soil and Nutrition Laboratory in Ithaca, New York, for analysis for copper and molybdenum. Hemoglobin and hematocrit values also were determined on the blood of these calves.

## EXPERIMENT IV

Six trials using copper-64 were carried out to observe the effects of adding molybdenum, sulfate sulfur, and molybdenum and sulfate sulfur on the copper absorption and metabolism of steers fed a diet low in copper, molybdenum, and sulfur.

The experimental design of these trials is found in Table 4. The animals were placed on the diet for each trial two months prior to the feeding of the radioactive sulfur. The day before feeding the isotope, the animals were placed in wooden metabolism stalls which were designed to collect urine and feces from steers. These stalls were isolated in a part of the barn so as not to be a health hazard when the radioisotope was in use.

Radioactive copper purchased from Union Carbide Corporation, at Oak Ridge, Tennessee, was prepared for administration behind lead bricks in the University of New Hampshire Central Counting Laboratory. In the first four trials 10 millicuries were purchased, and 20 millicuries were purchased for trials V and VI. The isotope was diluted and divided into two equal portions which were poured into two 10 oz. carbonated

TABLE 4

Design of Radioactive Copper Experiment

| Animal number | Age in months | Weight | Sulfate sulfur | Molybdenum |
|---|---|---|---|---|
| | | Trial I | | |
| 473 | 6 | 320 | 0.3% | 50 p.p.m. |
| 474 | 6 | 302 | 0.3% | None |
| | | Trial II | | |
| 473 | 8 | 415 | 0.3% | None |
| 474 | 8 | 400 | 0.3% | 50 p.p.m. |
| | | Trial III | | |
| 498 | 5-1/2 | 300 | None | 50 p.p.m. |
| 499 | 5-1/2 | 330 | None | None |
| | | Trial IV | | |
| 498 | 8 | 396 | None | None |
| 499 | 8 | 425 | None | 50 p.p.m. |
| | | Trial V | | |
| 473 | 16 | 781 | 0.3% | None |
| 474 | 16 | 751 | None | None |
| | | Trial VI | | |
| 498 | 13 | 665 | None | None |
| 499 | 13 | 707 | 0.3% | None |

beverage bottles. These bottles were then equipped with two milking machine teat cup liners which fit firmly over the neck of the bottle. These liners when forced in the mouth of the animal while its head was held up as far as possible, delivered the solution contained in the bottle into the esophagus. Care was taken to make sure all of the solution was swallowed before the head was relased.

Samples of blood, urine and feces were taken frequently for five days. Twenty-four hours after the administration of the isotope a liver biopsy was taken. Attempts to count the isotope with a windowless proportional flow counter model D-47 which was manufactured by Nuclear of Chicago Corporation for the first 24 hours of the first trial failed due to the weak $\beta$ -radiation. After this period, all samples were counted by use of a gamma well scintillation counter model DS5-5 which was manufactured by Nuclear of Chicago Corporation.

Five milliter samples of blood and urine were counted for 10 minutes. The samples of feces were wrapped in waxed paper and weighed on an analytical balance. Five minute counts were made on the feces samples because of high levels of the isotope. Liver samples also were wrapped in waxed paper and weighed on an analytical balance. These samples were counted for 20 minutes.

The animals were kept in the collection stalls for a period of 10 days and all feces and urine collected during this time were buried four feet below the surface of the

ground in accordance with Federal Register parts 20 and 30. When the six radioactive studies were completed, all four animals used in the trials were killed and buried according to regulations.

# RESULTS

## EXPERIMENT I

An average of the first three weekly body weights for each cow taken immediately after parturition and immediately after each seventy-five days on experiment are given in Table 4 in the Appenidx. From these values, weight changes for each period were calculated. The changes for the cows grouped by treatments are found in Table 5. These data indicate greater weight gains for all experimental groups than for the controls. This indicates that increased molybdenum in the diet did not cause poorer gains as was expected. The greater weight gains shown for the molybdenum supplemented animals were not significant due to the wide variations within each treatment.

Weekly body condition ratings were averaged to give a single rating for each cow during each experimental period. The average ratings are summarized by treatments in Table 6. The differences observed between treatments are not significant although animals receiving 50 p.p.m. of molybdenum showed a lower group average.

The condition of the hair of all animals on all treatments was found to be normal, showing no changes in hair color, distribution, or texture. The weekly observations made on consistency of feces revealed no evidence of looseness or

## TABLE 5

### Weight Changes
### (Experiment I)

| Animals by treatment | Periods I | II | III | IV | Totals |
|---|---|---|---|---|---|
| | Pounds | | | | |
| **Control** | | | | | |
| 282 | -8 | 52 | 21 | 70 | 135 |
| 290 | 21 | 35 | 22 | 41 | 119 |
| 293 | -71 | 11 | 20 | 76 | 36 |
| **Totals** | -58 | 98 | 63 | 187 | 290 |
| | | | | Average | 97 |
| Molybdenum 5 p.p.m. | | | | | |
| 285 | -2 | 30 | 51 | 95 | 174 |
| 288 | -37 | 43 | 33 | 62 | 101 |
| 292 | 22 | -4 | 45 | 19 | 82 |
| Totals | 17 | 69 | 129 | 176 | 357 |
| | | | | Average | 119 |
| **Molybdenum 10 p.p.m.** | | | | | |
| 283 | 30 | 76 | 19 | 47 | 172 |
| 287 | -37 | 43 | 33 | 62 | 101 |
| 291 | 6 | 19 | 88 | 67 | 180 |
| Totals | -1 | 138 | 140 | 176 | 453 |
| | | | | Average | 151 |
| Molybdenum 20 p.p.m. | | | | | |
| 281 | 31 | 37 | -7 | 55 | 116 |
| 289 | 39 | 50 | 57 | 79 | 225 |
| 295 | 34 | 23 | 51 | 82 | 190 |
| Totals | 104 | 110 | 101 | 216 | 531 |
| | | | | Average | 177 |
| **Molybdenum 50 p.p.m.** | | | | | |
| 284 | 28 | 31 | 33 | 76 | 168 |
| 286 | 14 | 41 | 17 | 22 | 94 |
| 294 | 33 | 38 | 6 | 84 | 161 |
| Totals | 75 | 110 | 56 | 182 | 423 |
| | | | | Average | 141 |

TABLE 6

Body Condition Ratings
(Experiment I)

| Animals by treatment | Periods I | II | III | IV | Average |
|---|---|---|---|---|---|
| **Control** | | | | | |
| 282 | FG | FG | F | F | FG |
| 290 | G | G | G | G | G |
| 293 | G+ | G+ | G+ | G+ | G+ |
| | | | | Average | G |
| **Molybdenum 5 p.p.m.** | | | | | |
| 285 | G | G | G | G | G |
| 288 | FG | FG | FG | FG | FG |
| 292 | G | G | G | G | G |
| | | | | Average | G |
| **Molybdenum 10 p.p.m.** | | | | | |
| 283 | FG | FG | FG | FG | FG |
| 287 | FG | FG | G | G | G |
| 291 | G | G | G+ | G+ | G+ |
| | | | | Average | G |
| **Molybdenum 20 p.p.m.** | | | | | |
| 281 | G | G | G+ | G+ | G+ |
| 289 | FG | G | G | G | G |
| 295 | G | G | G+ | G+ | G+ |
| | | | | Average | G+ |
| **Molybdenum 50 p.p.m.** | | | | | |
| 284 | FG | FG | FG | FG | FG |
| 286 | FG | FG | FG | FG | FG |
| 294 | G | G | G | G | G |
| | | | | Average | FG |

diarrhea for any animal, regardless of treatment.

Breeding records are given by treatments in Table 7. All but one animal had conceived successfully. The exception was 50 p.p.m. of molybdenum animal which was not pregnant after 5 services. A comparison of the number of days required to settle the other cows on this treatment with comparable data for the other treatments give no indication that the cause of breeding failure in the one cow was due to the treatment.

The average values for hemoglobin, hematocrit, inorganic phosphorus, and serum protein are recorded in Table 8 by treatments. The differences between treatments are not significant. This indicates among other things that molybdenum supplementation did not produce anemia or abnormal phosphorus metabolism.

The effects on appetite of added dietary molybdenum were studied by comparing each animal's total daily feed intake per hundred pounds of body weight. These values, as recorded in Table 9, show that there was greater variation among animals in each treatment than there was among treatments.

The milk and butterfat production records for each cow during each period of the experiment are given in Table 10. As might be expected there were large variations found among the animals on each treatment. The differences among treatments were not significant.

All of the animals in this experiment were considered

TABLE 7

Breeding Records
(Experiment I)

| Animals by treatment | Number of services | Number of days before conception |
|---|---|---|
| **Control** | | |
| 282 | 1 | 77 |
| 290 | 2 | 125 |
| 293 | 2 | 107 |
| Total | 5 | 309 |
| Average | 1.7 | 103 |
| **Molybdenum 5 p.p.m.** | | |
| 285 | 1 | 74 |
| 288 | 2 | 162 |
| 292 | 4 | 199 |
| Total | 7 | 435 |
| Average | 2.3 | 145 |
| **Molybdenum 10 p.p.m.** | | |
| 283 | 1 | 132 |
| 287 | 2 | 152 |
| 291 | 1 | 81 |
| Total | 4 | 365 |
| Average | 1.3 | 122 |
| **Molybdenum 20 p.p.m.** | | |
| 281 | 1 | 142 |
| 287 | 1 | 83 |
| 295 | 1 | 142 |
| Total | 3 | 367 |
| Average | 1 | 122 |
| **Molybdenum 50 p.p.m.** | | |
| 284 | 2 | 100 |
| 286 | 5+ (not preg.) | 300 |
| 294 | 2 | 83 |
| Total | 9+ | 483 |
| Average | 3+ | 161 |

TABLE 8

Average Composition of Blood
(Experiment I)

| Animals by treatment | Hemoglobin mg/100 ml | Hematocrits % | Phosphorus mg/100 ml | Serum protein % |
|---|---|---|---|---|
| Control | | | | |
| 282 | 9.64 | 30.7 | 6.9 | 7.5 |
| 290 | 10.18 | 33.7 | 6.1 | 7.3 |
| 293 | 8.86 | 30.7 | 6.0 | 7.1 |
| Totals | 28.68 | 95.1 | 19.0 | 21.9 |
| Average | 9.56 | 31.7 | 6.3 | 7.3 |
| Molybdenum 5 p.p.m. | | | | |
| 285 | 9.70 | 31.2 | 6.8 | 7.5 |
| 288 | 10.06 | 33.6 | 7.3 | 6.6 |
| 292 | 10.27 | 34.4 | 6.2 | 8.0 |
| Totals | 30.03 | 99.2 | 20.3 | 22.1 |
| Average | 10.01 | 33.1 | 6.8 | 7.4 |
| Molybdenum 10 p.p.m. | | | | |
| 283 | 9.21 | 29.9 | 7.0 | 8.1 |
| 287 | 10.19 | 33.8 | 7.0 | 8.0 |
| 291 | 9.72 | 33.6 | 6.4 | 7.4 |
| Totals | 29.12 | 97.3 | 20.4 | 23.5 |
| Average | 9.71 | 32.4 | 6.8 | 7.8 |
| Molybdenum 20 p.p.m. | | | | |
| 281 | 9.99 | 32.4 | 6.2 | 7.3 |
| 289 | 9.97 | 30.6 | 4.8 | 7.1 |
| 295 | 9.34 | 29.3 | 6.6 | 7.7 |
| Totals | 29.30 | 92.3 | 17.6 | 22.1 |
| Average | 9.77 | 30.8 | 5.9 | 7.4 |
| Molybdenum 50 p.p.m. | | | | |
| 284 | 9.90 | 31.9 | 6.4 | 7.5 |
| 286 | 9.16 | 29.8 | 8.0 | 7.7 |
| 294 | 9.33 | 30.5 | 6.8 | 7.8 |
| Totals | 28.39 | 92.2 | 21.2 | 23.0 |
| Average | 9.46 | 30.7 | 7.1 | 7.7 |

TABLE 9

Feed Consumed Daily per 100 Pounds Body Weight
(Experiment I)

| Animals by treatment | Periods I | II | III | IV | Total |
|---|---|---|---|---|---|
| | Pounds | | | | |
| **Control** | | | | | |
| 282 | 3.50 | 3.52 | 3.36 | 2.95 | 13.33 |
| 290 | 3.18 | 3.42 | 3.22 | 3.20 | 13.02 |
| 293 | 3.59 | 3.56 | 3.24 | 2.98 | 13.37 |
| | | | Total | | 39.72 |
| | | | Average | | 3.31 |
| **Molybdenum 5 p.p.m.** | | | | | |
| 285 | 3.08 | 3.04 | 2.82 | 2.73 | 11.67 |
| 288 | 2.99 | 2.95 | 2.75 | 2.67 | 11.36 |
| 292 | 3.30 | 3.55 | 3.49 | 3.32 | 13.66 |
| | | | Total | | 36.69 |
| | | | Average | | 3.06 |
| **Molybdenum 10 p.p.m.** | | | | | |
| 283 | 3.17 | 3.29 | 3.17 | 3.09 | 12.72 |
| 287 | 3.56 | 3.04 | 3.76 | 3.37 | 14.53 |
| 291 | 3.51 | 3.75 | 3.51 | 2.92 | 13.69 |
| | | | Total | | 40.94 |
| | | | Average | | 3.41 |
| **Molybdenum 20 p.p.m.** | | | | | |
| 281 | 3.53 | 3.49 | 3.46 | 3.29 | 13.77 |
| 289 | 3.22 | 3.41 | 3.25 | 2.92 | 12.80 |
| 295 | 3.09 | 3.17 | 3.01 | 2.76 | 12.03 |
| | | | Total | | 38.60 |
| | | | Average | | 3.22 |
| **Molybdenum 50 p.p.m.** | | | | | |
| 284 | 3.17 | 3.59 | 3.62 | 3.41 | 13.79 |
| 286 | 3.23 | 3.61 | 3.64 | 3.42 | 13.90 |
| 294 | 3.50 | 3.27 | 2.87 | 2.57 | 12.21 |
| | | | Total | | 39.90 |
| | | | Average | | 3.33 |

TABLE 10

Total Milk and Butterfat Production in Pounds
(Experiment I)

| Animals by treatment | Periods I | II | III | IV | Total milk | Total fat |
|---|---|---|---|---|---|---|
| Control | | | | | | |
| 282 milk | 2502.9 | 2118.6 | 1785.8 | 1524.4 | 7931.7 | |
| fat | 87.4 | 79.4 | 77.8 | 66.9 | | 311.5 |
| 290 milk | 2168.0 | 1772.0 | 1584.6 | 1627.8 | 7152.4 | |
| fat | 80.8 | 72.9 | 65.9 | 66.3 | | 285.9 |
| 293 milk | 2369.3 | 1841.6 | 1557.0 | 1098.2 | 6866.1 | |
| fat | 83.3 | 81.6 | 71.5 | 54.9 | | 291.3 |
| | | | Total | | 21950.2 | 888.7 |
| | | | Average | | 7316.7 | 296.2 |
| Molybdenum 5 p.p.m. | | | | | | |
| 285 milk | 1562.6 | 1399.5 | 1183.3 | 985.8 | 5131.2 | |
| fat | 53.5 | 54.9 | 48.9 | 42.9 | | 202.2 |
| 288milk | 1471.9 | 1249.3 | 1114.0 | 1081.1 | 4916.3 | |
| fat | 43.8 | 39.6 | 38.9 | 42.3 | | 163.6 |
| 292 milk | 2436.0 | 2403.3 | 2235.5 | 2190.4 | 9265.2 | |
| fat | 81.3 | 83.2 | 67.3 | 72.8 | | 304.6 |
| | | | Total | | 19312.7 | 670.4 |
| | | | Average | | 6437.6 | 223.5 |
| Molybdenum 10 p.p.m. | | | | | | |
| 283 milk | 1426.7 | 1285.9 | 1375.5 | 1022.7 | 5110.8 | |
| fat | 58.4 | 57.1 | 62.5 | 48.6 | | 226.6 |
| 287 milk | 2526.6 | 2292.1 | 2404.3 | 2231.6 | 9454.6 | |
| fat | 77.3 | 88.7 | 104.2 | 89.8 | | 360.0 |
| 291 milk | 2195.7 | 1986.9 | 1773.2 | 850.6 | 6806.4 | |
| fat | 72.2 | 76.4 | 63.2 | 35.3 | | 247.1 |
| | | | Total | | 21371.8 | 833.7 |
| | | | Average | | 7123.9 | 277.9 |

TABLE 10 (Cont.)

Total Milk and Butterfat Production in Pounds

(Experiment I)

| Animals by treatment | | Periods I | II | III | IV | Total milk | Total fat |
|---|---|---|---|---|---|---|---|
| Molybdenum 20 p.p.m. | | | | | | | |
| 281 | milk | 2909.8 | 2612.6 | 2154.7 | 1963.8 | 9640.9 | |
| | fat | 153.7 | 100.9 | 82.7 | 74.5 | | 411.8 |
| 289 | milk | 2106.9 | 2007.0 | 1770.7 | 1556.7 | 7441.3 | |
| | fat | 67.0 | 73.0 | 68.3 | 64.5 | | 272.8 |
| 295 | milk | 2405.1 | 2189.5 | 1871.7 | 1441.7 | 7908.2 | |
| | fat | 74.7 | 63.8 | 55.6 | 43.0 | | 237.1 |
| | | | | Total | | 24990.4 | 921.7 |
| | | | | Average | | 8330.1 | 307.2 |
| Molybdenum 50 p.p.m. | | | | | | | |
| 284 | milk | 1583.2 | 1365.9 | 1261.7 | 1192.1 | 5402.9 | |
| | fat | 62.9 | 63.2 | 56.2 | 53.2 | | 235.5 |
| 286 | milk | 2076.3 | 1951.9 | 1768.6 | 1769.6 | 7566.4 | |
| | fat | 61.9 | 71.5 | 64.4 | 54.3 | | 252.1 |
| 294 | milk | 2634.1 | 2208.8 | 1795.0 | 1176.0 | 7813.0 | |
| | fat | 86.8 | 75.5 | 58.1 | 49.9 | | 207.3 |
| | | | | Total | | 20783.2 | 694.9 |
| | | | | Average | | 6927.7 | 231.6 |

to have normal health except for mastitis. The incidence of this disease was quite high, but no treatment difference was noted. The reason for the unusually high mastitis infection was not discernible.

The copper analyses of the milk samples taken every 75 days during experiment are recorded in Table 10 of the Appendix. Although the levels of copper in these samples varied from 0.02 to 0.80 p.p.m., no treatment difference was found. All animals showed a significant change in the copper level of the milk as the lactation continued. The values for molybdenum analyses of the 75-day milk samples are recorded in Table 10 of the Appendix. The mean values calculated for each treatment indicate that the level of molybdenum in the milk increased with increased molybdenum in the diet. These values are plotted in Figure 1.

Copper and molybdenum levels of the blood serum are recorded in Table 11 of the Appendix. The mean values for each treatment calculated from these data are plotted in Figures 2 and 3. The mean levels of copper in the serum of all treatments decreased for the first 225 days of experiment. The decreases for the control group, however, was not as large as those for the treatments receiving molybdenum. After 225 days, the levels of serum copper for the controls increased to a mean level of 1.97 p.p.m. which was essentially the same as at the start of the experiment. The serum copper levels for the molybdenum treatments continued to decrease until at the end of 300 days on the experiment all molybdenum

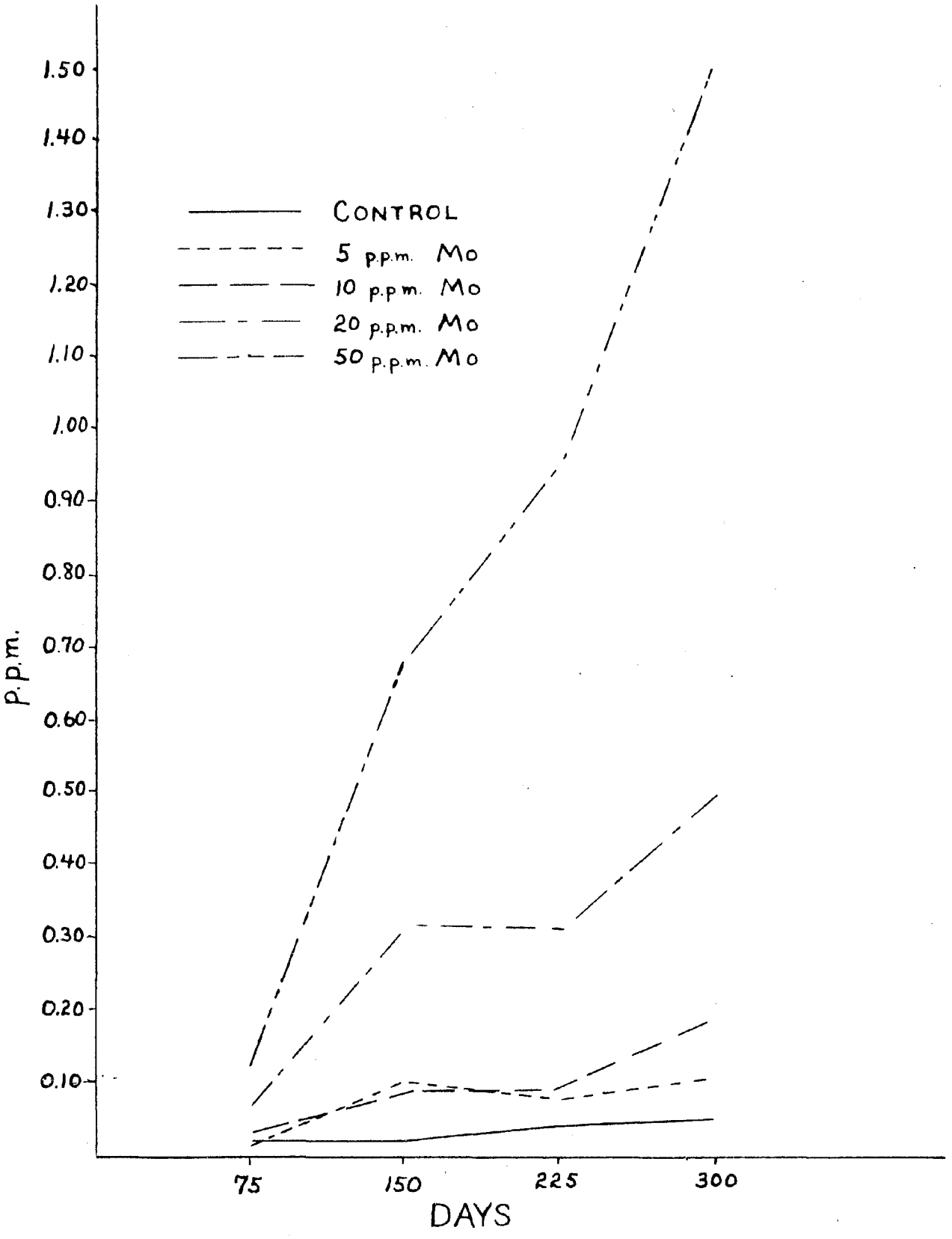


Fig. 1. Milk molybdenum levels for Experiment I.

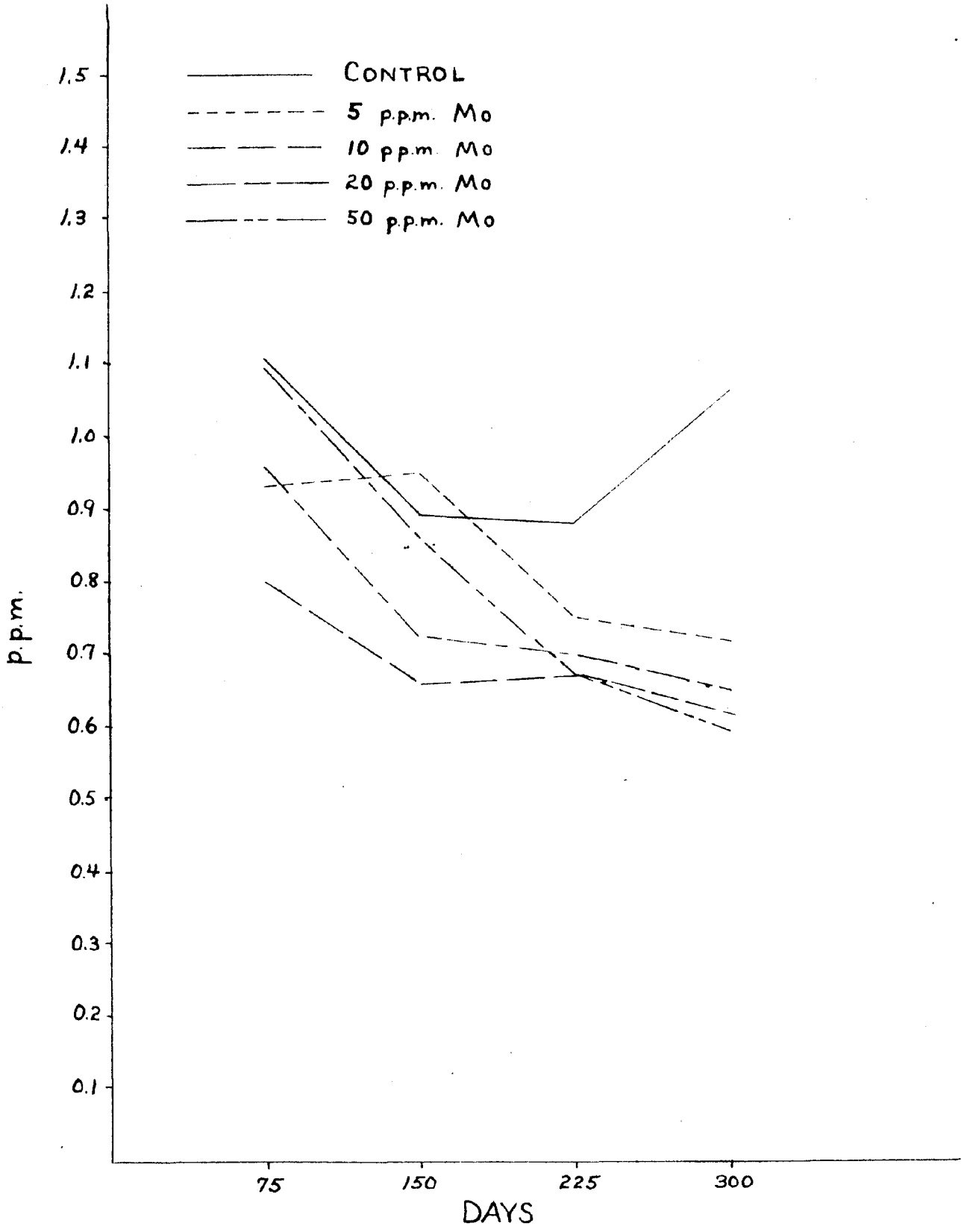


Fig. 2. Serum copper levels for Experiment I.

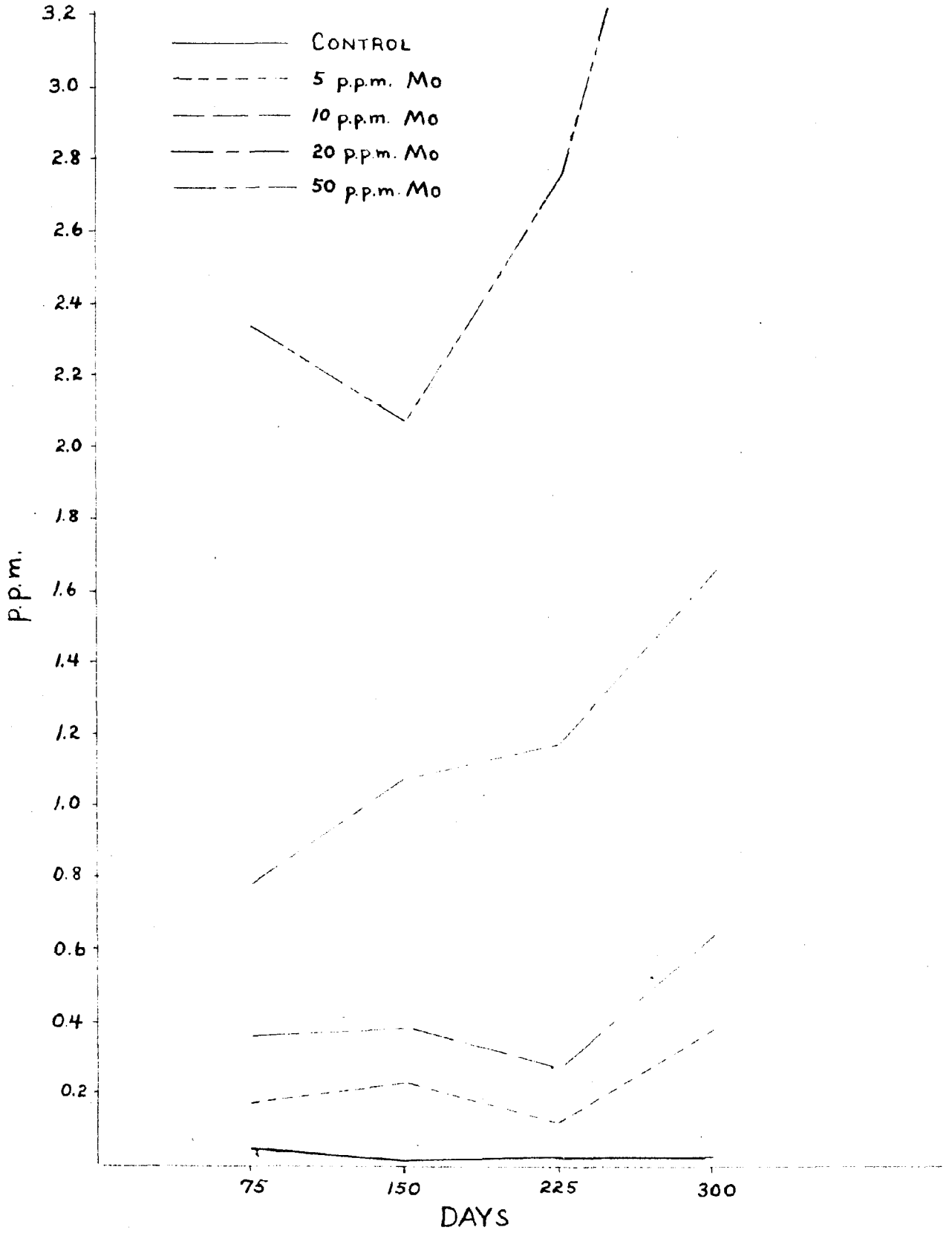


Fig. 3. Serum molybdenum levels for Experiment I.

treatments had serum copper levels from 0.6 to 0.72 p.p.m. The values, however, are still in the published normal range. Treatment means of serum molybdenum values indicate that the level of this element increased with increased molybdenum in the diet. These differences were significant at the 1% level.

Liver copper and molybdenum levels are recorded in Table 12. The mean liver copper values for each treatment are plotted in Figure 4. These values indicate that the liver copper levels for the animals which received molybdenum decreased during the period between 75 and 225 days and then showed an increase during the last 75 days of the experiment. The liver copper levels for the controls increased as the experiment continued. At the end of 300 days on experiment the liver copper levels for the control group were 60 p.p.m. greater than for the other treatments. The mean liver molybdenum values are shown in Figure 5. These data indicate that the molybdenum content of the liver increased with the level of molybdenum in the diet, but at each level of intake the liver seemed to stabilize after 150 days of experiment.

## EXPERIMENT II

Average body weights at the start of the experiment and at the end of each seventy-five days were calculated in the same manner as described for Experiment I. From these values, which are recorded in Table 15 in the Appendix, body weight changes for each period were calculated. A comparison of these weight changes, as found in Table 11, showed no

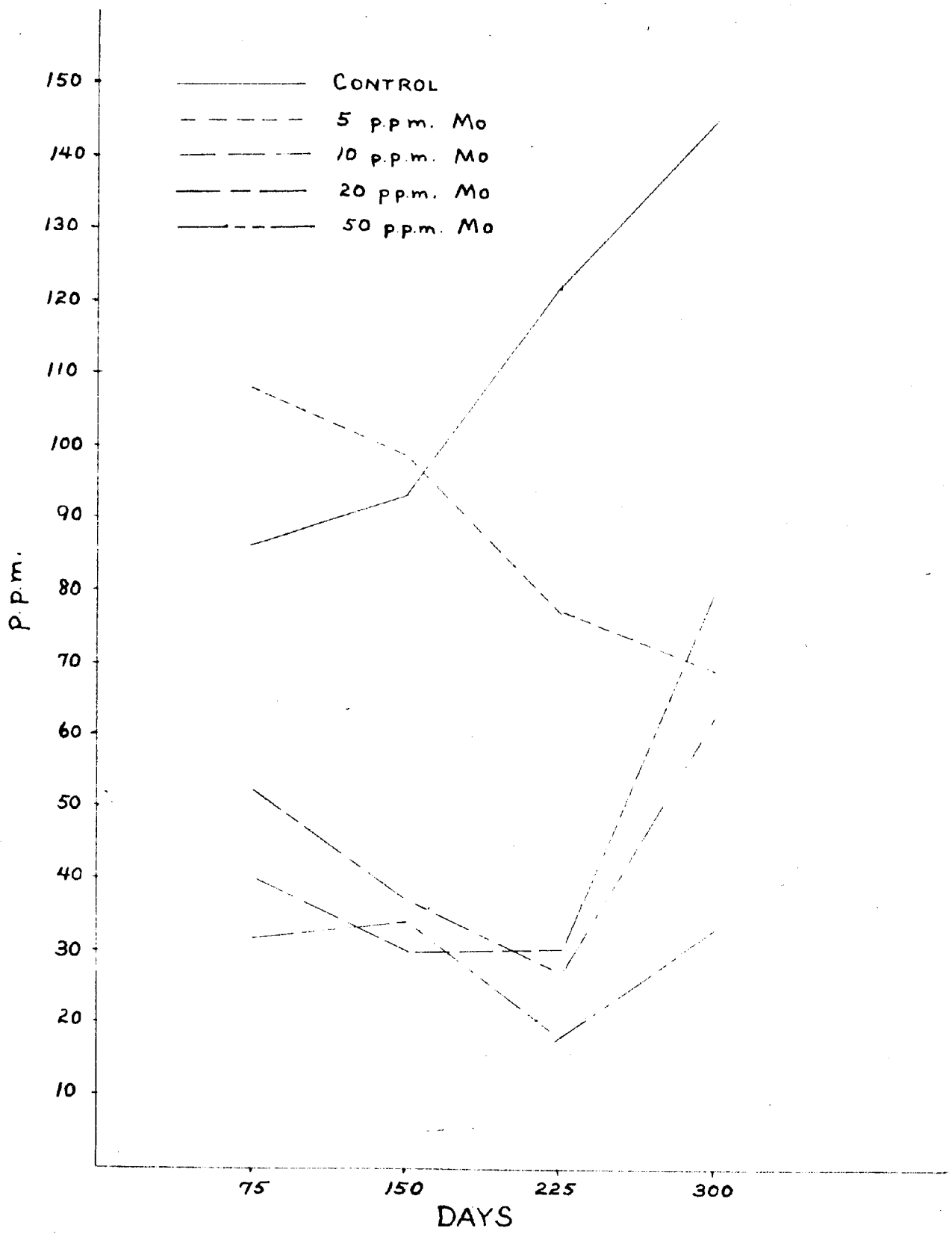


Fig. 4. Liver copper levels for Experiment I.

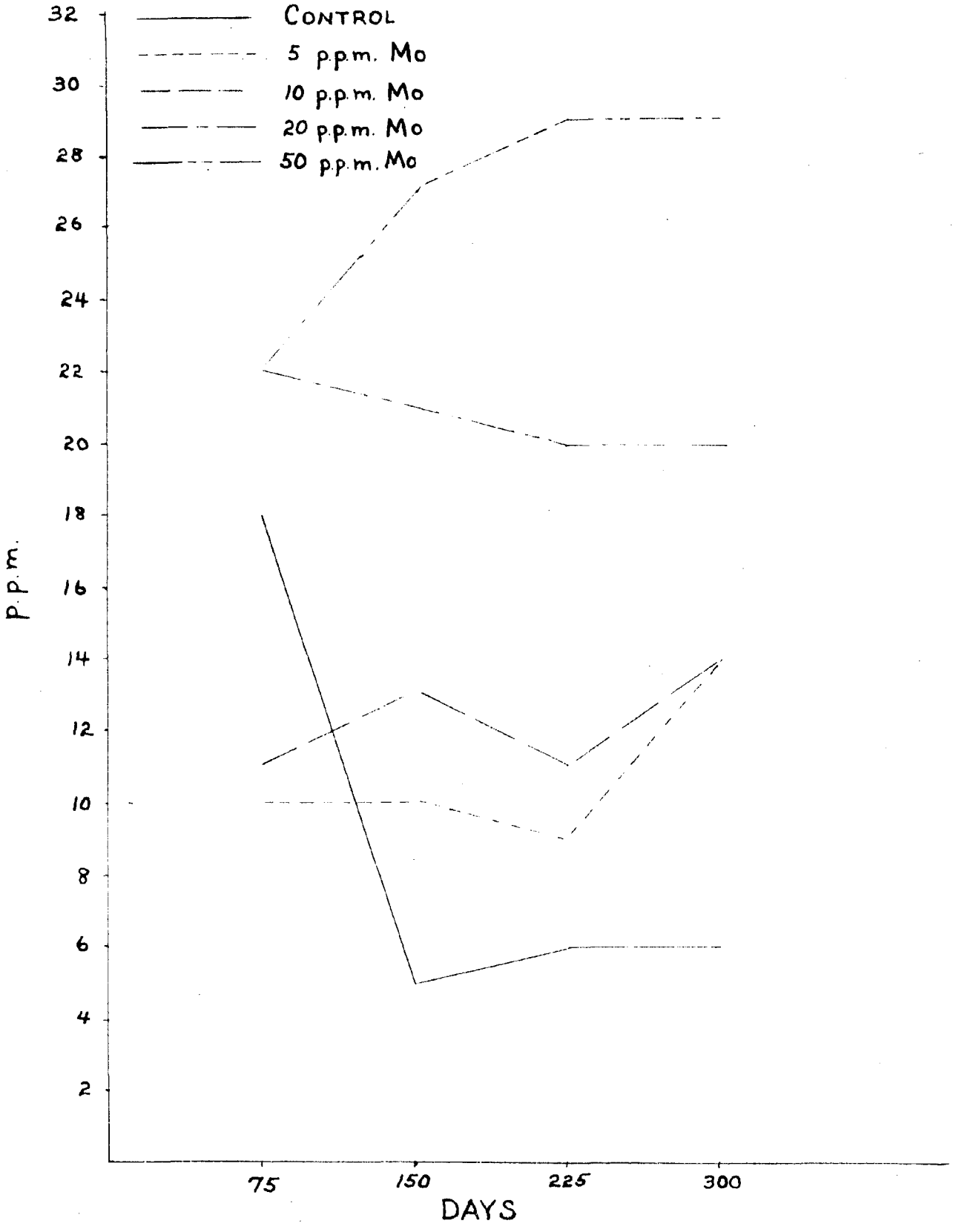


Fig. 5. Liver molybdenum levels for Experiment I.

TABLE 11

Weight Changes
(Experiment II)

| Animals by treatment | Periods I | II | III | IV | Totals |
|---|---|---|---|---|---|
| | Pounds | | | | |
| **Sulfate sulfur 0.3%** | | | | | |
| 386 | 3 | -2 | -24 | 69 | 46 |
| 395 | -29 | -5 | 5 | 66 | 37 |
| 398 | -3 | 0 | -20 | -2 | -25 |
| Totals | -29 | -7 | -39 | 133 | 58 |
| | | | | Average | 19 |
| **Sulfate sulfur 0.3%, molybdenum 5 p.p.m.** | | | | | |
| 390 | -3 | 19 | | | |
| 392 | 34 | -9 | | | |
| 399 | -50 | 10 | | | |
| **Totals** | -19 | 18 | | | |
| **Sulfate sulfur 0.3%, molybdenum 10 p.p.m.** | | | | | |
| 388 | 25 | 6 | 19 | | |
| 391 | 10 | -2 | 7 | | |
| 396 | -8 | -32 | 2 | | |
| Totals | 27 | -28 | 28 | | |
| **Sulfate sulfur 0.3%, molybdenum 20 p.p.m.** | | | | | |
| 389 | 7 | -47 | 20 | -38 | -58 |
| 393 | -3 | 6 | 74 | 76 | 153 |
| 397 | 31 | 16 | -37 | 6 | 16 |
| Totals | 34 | -25 | 57 | 44 | 111 |
| | | | | Average | 37 |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m.** | | | | | |
| 387 | 19 | 37 | 3 | -3 | 56 |
| 394 | 38 | -9 | -11 | 39 | 57 |
| 400 | 20 | -14 | 27 | 45 | 78 |
| Totals | 77 | 14 | 19 | 81 | 191 |
| | | | | Average | 64 |

TABLE 11 (Cont.)

Weight Changes
(Experiment II)

| Animals by treatment | Periods I | II | III | IV | Totals |
|---|---|---|---|---|---|
| | Pounds | | | | |
| **Sulfate sulfur 0.3%, molybdenum 100 p.p.m.** | | | | | |
| 388 | | | | -105 | |
| 392 | | | | -50 | |
| Totals | | | | -155 | |
| | | | | Average | -78 |
| **Sulfate Sulfur 0.3%, molybdenum 200 p.p.m.** | | | | | |
| 391 | | | | -210 | |
| 396 | | | | -133 | |
| Total | | | | -343 | |
| | | | | Average | -172 |
| **Animals Continued Through A Second Lactation** | | | | | |
| **Sulfate sulfur 0.3%** | | | | | |
| 386 | -32 | -6 | 17 | 139 | 118 |
| 395 | -31 | 20 | -43 | 4 | -50 |
| Totals | -63 | 14 | -26 | 143 | 68 |
| | | | | Average | 34 |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m.** | | | | | |
| 387 | -5 | 0 | -43 | 28 | -20 |
| 394 | -3 | -4 | 41 | -90 | -56 |
| Totals | -8 | -4 | -2 | -62 | -76 |
| | | | | Average | -38 |

effect from feeding molybdenum for 300 days at levels as high as 50 p.p.m. Although two cows which received 50 p.p.m. molybdenum and 0.3% sulfate sulfur showed a weight loss of 38 pounds, this had no significance because one of the two animals which received 0.3% sulfate sulfur showed a weight loss of 50 pounds during the second lactation. The animals which were fed 100 and 200 p.p.m. molybdenum for one period of seventy-five days, however, showed weight losses of 78 and 172 pounds respectively. These exceptionally high weight losses resulted from the feeding of molybdates.

Body condition ratings for each period are summarized in Table 12. Molybdenum levels as high as 50 p.p.m. caused no lowering of body condition, as is shown by these data. Similarly, the two cows retained on the treatment of 50 p.p.m. molybdenum and 0.3% sulfate sulfur for a second lactation. showed no loss of body condition during the second lactation. The animals receiving 100 and 200 p.p.m. for seventy-five days, however, showed rapid deterioration of body condition.

A description of hair condition changes noted during this experiment is found in Table 13. No changes in hair color, texture, or distribution were noted for treatments containing 0.3% sulfate sulfur and up to 20 p.p.m. of molybdenum. After 225 days on experiment, animals receiving 0.3% sulfate sulfur and 50 p.p.m. molybdenum developed achromotrichia, particularly around the face and eyes, alopecia around the eyes and muzzle, and the hair texture became soft and lacked body. The two animals receiving this treatment which were continued

TABLE 12

Body Condition Ratings
(Experiment II)

| Animals by treatment | Periods I | II | III | IV | Average |
|---|---|---|---|---|---|
| **Sulfate sulfur 0.3%** | | | | | |
| 386 | FG | FG | FG | FG | FG |
| 395 | G | G | G | G | G |
| 398 | FG | FG | FG | FG | FG |
| | | | | Average | FG |
| **Sulfate sulfur 0.3%, molybdenum 5 p.p.m.** | | | | | |
| 390 | FG | G | | | |
| 392 | G | G+ | G+ | | |
| 399 | FG | FG | | | |
| **Sulfate sulfur 0.3%, molybdenum 10 p.p.m.** | | | | | |
| 388 | G | G+ | G+ | | |
| 391 | G | G | G | | |
| 396 | G | G | G | | |
| **Sulfate sulfur, molybdenum 20 p.p.m.** | | | | | |
| 389 | G+ | G+ | G+ | G | G+ |
| 393 | G | G+ | G+ | G+ | G+ |
| 397 | FG | G | FG | FG | FG |
| | | | | Average | G |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m.** | | | | | |
| 387 | G | G | G+ | G | G |
| 394 | G+ | G+ | G+ | G+ | G+ |
| 400 | G | G | FG | FG | FG |
| | | | | Average | G |
| **Sulfate sulfur 0.3%, molybdenum 100 p.p.m.** | | | | | |
| 388 | FG | | | | |
| 392 | G | | | | |
| **Sulfate sulfur 0.3%, molybdenum 200 p.p.m.** | | | | | |
| 391 | F | | | | |
| 396 | F | | | | |

Animals Continued Through A Second Lactation

| | | | | | |
|---|---|---|---|---|---|
| **Sulfate sulfur 0.3%** | | | | | |
| 386 | G+ | G | G | G+ | G |
| 395 | G+ | G | G | G | G |
| | | | | Average | G |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m.** | | | | | |
| 387 | G | G | G | G | G |
| 394 | G | G | G | G | G |
| | | | | Average | G |

TABLE 13

Hair Condition
(Experiment II)

| Animals by treatment | Hair color change | Distribution | Hair texture |
|---|---|---|---|
| Sulfate sulfur 0.3% | | | |
| 386 | None | Normal | Normal |
| 395 | " | " | " |
| 398 | " | " | " |
| Sulfate sulfur 0.3%, molybdenum 5 p.p.m. | | | |
| 390 | None | Normal | Normal |
| 392 | " | " | " |
| 399 | " | " | " |
| Sulfate sulfur 0.3%, molybdenum 10 p.p.m. | | | |
| 388 | None | Normal | Normal |
| 391 | " | " | " |
| 396 | " | " | " |
| Sulfate sulfur 0.3%, molybdenum 20 p.p.m. | | | |
| 389 | None | Normal | Normal |
| 393 | " | " | " |
| 397 | " | " | " |
| Sulfate sulfur 0.3%, molybdenum 50 p.p.m. | | | |
| 387 | Black hair turned brown, around eyes & muzzle changed to gray after 250 days | Some alopecia around eyes | Hair very soft and fine |
| 394 | Black hair turned brown over most of the body & gray around eyes. White became silver after 225 days | Some alopecia around eyes | Very light and soft |
| 400 | Black hair turned brown & gray over most of body after 225 days. Black was completely changed to copper color at 300 days. White became silver | Alopecia around eyes | Hair soft and cotton-like |

TABLE 13 (Cont.)

Hair Condition
(Experiment II)

| Animals by treatment | Hair color change | Distribution | Hair texture |
|---|---|---|---|
| Sulfate sulfur 0.3%, molybdenum 100 p.p.m. | | | |
| 388 | Slight graying around eyes after 75 days | Normal | Normal |
| 392 | Slight graying around eyes after 75 days. White hair interspersed in black | Normal | Normal |
| Sulfate sulfur 0.3%, molybdenum 200 p.p.m. | | | |
| 391 | White hair interspersed in black after 30 days. Gray around eyes | Normal | Normal |
| 396 | White & golden hair interspersed in black areas after 40 days. Gray around eyes | Normal | Normal |
| | Animals continued Through A Second Lactation | | |
| Sulfate sulfur 0.3% | | | |
| 386 | None | Normal | Normal |
| 395 | None | Normal | Normal |
| Sulfate sulfur 0.3%, molybdenum 50 p.p.m. | | | |
| 387 | Achromotrichis of black hair became more complete after parturition | Alopecia severe over face. Some on body | Hair very soft and lacked body |
| 394 | Achromotrichia of black hair became more complete after parturition. White hair remained white | Some alopecia around eyes | Hair soft and lacked body |

for a second lactation developed these changes more completely during the second lactation. The animals which received 100 and 200 p.p.m. molybdenum developed a condition where white hairs were interspersed throughout the black hair on the body. This change was noted as early as 30 days after the high level treatments were started.

Weekly observations revealed that the feces were higher in water content for cows in this experiment than for Experiment I. These feces were described as being slightly loose to loose, but no differences were noted among treatments. The four animals which received 100 and 200 p.p.m. molybdenum developed severe diarrhea within two weeks from the start of the treatments. The two animals which received the 100 p.p.m. level tended to overcome the diarrhea condition when a decrease in feed consumption lowered appreciably the amount of molybdate administered. The animals on the 200 p.p.m. level, however, continued to have debilitating scours during the entire time of treatment.

The breeding records for this experiment are recorded by treatments in Table 14. The rate of conception was low. Seven animals, representing every treatment, did not conceive at the end of 300 days on experiment. For many of the animals heat periods were not observed by the herdsmen and thus were not bred. Part of this problem may have been caused by limiting the exercise to prevent the spread of brucellosis. It was noted further that heat periods were more frequently missed during the period from December through February and more fre-

TABLE 14

Breeding Records
(Experiment II)

| Animals by treatment | Number of services | Number of days before conception |
|---|---|---|
| **Sulfate sulfur 0.3%** | | |
| 386 | 2 | 210 |
| 395 | 1 | 77 |
| 398 | 1 (not preg.) | 300 |
| Total | 4 | 587 |
| Average | 1.3 | 196 |
| **Sulfate sulfur 0.3%, molybdenum 5 p.p.m.** | | |
| 390 | 3 | 106 |
| 392 | 2 | 110 |
| 399 | 0 (not preg.) | 300 |
| Total | 5+ | 516 |
| Average | 1.7 | 166 |
| **Sulfate sulfur 0.3%, molybdenum 10 p.p.m.** | | |
| 388 | 2 | 159 |
| 391 | 4 (not preg.) | 300 |
| 396 | 1 (not preg.) | 300 |
| Total | 7+ | 759 |
| Average | 2.3 | 253 |
| **Sulfate sulfur 0.3%, molybdenum 20 p.p.m.** | | |
| 389 | 4+ (not preg.) | 300 |
| 393 | 1 | 76 |
| 397 | 0 (not preg.) | 300 |
| Total | 5+ | 676 |
| Average | 1.7 | 225 |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m.** | | |
| 387 | 2 | 180 |
| 394 | 1 | 71 |
| 400 | 1 (not preg.) | 300 |
| Total | 4+ | 551 |
| Average | 1.3 | 164 |
| **Animals Continued Through A Second Lactation** | | |
| **Sulfate sulfur 0.3%** | | |
| 386 | 1 | 152 |
| 395 | 2 | 239 |
| Total | 3 | 391 |
| Average | 1.5 | 186 |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m.** | | |
| 387 | 5+ (not preg.) | 375 |
| 394 | 4 | 279 |
| Total | 9+ | 654 |
| Average | 4.5 | 316 |

quently for those animals kept in the darker areas of the barn.

The average hemoglobin and hematocrit values for each period are found in Tables 15 and 16, respectively. These values showed more variation within each treatment than among treatments thus indicating no difference due to treatment. Because values were all in the normal range, there was no anemia. Values for serum protein determinations are recorded in Table 17. There are no differences noted among treatments for these values.

The effects of added molybdenum on appetite were studied on the basis of total feed consumption as in Experiment I. These values, which are recorded in Table 18 show no differences due to treatment except for animals which received 0.3% sulfate sulfur with 100 and 200 p.p.m. molybdenum. The four animals which were on these treatments went off feed after two weeks and showed poor appetite until the high levels of molybdate were removed.

The milk and butterfat production values for the animals on this experiment are shown in Table 19. There appears to be no differences due to treatment. Production levels varied greatly in all treatments. Such variations should be expected under the conditions by which the animals were selected for the experiment. The feeding of 100 and 200 p.p.m. molybdenum with 0.3% sulfate sulfur, however, produced an abrupt drop in milk production when toxicity symptoms were observed. Evidence that the high molybdenum levels

TABLE 15

Average Hemoglobin Values
(Experiment II)

| Animals by treatment | Periods I | II | III | IV | Totals |
|---|---|---|---|---|---|
| | mg/100 ml of blood | | | | |
| **Sulfate sulfur 0.3%** | | | | | |
| 386 | 9.87 | 9.49 | 9.31 | 9.29 | 37.96 |
| 395 | 9.88 | 9.30 | 8.46 | 8.44 | 36.08 |
| 398 | 9.26 | 8.46 | 8.59 | 8.45 | 34.76 |
| | | | | Total | 108.80 |
| | | | | Average | 9.07 |
| **Sulfate sulfur 0.3%, molybdenum 5 p.p.m.** | | | | | |
| 390 | 10.19 | 9.37 | | | 19.56 |
| 392 | 10.38 | 9.81 | 9.73 | | 29.92 |
| 399 | 9.78 | 9.41 | | | 19.19 |
| | | | | Total | 68.67 |
| | | | | Average | 9.81 |
| **Sulfate sulfur 0.3%, molybdenum 10 p.p.m.** | | | | | |
| 388 | 9.38 | 9.21 | 9.24 | | 27.83 |
| 391 | 10.46 | 8.74 | 9.77 | | 28.97 |
| 396 | 9.60 | 9.21 | 8.87 | | 27.68 |
| | | | | Total | 84.48 |
| | | | | Average | 9.39 |
| **Sulfate sulfur 0.3%, molybdenum 20 p.p.m.** | | | | | |
| 389 | 10.10 | 9.87 | 9.91 | 10.07 | 39.95 |
| 393 | 10.53 | 9.53 | 10.09 | 9.10 | 39.25 |
| 397 | 9.21 | 8.53 | 8.18 | 8.18 | 34.10 |
| | | | | Total | 113.30 |
| | | | | Average | 9.44 |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m.** | | | | | |
| 387 | 10.32 | 9.94 | 9.57 | 9.25 | 39.08 |
| 394 | 9.47 | 8.50 | 8.37 | 8.30 | 34.64 |
| 400 | 10.26 | 9.35 | 8.92 | 8.99 | 37.52 |
| | | | | Total | 111.24 |
| | | | | Average | 9.27 |
| **Sulfate sulfur 0.3%, molybdenum 100 p.p.m.** | | | | | |
| 388 | | | | 9.00 | |
| 392 | | | | 8.95 | |
| | | | | Total | 17.95 |
| | | | | Average | 8.96 |
| **Sulfate sulfur 0.3%, molybdenum 200 p.p.m.** | | | | | |
| 391 | | | | 9.06 | |
| 396 | | | | 8.44 | |
| | | | | Total | 17.50 |
| | | | | Average | 8.75 |

TABLE 15 (Cont.)

Average Hemoglobin Values
(Experiment II)

| Animals by treatment | Periods I | II | III | IV | Totals |
|---|---|---|---|---|---|
| | mg/100 ml of blood | | | | |
| **Animals Continued Through A Second Lactation** | | | | | |
| **Sulfate sulfur 0.3%** | | | | | |
| 386 | 9.89 | 9.50 | 9.89 | 10.24 | 39.52 |
| 395 | 9.41 | 8.80 | 9.75 | 9.45 | 37.41 |
| | | | | Total | 76.93 |
| | | | | Average | 9.62 |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m.** | | | | | |
| 387 | 10.88 | 9.98 | 10.59 | 9.62 | 41.07 |
| 394 | 8.85 | 8.82 | 9.49 | 9.21 | 36.37 |
| | | | | Total | 77.44 |
| | | | | Average | 9.68 |

TABLE 16

Average Hematocrit Values
(Experiment II)

| Animals by treatment | Periods I | II | III | IV | Totals |
|---|---|---|---|---|---|
| | | | % | | |
| **Sulfate sulfur 0.3%** | | | | | |
| 386 | 30.6 | 29.5 | 29.5 | 29.4 | 119.0 |
| 395 | 31.7 | 29.9 | 26.7 | 26.8 | 115.1 |
| 398 | 28.1 | 27.2 | 26.1 | 26.0 | 107.4 |
| | | | | Total | 341.5 |
| | | | | Average | 28.5 |
| **Sulfate sulfur 0.3%, molybdenum 5 p.p.m.** | | | | | |
| 390 | 33.0 | 32.1 | | | 65.1 |
| 392 | 34.0 | 32.6 | 33.4 | | 100.0 |
| 399 | 32.4 | 31.5 | | | 63.9 |
| | | | | Total | 229.0 |
| | | | | Average | 32.7 |
| **Sulfate sulfur 0.3%, molybdenum 10 p.p.m.** | | | | | |
| 388 | 30.6 | 29.2 | 31.5 | | 91.3 |
| 391 | 33.2 | 29.5 | 32.4 | | 95.1 |
| 396 | 30.0 | 29.2 | 28.2 | | 87.4 |
| | | | | Total | 273.8 |
| | | | | Average | 30.4 |
| **Sulfate sulfur 0.3%, molybdenum 20 p.p.m.** | | | | | |
| 389 | 32.2 | 31.8 | 32.8 | 32.0 | 128.8 |
| 393 | 33.2 | 31.8 | 34.1 | 30.2 | 129.3 |
| 397 | 29.6 | 26.5 | 27.0 | 23.8 | 106.9 |
| | | | | Total | 365.0 |
| | | | | Average | 30.4 |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m.** | | | | | |
| 387 | 33.0 | 32.5 | 31.2 | 28.8 | 125.5 |
| 394 | 29.5 | 27.0 | 27.5 | 26.2 | 110.2 |
| 400 | 32.3 | 30.7 | 29.3 | 29.1 | 121.4 |
| | | | | Total | 357.1 |
| | | | | Average | 29.8 |
| **Sulfate sulfur, 0.3%, molybdenum 100 p.p.m.** | | | | | |
| 388 | | | | 27.4 | |
| 392 | | | | 30.8 | |
| | | | | Total | 58.2 |
| | | | | Average | 29.1 |
| **Sulfate sulfur, 0.3%, molybdenum 200 p.p.m.** | | | | | |
| 391 | | | | 29.1 | |
| 396 | | | | 25.5 | |
| | | | | Total | 54.6 |
| | | | | Average | 27.3 |

TABLE 16 (Cont.)

Average Hematocrit Values
(Experiment II)

| Animals by treatment | Periods I | II | III | IV | Totals |
|---|---|---|---|---|---|
| | | | % | | |
| **Animals continued Through A Second Lactation** | | | | | |
| **Sulfate sulfur 0.3%** | | | | | |
| 386 | 29.5 | 29.3 | 30.3 | 31.8 | 120.9 |
| 395 | 30.0 | 30.3 | 30.0 | 29.6 | 119.9 |
| | | | | Total | 240.8 |
| | | | | Average | 30.10 |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m.** | | | | | |
| 387 | 32.3 | 31.7 | 33.6 | 29.5 | 127.1 |
| 394 | 27.5 | 27.0 | 29.1 | 28.8 | 112.4 |
| | | | | Total | 239.5 |
| | | | | Average | 29.94 |

TABLE 17

Serum Protein Values
(Experiment II)

| Animals by treatment | Days 0 | 75 | 150 | 225 | 300 | Total |
|---|---|---|---|---|---|---|
| | % | | | | | |
| **Sulfate sulfur 0.3%** | | | | | | |
| 386 | 7.8 | 8.6 | 8.0 | 8.3 | 7.7 | 40.4 |
| 395 | 7.4 | 8.6 | 7.2 | 7.2 | 7.0 | 37.4 |
| 398 | 7.5 | 8.1 | 6.6 | 7.2 | 7.4 | 36.8 |
| | | | | Total | | 114.6 |
| | | | | Average | | 7.6 |
| **Sulfate sulfur 0.3%, molybdenum 5 p.p.m.** | | | | | | |
| 390 | 6.7 | 7.1 | 7.9 | | | 21.7 |
| 392 | 6.6 | 7.8 | 7.5 | 7.0 | | 28.9 |
| 399 | 7.3 | 8.2 | 7.4 | | | 22.9 |
| | | | | Total | | 73.5 |
| | | | | Average | | 7.4 |
| **Sulfate sulfur 0.3%, molybdenum 10 p.p.m.** | | | | | | |
| 388 | 8.1 | 7.7 | 7.7 | 7.3 | | 30.8 |
| 391 | 7.4 | 8.8 | 7.8 | 8.0 | | 32.0 |
| 396 | 7.0 | 7.3 | 7.6 | 8.0 | | 29.9 |
| | | | | Total | | 92.7 |
| | | | | Average | | 7.7 |
| **Sulfate sulfur 0.3%, molybdenum 20 p.p.m.** | | | | | | |
| 389 | 6.0 | 6.2 | 7.4 | 7.0 | 6.7 | 33.3 |
| 393 | 6.4 | 7.0 | 7.7 | 7.3 | 7.0 | 35.4 |
| 397 | 7.0 | 7.4 | 7.8 | 8.0 | 7.3 | 37.5 |
| | | | | Total | | 106.2 |
| | | | | Average | | 7.1 |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m.** | | | | | | |
| 387 | 7.0 | 8.1 | 8.0 | 7.0 | 6.8 | 36.9 |
| 394 | 6.0 | 7.6 | 7.5 | 7.7 | 7.5 | 36.3 |
| 400 | 7.8 | 8.0 | 7.6 | 7.8 | 7.9 | 39.1 |
| | | | | Total | | 112.3 |
| | | | | Average | | 7.5 |

TABLE 17 (Cont.)

Serum Protein Values
(Experiment II)

| Animals by treatment | Days 0 | 75 | 150 | 225 | 300 | Total |
|---|---|---|---|---|---|---|
| | % | | | | | |
| **Sulfate sulfur 0.3%, molybdenum 100 p.p.m.** | | | | | | |
| 388 | | | | | 7.0 | |
| 392 | | | | | 6.8 | |
| Total | | | | | | 13.8 |
| Average | | | | | | 6.9 |
| **Sulfate sulfur 0.3%, molybdenum 200 p.p.m.** | | | | | | |
| 391 | | | | | 6.4 | |
| 396 | | | | | 7.3 | |
| Total | | | | | | 13.7 |
| Average | | | | | | 6.9 |
| **Animals Continued Through A Second Lactation** | | | | | | |
| **Sulfate sulfur 0.3%** | | | | | | |
| 386 | 8.2 | 8.6 | 8.1 | 8.5 | 8.3 | 41.7 |
| 395 | 7.1 | 8.2 | 7.7 | 8.0 | 7.8 | 38.8 |
| Total | | | | | | 80.5 |
| Average | | | | | | 8.05 |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m.** | | | | | | |
| 387 | 7.0 | 7.9 | 7.9 | 8.1 | 7.2 | 38.1 |
| 394 | 7.6 | 7.4 | 7.4 | 8.0 | 9.0 | 39.4 |
| Total | | | | | | 77.5 |
| Average | | | | | | 7.75 |

TABLE 18

Feed Consumed Daily per 100 Pounds Body Weight
(Experiment II)

| | Periods | | | | |
|---|---|---|---|---|---|
| | I | II | III | IV | Total |
| Animals by treatment | | | Pounds | | |
| **Sulfate sulfur 0.3%** | | | | | |
| 386 | 3.64 | 4.01 | 3.73 | 3.47 | 14.85 |
| 395 | 3.68 | 3.89 | 3.53 | 3.09 | 14.19 |
| 398 | 3.39 | 3.05 | 2.80 | 2.61 | 11.85 |
| | | | | Total | 40.89 |
| | | | | Average | 3.41 |
| **Sulfate sulfur 0.3%, molybdenum 5 p.p.m.** | | | | | |
| 390 | 4.07 | 3.79 | | | 7.86 |
| 392 | 3.51 | 3.77 | 3.57 | | 10.85 |
| 399 | 3.84 | 3.44 | | | 7.28 |
| | | | | Total | 25.99 |
| | | | | Average | 3.71 |
| **Sulfate sulfur 0.3%, molybdenum 10 p.p.m.** | | | | | |
| 388 | 3.55 | 3.33 | 3.29 | | 10.17 |
| 391 | 3.55 | 3.45 | 3.08 | | 10.08 |
| 396 | 4.01 | 3.48 | 2.69 | | 10.18 |
| | | | | Total | 30.43 |
| | | | | Average | 3.38 |
| **Sulfate sulfur 0.3%, molybdenum 20 p.p.m.** | | | | | |
| 389 | 3.65 | 3.87 | 3.47 | 3.09 | 14.08 |
| 393 | 3.77 | 3.65 | 3.28 | 2.77 | 13.47 |
| 397 | 3.25 | 3.05 | 2.68 | 2.29 | 11.27 |
| | | | | Total | 38.82 |
| | | | | Average | 3.24 |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m.** | | | | | |
| 387 | 3.37 | 3.05 | 2.81 | 2.48 | 11.71 |
| 394 | 3.39 | 3.12 | 2.83 | 2.89 | 12.23 |
| 400 | 4.40 | 4.11 | 3.11 | 3.27 | 14.89 |
| | | | | Total | 38.83 |
| | | | | Average | 3.24 |
| **Sulfate sulfur 0.3%, molybdenum 100 p.p.m.** | | | | | |
| 388 | | | | 2.39 | |
| 392 | | | | 2.47 | |
| | | | | Total | 4.86 |
| | | | | Average | 2.43 |
| **Sulfate sulfur 0.3%, molybdenum 200 p.p.m.** | | | | | |
| 391 | | | | 1.87 | |
| 396 | | | | 2.47 | |
| | | | | Total | 4.34 |
| | | | | Average | 2.17 |

TABLE 19

Total Milk and Butterfat Production in Pounds
(Experiment II)

| Animals by treatment | | Periods I | II | III | IV | Total milk | Total fat |
|---|---|---|---|---|---|---|---|
| **Sulfate sulfur 0.3%** | | | | | | | |
| 386 | milk | 2147.1 | 2747.4 | 1671.8 | 1480.4 | 8046.7 | |
| | fat | 65.0 | 65.9 | 60.6 | 51.7 | | 243.2 |
| 395 | milk | 2842.0 | 2476.4 | 1866.5 | 1221.3 | 8406.2 | |
| | fat | 115.9 | 107.4 | 73.9 | 60.6 | | 357.8 |
| 398 | milk | 1787.7 | 1365.5 | 1123.6 | 795.4 | 5072.2 | |
| | fat | 77.8 | 53.1 | 42.2 | 25.9 | | 199.0 |
| | | | | Total | | 21325.1 | 800.0 |
| | | | | Average | | 7175.0 | 266.6 |
| **Sulfate sulfur 0.3%, molybdenum 5 p.p.m.** | | | | | | | |
| 390 | milk | 1856.2 | 1457.1 | | | | |
| | fat | 76.7 | 59.6 | | | | |
| 392 | milk | 2356.3 | 1843.1 | 1479.1 | | | |
| | fat | 77.6 | 62.9 | 50.7 | | | |
| 399 | milk | 2811.0 | 2052.9 | | | | |
| | fat | 128.5 | 76.3 | | | | |
| **Sulfate sulfur 0.3%, molybdenum 10 p.p.m.** | | | | | | | |
| 388 | milk | 1703.7 | 1429.0 | 1412.0 | | | |
| | fat | 54.8 | 47.9 | 48.3 | | | |
| 391 | milk | 2038.6 | 1695.7 | 1526.4 | | | |
| | fat | 76.76 | 65.0 | 53.5 | | | |
| 396 | milk | 2778.0 | 2181.0 | 1683.2 | | | |
| | fat | 105.6 | 85.3 | 71.55 | | | |
| **Sulfate sulfur 0.3%, molybdenum 20 p.p.m.** | | | | | | | |
| 389 | milk | 2361.5 | 1945.1 | 1552.8 | 1090.4 | 6949.8 | |
| | fat | 81.2 | 69.0 | 59.7 | 42.6 | | 252.5 |
| 393 | milk | 1970.4 | 1667.0 | 1221.3 | 310.9 | 5169.6 | |
| | fat | 62.3 | 55.7 | 40.0 | 12.3 | | 170.3 |
| 397 | milk | 2268.3 | 1702.8 | 1210.9 | 1025.6 | 6207.6 | |
| | fat | 64.5 | 55.2 | 38.9 | 34.4 | | 193.0 |
| | | | | Total | | 18327.0 | 615.8 |
| | | | | Average | | 6109.0 | 205.3 |

TABLE 19 (Cont.)

Total Milk and Butterfat Production in Pounds
(Experiment II)

| Animals by treatment | Periods I | II | III | IV | Total milk | Total fat |
|---|---|---|---|---|---|---|
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m.** | | | | | | |
| 387 milk | 1763.4 | 1169.0 | 657.2 | 150.1 | 3739.7 | |
| fat | 55.2 | 39.4 | 25.8 | 12.9 | | 133.3 |
| 394 milk | 2766.3 | 2276.0 | 1811.3 | 1239.2 | 8092.8 | |
| fat | 92.5 | 85.9 | 60.1 | 46.5 | | 285.0 |
| 400 milk | 3494.2 | 2691.6 | 1801.4 | 1349.9 | 9337.1 | |
| fat | 107.7 | 78.2 | 54.0 | 37.8 | | 277.1 |
| Total | | | | | 21169.6 | 695.4 |
| Average | | | | | 7056.6 | 231.8 |
| **Sulfate sulfur 0.3%, molybdenum 100 p.p.m.** | | | | | | |
| 388 milk | | | | 651.5 | | |
| fat | | | | 24.1 | | |
| 392 milk | | | | 474.0 | | |
| fat | | | | 19.4 | | |
| **Sulfate sulfur 0.3%, molybdenum 200 p.p.m.** | | | | | | |
| 391 milk | | | | 657.9 | | |
| fat | | | | 25.2 | | |
| 396 milk | | | | 591.7 | | |
| fat | | | | 28.5 | | |
| **Animals Continued Through A Second Lactation** | | | | | | |
| **Sulfate sulfur 0.3%** | | | | | | |
| 386 milk | 3364.7 | 3217.9 | 2712.0 | 2209.8 | 11504.4 | |
| fat | 110.4 | 99.1 | 92.9 | 86.2 | | 388.6 |
| 395 milk | 3165.1 | 2308.6 | 2239.0 | 2199.5 | 9912.3 | |
| fat | 130.5 | 140.6 | 104.8 | 93.4 | | 469.3 |
| Total | | | | | 21416.7 | 855.9 |
| Average | | | | | 10708.4 | 428.0 |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m.** | | | | | | |
| 387 milk | 2681.7 | 1265.2 | 247.1 | | 4194.0 | |
| fat | 95.8 | 58.4 | 13.6 | | | 167.8 |
| 394 milk | 2716.9 | 2553.3 | 1747.3 | 1467.9 | 8485.4 | |
| fat | 97.6 | 93.0 | 70.9 | 62.5 | | 324.0 |
| Total | | | | | 12679.4 | 491.8 |
| Average | | | | | 6339.7 | 245.9 |

caused this drop was provided by the recovery of animals when removed from these treatments. Although these animals were producing less than 2 pounds of milk per day, production levels returned to 16 and 18 pounds when the molybdenum feeding was discontinued.

The health of all the animals receiving levels of not over 50 p.p.m. molybdenum was considered normal, except for brucellosis and mastitis. The occurrence of brucellosis in two animals cannot be considered to have had any connection with the experimental treatment the animals received. An unusually high incidence of mastitis was not confined to any particular treatment and no adequate explanation could be found for the occurrence of the disease. Animals receiving 100 and 200 p.p.m. molybdenum with 0.3% sulfate sulfur exhibited a very toxic condition after three weeks on experiment. These animals became emaciated and appeared lethargic. On several occasions they lost nervous control of their rear quarters and help was required to move them to the scales for weighing. When the animals showed these conditions, molybdenum feeding was suspended for one week. Upon resumption of the treatment it appeared the animals adjusted somewhat to these high levels and nervous symptoms did not recur.

The health of the animals continued through a second lactation on a level of 50 p.p.m. molybdenum and 0.3% sulfate sulfur was not shown to be impaired. Except for changes in hair condition the two animals performed comparably to the two animals which received 0.3% sulfate sulfur. All four

animals showed no problems during parturition and gave birth to normal size calves. These calves appeared normal in every respect except that both calves from the 50 p.p.m. molybdenum animals had brown and white coats whereas the calves of the control animals had normal black and white coats.

Table 20

Composition of Serum, Liver, and Spleen of Calves Born to Cows Continued Through a Second Lactation

| Calf by dam's treatment | Copper content p.p.m. | | | Molybdenum content p.p.m. | | |
|---|---|---|---|---|---|---|
| | Serum | Liver | Spleen | Serum | Liver | Spleen |
| Sulfate sulfur 0.3% | | | | | | |
| Calf of 386 | 0.30 | 118 | 0.60 | 0.05 | 0.0 | 0.0 |
| Calf of 395 | 0.16 | 127 | 1.4 | 0.00 | 0.0 | 0.0 |
| Sulfate sulfur 0.3% and 50 p.p.m. molybdenum | | | | | | |
| Calf of 387 | 0.00 | 1.9 | 0.91 | 0.13 | 0.0 | 0.0 |
| Calf of 394 | 0.05 | 1.70 | 0.83 | 0.46 | 0.0 | 0.0 |

The copper and molybdenum analyses of serum, liver, and spleen taken from these calves are recorded in Table 20. The serum molybdenum values for these calves indicate that a small amount of molybdenum was transfered across the fetal membranes in the cows receiving molybdenum. Levels of molybdenum in the liver and spleen of these calves were undetectable.

The copper level in the serum and liver of the calves born to the cows which received 50 p.p.m. molybdenum with 0.3% sulfate sulfur indicated these calves were extremely copper deficient. Although the serum copper levels of the two calves from the cows which received sulfate were below normal, these

calves had considerable higher levels than the calves born to the cows which received molybdenum and sulfate.

The values for the copper and molybdenum analyses of milk samples for this experiment are recorded in Table 20 of the Appendix. Milk copper levels were not affected by the level of molybdenum in the diet but were significantly lower in this experiment than in Experiment I. This difference between Experiments I and II indicated that sulfate had an effect of lowering the level of copper in the milk. The mean milk molybdenum levels are plotted in Figure 6 by treatments. In contrast to Experiment I, the molybdenum levels of the milk in this experiment showed only slight increases with increased dietary molybdenum. These data indicate that the addition of 0.3% sulfate sulfur to the diet decreased the molybdenum excretion in the milk.

Serum molybdenum and copper levels for Experiment II are recorded in Table 21 of the Appendix. The mean serum copper values are plotted in Figure 7. Animals receiving 5, 10, and 20 p.p.m. of molybdenum with 0.3% sulfate sulfur added to the diet showed similar results to the 5, 10, and 20 p.p.m. of molybdenum treatments in Experiment I. For these treatments the serum copper decreased as the experiment progressed. Animals which received 50 p.p.m. of molybdenum with 0.3% sulfate sulfur had an increase in serum copper for the first 150 days of experiment and then showed a sharp decrease during the last 150 days of experiment. The animals on the 0.3% sulfate sulfur treatment showed only slight changes in serum copper

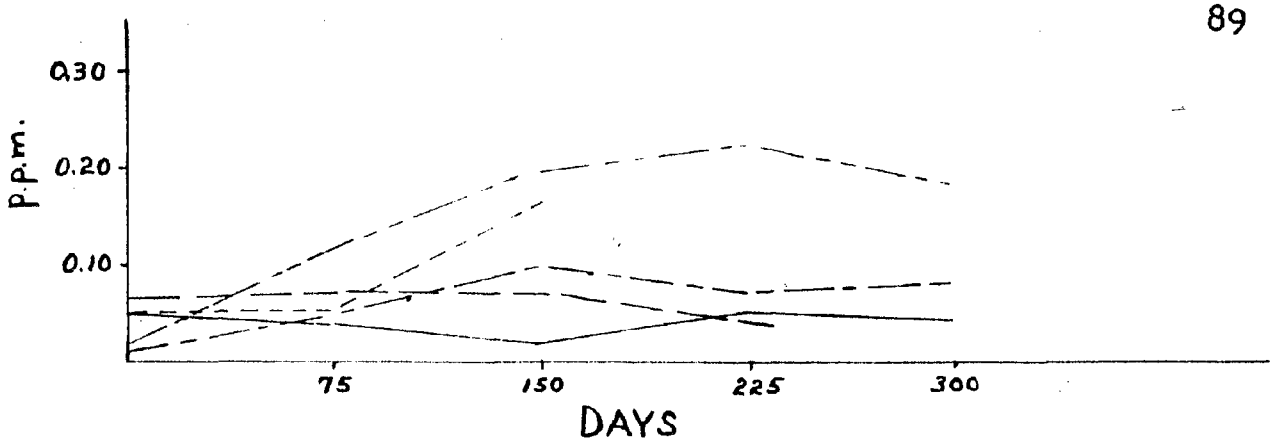


Fig. 6. Milk molybdenum levels for Experiment II.

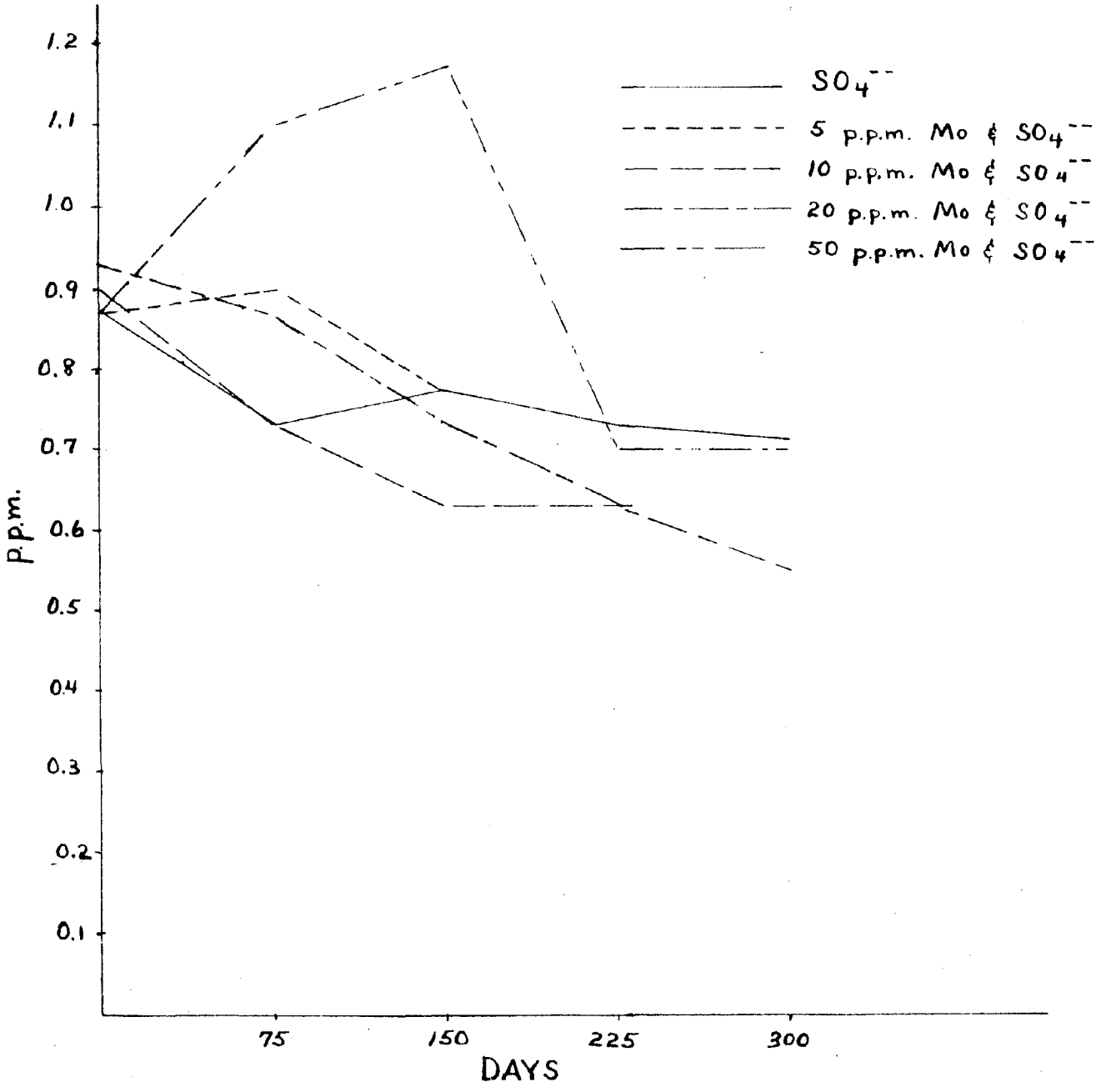


Fig. 7. Serum Copper levels for Experiment II

as the experiment progressed and did not differ significantly from the other treatments at the end of the experiment. The mean serum molybdenum values are plotted in Figure 8. These values increased with increased molybdenum in the diet. These data show, however, that the response to added dietary molybdenum was much less in this experiment than for Experiment I. This would indicate that the addition of sulfate sulfur diminished this response.

Liver copper and molybdenum levels for Experiment II are recorded in Table 22 of the Appendix. The mean liver copper values, which are plotted in Figure 9, show a rapid decrease during the first 225 days of experiment for all molybdenum treatments. After 225 days these values remained at this low level in contrast to Experiment I where these treatments showed an increase. The animals receiving 0.3% sulfate sulfur, without added molybdenum which had a lower value at the start of the experiment, showed a general increase in liver copper as the experiment progressed. The mean liver molybdenum values are plotted in Figure 10. In contrast to Experiment I, the liver molybdenum levels did not show consistent trends. It appears that the addition of 0.3% sulfate sulfur to the diet had an effect on the metabolism of molybdenum and that it was of a heterogeneous nature.

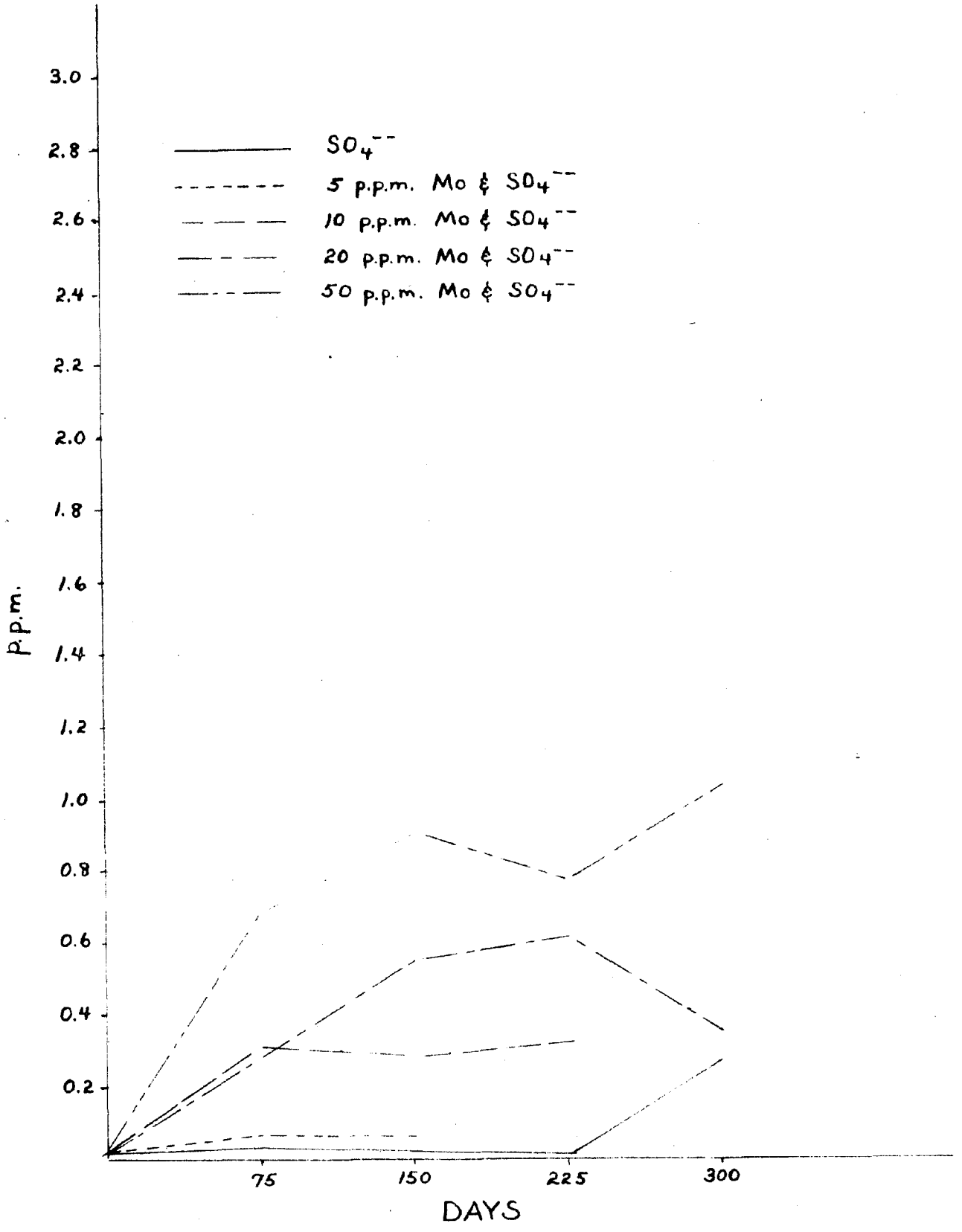


Fig. 8. Serum molybdenum levels for Experiment II.

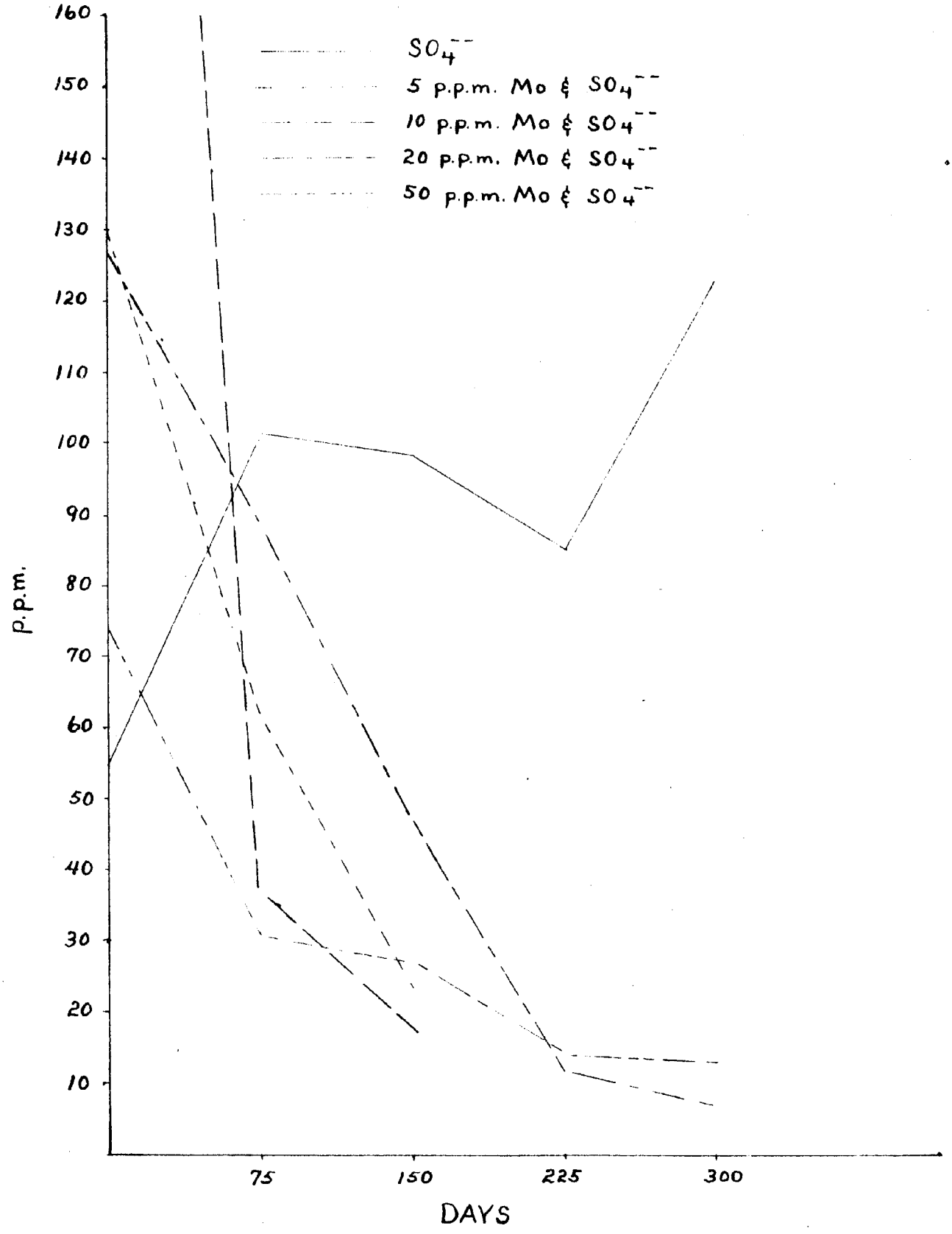


Fig. 9. Liver copper levels for Experiment II.

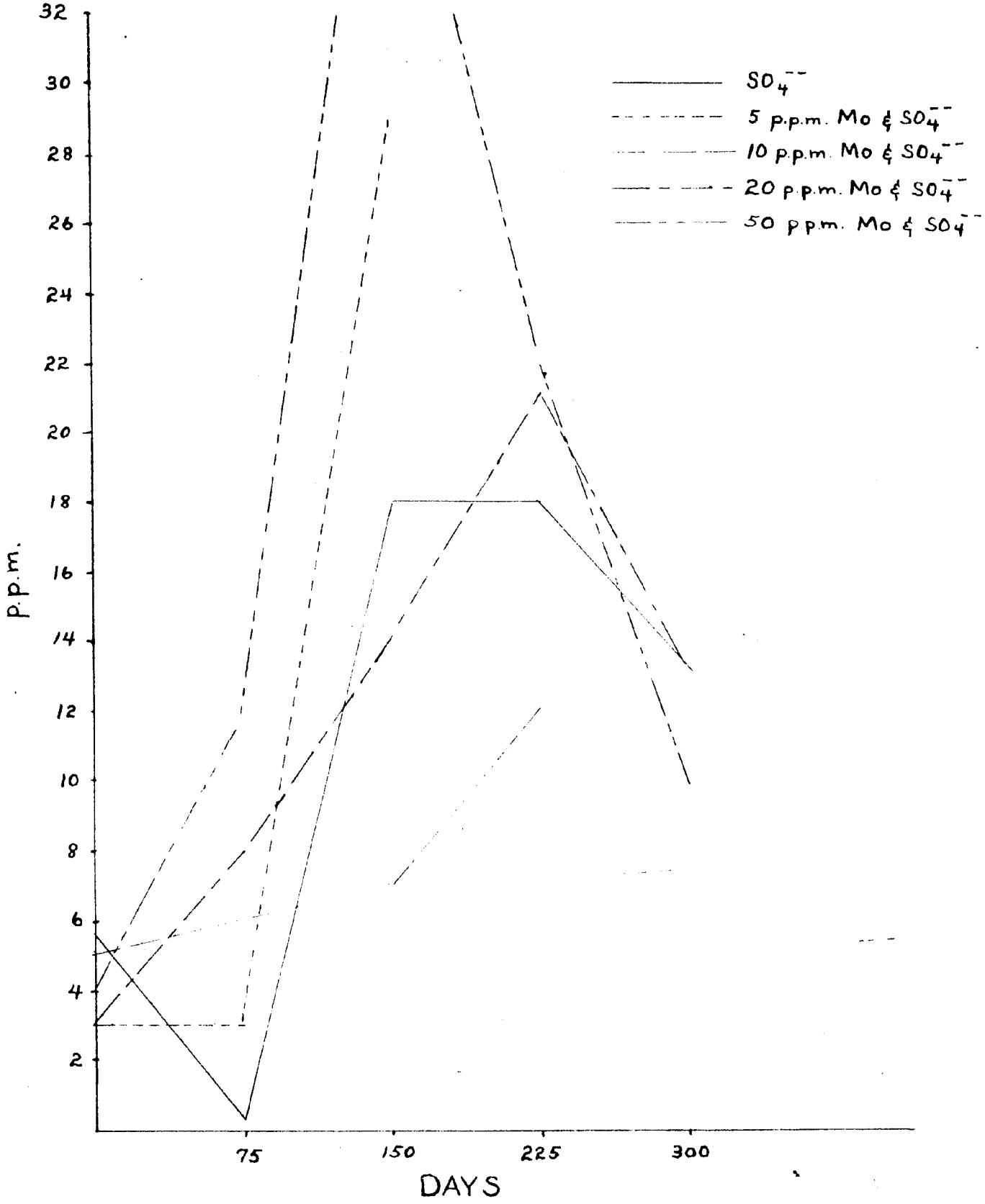


Fig. 10. Liver molybdenum levels for Experiment II.

## EXPERIMENT III

Body weights at the start of the experiment and at the end of each seventy-five day period were calculated in the same manner as described for Experiments I and II. These values are recorded in Table 25 in the Appendix. A summary of weight changes calculated from these values are recorded by treatments in Table 21. The average weight gains for the animals which received the treatments of 0.3% sulfate sulfur, outdoors, and 50 p.p.m. molybdenu, outdoors, were 148 pounds and 105 pounds, respectively. These average weight gains were considerably higher than the average gain of 11 pounds for the animals receiving the treatment of 50 p.p.m. molybdenum and 0.3% sulfate sulfur, outdoors. Both the treatment receiving 50 p.p.m. molybdenum and 0.3% sulfate sulfur, indoors, and the treatment receiving 50 p.p.m. molybdenum, 5 p.p.m. arsenic, 5 p.p.m. lead, 100 p.p.m. rubidium and 0.3% sulfate sulfur indoors, showed average weight losses during the experiment. The difference of the average weight loss for these treatments was 100 pounds which would indicate there was no effect on body weight from adding arsenic, lead, and rubidium to a diet high in molybdenum and sulfate. The difference found between the treatments outdoors and indoors are very large and appear to result from the difference of treatment.

Body condition ratings for each animal by periods are recorded in Table 22. Animals in the treatments 50 p.p.m.

TABLE 21

Weight Changes
(Experiment III)

| Animals by treatment | Periods I | II | III | IV | Totals |
|---|---|---|---|---|---|
| | Pounds | | | | |
| **Sulfate sulfur 0.3% (outdoors)** | | | | | |
| 477 | 54 | -77 | 71 | 35 | 83 |
| 482 | 45 | 64 | 77 | 29 | 215 |
| 488 | 15 | 33 | 57 | 42 | 147 |
| Totals | 114 | 20 | 205 | 106 | 445 |
| | | | | Average | 148.3 |
| Molybdenum 50 p.p.m. (outdoors) | | | | | |
| 479 | -3 | 14 | 32 | 67 | 110 |
| 481 | 7 | 66 | 12 | 50 | 135 |
| 489 | -20 | 21 | 21 | 47 | 69 |
| Totals | -16 | 101 | 65 | 164 | 314 |
| | | | | Average | 104.7 |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m. (outdoors)** | | | | | |
| 480 | 15 | 12 | 9 | 12 | 48 |
| 483 | -80 | -28 | 51 | 24 | -33 |
| 490 | -38 | -7 | -20 | 82 | 17 |
| Totals | -103 | -23 | 40 | 118 | 32 |
| | | | | Average | 10.7 |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m. (indoors)** | | | | | |
| 476 | 21 | 13 | 32 | -29 | 37 |
| 485 | 14 | -8 | -7 | -17 | -18 |
| 487 | 0 | -20 | 6 | -97 | -111 |
| Totals | 35 | -15 | 31 | -143 | -92 |
| | | | | Average | -30.7 |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m., arsenic 5 p.p.m., rubidium 100 p.p.m., and lead 5 p.p.m. (indoors)** | | | | | |
| 478 | 10 | 67 | -101 | 62 | 38 |
| 484 | -3 | -39 | -34 | 21 | -55 |
| 486 | 2 | -15 | -3 | -31 | -47 |
| Totals | 9 | 13 | -138 | 52 | -64 |
| | | | | Average | -21.3 |

TABLE 22

Body Condition Ratings
(Experiment III)

| Animals by treatment | Periods I | II | III | IV | Average |
|---|---|---|---|---|---|
| **Sulfate sulfur 0.3% (outdoors)** | | | | | |
| 477 | G | G+ | G | G+ | G+ |
| 482 | G | G | G+ | G+ | G |
| 488 | G | G | G | G | G |
| | | | Treatment average | | G |
| **Molybdenum 50 p.p.m. (outdoors)** | | | | | |
| 479 | G | G+ | G+ | G+ | G+ |
| 481 | G | G+ | G+ | VG | G+ |
| 489 | G | G | G | G | G |
| | | | Treatment average | | G+ |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m. (outdoors)** | | | | | |
| 480 | G+ | G+ | G+ | G+ | G+ |
| 483 | G+ | G | G | FG | G |
| 490 | G | G | F | F | FG |
| | | | Treatment average | | G |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m. (indoors)** | | | | | |
| 476 | F | F | FG | FG | F |
| 485 | G | G | G | G | G |
| 487 | FG | FG | F | P | F |
| | | | Treatment average | | F |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m., arsenic 5 p.p.m., rubidium 100 p.p.m. and lead 5 p.p.m. (indoors)** | | | | | |
| 478 | FG | G | FG | FG | FG |
| 484 | G+ | G+ | G | G | G |
| 486 | G | FG | FG | F | FG |
| | | | Treatment average | | FG |

molybdenum, outdoors, and 0.3% sulfate sulfur, outdoors, had higher ratings than the treatment 50 p.p.m. molybdenum with 0.3% sulfate sulfur, outdoors. All the treatments housed outdoors, however showed consistently higher body condition ratings than the treatments housed indoors. There was no advantage in body condition seen for either treatment indoors which indicated that the addition of arsenic, lead, and rubidium had no additional effect in lowering body condition.

A record of the hair condition ratings during Experiment III are found in Table 23. The treatments of 50 p.p.m. molybdenum, outdoors and 0.3% sulfate sulfur, outdoors, showed no changes in hair condition throughout the experiment. All other treatments, however, showed achromotrichia, alopecia, and hair texture changes which started after 150 days on experiment. It is evident from these observations that both a high level of molybdenum and a high level of inorganic sulfate were necessary to cause these hair changes. Four cows which received 50 p.p.m. molybdenum with 0.3% sulfate sulfur for 300 days, showed complete recovery of hair condition abnormalities after receiving 10 and 20 p.p.m. copper added to this diet. These data indicated a copper deficiency was the cause of these hair changes.

Weekly observations indicated that all animals receiving 0.3% sulfate sulfur in the diet had a higher water content in the feces than the three animals not receiving any sulfate sulfur. The feces of the animals which received 0.3% sulfate sulfur were described as being slightly loose to very

TABLE 23

Hair Condition
(Experiment III)

| Animals by treatment | Hair color change | Distribution | Hair texture |
|---|---|---|---|
| Sulfate sulfur 0.3% (outdoors) | | | |
| 477 | None | Normal | Normal |
| 482 | " | " | " |
| 488 | " | " | " |
| Molybdenum 50 p.p.m. (outdoors) | | | |
| 479 | None | Normal | Normal |
| 481 | " | " | " |
| 489 | " | " | " |
| Sulfate sulfur 0.3%, molybdenum 50 p.p.m. (outdoors) | | | |
| 480 | White hair became silver white at 230 days on experiment | Alopecia around eyes | Hair lacked body and became very soft at 200 days |
| 483 | Achromotrichia of black hair & white hair became silver white at 160 days on experiment | Alopecia around eyes and under jaw | Hair very soft, lacked body after 160 days |
| 490 | Achromotrichia of black hair. White hair turned silver white at 125 days on experiment | Alopecia on face very pronounced, and moderate over body | Hair lacked body. Soft cotton texture after 150 days |
| Sulfate sulfur 0.3%, molybdenum 50 p.p.m. (indoors) | | | |
| 476 | Achromotrichia of black hair at 150 days on experiment | Alopecia prominent around eyes, and to less degree over body | Hair very soft and loss of body |
| 485 | Black hair turned to brown at 200 days | Slight alopecia around eyes | Normal |
| 487 | Achromotrichia of black hair at 150 days | Wide spread alopecia over whole body | Hair very soft, having no body |

TABLE 23 (Cont.)

Hair Condition
(Experiment III)

| Animals by treatment | Hair color change | Distribution | Hair texture |
|---|---|---|---|
| Sulfate sulfur 0.3%, molybdenum 50 p.p.m., arsenic 5 p.p.m., rubidium 100 p.p.m., and lead 5 p.p.m. (indoors) | | | |
| 478 | Moderate amount of black hair turning to brown. White hair became silver white | Slight alopecia around eyes | Normal |
| 484 | White hair turned to silver white | Wide-spread alopecia over face and body | Hair became softer with less body at 225 days |
| 486 | Achromotrichia of black hair. White hair became silver white at 170 days | Wide-spread alopecia over entire body | Hair soft cotton-like texture after 200 days |

loose. Definite diarrhea was not observed for any animals while housed indoors or outdoors in the pole barn. The feces of the animals which were placed on pasture for 75 days showed an even greater water content. At times individual animals had bowel movements which approached a condition of diarrhea. At no time, however, was such a condition debilitating. Such looseness was present in all treatments of animals on pasture. It is interesting to note that the administration of copper sulfate at 10 and 20 p.p.m. to animals which had finished 300 days on experiment but were still receiving the assigned treatment showed a great lowering of the water content in the feces.

The breeding records are recorded in Table 24. All the animals housed outdoors conceived with little delay. One of these animals aborted at the end of 300 days on experiment, but tests for brucellosis and vibrio fedus were negative. Three animals from the treatments indoors were not pregnant at the end of 300 days. Two of these were bred more than four times, this indicating that heat periods were observed. The third animal was bred only twice, however, because heat periods were not observed. All the animals which did not conceive occupied stanchions in the darkest part of the barn.

The average hemoglobin and hematocrit values for each period are recorded in Tables 25 and 26, respectively. There is wider variation among animals in each treatment than there is among treatments, this indicating no treatment effect. Because all the animals, with one exception, maintained a normal range of values there was no evidence of anemia. The one ex-

TABLE 24

Breeding Records
(Experiment III)

| Animals by treatment | Number of services | Number of days before conception |
|---|---|---|
| **Sulfate sulfur 0.3% (outdoors)** | | |
| 477 | 2 | 269 |
| 482 | 2 | 158 |
| 488 | 1 | 112 |
| Total | 5 | 539 |
| Average | 1.7 | 180 |
| **Molybdenum 50 p.p.m. (outdoors)** | | |
| 479 | 1 | 180 |
| 481 | 1 | 117 |
| 489 | 4 | 206 |
| Total | 6 | 503 |
| Average | 2 | 166 |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m. (outdoors)** | | |
| 480 | 1 (Aborted) | 162 |
| 483 | 1 | 110 |
| 490 | 1 | 129 |
| Total | 3 | 401 |
| Average | 1 | 134 |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m. (indoors)** | | |
| 476 | 4 (not preg.) | 300 |
| 485 | 1 | 144 |
| 487 | 1 | 166 |
| Total | 6 | 610 |
| Average | 2+ | 203 |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m., arsenic 5 p.p.m., rubidium 100 p.p.m., and lead 5 p.p.m. (indoors)** | | |
| 478 | 1 | 177 |
| 484 | 7 (not preg.) | 300 |
| 486 | 2 (not preg.) | 300 |
| Total | 10 | 777 |
| Average | 3.3+ | 259 |

TABLE 25

**Average Hemoglobin Values**
**(Experiment III)**

| Animals by treatment | Periods I | II | III | IV | 300 day averages |
|---|---|---|---|---|---|
| | mg/100 ml of blood | | | | |
| **Sulfate sulfur 0.3% (outdoors)** | | | | | |
| 477 | 11.03 | 10.90 | 11.09 | 11.88 | 11.23 |
| 482 | 9.80 | 10.41 | 10.41 | 9.73 | 10.09 |
| 488 | 10.87 | 10.14 | 10.51 | 9.62 | 10.29 |
| Totals | 31.70 | 31.45 | 32.01 | 31.23 | 31.61 |
| Averages | 10.57 | 10.48 | 10.67 | 10.41 | 10.54 |
| **Molybdenum 50 p.p.m. (outdoors)** | | | | | |
| 479 | 8.80 | 8.56 | 8.78 | 8.52 | 8.67 |
| 481 | 9.92 | 10.88 | 10.48 | 10.25 | 10.38 |
| 489 | 10.21 | 11.10 | 11.02 | 9.25 | 10.40 |
| Totals | 28.93 | 30.54 | 30.28 | 28.02 | 29.45 |
| Averages | 9.64 | 10.18 | 10.09 | 9.34 | 9.82 |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m. (outdoors)** | | | | | |
| 480 | 10.12 | 9.65 | 9.56 | 10.32 | 9.91 |
| 483 | 9.46 | 9.43 | 9.03 | 8.87 | 9.20 |
| 490 | 11.04 | 11.22 | 10.16 | 9.50 | 10.48 |
| Totals | 30.62 | 30.30 | 28.75 | 28.69 | 29.59 |
| Averages | 10.21 | 10.10 | 9.58 | 9.56 | 9.86 |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m. (indoors)** | | | | | |
| 476 | 9.35 | 9.90 | 10.33 | 9.33 | 9.73 |
| 485 | 9.32 | 9.97 | 10.88 | 9.87 | 10.01 |
| 487 | 10.51 | 10.73 | 10.25 | 7.80 | 9.82 |
| Totals | 29.18 | 30.60 | 31.46 | 27.00 | 29.56 |
| Averages | 9.73 | 10.20 | 10.49 | 9.00 | 9.85 |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m., arsenic 5 p.p.m., rubidium 100 p.p.m., and lead 5 p.p.m.** | | | | | |
| 478 | 9.70 | 10.01 | 10.43 | 10.82 | 10.24 |
| 484 | 10.40 | 10.54 | 11.17 | 9.73 | 10.46 |
| 486 | 9.63 | 9.96 | 9.52 | 8.76 | 9.47 |
| Totals | 29.73 | 30.51 | 31.12 | 29.31 | 30.17 |
| Averages | 9.91 | 10.17 | 10.37 | 9.77 | 10.06 |

TABLE 26

Average Hematocrit Values
(Experiment III)

| Animals by treatment | Periods I | II | III | IV | 300 day Averages |
|---|---|---|---|---|---|
| | | | % | | |
| **Sulfate sulfur 0.3% (outdoors)** | | | | | |
| 477 | 34.0 | 33.8 | 35.4 | 38.8 | 35.50 |
| 482 | 29.6 | 29.8 | 33.4 | 31.9 | 31.17 |
| 488 | 33.7 | 33.1 | 32.6 | 31.4 | 32.70 |
| Totals | 97.3 | 96.7 | 101.4 | 102.1 | 99.37 |
| Averages | 32.4 | 32.2 | 33.8 | 34.0 | 33.12 |
| **Molybdenum 50 p.p.m. (outdoors)** | | | | | |
| 479 | 28.0 | 25.1 | 26.8 | 25.9 | 26.45 |
| 481 | 30.0 | 30.5 | 31.2 | 31.0 | 30.68 |
| 489 | 32.8 | 33.9 | 34.2 | 28.9 | 32.45 |
| Totals | 90.8 | 89.5 | 92.2 | 85.8 | 89.58 |
| Averages | 30.3 | 29.8 | 30.7 | 28.6 | 29.86 |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m. (outdoors)** | | | | | |
| 480 | 31.9 | 30.4 | 28.6 | 31.8 | 30.68 |
| 483 | 29.7 | 29.8 | 27.6 | 28.3 | 28.85 |
| 490 | 33.6 | 36.0 | 32.8 | 28.9 | 32.82 |
| Totals | 95.2 | 96.2 | 89.0 | 89.0 | 92.35 |
| Averages | 31.7 | 32.1 | 29.7 | 29.7 | 30.78 |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m. (indoors)** | | | | | |
| 476 | 29.0 | 31.7 | 33.4 | 29.5 | 30.90 |
| 485 | 30.7 | 31.8 | 32.8 | 33.0 | 32.08 |
| 487 | 32.7 | 32.2 | 30.6 | 24.4 | 29.98 |
| Totals | 92.4 | 95.7 | 96.8 | 86.9 | 92.96 |
| Averages | 30.8 | 31.9 | 32.3 | 29.0 | 30.99 |
| **Sulfate sulfur 0.3%, molybdenum 5 p.p.m., arsenic 5 p.p.m., rubidium 100 p.p.m., and lead 5 p.p.m. (indoors)** | | | | | |
| 478 | 30.9 | 31.9 | 35.4 | 38.8 | 34.25 |
| 484 | 34.2 | 35.2 | 35.6 | 31.9 | 34.23 |
| 486 | 31.0 | 31.3 | 30.7 | 27.6 | 30.15 |
| Totals | 96.1 | 98.4 | 101.7 | 98.3 | 98.63 |
| Averages | 32.0 | 32.8 | 33.9 | 32.8 | 32.88 |

ception, animal 487, received the treatment of 50 p.p.m. molybdenum and 0.3% sulfate sulfur indoors. This animal's hemoglobin and hematocrit values dropped from 10.48 to 6.42 and 29.5 to 20.0 respectively within a two month period. Serum protein values are recorded in Table 27. The values for all treatments are similar.

Milk and butterfat production are recorded in Table 28. The average milk production of the animals housed outdoors was found to greatly exceed the average production of the animals housed indoors. As might be expected the animals within each treatment vary in production due to the manner in which they were selected.

Outside of mastitis and flyspray burn, the health of all the animals with one exception was considered normal. The exception was animal 487, which received 50 p.p.m. molybdenum and 0.3% sulfate sulfur and was maintained indoors. This animal showed a loss of appetite during the third and fourth periods of experiment and on several occasions went off feed entirely. She continued to lose weight during period four and had a dull appearance. At 280 days on experiment, when anemia was noted, the animal was given 2 g. of copper sulfate per day. After two weeks with no sign of improvement the animal was slaughtered and the carcass was examined. The only abnormality found was that the liver had large white areas of infection interspersed throughout the tissues of the organ. The incidence of mastitis was unusually high, particularly among the animals housed indoors, but no treatment differences were seen. The animals housed outdoors were relatively free

TABLE 27

Serum Protein
(Experiment III)

| Animals by treatment | Days 0 | 75 | 150 | 225 | 300 | Total |
|---|---|---|---|---|---|---|
| | % | | | | | |
| **Sulfate sulfur 0.3% (outdoors)** | | | | | | |
| 477 | 7.2 | 7.6 | 8.8 | 7.9 | 7.9 | 39.4 |
| 482 | 7.3 | 7.4 | 7.4 | 7.6 | 7.4 | 37.1 |
| 488 | 7.5 | 8.3 | 7.5 | 7.4 | 7.6 | 38.3 |
| | | | | Total | | 114.8 |
| | | | | Average | | 7.65 |
| **Molybdenum 50 p.p.m. (outdoors)** | | | | | | |
| 479 | 6.4 | 7.4 | 6.8 | 7.0 | 7.7 | 35.3 |
| 481 | 7.9 | 8.2 | 8.6 | 8.1 | 8.4 | 41.2 |
| 489 | 7.5 | 8.0 | 7.2 | 7.4 | 7.2 | 37.3 |
| | | | | Total | | 113.8 |
| | | | | Average | | 7.59 |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m. (outdoors)** | | | | | | |
| 480 | 7.9 | 8.0 | 7.4 | 7.4 | 7.8 | 38.5 |
| 483 | 6.4 | 7.2 | 7.1 | 7.0 | 7.8 | 35.5 |
| 490 | 7.9 | 8.0 | 6.9 | 6.6 | 7.4 | 36.8 |
| | | | | Total | | 110.8 |
| | | | | Average | | 7.39 |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m. (indoors)** | | | | | | |
| 476 | 7.5 | 7.8 | 7.6 | 7.0 | 7.0 | 36.9 |
| 485 | 7.0 | 7.8 | 7.7 | 7.8 | 7.6 | 37.9 |
| 487 | 7.4 | 7.8 | 7.8 | 7.3 | 9.1 | 39.4 |
| | | | | Total | | 114.2 |
| | | | | Average | | 7.61 |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m., arsenic 5 p.p.m., rubidium 100 p.p.m., and lead 5 p.p.m. (indoors)** | | | | | | |
| 478 | 7.8 | 8.2 | 7.5 | 7.4 | 7.9 | 38.8 |
| 484 | 6.7 | 7.5 | 7.8 | 7.2 | 7.4 | 36.6 |
| 486 | 7.3 | 6.4 | 7.0 | 6.7 | 6.8 | 34.2 |
| | | | | Total | | 109.6 |
| | | | | Average | | 7.31 |

TABLE 28

Total Milk and Butterfat Production in Pounds
(Experiment III)

| Animals by treatment | | Periods I | II | III | IV | Total milk | Total fat |
|---|---|---|---|---|---|---|---|
| **Sulfate sulfur 0.3% (outdoors)** | | | | | | | |
| 477 | milk | 3088.1 | 2877.0 | 2061.4 | 2386.8 | 10413.3 | |
| | fat | 146.0 | 104.4 | 79.9 | 87.5 | | 417.8 |
| 482 | milk | 1997.3 | 1993.1 | 1692.6 | 1282.9 | 6965.9 | |
| | fat | 92.2 | 92.2 | 75.4 | 62.3 | | 322.8 |
| 488 | milk | 2768.4 | 2541.8 | 1998.8 | 1512.3 | 8821.3 | |
| | fat | 82.7 | 79.2 | 60.7 | 63.8 | | 286.4 |
| | | | | Total | | 26200.5 | 1027.0 |
| | | | | Average | | 8733.5 | 342.3 |
| **Molybdenum 50 p.p.m. (outdoors)** | | | | | | | |
| 479 | milk | 2972.9 | 2513.7 | 1854.5 | 1543.9 | 8885.0 | |
| | fat | 96.9 | 95.5 | 69.2 | 59.0 | | 320.6 |
| 481 | milk | 2769.4 | 2213.5 | 1845.1 | 1329.2 | 8157.2 | |
| | fat | 99.5 | 92.4 | 64.7 | 48.3 | | 304.9 |
| 489 | milk | 3482.6 | 2722.9 | 2140.4 | 1593.3 | 9939.2 | |
| | fat | 113.5 | 83.6 | 66.4 | 52.5 | | 316.0 |
| | | | | Total | | 26981.4 | 941.5 |
| | | | | Average | | 8994.0 | 313.8 |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m. (outdoors)** | | | | | | | |
| 480 | milk | 2913.8 | 2462.4 | 1899.0 | 1414.5 | 8689.7 | |
| | fat | 105.4 | 96.4 | 78.0 | 57.6 | | 337.4 |
| 483 | milk | 3296.0 | 2473.1 | 1516.2 | 1455.1 | 8740.4 | |
| | fat | 108.4 | 88.4 | 57.1 | 61.3 | | 315.2 |
| 490 | milk | 2496.7 | 1279.3 | 678.2 | 189.1 | 4643.3 | |
| | fat | 79.9 | 41.2 | 20.4 | 8.5 | | 150.0 |
| | | | | Total | | 22073.4 | 802.6 |
| | | | | Average | | 7357.8 | 267.5 |

TABLE 28 (Cont.)

Total Milk and Butterfat Production in Pounds
(Experiment III)

| Animals by treatment | Periods I | II | III | IV | Total milk | Total fat |
|---|---|---|---|---|---|---|
| Sulfate sulfur 0.3%, molybdenum 50 p.p.m. (indoors) | | | | | | |
| 476 milk | 1456.9 | 1746.6 | 1029.5 | 806.4 | 5039.4 | |
| fat | 55.9 | 70.7 | 44.5 | 31.1 | | 202.2 |
| 485 milk | 2668.5 | 2001.0 | 1257.7 | 1083.5 | 7010.1 | |
| fat | 85.3 | 79.3 | 42.8 | 41.1 | | 248.5 |
| 487 milk | 2468.0 | 1730.2 | 1050.3 | 466.4 | 5714.9 | |
| fat | 74.0 | 56.7 | 32.6 | 13.4 | | 176.7 |
| | | | Totals | | 17764.4 | 627.4 |
| | | | Average | | 5921.5 | 209.1 |
| Sulfate sulfur 0.3%, molybdenum 50 p.p.m., arsenic 5 p.p.m., rubidium 100 p.p.m., lead 5 p.p.m. (indoors) | | | | | | |
| 478 milk | 2659.5 | 2391.0 | 1603.3 | 1997.5 | 8651.3 | |
| fat | 105.4 | 98.0 | 66.7 | 79.9 | | 350.0 |
| 484 milk | 2372.9 | 1796.9 | 1206.5 | 1117.8 | 6494.1 | |
| fat | 93.3 | 78.0 | 50.8 | 41.3 | | 263.4 |
| 486 milk | 2302.5 | 1639.7 | 1077.2 | 736.8 | 5756.2 | |
| fat | 56.7 | 49.0 | 37.7 | 28.1 | | 171.5 |
| | | | Totals | | 20901.6 | 784.9 |
| | | | Average | | 6967.2 | 261.6 |

of the disease. During the summer, when all the animals were sprayed with an insecticide, several cows which received 50 p.p.m. molybdenum with 0.3% sulfate sulfur developed severe skin burns. These skin conditions healed very slowly. It was unusual that animals from other treatments were not effected.

The milk copper levels for Experiment III are recorded in Table 28 of the Appendix. Except for the values of the 150 day samples, the milk copper levels were in good agreement with those of Experiment I and II. The values for the 150 day samples for all animals appear to be inaccurate due to errors in analysis. The reason for these errors is not evident. The levels for milk molybdenum for this experiment are recorded in Table 28 of the Appendix. The means of these values for each treatment are plotted in Figure 11. The animals which received 50 p.p.m. of molybdenum with no sulfate had much higher milk molybdenum levels than those which received 50 p.p.m. of molybdenum with 0.3% sulfate sulfur. These results are in complete agreement with a comparison of Experiments I and II. It provides further evidence that increased sulfate sulfur in the diet reduces the excretion of molybdenum in the milk.

The values for serum copper and molybdenum for this experiment are recorded in Table 29 of the Appendix. The mean serum copper values, which are plotted in Figure 12, show that serum copper decreased during the course of the experiment for all treatments. Both of the treatments which received molybdenum with sulfate sulfur had serum copper levels which were subnormal at the end of the experiment. The mean serum

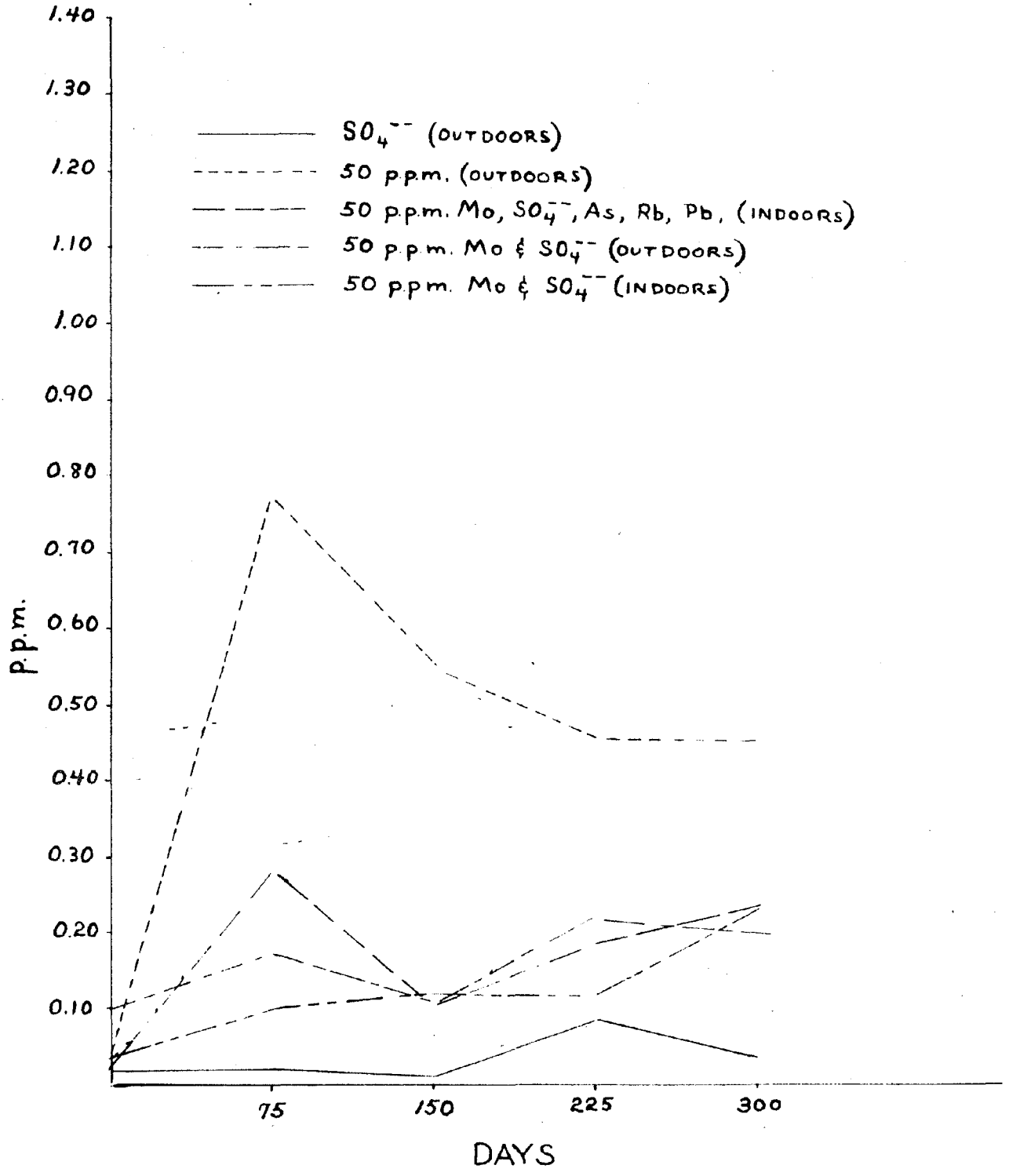


Fig. 11. Milk molybdenum levels for Experiment III.

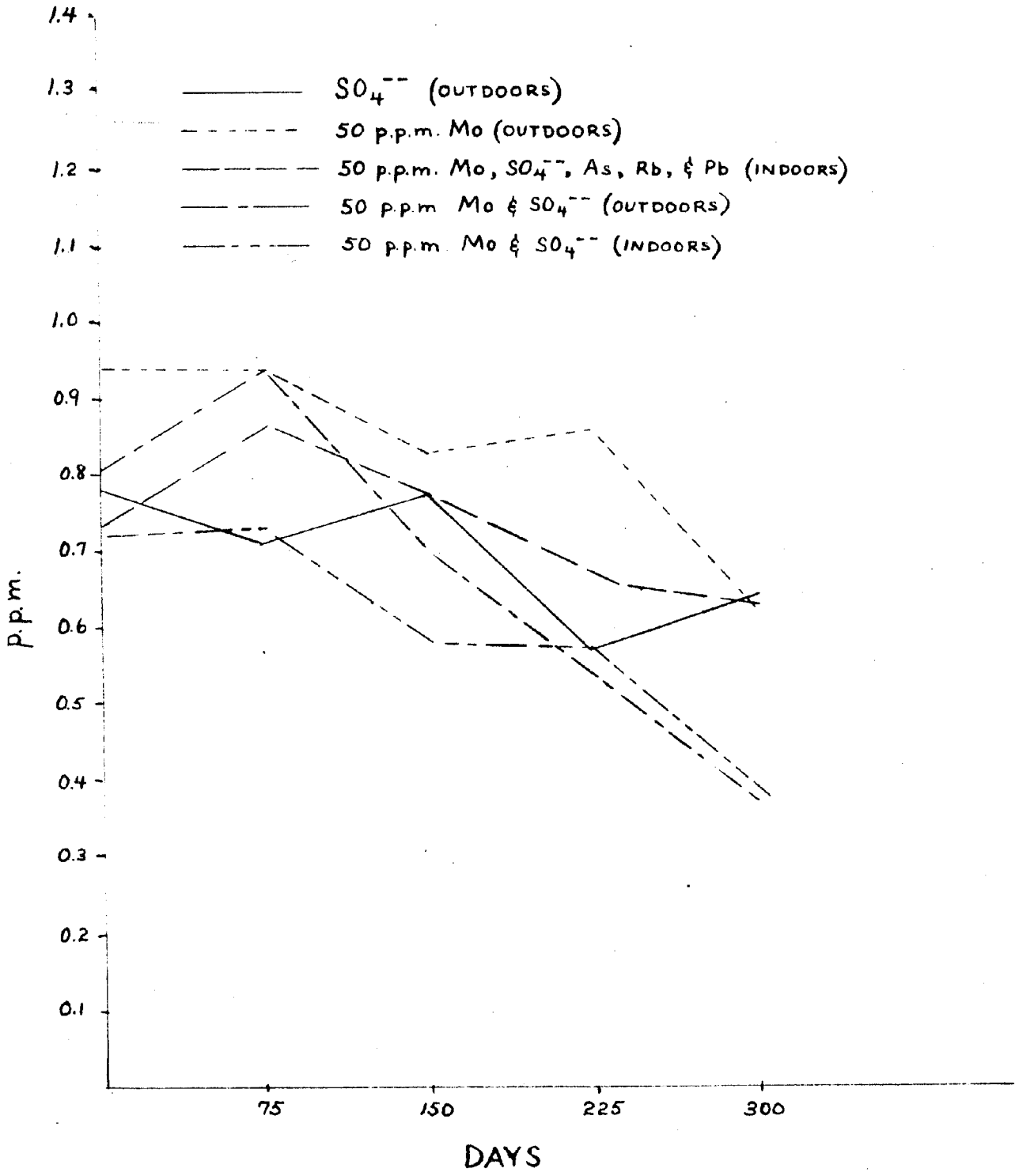


Fig. 12. Serum copper levels for Experiment III.

molybdenum levels are plotted in Figure 13. The level for the treatment which received 50 p.p.m. of molybdenum was higher than for the treatments which received 50 p.p.m. of molybdenum with 0.3% sulfate sulfur. These results are in agreement with the results of Experiments I and II, and indicate that the sulfate level of diet has an effect on the serum molybdenum level.

Liver molybdenum and copper levels are recorded in Table 30 of the Appendix. The mean liver copper levels are plotted in Figure 14. There was a general decrease in the levels of liver copper for all levels of molybdenum intake through the first 225 days of the experiment. During the last 75 days both outdoor treatments of molybdenum showed unexplained increases of liver copper. The treatment which received 0.3% sulfate sulfur showed an increase for the first 150 days of experiment and then a decrease for the remaining 150 days. The mean liver molybdenum levels, which are plotted in Figure 15, show that the 50 p.p.m. of molybdenum treatment had a higher liver molybdenum level than the treatments which received 50 p.p.m. of molybdenum with 0.3% sulfur. This comparison shows the depressing effect sulfate has on molybdenum deposition in the tissues. The level of molybdenum in the liver of the treatment which received 0.3% sulfate sulfur increased as the experiment progressed.

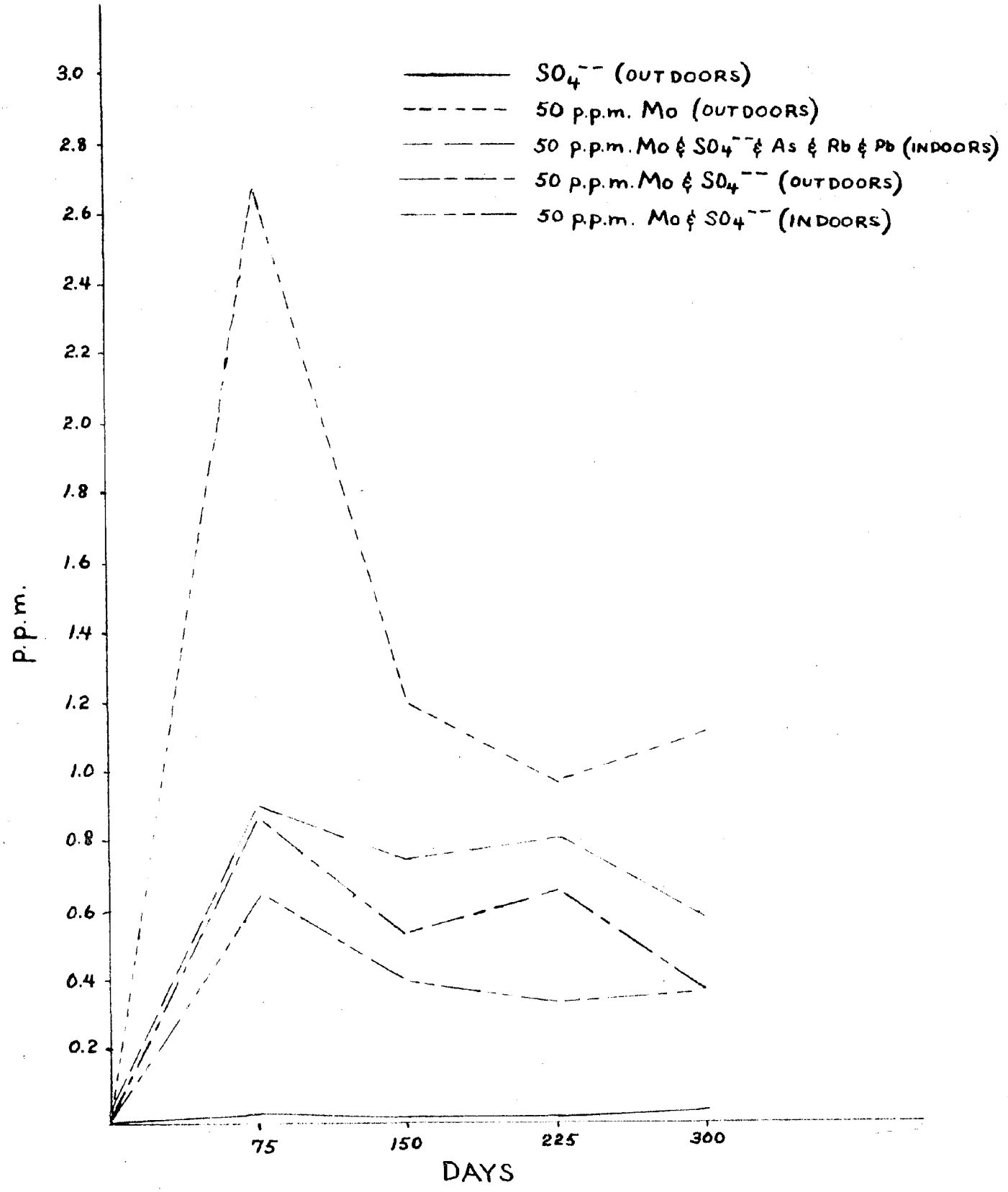


Fig. 13. Serum molybdenum levels for Experiment III.

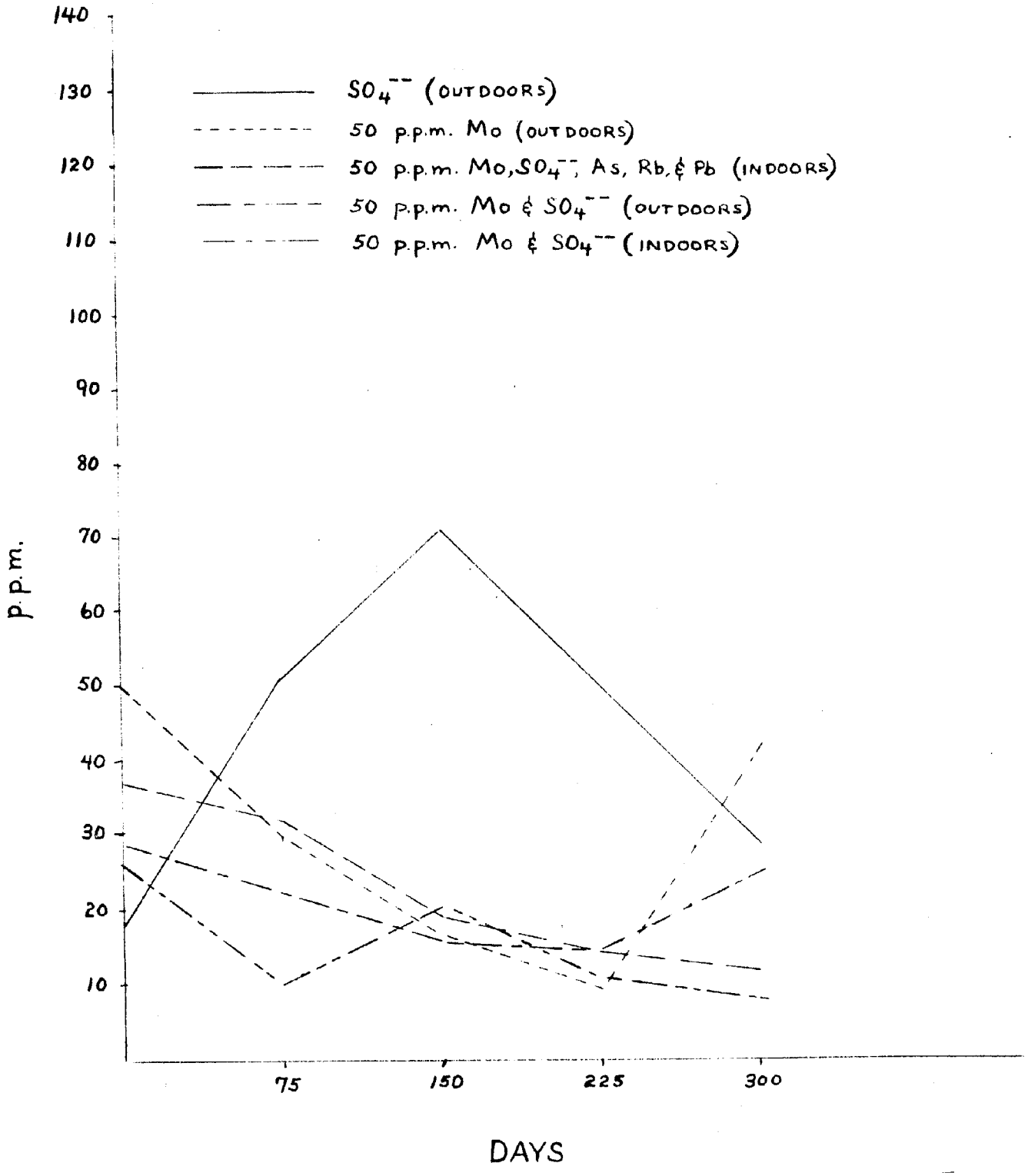


Fig. 14. Liver copper levels for Experiment III.

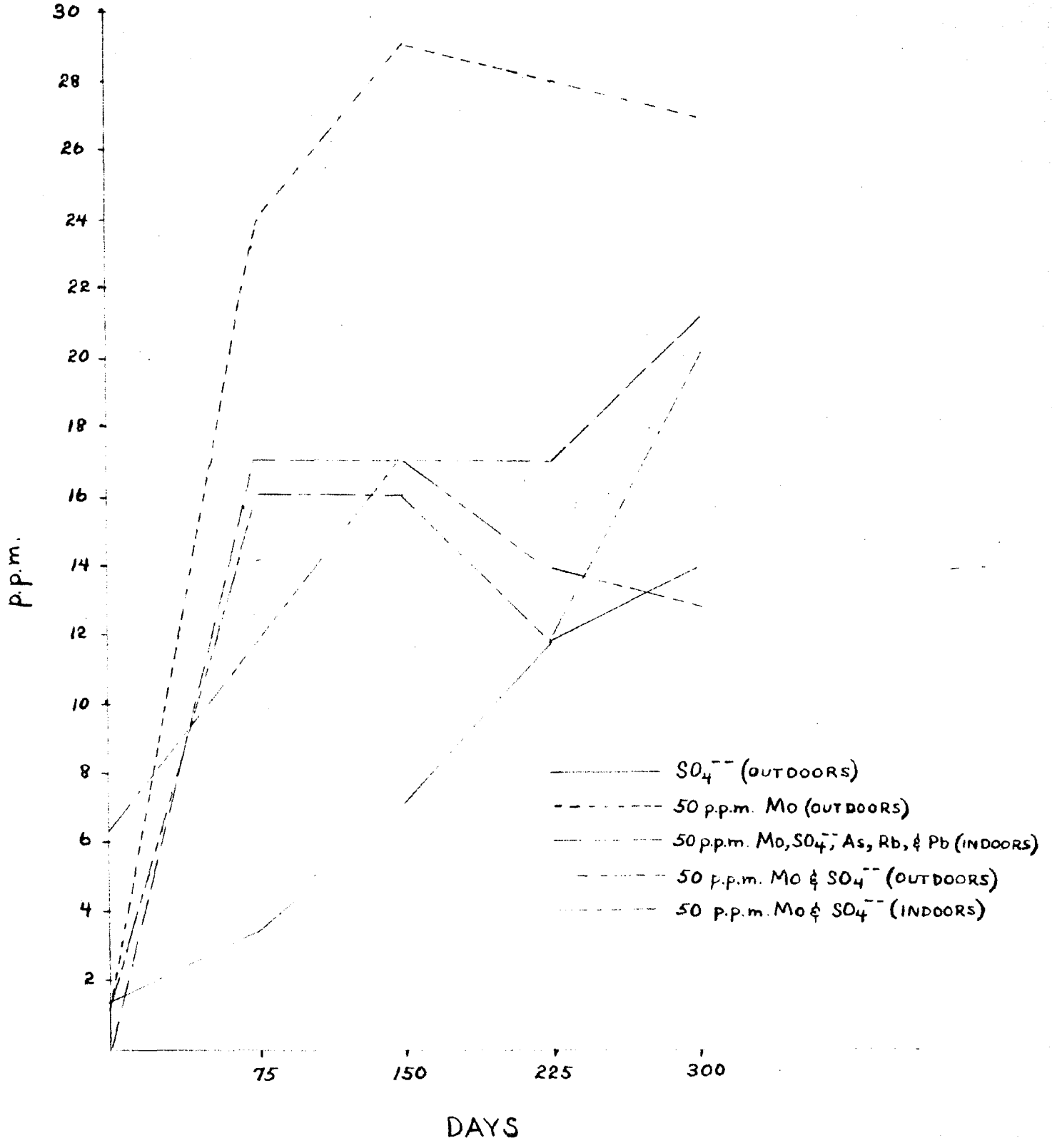


Fig. 15. Liver molybdenum levels for Experiment III.

## EXPERIMENT IV

Attempts to measure the absorption of copper-64 by the blood with a windowless proportional flow counter were not successful due to the low energy of the Beta radiation of this isotope. As a result accurate counts for the first 24 hours of experiment were not obtained during the first trial. The gamma well scintillation counter was used in all counting thereafter and was found to be very reliable. In Figure 16 the levels of copper-64 obtained from blood for Trials I and II are plotted. In both trials the steer which received 50 p.p.m. with 0.3% sulfate sulfur had a slightly higher level of copper-64 in the blood than was found for the steer which received 0.3% sulfate sulfur. The levels of copper-64 found in the blood for Trials III and IV are plotted in Figure 17. In Trial III steer 498 which received 50 p.p.m. molybdenum showed considerably higher levels of copper-64 in the blood than were found for the control steer 499. In Trial IV with the treatments reversed, both steers, 498 and 499, showed similar levels of copper-64 in the blood for the first 12 hours. After this time, however, steer 498, which received no molybdenum, showed higher levels than steer 499 which received 50 p.p.m. molybdenum. The levels ofcopper-64 found in the blood for Trials V and VI are plotted in Figure 18. No differences were found between the blood levels of the steer which received 0.3% sulfate sulfur and the control steer in either Trials V or VI.

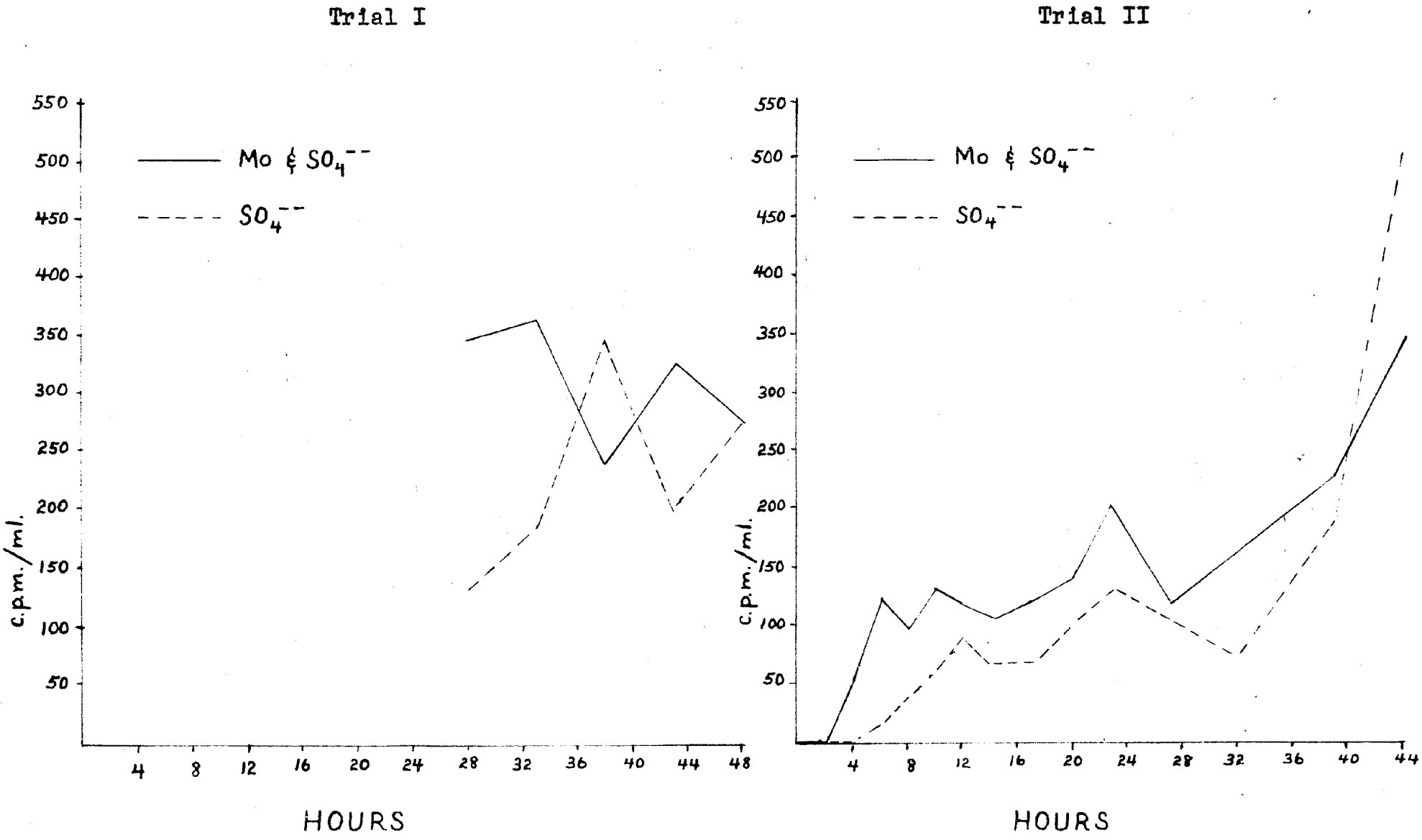


Fig. 16. Levels of copper-64 in blood for Trials I and II.

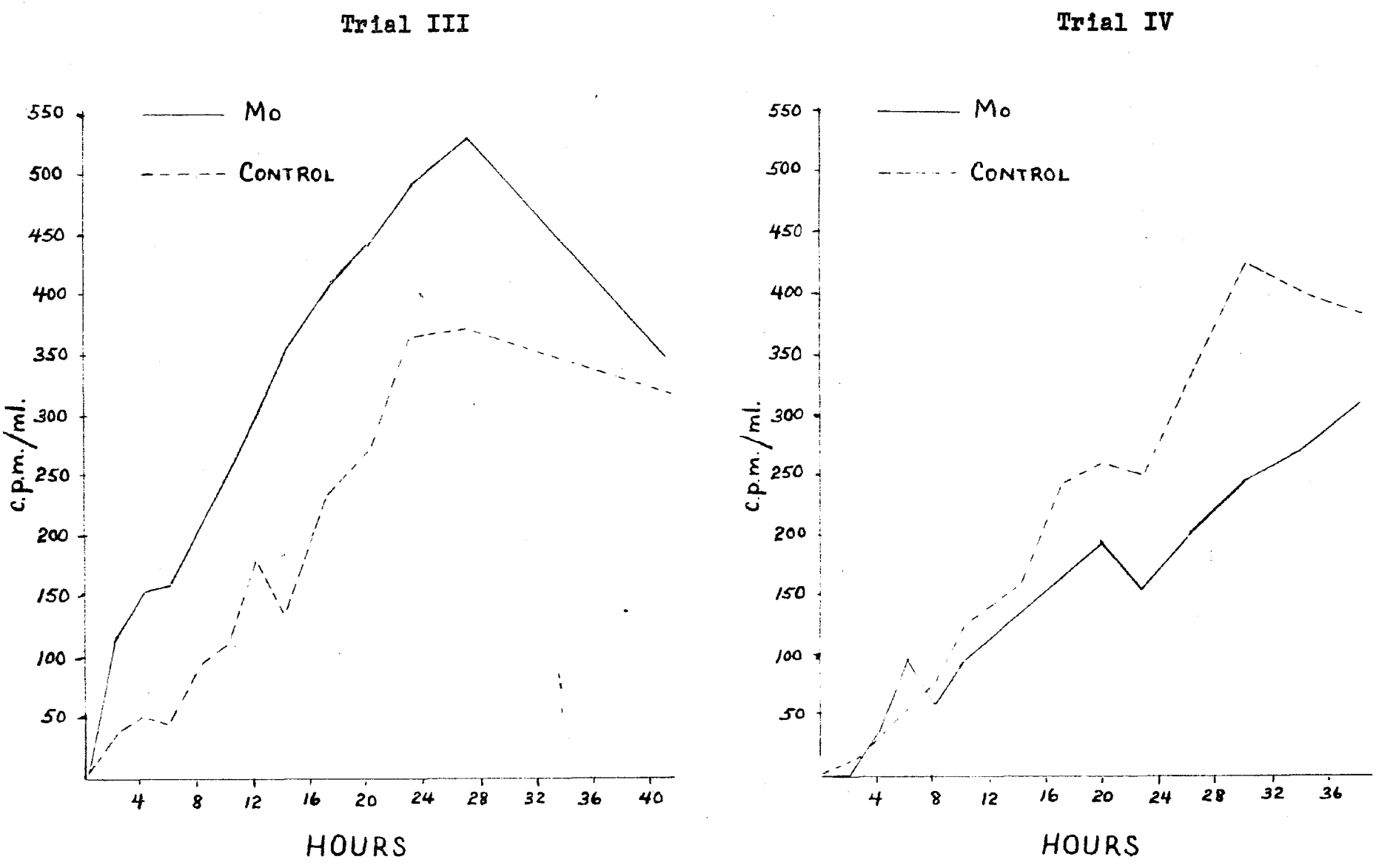


Fig. 17. Levels of copper-64 in Blood for Trials III and IV.

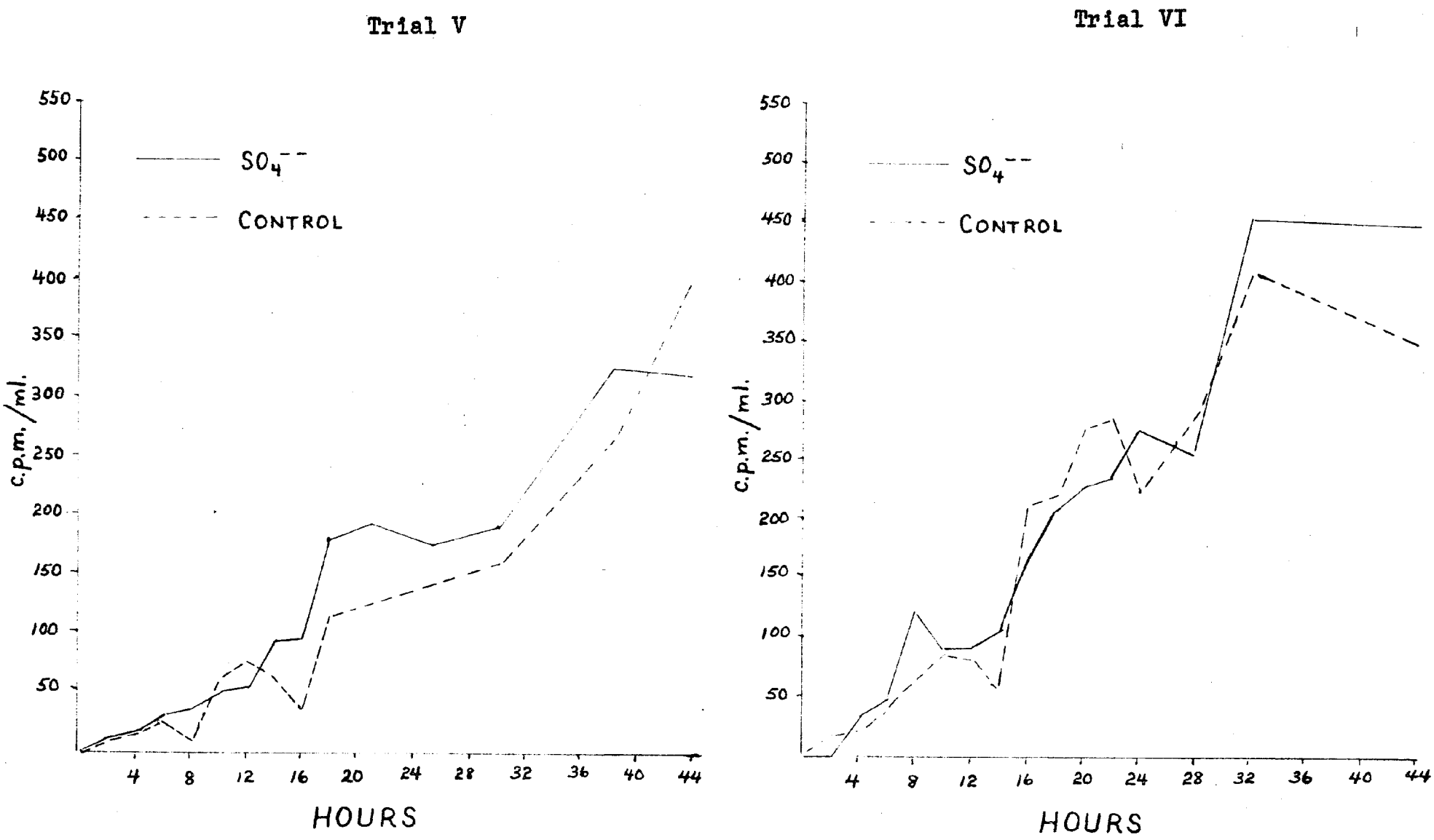


Fig. 18. Levels of copper-64 in blood for Trials V and VI.

A very surprising fact discovered was the unusually low absorption of the copper-64 by the steers. In all six trials, the recovery of copper-64 in the feces was greater than 96.5%. Copper-64 was recovered in the feces as early as six hours after administration and continued to be excreted for five days. The level of copper-64 found in the urine was not sufficient to count accurately without concentrating the sample. Results from measuring the activity of evaporated urine samples indicated less than 0.5% of the copper-64 administered was excreted by this pathway. Calculation of the level of copper-64 in the urine was hampered by feces contaminations on several occasions.

The copper-64 contents of liver samples obtained 24 hours after administration of the isotope are recorded by trials in Table 29. In Trial I the steer which received 50 p.p.m. molybdenum and 0.3% sulfate sulfur deposited only 30 per cent as much copper-64 in the liver as the steer which received 0.3% sulfate. Similar results were found in Trial II where the steer which received 50 p.p.m. molybdenum with 0.3% sulfate sulfur deposited 13 per cent of the level of copper -64 in the liver of the steer which received 0.3% sulfate sulfur. In Trials III and IV, where sulfate was removed from the ration, the deposition of copper-64 in the livers of the steers which received 50 p.p.m. molybdenum was respectively 21 and 6 per cent of the level found in the steers which did not receive molybdenum. In comparing the results of Trials V and VI, which was designed to test the

TABLE 29

Copper-64 levels in liver
(Experiment IV)

| Trial | Animal | Dose | Accumulation of copper-64 in liver during 24 hrs. counts per minute/gr.* | Treatment |
|---|---|---|---|---|
| I | 473 | 5 mc. | 1405 | $SO_4^=$ + Mo. |
| | 474 | 5 mc. | 4690 | $SO_4^=$ |
| II | 473 | 5 mc. | 5894 | $SO_4^=$ |
| | 474 | 5 mc. | 766 | $SO_4^=$ + Mo. |
| III | 498 | 5 mc. | 1887 | Mo. |
| | 499 | 5 mc. | 9048 | ——— |
| IV | 498 | 5 mc. | 4945 | ——— |
| | 499 | 5 mc. | 315 | Mo. |
| V | 473 | 10 mc. | 1440 | $SO_4^=$ |
| | 474 | 10 mc. | 1520 | ——— |
| VI | 498 | 10 mc. | 4306 | ——— |
| | 499 | 10 mc. | 4242 | $SO_4^=$ |

* Corrected to time when copper-64 level was equal to dose.

effect of adding sulfate to the diet on the deposition of copper-64 in the liver, differences of 5 and 1 per cent greater deposition occurred in the controls, respectively.

The data obtained from the analysis of serum, liver and milk were tested for statistical significance by Professor Owen B. Durgin, Agriculture Experiment Station Statistician. A summary of the results of this statistical analysis is found in the Appendix. These data were used as a guide for the reporting of results in this thesis.

# DISCUSSION

Although Davis (39), Cunningham (31), Miller (118), and Dick (52) reported that symptoms of molybdenum toxicity occurred when sheep and cattle were fed rations containing levels of from 6 to 36 p.p.m. of molybdenum, similar results were not obtained on this experiment. Levels of 5 to 50 p.p.m. were fed without developing the usual symptoms of toxicity such as loss of body weight, diarrhea, achromotrichia, anemia, and nervous disorders.

The analysis of milk, serum, and liver for copper and molybdenum, however, revealed some important changes which were the result of treatment. Molybdenum levels were shown to increase in the milk, serum and liver with increased feeding of sodium molybdate. These data indicate that molybdenum is absorbed at a rate which is dependent on the level of this element in the diet. The fact that all of the treatments showed stabilization of these molybdenum levels indicates that the excretion of this element is a function of its absorption. The data obtained from analysis of liver and serum are consistent with work reported by Dick (52) with sheep and Cunningham (37) with cattle. Similarly, the levels of molybdenum in the milk are in good agreement with work reported by Archibald (9) in which four cows were fed an estimated level of 10 p.p.m. molybdenum.

The analysis of milk, liver, and serum for copper is of even greater interest because molybdenum toxicity has been

described by Cunningham (31), Davis (39), and Dick (52) as complicated copper deficiency. In this experiment the copper levels of the milk showed a change due to time but not to treatment. Apparently, copper secretion in milk varies with stage of lactation. No supporting evidence of this fact could be found in the literature. The copper contents of both serum and liver were shown to decrease with an increase of molybdenum in the diet. The serum copper levels for animals which received from 5 to 50 p.p.m. of molybdenum were just under the normal range of 80 to 120 micrograms per 100 ml. of serum given by Underwood (152) and Marston (112). Adams et al. (1) have reported, however, finding levels of serum copper in cattle which were below this range with no apparent copper deficiency. It should be noted also that the level of copper found for these animals was from 10 to 20 micrograms per 100 ml. serum above the levels reported by Cunningham (31), Marston (112), and Davis (39) to be present in copper deficient animals.

At the end of Experiment I, several animals which received supplemental molybdenum had levels of liver copper as low as 9 to 12 p.p.m. This level of copper is far below the normal range of 100 to 400 p.p.m. reported by Underwood (152) and Marston (112). These levels are as low as those reported to occur in copper deficiency by Bennetts (14), Cunningham (31), Davis (39), and Marston (112). The existence of such low liver copper levels with no evidence of deficiency in these cows indicates that this species can tolerate

a wider range than indicated by the above investigators.

It is evident from these data that the feeding of molybdate lowered the liver copper and serum copper levels. These results are in good agreement with those reported by Cunningham (37) and Mylrea (128) in cattle. Dick (52) working with sheep reported that molybdenum had no effect on serum and liver copper levels unless sufficient inorganic sulfate was present in the diet. Analysis of hay fed during Experiment I revealed a copper content of 1.85 to 3.40 p.p.m., and a sulfur content of 0.16 to 0.26 per cent. The grain used in Experiment I had a copper content of 8.9 to 10.3 p.p.m., and a sulfur content of 0.28 to 0.34 per cent. The analysis of water showed only a trace of copper which was negligible in relation to the amount found in the feed. The overall estimates in the diet were for copper of 4.5 to 6.0 p.p.m. and for sulfur of 0.25 to 0.31 per cent. This level of copper was considered just adequate to maintain normal health in dairy cattle. The sulfur level was considered more than adequate but not unusually high. Unfortunately, the amount of sulfur as inorganic sulfate was not determined and therefore no comparison can be made with the work of Dick (52).

Because both liver and serum copper levels were lowered, it could be reasoned that sufficient inorganic sulfate was present to effect the molybdenum-copper interaction. If this were so, however, molybdenum toxicity symptoms should have been found in these cattle, but no such symptoms were observed. Cunningham (37) and Mylrea (128) reported similar

results from feeding levels up to 16 p.p.m. of molybdenum.

These data indicate that other unidentified conditions necessary for development of molybdenum toxicity were not present. It could be reasoned that the level of inorganic sulfate was not high enough to alter liver and serum copper to a point where toxicity symptoms could occur. A second possibility is that molybdenum was not in the proper form to cause the toxicity. Ferguson et al. (63) reported that 70 to 80 per cent of the molybdenum found in pasture plants was in the form of soluble molybdate. This information indicates that the molybdate salt should be equally as effective as plant molybdenum in causing the toxicity.

Experiment II was designed identically to Experiment I except that 0.3% sulfate sulfur was added to the basal diet. The development of achromotrichia and alopecia by the animals which received 50 p.p.m. molybdenum indicated that additional inorganic sulfate was a factor in producing molybdenum toxicity. Analysis of serum, liver, and milk for copper and molybdenum do not explain the role of this inorganic sulfate. Although the serum, liver and milk molybdenum levels of Experiment II were increased due to molybdenum feeding, these increases were small compared to those received in Experiment I. This comparison makes it evident that inorganic sulfate added to the diet alters molybdenum metabolism. Dick (52) has reported identical results from the analysis of serum and liver samples from sheep. Cunningham (37) also showed this effect of sulfate on the serum and liver molybdenum levels in cattle. Ac-

cording to Dick (52), inorganic sulfate in the diet lowers the absorption of molybdenum, thus accounting for the lower levels of molybdenum found in the serum and liver.

The copper levels found in the serum and liver samples of this experiment are of even greater interest because achromotrichia and alopecia occurred in the 50 p.p.m. of molybdenum animals. Contrary to the expected results, the liver and serum copper levels of the animals in Experiment II were as high or higher than those of Experiment I. Further evidence that lower serum and liver copper levels are not responsible for the appearance of molybdenum toxicity symptoms is found by comparing the treatments in Experiment II. At the end of 300 days the levels of serum copper and liver copper are about the same for the 20 and 50 p.p.m. of molybdenum with 0.3% sulfate sulfur treatments, while only the 50 p.p.m. of molybdenum with sulfate showed achromotrichia and alopecia. Of even greater significance is the fact that both 100 and 200 p.p.m. of molybdenum treatments had high serum copper levels and not unusually low liver copper levels (10 to 47 p.p.m.) but showed all of the symptoms of severe molybdenum toxicity.

Except for the treatments of 100 and 200 p.p.m. of molybdenum, which are not typical of those found under natural conditions, only mild symptoms of molybdenum toxicity were observed and these occurred only in the 50 p.p.m. of molybdenum treatment. Evidence that additional time was not a factor in the lack of severity this toxicity was provided by

continuing two of the 50 p.p.m. of molybdenum with sulfate animals through a second lactation. These animals developed more marked changes in hair condition, but showed no further evidence of toxicity. These data indicate that factors other than inorganic sulfate are responsible for increasing the toxicity of molybdenum because the level of inorganic sulfate in this experiment far exceeds that found in plants.

In Experiment III the effect of stress and the addition of some ions not found in New Hampshire hays were tested. In many diseases stress is known to be an important factor in causing the onset of symptoms of the malady. Cows were subjected to extreme cold by housing them in a pole type barn and feeding them in an open lot. The results of this experiment indicated that this type of stress had no additional effect on molybdenum toxicity. All animals which received 50 p.p.m. of molybdenum with 0.3% sulfate sulfur developed achromotrichia and alopecia, while animals receiving either 50 p.p.m. of molybdenum or 0.3% sulfate sulfur did not. These results are in complete agreement with those found in Experiments I and II. The addition of lead, arsenic, and rubidium to the diet produced no further increase in molybdenum toxicity, thus indicating that none of these ions are complicating factors.

The development of toxicity symptoms other than achromotrichia and alopecia occurred in one 50 p.p.m. of molybdenum with 0.3% sulfate animal housed indoors. No good explanation for this occurrence can be found. This animal was

small and less rugged looking than the other animals at the start of the experiment, and it is believed that a lack of desirable constitution and vigor was the reason for the greater susceptibility to the experimental treatment. Evidence that the constitution of an animal can influence the incidence of toxicity is reported by Hayashi (82). This worker found that Holstein calves showed less susceptibility to molybdenum toxicity than the native cattle of Japan. The ability of some animals to withstand larger doses of molybdenum than others may account for part of the difference in the occurrence of molybdenum toxicity under natural conditions and experimental conditions. Frequently, molybdenum toxicity has occurred in range areas where less attention has been devoted to animal vigor, whereas animals selected for experimental purposes are generally of good constitution.

In trying to find a reason for differences in the level of molybdenum required to cause toxicity in these experiments and in areas where the malady occurs naturally, several other factors should be considered. One strong possibility is the effect of internal parasites. Bremner (19), Gibson (71), and Seekles (143) have reported a lowering of serum and liver copper by heavy infestations of internal parasites. In many of the areas where molybdenum toxicity has occurred, the climate also has been favorable for internal parasites. Kretschmer and Beardsley (91) reported that many cattle suspected of molybdenum toxicity in Florida responded to a treatment of phenothiazine. Animals in cooler climates and especially if confined in a barn are not normally infected

with parasites. An examination of all the animals on this experiment for internal parasites was negative.

The general state of nutrition may be another factor involved in molybdenum toxicity. Because molybdenum toxicity has occurred frequently in range areas where cattle are fed only grass, a shortage of protein may have been a complicating factor. Miller and Engel reported that the protein level of the diet greatly influenced the development of molybdenum toxicity in rats. The use of a 17% grain mixture in the present experiments provided more than enough protein to meet the animals' requirements. Recently, Davis (40) reported the isolation of an organic complexing agent from grass which reduced the availability of copper to the animal. It is possible that such a complexing agent could reduce the copper absorption and thereby increase molybdenum toxicity.

Extensive analysis of forage from areas where molybdenum toxicity occurs occasionally has shown high levels of zinc. Work with rats reported by Gray and Ellis (73) has shown that this element will cause anemia which can be corrected by copper administration. Because the zinc ion has a size and electronic structure similar to copper, it may act as a biological antagonist to copper. There also is a possibility that some other element or ion present in these forages increases the toxicity of molybdenum.

The question of whether molybdenum without the presence of any inorganic sulfate has an effect on copper metabolism is still not clear. Only Dick (52) has reported that molybdenum without inorganic sulfate does not change serum or liver copper

levels. Because it is very difficult to compound a ration with no inorganic sulfate, this question will be difficult to answer. That inorganic sulfate increases the toxicity of molybdenum has been shown by these experiments and by the work of Dick (52), Cunningham (37), and Mylrea (128). The mechanism by which sulfate brings about this effect is not clear.

The results of Experiment IV indicate that Dick's (52) theory of sulfate prohibiting the transport of molybdenum and copper across a membrane is in error. In four paired trials neither sulfate nor sulfate with molybdenum was shown to have any effect on the absorption of orally administered copper-64 as measured by the level of radioactivity in the blood. These trials indicated that molybdenum reduced the deposition of copper-64 in the liver.

The results of these trials also tend to disprove the theory proposed by Halverson et al (178), that large amounts of sulfide formed from sulfate reduction in the rumen combines with copper to form insoluble copper sulfide, which is unavailable to the animal. Because copper-64 levels of the blood were not affected by sulfate addition to the diet, it appears that the formation of insoluble copper sulfide must have been small. The results of sulfide determinations made on rumen samples during Experiment III indicated large amounts of the ion present in the rumen liquid for the first two hours following the feeding of sulfate. Treatments which received 0.3% sulfate sulfur had higher levels of sulfide

than the treatment which received no sulfate. Differences were not found to be due to addition of molybdenum to the diet, however. In all treatments sulfide in rumen liquid was undetectable a few hours after feeding, and only a trace of sulfide was detectable in the solid portion of rumen content.

It would seem quite probable that much of the copper found in plants would be released after two hours of digestion. If such were the case the formation of copper sulfide would not be appreciable. If this is the case a decrease in the copper level of liver would reflect a negative balance of copper. Using this line of reasoning many of the conditions found in this experiment can be explained. In Experiment I, the liver copper level for all molybdenum treatments was shown to increase during the last 75 days of experiment. These results are consistent because milk production was decreased and therefore less copper was excreted from the body. Similar results were obtained with the animals receiving 50 p.p.m. molybdenum in Experiment III. The liver's serving as a storage organ also would explain why serum copper levels did not drop as rapidly as the liver copper levels. It would not be until the liver had been depleted of all the copper stores that an appreciable decrease of serum copper would be observed.

If liver copper can serve as an indicator of the animal's balance of copper, it may help explain the action of molybdenum on copper. It does not seem probable that molyb-

denum acts as a biological antagonist to copper because chemically these two elements are quite different. Recent work reported by Scaife (139) indicates that molybdates inhibit the activity of certain copper containing enzymes. With such loss of enzyme activity it would seem probable that the body would increase the production of these copper enzymes and thus place a greater demand on the body for copper. The sudden increase in serum copper reported by Dick (52) when molybdenum was fed would indicate that such a body response was taking place. If these enzymes are blocked due to the action of molybdate they probably would be gradually excreted from the body and thus deplete the levels of copper in the body.

Recent reports by Singer (145) indicate a similar blocking of copper containing enzymes by both sulfide and sulfate ions. This action can be used to explain the effect inorganic sulfate has on increasing molybdenum toxicity. Because molybdenum has been shown by Mills et al. (124) to reduce the production of sulfide oxidase, abnormally high levels of sulfide ions could build up in the blood and tie up copper enzymes. The animal would respond to such a loss of enzyme activity by an increase in the production of these enzymes and thus create a greater demand for copper. If such an increased demand for copper placed the animal in a negative balance, the animal would develop copper deficiency.

This mechanism also could be used to explain why high levels of molybdenum cause the symptoms of molybdenum toxicity

to appear before the liver and serum copper levels are below normal. In such conditions the high concentration of both sulfide and molybdenum would tie up the copper enzymes in the body at a rate faster than the body could produce them and hence the symptoms of copper deficiency would appear. At the same time the excretion of these now useless copper containing enzymes may be slower than the rate of blocking and therefore serum and liver copper levels would still be normal or even above normal. The animals which received 100 and 200 p.p.m. of molybdenum in Experiment II were examples of animals showing molybdenum toxicity while having high serum and liver copper levels.

The appearance of achromotrichia and alopecia in animals on a diet of 50 p.p.m. molybdenum with 0.3% sulfate sulfur might be described as the first defense mechanism of the animals. Hair production being a less essential body process may be neglected by the body in preference to more important functions. This condition therefore presents a good indication of the stage of copper deficiency and may be a better index than the level of serum copper. It would appear that the low level of copper found in the liver and serum of calves born to the cows continued through a second lactation while receiving molybdenum and sulfate, was also an effort to conserve copper for vital body processes.

According to such an explanation, molybdenum toxicity causes an increase in rate of copper metabolism which results in a deficiency of this element. This scheme would account

for the fact that copper administered to animals suffering from this malady brings about complete recovery, as was shown in Experiment III.

Evidence against such a mechanism being responsible for the copper-molybdenum-sulfate interrelationship has been reported by Cox et al. (28). These workers fed high dietary levels of molybdenum to rats and calves and determined the effect of this treatment on liver and heart enzymes. The results of these experiments indicated no difference in liver tyrosine oxidase and heart cytochrome oxidase activity between control and experimental groups. It should be pointed out, however, that no sulfate was added to the diets of these animals and there were no toxicity symptoms recorded for the calves. Furthermore, the levels of liver copper in all experimental groups were extremely high, thus indicating that these animals were not suffering from copper deficiency.

In view of the results obtained by Scaife (139) and Mills et al. (124) with molybdenum inhibiting the activity of copper containing enzymes, more research on the effects of both molybdate and ions of sulfur on copper containing enzymes would seem most promising in explaining this interrelationship.

# SUMMARY AND CONCLUSIONS

Four experiments designed to investigate the copper-molybdenum-sulfate sulfur interrelationships in cattle were carried out. In ExperimentI, fifteen Holstein first calf heifers were used to observe the effects of adding from 5 to 50 p.p.m. of molybdenum to a diet low in copper, molybdenum, and sulfur. At the end of 300 days on experiment, symptoms of molybdenum toxicity such as achromotrichia, alopecia, diarrhea, emaciation, and anemia were not found for any level of treatment. Animal production as measured by milk production and changes in body weight was not measurably affected by treatment. Analysis of serum, liver, and milk samples from these animals for copper and molybdenum indicated the following effects due to treatment.

1. The serum, liver, and milk molybdenum levels increased with increased molybdenum in the diet.

2. The feeding of molybdenum did not have any effect on the level of copper in the milk.

3. Treatments which received supplemental molybdenum showed decreased liver copper levels during the first 225 days of the experiment. During the last 75 days of experiment these animals showed increases in liver copper which were presumed caused by a lower molybdenum intake due to decreased feed consumption and less copper loss from the body due to decreased milk production.

4. Although the serum copper levels of all molybdenum

treatments decreased during the experimental period, all treatment means were still in the published normal range.

5. The control animals showed increases of serum and liver copper during the experiment.

In Experiment II fifteen first calf heifers were used to observe the effects of adding 0.3% sulfate sulfur to the treatments used in Experiment I. At the end of 225 days on experiment the cows receiving 50 p.p.m. molybdenum developed achromotrichia and alopecia. Continuation of the treatment for the remainder of the first lactation and for an additional lactation for two animals produced no further symptoms of molybdenum toxicity. The treatments receiving less than the 50 p.p.m. molybdenum level did not develop any molybdenum toxicity symptoms. The analysis of liver, serum, and milk for copper and molybdenum indicated the following effects due to adding sulfate to diets containing various levels of molybdenum.

1. The serum, liver, and milk molybdenum levels were lower for each treatment level of molybdenum than were observed in Experiment I. This indicated lower molybdenum absorption.

2. Milk copper levels were significantly lower in this experiment than in Experiment I, but there were no differences among molybdenum treatments.

3. Treatments receiving molybdenum continuously lost liver copper until they reached a low level and these low levels persisted until the end of the experiment. Because

no increase was observed during the latter part of the lactation as was observed in Experiment I, the addition of sulfate appeared to increase the copper deficiency caused by high levels of molybdenum.

4. Serum copper levels for all treatments receiving molybdenum decreased during the experiment, while the level for the treatment which did not receive molybdenum remained constant.

5. Four animals which were fed levels of 100 and 200 p.p.m. of molybdenum with 0.3% sulfate sulfur for 75 days developed archromotrichia, diarrhea, loss of nervous control, and emaciation, and milk production stopped. These symptoms occurred while serum copper levels were high and liver copper levels were not unusually low. These animals did not develop anemia.

6. Calves born to dams showing achromotrichia and alopecia had unusually low serum and liver copper levels. Although these calves appeared healthy, they had brown and white coats instead of black and white.

In Experiment III, fifteen Holstein first calf heifers were used to test the effect of stress and other possible complicating factors on the development of molybdenum toxicity. Nine animals were subjected to extreme cold by being housed in a pole barn and fed in an open lot for 300 days on experiment. The stress of extreme cold had no additional effect on the development of molybdenum toxicity. These nine animals, which were divided into the three treatment, 0.3% sulfate

sulfur, 50 p.p.m. molybdenum with 0.3% sulfate sulfur, and 50 p.p.m. molybdenum, showed identical results with those obtained from Experiment I and II. The results obtained from adding lead, arsenic, and rubidium to a diet containing 50 p.p.m. molybdenum with 0.3% sulfate sulfur indicated that these elements caused no additional effect on the development of molybdenum toxicity. During Experiment III one animal which received 50 p.p.m. molybdenum with 0.3% sulfate sulfur went off feed, became emaciated, and developed anemia after 275 days on experiment. This animal became progressively worse and, after showing no improvement when fed 20 p.p.m. copper for one week, was slaughtered. This occurrence was believed due to a lack of constitution and vigor in the animal. The analyses of serum, liver, and milk for copper and molybdenum were in good agreement with the results obtained from Experiments I and II.

In Experiment IV a series of six trials using copper-64 were carried out to study the effects of sulfate sulfur, molybdenum, and molybdenum with sulfate sulfur on copper metabolism. The data from these trials indicated the following:

1. The level of copper-64 absorbed into the blood is small and is not affected by the presence of high levels of sulfate sulfur, molybdenum, or molybdenum with sulfate sulfur in the diet.

2. The addition of molybdenum to the diet of steers reduced by 70 to 94% the amount of copper-64 deposited in the

in the liver.

3. The amount of copper-64 recovered in the feces of these steers was greater than 96.5% of that fed, while amounts found in the urine was less than 0.5%.

4. Copper-64 appeared in the feces as early as six hours after administration and continued to be excreted for five days.

Several factors are suggested as possible explanation as to why much higher levels of molybdenum are required to produce molybdenum toxicity experimentally than are found in areas where the toxicity occurs, under farm conditions. These factors are low levels of protein in the diet, internal parasites, high levels of zinc, and the presence of complexing agents which would render plant copper unavailable to the animal.

The theory that molybdenum toxicity in ruminants is a conditioned copper deficiency is quite well established. At this time, however, the mechanism by which molybdenum and sulfate sulfur cause this conditioned copper deficiency is not understood. Evidence that copper absorption is not affected by high levels of sulfate and molybdenum in the diet was obtained from the results of Experiment IV. This tends to disprove Dick's (52) theory that sulfate prohibits the transport of molybdenum and copper across membranes. Similarly, the theory of Halverson (78) that copper absorption is reduced by the formation of insoluble copper sulfide also appears to be in error.

Recent evidence by Mills et al. (124), Scaife (139), and Mason (113), indicating that copper-containing enzymes are inactivated by molybdate and sulfide ions, appears much more tenable. A theory based on the assumption of such blocking of copper enzymes by these ions appears to explain the observed facts of molybdenum toxicity.

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# APPENDIX

## TABLE 1

### Consumption of Sodium Molybdate* (Experiment I)

| Animals by treatment | Theoretical quantity to be fed in grams | Actual quantity fed in grams | Per cent error |
|---|---|---|---|
| Control | | | |
| 282 | 0 | 0 | 0 |
| 290 | 0 | 0 | 0 |
| 293 | 0 | 0 | 0 |
| Molybdenum 5 p.p.m. | | | |
| 285 | 50.7 | 52.3 | 3.1 |
| 288 | 45.9 | 46.6 | 1.4 |
| 292 | 57.4 | 57.0 | 0.6 |
| Molybdenum 10 p.p.m. | | | |
| 283 | 100.8 | 104.1 | 3.3 |
| 287 | 128.9 | 129.9 | 0.8 |
| 291 | 116.6 | 115.7 | 0.8 |
| Molybdenum 20 p.p.m. | | | |
| 281 | 273.9 | 272.8 | 0.4 |
| 289 | 234.3 | 235.0 | 0.3 |
| 295 | 213.2 | 219.9 | 3.1 |
| Molybdenum 50 p.p.m. | | | |
| 284 | 559.3 | 551.5 | 1.4 |
| 286 | 551.3 | 555.6 | 0.8 |
| 294 | 500.2 | 497.8 | 0.5 |

*Total for 300 days

## TABLE 2

### 17% Cottonseed Ration (Experiments I, II, III, and IV)

| | |
|---|---|
| 800 lb. corn meal | 20 lb. dicalcium phosphate |
| 500 lb. crimped oats | 20 lb. iodized salt |
| 200 lb. molasses | 1 lb. dry vitamin A (5000 units/g) |
| 400 lb. cottonseed meal | 1 gm. cobalt carbonate |
| 200 lb. wheat bran | 40 gm. irradiated yeast (142F) |

TABLE 3

Body Weight in Pounds
(Experiment I)

| Week | Animal number 281 | 282 | 283 | 284 | 285 | 286 | 287 | 288 | 289 | 290 | 291 | 292 | 293 | 294 | 295 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| 1 | 1083 | 871 | 850 | 862 | 948 | 835 | 960 | 913 | 979 | 934 | 928 | 915 | 853 | 985 | 1025 |
| 2 | 1128 | 900 | 852 | 873 | 957 | 825 | 953 | 924 | 934 | 914 | 928 | 940 | 828 | 957 | 1019 |
| 3 | 1087 | 862 | 800 | 880 | 959 | 818 | 919 | 918 | 983 | 930 | 932 | 955 | 792 | 928 | 999 |
| 4 | 1078 | 878 | 819 | 881 | 969 | 825 | 926 | 935 | 990 | 926 | 925 | 954 | 773 | 939 | 988 |
| 5 | 1095 | 877 | 825 | 900 | 965 | 824 | 929 | 930 | 995 | 924 | 920 | 964 | 779 | 898 | 973 |
| 6 | 1073 | 835 | 832 | 895 | 968 | 832 | 935 | 930 | 1000 | 919 | 919 | 952 | 792 | 927 | 977 |
| 7 | 1078 | 855 | 842 | 896 | 977 | 840 | 940 | 914 | 1005 | 924 | 893 | 960 | 791 | 908 | 949 |
| 8 | 1080 | 883 | 858 | 930 | 958 | 843 | 952 | 922 | 982 | 932 | 940 | 976 | 759 | 928 | 977 |
| 9 | 1122 | 870 | 853 | 909 | 960 | 829 | 967 | 889 | 974 | 939 | 960 | 948 | 744 | 930 | 980 |
| 10 | 1140 | 890 | 864 | 904 | 966 | 845 | 966 | 861 | 1008 | 958 | 930 | 973 | 788 | 911 | 960 |
| 11 | 1140 | 872 | 875 | 895 | 930 | 862 | 962 | 894 | 1029 | 945 | 916 | 970 | 789 | 923 | 1000 |
| 12 | 1132 | 883 | 869 | 872 | 984 | 863 | 984 | 878 | 1024 | 959 | 930 | 1002 | 789 | 937 | 982 |
| 13 | 1142 | 914 | 900 | 911 | 996 | 863 | 938 | 934 | 1034 | 961 | 949 | 965 | 780 | 925 | 996 |
| 14 | 1127 | 919 | 919 | 942 | 984 | 893 | 1006 | 916 | 1043 | 971 | 951 | 981 | 775 | 945 | 986 |
| 15 | 1138 | 915 | 910 | 915 | 992 | 890 | 983 | 904 | 1055 | 971 | 959 | 964 | 800 | 952 | 1000 |

TABLE 3 (Cont.)

Body Weight in Pounds
(Experiment I)

| | Animal number | | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Week | 281 | 282 | 283 | 284 | 285 | 286 | 287 | 288 | 289 | 290 | 291 | 292 | 293 | 294 | 295 |
| 16 | 1171 | 919 | 919 | 926 | 1002 | 887 | 992 | 910 | 1049 | 973 | 961 | 985 | 773 | 949 | 986 |
| 17 | 1187 | 930 | 926 | 910 | 988 | 892 | 1011 | 917 | 1056 | 981 | 976 | 996 | 763 | 963 | 995 |
| 18 | 1150 | 938 | 923 | 944 | 983 | 896 | 1005 | 910 | 1049 | 1000 | 974 | 968 | 785 | 960 | 1005 |
| 19 | 1156 | 930 | 942 | 919 | 977 | 891 | 1037 | 923 | 1056 | 990 | 960 | 982 | 780 | 951 | 1032 |
| 20 | 1181 | 918 | 926 | 933 | 975 | 901 | 1007 | 925 | 1054 | 983 | 941 | 932 | 784 | 950 | 972 |
| 21 | 1176 | 951 | 943 | 934 | 974 | 893 | 1030 | 918 | 1045 | 990 | 962 | 950 | 792 | 965 | 1008 |
| 22 | 1169 | 920 | 952 | 934 | 991 | 895 | 1025 | 930 | 1064 | 972 | 959 | 977 | 793 | 971 | 1029 |
| 23 | 1187 | 924 | 945 | 934 | 1006 | 912 | 1039 | 916 | 1056 | 968 | 977 | 982 | 771 | 932 | 1023 |
| 24 | 1163 | 933 | 950 | 957 | 1004 | 907 | 1039 | 925 | 1057 | 990 | 996 | 970 | 770 | 947 | 1019 |
| 25 | 1153 | 943 | 938 | 942 | 990 | 905 | 1034 | 923 | 1074 | 991 | 1000 | 977 | 790 | 931 | 1045 |
| 26 | 1156 | 926 | 942 | 946 | 1010 | 921 | 1059 | 940 | 1074 | 1005 | 1012 | 971 | 780 | 961 | 1039 |
| 27 | 1165 | 951 | 961 | 962 | 1011 | 917 | 1047 | 946 | 1075 | 980 | 987 | 992 | 792 | 955 | 1033 |
| 28 | 1174 | 952 | 955 | 955 | 1030 | 913 | 982 | 947 | 1057 | 992 | 983 | 1004 | 802 | 937 | 1052 |
| 29 | 1196 | 941 | 975 | 957 | 995 | 891 | 1027 | 929 | 1080 | 1014 | 1009 | 1000 | 794 | 975 | 1065 |
| 30 | 1174 | 920 | 960 | 967 | 1006 | 911 | 1094 | 962 | 1084 | 1013 | 1012 | 1000 | 802 | 972 | 1065 |

TABLE 3 (Cont.)

Body Weight in Pounds
(Experiment I)

| | Animal number | | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Week | 281 | 282 | 283 | 284 | 285 | 286 | 287 | 288 | 289 | 290 | 291 | 292 | 293 | 294 | 295 |
| 31 | 1166 | 951 | 976 | 972 | 1038 | 919 | 1092 | 944 | 1095 | 1007 | 1044 | 1006 | 805 | 971 | 1061 |
| 32 | 1152 | 955 | 981 | 963 | 1054 | 908 | 1087 | 964 | 1130 | 1000 | 1031 | 995 | 803 | 952 | 1040 |
| 33 | 1172 | 973 | 919 | 975 | 1084 | 911 | 1104 | 963 | 1107 | 1006 | 1048 | 1009 | 807 | 981 | 1060 |
| 34 | 1188 | 996 | 930 | 977 | 1071 | 902 | 1092 | 970 | 1140 | 1008 | 1034 | 1007 | 832 | 955 | 1055 |
| 35 | 1185 | 985 | 925 | 1003 | 1084 | 911 | 1097 | 982 | 1106 | 1021 | 1045 | 983 | 824 | 971 | 1053 |
| 36 | 1207 | 983 | 914 | 994 | 1086 | 913 | 1105 | 968 | 1141 | 1008 | 1044 | 991 | 835 | 980 | 1072 |
| 37 | 1225 | 980 | 927 | 994 | 1080 | 918 | 1113 | 965 | 1117 | 1014 | 1062 | 995 | 854 | 922 | 1085 |
| 38 | 1211 | 976 | 935 | 1000 | 1085 | 909 | 1096 | 992 | 1168 | 1023 | 1061 | 985 | 835 | 1000 | 1093 |
| 39 | 1016 | 998 | 953 | 1013 | 1109 | 918 | 1139 | 1018 | 1148 | 1004 | 1090 | 982 | 861 | 994 | 1090 |
| 40 | 1206 | 993 | 967 | 1016 | 1115 | 912 | 1137 | 999 | 1141 | 1023 | 1086 | 1015 | 865 | 1009 | 1112 |
| 41 | 1222 | 979 | 1000 | 1029 | 1126 | 930 | 1162 | 1009 | 1189 | 1023 | 1075 | 1009 | 891 | 1039 | 1120 |
| 42 | 1235 | 1013 | 993 | 1033 | 1133 | 929 | 1155 | 1000 | 1168 | 1060 | 1126 | 1032 | 875 | 1032 | 1143 |
| 43 | 1200 | 1028 | 1011 | 1038 | 1124 | 935 | 1129 | 1015 | 1198 | 1053 | 1126 | | 880 | 1067 | 1146 |
| 44 | 1229 | 1020 | 1013 | | | 940 | 1164 | 1042 | 1204 | 1080 | 1160 | | 888 | 1056 | |
| 45 | | | | | | 955 | 1147 | | | | | | 867 | | |

TABLE 4

Average Body Weights in Pounds
(Experiment I)

| Animal Number | Days | | | | |
|---|---|---|---|---|---|
| | 0 | 75 | 150 | 225 | 300 |
| 281 | 1103 | 1134 | 1171 | 1164 | 1219 |
| 282 | 885 | 877 | 929 | 950 | 1020 |
| 283 | 834 | 864 | 940 | 959 | 1006 |
| 284 | 875 | 903 | 934 | 967 | 1033 |
| 285 | 954 | 952 | 982 | 1033 | 1128 |
| 286 | 831 | 845 | 896 | 913 | 935 |
| 287 | 958 | 965 | 1021 | 1094 | 1149 |
| 288 | 918 | 881 | 924 | 957 | 1019 |
| 289 | 965 | 1004 | 1054 | 1111 | 1190 |
| 290 | 926 | 947 | 982 | 1004 | 1045 |
| 291 | 929 | 935 | 954 | 1042 | 1109 |
| 292 | 937 | 959 | 955 | 1000 | 1019 |
| 293 | 845 | 774 | 785 | 805 | 881 |
| 294 | 957 | 924 | 962 | 968 | 1052 |
| 295 | 1014 | 980 | 1003 | 1054 | 1136 |

TABLE 5

Total Hay and Grain Consumption in Pounds
(Experiment I)

| Animals by treatment | Days 75 | 150 | 225 | 300 |
|---|---|---|---|---|
| **Control** | | | | |
| 282 Hay | 1342.6 | 1617.0 | 1605.7 | 1549.2 |
| Grain | 941.8 | 821.7 | 771.1 | 655.3 |
| 290 Hay | 1462.0 | 1723.9 | 1783.6 | 1797.6 |
| Grain | 757.7 | 781.0 | 623.1 | 667.7 |
| 293 Hay | 1206.3 | 1250.1 | 1254.5 | 1240.2 |
| Grain | 924.0 | 823.4 | 672.1 | 677.2 |
| **Molybdenum 5 p.p.m.** | | | | |
| 285 Hay | 1634.2 | 1695.2 | 1722.9 | 1786.1 |
| Grain | 582.6 | 554.2 | 422.1 | 461.3 |
| 288 Hay | 1510.4 | 1602.8 | 1599.9 | 1608.9 |
| Grain | 548.7 | 420.3 | 342.5 | 386.3 |
| 292 Hay | 1496.0 | 1705.8 | 1737.0 | 1787.6 |
| Grain | 863.9 | 878.2 | 848.8 | 704.9 |
| **Molybdenum 10 p.p.m.** | | | | |
| 283 Hay | 1423.2 | 1750.7 | 1722.3 | 1692.7 |
| Grain | 582.9 | 522.5 | 547.2 | 534.0 |
| 287 Hay | 1680.8 | 1975.2 | 1977.1 | 1933.9 |
| Grain | 849.3 | 903.8 | 998.1 | 905.3 |
| 291 Hay | 1661.6 | 1905.7 | 1967.9 | 1953.7 |
| Grain | 775.5 | 789.6 | 684.1 | 413.3 |
| **Molybdenum 20 p.p.m.** | | | | |
| 281 Hay | 1849.5 | 1980.2 | 2151.2 | 2208.9 |
| Grain | 1057.7 | 1044.0 | 886.7 | 767.3 |
| 289 Hay | 1651.4 | 1920.9 | 1936.3 | 1890.0 |
| Grain | 733.4 | 755.1 | 692.8 | 645.5 |
| 295 Hay | 1386.6 | 1623.0 | 1693.0 | 1765.0 |
| Grain | 902.5 | 752.5 | 667.7 | 509.4 |
| **Molybdenum 50 p.p.m.** | | | | |
| 284 Hay | 1509.9 | 1877.9 | 1999.7 | 2089.6 |
| Grain | 610.5 | 602.0 | 592.1 | 479.5 |
| 286 Hay | 1456.8 | 1715.7 | 1750.4 | 1747.4 |
| Grain | 710.8 | 693.9 | 734.0 | 612.0 |
| 294 Hay | 1423.4 | 1390.8 | 1376.2 | 1435.4 |
| Grain | 1022.3 | 902.4 | 676.2 | 502.7 |

TABLE 6

Hemoglobins
(Experiment I)

| Biweekly values | Animal number 281 | 282 | 283 | 284 | 285 | 286 | 287 | 288 | 289 | 290 | 291 | 292 | 293 | 294 | 295 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | mg/100 ml blood | | | | | | | | | | | | | | |
| 1 | 11.33 | 11.09 | 10.47 | 10.67 | 10.72 | 9.82 | 11.09 | 10.67 | 10.54 | 11.21 | 10.00 | 9.64 | 9.55 | 10.54 | 9.33 |
| 2 | 10.24 | 9.82 | 9.82 | 9.76 | 9.39 | 9.39 | 10.61 | 10.12 | 10.24 | 10.24 | 9.94 | 10.67 | 9.03 | 9.45 | 9.39 |
| 3 | 9.76 | 9.64 | 9.76 | 9.76 | 10.12 | 9.76 | 10.24 | 9.09 | 9.70 | 11.24 | 9.58 | 9.76 | 8.67 | 9.15 | 8.48 |
| 4 | 10.00 | 9.82 | 9.39 | 9.27 | 9.33 | 8.85 | 10.18 | 10.12 | 9.33 | 9.94 | 8.67 | 9.64 | 8.18 | 9.27 | 9.45 |
| 5 | 10.18 | 9.58 | 8.79 | 9.39 | 9.21 | 8.54 | 9.70 | 9.21 | 9.53 | 9.33 | 8.52 | 10.00 | 8.48 | 9.15 | 9.64 |
| 6 | 9.58 | 8.73 | 8.79 | 9.27 | 9.27 | 8.71 | 9.88 | 9.27 | 9.58 | 9.58 | 9.30 | 9.94 | 8.48 | 8.85 | 10.00 |
| 7 | 9.64 | 8.79 | 8.91 | 9.27 | 9.94 | 8.97 | 9.88 | 8.12 | 9.82 | 9.58 | 9.45 | 9.94 | 8.85 | 8.85 | 9.52 |
| 8 | 10.42 | 9.39 | 8.79 | 9.52 | 9.76 | 8.79 | 10.00 | 9.64 | 10.18 | 9.58 | 9.58 | 10.61 | 8.54 | 8.61 | 9.21 |
| 9 | 10.10 | 9.50 | 8.87 | 10.42 | 9.52 | 8.91 | 10.48 | 10.12 | 10.61 | 9.33 | 9.21 | 9.94 | 8.54 | 9.21 | 9.21 |
| 10 | 9.88 | 9.88 | 8.97 | 10.85 | 10.30 | 8.97 | 10.24 | 10.49 | 10.00 | 9.76 | 9.97 | 10.18 | 8.91 | 9.33 | 9.15 |
| 11 | 9.88 | 9.88 | 9.70 | 10.00 | 9.82 | 8.97 | 9.94 | 10.30 | 10.06 | 10.16 | 10.30 | 10.48 | 8.91 | 9.03 | 9.21 |
| 12 | 10.00 | 9.58 | 9.58 | 10.00 | 9.52 | 9.15 | 10.24 | 10.61 | 10.48 | 10.30 | 10.30 | 10.00 | 8.97 | 9.21 | 9.58 |
| 13 | 9.88 | 9.94 | 9.88 | 10.18 | 9.52 | 9.21 | 10.24 | 10.65 | 10.48 | 10.30 | 9.21 | 10.30 | 10.48 | 9.58 | 9.50 |
| 14 | 9.94 | 9.76 | 9.58 | 9.88 | 9.21 | 9.21 | 10.06 | 10.61 | 10.42 | 10.91 | 10.30 | 11.09 | 9.09 | 9.70 | 9.33 |
| 15 | 9.94 | 9.64 | 9.21 | 10.18 | 10.36 | 9.70 | 10.73 | 10.73 | 10.00 | 10.91 | 10.61 | 10.61 | 9.52 | 9.33 | 9.17 |
| 16 | 10.30 | 9.88 | 9.88 | 10.48 | 10.12 | 9.09 | 11.03 | 10.67 | 10.18 | 10.61 | 10.48 | 10.48 | 8.79 | 9.39 | 9.21 |
| 17 | 9.39 | 9.39 | 9.39 | 9.70 | 9.52 | 9.52 | 10.00 | 10.36 | 9.88 | 10.30 | 9.94 | 10.48 | 8.54 | 9.39 | 9.27 |
| 18 | 9.64 | 8.61 | 8.85 | 9.52 | 10.06 | 9.52 | 10.18 | 10.18 | 10.00 | 10.39 | 9.88 | 10.30 | 8.61 | 9.45 | 9.64 |
| 19 | 10.12 | 9.94 | 7.21 | 10.06 | 9.33 | 9.33 | 10.24 | 10.30 | 9.58 | 10.30 | 9.52 | 10.61 | 9.09 | 9.58 | 9.09 |
| 20 | 9.82 | 9.78 | 8.54 | 9.88 | 9.21 | 8.79 | 9.52 | 10.30 | 9.15 | 9.70 | 9.70 | 10.79 | 8.42 | 9.68 | 9.09 |
| 21 | 9.82 | 9.70 | 9.03 | 9.76 | 9.39 | 9.21 | 10.48 | 9.70 | 9.70 | 10.00 | 9.70 | 10.30 | 8.36 | 9.21 | 9.39 |

## TABLE 7

### Hematocrits
### (Experiment I)

| Biweekly values Period IV | Animal number 281 | 282 | 283 | 284 | 285 | 286 | 287 | 288 | 289 | 290 | 291 | 292 | 293 | 294 | 295 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | % | | | | | | | | | | | | | | |
| 1 | 31.5 | 32.9 | 30.6 | 34.2 | 29.3 | 31.0 | 33.0 | 34.0 | 32.0 | 34.0 | 34.0 | 32.9 | 31.5 | 30.6 | 29.3 |
| 2 | 34.0 | 29.8 | 29.0 | 32.3 | 34.0 | 30.0 | 36.5 | 37.4 | 29.8 | 32.0 | 33.0 | 33.0 | 30.5 | 30.0 | 29.0 |
| 3 | 31.5 | 30.0 | 29.0 | 30.5 | 31.5 | 29.5 | 37.0 | 34.0 | 32.0 | 36.5 | 34.5 | 37.5 | 33.0 | 30.0 | 29.5 |
| 4 | 31.5 | 31.0 | 30.0 | 31.0 | 31.5 | 29.0 | 34.0 | 33.0 | 32.0 | 33.0 | 33.0 | 35.0 | 31.0 | 30.0 | 29.0 |
| 5 | 32.0 | 31.5 | 33.0 | 32.5 | 31.5 | 30.0 | 33.5 | 32.0 | 30.5 | 33.5 | 35.0 | 34.5 | 32.0 | 30.0 | 29.0 |
| 6 | 32.5 | 30.0 | 29.0 | 31.0 | 30.5 | 30.0 | 31.0 | 32.0 | 29.5 | 33.5 | 33.0 | 34.0 | 30.0 | 31.5 | 29.5 |
| 7 | 33.5 | 30.0 | 28.5 | 31.5 | 30.0 | 29.0 | 31.5 | 33.0 | 28.5 | 33.5 | 32.5 | 34.0 | 27.0 | 31.5 | 30.0 |

## TABLE 8

### Serum Protein
### (Experiment I)

| Monthly values period IV | Animal number 281 | 282 | 283 | 284 | 285 | 286 | 287 | 288 | 289 | 290 | 291 | 292 | 293 | 294 | 295 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | % | | | | | | | | | | | | | | |
| 1 | 7.8 | 7.3 | 7.0 | 7.8 | 7.4 | 8.0 | 8.0 | 6.4 | 7.2 | 7.2 | 7.2 | 7.9 | 7.0 | 7.9 | 7.8 |
| 2 | 7.1 | 7.4 | 9.2 | 7.4 | 7.5 | 7.4 | 8.0 | 6.6 | 7.2 | 7.0 | 7.4 | 7.9 | 7.0 | 8.0 | 7.8 |
| 3 | 7.1 | 7.8 | 8.2 | 7.3 | 7.5 | 7.7 | 8.0 | 6.7 | 7.0 | 7.6 | 7.5 | 8.2 | 7.4 | 7.6 | 7.6 |

TABLE 9

Inorganic Phosphorous

(Experiment I)

| Biweekly values Periods III & IV | Animal number 281 | 282 | 283 | 284 | 285 | 286 | 287 | 288 | 289 | 290 | 291 | 292 | 293 | 294 | 295 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | mg/100 ml blood | | | | | | | | | | | | | | |
| 1 | 5.2 | 6.3 | 6.8 | 5.4 | 5.8 | 7.8 | 6.2 | 6.7 | 3.2 | 5.3 | 4.5 | 4.5 | 5.4 | 7.0 | 6.3 |
| 2 | 6.0 | 6.8 | 6.8 | 6.0 | 7.2 | 8.8 | 6.4 | 7.1 | 5.0 | 5.5 | 6.1 | 7.5 | 6.0 | 6.3 | 5.8 |
| 3 | 6.4 | 6.3 | 6.5 | 6.0 | 7.3 | 7.8 | 7.0 | 7.1 | 5.0 | 5.7 | 6.2 | 7.0 | 6.5 | 6.2 | 6.0 |
| 4 | 5.8 | 6.3 | 6.7 | 6.0 | 7.0 | 7.3 | 6.9 | 6.1 | 4.5 | 5.8 | 6.3 | 6.1 | 6.8 | 5.8 | 6.0 |
| 5 | 6.2 | 6.4 | 6.3 | 6.7 | 6.8 | 7.4 | 7.3 | 7.5 | 5.2 | 5.9 | 6.8 | 6.4 | 6.0 | 5.7 | 6.5 |
| 6 | 6.8 | 7.2 | 6.3 | 8.0 | 7.4 | 8.0 | 7.2 | 8.0 | 5.6 | 7.8 | 7.0 | 5.8 | 5.8 | 7.0 | 6.8 |
| 7 | 6.2 | 7.4 | 8.0 | 6.4 | 6.2 | 8.2 | 7.4 | 7.6 | 5.8 | 6.2 | 6.8 | 6.6 | 6.4 | 8.4 | 7.4 |
| 8 | 6.8 | 8.2 | 8.2 | 6.4 | 7.0 | 8.8 | 7.2 | 8.2 | 4.0 | 6.4 | 7.6 | 6.0 | 5.4 | 8.0 | 7.6 |

TABLE 10

Composition of Milk
(Experiment I)

| | Milk copper | | | | Milk molybdenum | | | |
|---|---|---|---|---|---|---|---|---|
| Control | 75 | 150 | 225 | 300 | 75 | 150 | 225 | 300 |
| 282 | --- | 0.11 | 0.33 | 0.07 | --- | 0.00 | 0.04 | 0.06 |
| 290 | 0.38 | 0.18 | 0.08 | 0.04 | 0.05 | 0.02 | 0.04 | 0.04 |
| 293 | 0.18 | 0.52 | 0.07 | 0.06 | 0.01 | 0.05 | 0.04 | 0.04 |
| Molybdenum 5 p.p.m. | | | | | | | | |
| 285 | --- | 0.16 | 0.38 | 0.10 | --- | 0.07 | 0.07 | 0.04 |
| 288 | 0.17 | 0.20 | 0.80 | 0.05 | 0.02 | 0.17 | 0.10 | 0.17 |
| 292 | 0.16 | 0.18 | 0.06 | 0.04 | 0.01 | 0.05 | 0.07 | 0.11 |
| Molybdenum 10 p.p.m. | | | | | | | | |
| 283 | --- | 0.13 | 0.32 | 0.05 | --- | 0.02 | 0.11 | 0.21 |
| 287 | 0.16 | 0.12 | 0.80 | 0.04 | 0.02 | 0.13 | 0.06 | 0.10 |
| 291 | 0.14 | 0.23 | 0.10 | 0.07 | 0.06 | 0.11 | 0.11 | 0.27 |
| Molybdenum 20 p.p.m. | | | | | | | | |
| 281 | --- | 0.11 | 0.18 | 0.06 | --- | 0.20 | 0.36 | 0.48 |
| 289 | 0.17 | 0.14 | 0.07 | 0.04 | 0.05 | 0.21 | 0.20 | 0.18 |
| 295 | 0.25 | 0.21 | 0.04 | 0.06 | 0.15 | 0.56 | 0.36 | 0.84 |
| Molybdenum 50 p.p.m. | | | | | | | | |
| 284 | --- | 0.12 | 0.44 | 0.04 | --- | 0.51 | 1.41 | 1.05 |
| 286 | 0.12 | 0.16 | 0.79 | 0.02 | 0.18 | 0.72 | 0.50 | 0.66 |
| 294 | 0.17 | 0.21 | 0.07 | 0.06 | 0.19 | 0.80 | 0.94 | 2.91 |

**TABLE 11**

**Composition of Serum**
**(Experiment I)**

| | Serum copper (p.p.m.) | | | | Serum molybdenum (p.p.m.) | | | |
|---|---|---|---|---|---|---|---|---|
| Control | 75 | 150 | 225 | 300 | 75 | 150 | 225 | 300 |
| 282 | 1.11 | 0.95 | 0.88 | 0.69 | 0.01 | 0.03 | 0.01 | 0.03 |
| 290 | 1.35 | 0.89 | 0.90 | 1.00 | 0.08 | 0.01 | 0.04 | 0.05 |
| 293 | 0.90 | 0.83 | 0.84 | 1.53 | 0.11 | 0.06 | 0.02 | 0.02 |
| Molybdenum 5 p.p.m. | | | | | | | | |
| 285 | 1.24 | 1.10 | 0.74 | 0.76 | 0.24 | 0.12 | 0.12 | 0.21 |
| 288 | 0.63 | 0.85 | 0.78 | 0.66 | 0.16 | 0.45 | 0.16 | 0.65 |
| 292 | 0.93 | 0.89 | 0.73 | 0.82 | 0.12 | 0.13 | 0.11 | 0.29 |
| Molybdenum 10 p.p.m. | | | | | | | | |
| 283 | 0.93 | 0.40 | 0.54 | 0.44 | 0.42 | 0.41 | 0.31 | 0.86 |
| 287 | 0.43 | 0.60 | 0.61 | 0.58 | 0.16 | 0.43 | 0.16 | 0.30 |
| 291 | 1.05 | 0.98 | 0.85 | 0.83 | 0.51 | 0.34 | 0.34 | 0.75 |
| Molybdenum 20 p.p.m. | | | | | | | | |
| 281 | 1.11 | 0.73 | 0.68 | 0.49 | 0.69 | 0.50 | 1.25 | 1.96 |
| 289 | 0.83 | 0.70 | 0.69 | 0.69 | 0.42 | 0.72 | 0.91 | 1.00 |
| 295 | 0.95 | 0.78 | 0.73 | 0.78 | 1.23 | 1.98 | 1.34 | 1.96 |
| Molybdenum 50 p.p.m. | | | | | | | | |
| 284 | 1.70 | 1.08 | 0.85 | 0.64 | 3.90 | 1.50 | 4.08 | 3.95 |
| 286 | 0.60 | 0.65 | 0.52 | 0.50 | 1.50 | 2.40 | 1.76 | 2.24 |
| 294 | 1.00 | 0.85 | 0.67 | 0.66 | 1.60 | 2.30 | 2.41 | 5.65 |

TABLE 12

Composition of Liver
(Experiment I)

| | Liver copper (p.p.m.) | | | | Liver molybdenum (p.p.m.) | | | |
|---|---|---|---|---|---|---|---|---|
| Control | 75 | 150 | 225 | 300 | 75 | 150 | 225 | 300 |
| 282 | 24 | 28 | 44 | 51 | 35 | 4 | 6 | 6 |
| 290 | 115 | 129 | 175 | 162 | 6 | 7 | 8 | 7 |
| 293 | 119 | 123 | 148 | 222 | 12 | 3 | 5 | 5 |
| Molybdenum 5 p.p.m. | | | | | | | | |
| 285 | 206 | 197 | 165 | 158 | 15 | 9 | 8 | 5 |
| 288 | 102 | 79 | 55 | 29 | 5 | 13 | 10 | 9 |
| 292 | 17 | 21 | 12 | 21 | 10 | 7 | 10 | 29 |
| Molybdenum 10 p.p.m. | | | | | | | | |
| 283 | 7 | 10 | 12 | 189 | 8 | 16 | 14 | 23 |
| 287 | 27 | 12 | 21 | 14 | 7 | 10 | 9 | 8 |
| 291 | 87 | 67 | 56 | 34 | 17 | 12 | 10 | 12 |
| Molybdenum 20 p.p.m. | | | | | | | | |
| 281 | 25 | 17 | 20 | 22 | 26 | 18 | 21 | 23 |
| 289 | 26 | 12 | -- | 9 | 20 | 19 | -- | 13 |
| 295 | 106 | 81 | 33 | 157 | 19 | 25 | 19 | 24 |
| Molybdenum 50 p.p.m. | | | | | | | | |
| 284 | 34 | 20 | 15 | 16 | 23 | 27 | 31 | 21 |
| 286 | 20 | 54 | 24 | 12 | 16 | 27 | 22 | 23 |
| 294 | 42 | 27 | 14 | 72 | 28 | 35 | 33 | 44 |

TABLE 13

Administration of Sodium Molybdate
(Experiment II)

| Animals by treatment | Theoretical quantity to be fed in grams | Actual quantity fed in grams | Per cent error |
|---|---|---|---|
| **Control** | | | |
| 386 | 0 | 0 | 0 |
| 395 | 0 | 0 | 0 |
| 398 | 0 | 0 | 0 |
| **Molybdenum 5 p.p.m.** | | | |
| 390 | 29.2 ** | 27.7 | 5.0 |
| 392 | 48.7 *** | 46.1 | 5.4 |
| 399 | 27.7 ** | 26.3 | 4.9 |
| **Molybdenum 10 p.p.m.** | | | |
| 388 | 80.3 | 78.5 | 2.2 |
| 391 | 80.0 | 77.2 | 3.4 |
| 396 | 86.1 | 88.5 | 2.8 |
| **Molybdenum 20 p.p.m.** | | | |
| 389 | 238.2 | 232.1 | 2.6 |
| 393 | 216.7 | 206.9 | 4.5 |
| 397 | 175.5 | 177.0 | 0.9 |
| **Molybdenum 50 p.p.m.** | | | |
| 387 | 486.6 | 490.9 | 0.9 |
| 394 | 631.5 | 614.7 | 2.7 |
| 400 | 611.5 | 625.6 | 2.3 |
| **Molybdenum 100 p.p.m.** | | | |
| 388 | 176.4* | 162.6 | 7.8 |
| 392 | 210.7* | 193.0 | 8.4 |
| **Molybdenum 200 p.p.m.** | | | |
| 391 | 278.8* | 280.3 | 0.5 |
| 396 | 343.0* | 410.3 | 19.6 |

* one period
** two periods
*** three periods

TABLE 14

Body Weight in Pounds
(Experiment II)

| | Animal number | | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Week | 386 | 387 | 388 | 389 | 390 | 391 | 392 | 393 | 394 | 395 | 396 | 397 | 398 | 399 | 400 |
| 1 | 1006 | 967 | 886 | 958 | 880 | 919 | 1025 | 900 | 1180 | 1060 | 1010 | 872 | 875 | 959 | 969 |
| 2 | 1014 | 955 | 883 | 1029 | 851 | 894 | 1011 | 891 | 1144 | 1055 | 1011 | 887 | 877 | 924 | 974 |
| 3 | 1048 | 979 | 900 | 1035 | 865 | 925 | 1034 | 915 | 1144 | 1028 | 981 | 874 | 865 | 891 | 946 |
| 4 | 1010 | 937 | 877 | 1010 | 851 | 915 | 1060 | 914 | 1151 | 1006 | 975 | 894 | 835 | 901 | 974 |
| 5 | 1022 | 958 | 900 | 1045 | 869 | 913 | 1042 | 920 | 1060 | 982 | 985 | 890 | 876 | 868 | 954 |
| 6 | 1030 | 954 | 910 | 1037 | 825 | 914 | 1150 | 906 | 1162 | 1000 | 996 | 888 | 866 | 883 | 979 |
| 7 | 998 | 928 | 910 | 1036 | 864 | 914 | 1018 | 881 | 1164 | 1030 | 974 | 889 | 875 | 865 | 941 |
| 8 | 1022 | 936 | 919 | 1034 | 882 | 909 | 1050 | 881 | 1155 | 1011 | 1010 | 911 | 852 | 860 | 975 |
| 9 | 926 | 943 | 908 | 918 | 857 | 904 | 1050 | 879 | 1198 | 1013 | 998 | 907 | 873 | 877 | 995 |
| 10 | 1048 | 950 | 923 | 1013 | 860 | 921 | 1053 | 885 | 1215 | 1005 | 1005 | 920 | 867 | 869 | 989 |
| 11 | 1050 | 959 | 900 | 1027 | 859 | 927 | 1053 | 898 | 1184 | 1044 | 987 | 909 | 885 | 876 | 953 |
| 12 | 979 | 936 | 921 | 1003 | 868 | 921 | 1066 | 915 | 1184 | 1007 | 988 | 906 | 856 | 882 | 1006 |
| 13 | 1052 | 994 | 932 | 1035 | 868 | 947 | 1023 | 872 | 1180 | 1001 | 994 | 927 | 875 | 868 | 950 |
| 14 | 1011 | 974 | 936 | 1010 | 870 | 916 | 1038 | 873 | 1138 | 1008 | 983 | 925 | 842 | 874 | 957 |
| 15 | 1050 | 991 | 932 | 1018 | 860 | 921 | 1046 | 896 | 1156 | 1017 | 981 | 945 | 868 | 886 | 959 |

TABLE 14 (Cont.)

Body Weight in Pounds
(Experiment II)

| | Animal number | | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Week | 386 | 387 | 388 | 389 | 390 | 391 | 392 | 393 | 394 | 395 | 396 | 397 | 398 | 399 | 400 |
| 16 | 1027 | 962 | 877 | 982 | 889 | 929 | 1030 | 886 | 1171 | 996 | 1003 | 955 | 855 | 877 | 977 |
| 17 | 1006 | 970 | 915 | 996 | 873 | 900 | 1035 | 890 | 1175 | 1012 | 962 | 921 | 847 | 867 | 947 |
| 18 | 1021 | 991 | 924 | 1004 | 848 | 918 | 1040 | 898 | 1183 | 991 | 993 | 950 | 835 | 888 | 985 |
| 19 | 1036 | 967 | 918 | 998 | 883 | 920 | 1044 | 900 | 1175 | 1022 | 991 | 931 | 858 | 903 | 955 |
| 20 | 1031 | 982 | 923 | 980 | 882 | 912 | 1047 | 902 | 1188 | 1007 | 950 | 940 | 862 | | 970 |
| 21 | 1050 | 991 | 925 | 973 | 880 | 936 | 1050 | 911 | 1160 | 1020 | 952 | 920 | 873 | | 977 |
| 22 | 991 | 982 | 915 | 947 | 881 | 916 | 1048 | 903 | 1206 | 1014 | 981 | 923 | 873 | | 956 |
| 23 | 1033 | 969 | 918 | 975 | | 927 | 1041 | 817 | 1189 | 1017 | 963 | 917 | 838 | | 971 |
| 24 | 1011 | 986 | 910 | 953 | | 920 | 1038 | 920 | 1198 | 1025 | 1008 | 919 | 867 | | 939 |
| 25 | 1025 | 979 | 934 | 975 | | 938 | 1058 | 937 | 1110 | 1051 | 971 | 927 | 833 | | 916 |
| 26 | 988 | 968 | 926 | 963 | | 953 | 1065 | 947 | 1227 | 1003 | 969 | 909 | 851 | | 924 |
| 27 | 1002 | 996 | 954 | 985 | | 943 | 1040 | 956 | 1224 | 1021 | 971 | 934 | 849 | | 951 |
| 28 | 1046 | 1007 | 945 | 995 | | 935 | 952 | 964 | 1198 | 1024 | 967 | 898 | 852 | | 955 |
| 29 | 1035 | 1016 | 954 | 1010 | | 940 | 1050 | 955 | 1223 | 998 | 930 | 892 | 857 | | 940 |
| 30 | 983 | 972 | 931 | 984 | | 930 | 1058 | 978 | 1161 | 982 | 959 | 882 | 870 | | 950 |

TABLE 14 (Cont.)

Body Weight in Pounds
(Experiment II)

| | Animal number | | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Week | 386 | 387 | 388 | 389 | 390 | 391 | 392 | 393 | 394 | 395 | 396 | 397 | 398 | 399 | 400 |
| 31 | 1026 | 998 | 943 | 997 | | 917 | 1044 | 950 | 1198 | 1033 | 965 | 900 | 845 | | 939 |
| 32 | 991 | 995 | 945 | 980 | | 939 | 1042 | 1008 | 1213 | 1041 | 965 | 890 | 833 | | 938 |
| 33 | 1001 | 950 | 918 | 952 | | 955 | 1047 | 1013 | 1217 | 1011 | 945 | 915 | 864 | | 933 |
| 34 | 1005 | 900 | 932 | 937 | | 915 | 1060 | 966 | 1212 | 1040 | 861 | 883 | 858 | | 935 |
| 35 | 1029 | 934 | 932 | 957 | | 904 | 1051 | 1020 | 1230 | 1038 | 926 | 884 | 842 | | 945 |
| 36 | 1020 | 932 | 914 | 940 | | 769 | 983 | 1016 | 1210 | 1044 | 900 | 877 | 855 | | 933 |
| 37 | 1022 | 924 | 909 | 967 | | 815 | 845 | 1008 | 1185 | 1065 | 884 | 876 | 832 | | 930 |
| 38 | 1035 | 949 | 845 | 957 | | 839 | 998 | 1014 | 1192 | 1045 | 846 | 885 | 834 | | 950 |
| 39 | 1048 | 955 | 832 | 963 | | 785 | 1001 | 1029 | 1228 | 1058 | 848 | 866 | 835 | | 922 |
| 40 | 1053 | 970 | 848 | 977 | | 768 | 977 | 1038 | 1225 | 1095 | 842 | 864 | 833 | | 991 |
| 41 | 1073 | 959 | 860 | 961 | | 723 | 971 | 1063 | 1233 | 1072 | 818 | 904 | 862 | | 970 |
| 42 | 1077 | 1007 | 840 | 965 | | 697 | 1000 | 1036 | 1210 | 1075 | 826 | 891 | 845 | | 990 |
| 43 | 1057 | 988 | 804 | 922 | | 735 | 1024 | 1066 | 1197 | 1107 | 847 | 895 | | | 1000 |

TABLE 14 (Cont.)

Body Weight in Pounds
Animals Continued Through a Second Lactation
(Experiment II)

| Week | Animal number 386 | 387 | 394 | 395 | Week | Animal number 386 | 387 | 394 | 395 |
|---|---|---|---|---|---|---|---|---|---|
| 1 | | 1026 | 1158 | 1109 | 23 | 1145 | 982 | 1168 | 1085 |
| 2 | 1177 | 1004 | 1190 | 1109 | 24 | 1180 | 951 | 1209 | 1054 |
| 3 | 1154 | 1012 | 1218 | 1085 | 25 | 1129 | 975 | 1195 | 1072 |
| 4 | 1113 | 1022 | 1175 | 1092 | 26 | 1127 | 970 | 1178 | 1074 |
| 5 | 1127 | 1031 | 1187 | 1085 | 27 | 1127 | 979 | 1229 | 1099 |
| 6 | 1137 | 1017 | 1174 | 1102 | 28 | 1144 | 982 | 1190 | 1083 |
| 7 | 1140 | 1008 | 1182 | 1092 | 29 | 1123 | 967 | 1244 | 1109 |
| 8 | 1130 | 1010 | 1198 | 1067 | 30 | 1133 | 965 | 1214 | 1100 |
| 9 | 1156 | 1008 | 1195 | 1053 | 31 | 1141 | 938 | 1212 | 1009 |
| 10 | 1153 | 994 | 1208 | 1055 | 32 | 1197 | 994 | 1242 | 1032 |
| 11 | 1134 | 1027 | 1175 | 1069 | 33 | 1223 | 986 | 1218 | 1049 |
| 12 | 1114 | 1006 | 1176 | 1085 | 34 | 1200 | 986 | 1243 | 1071 |
| 13 | 1123 | 1032 | 1183 | 1036 | 35 | 1210 | 998 | 1158 | 1120 |
| 14 | 1143 | 1030 | 1183 | 1073 | 36 | 1230 | 988 | 1177 | 1129 |
| 15 | 1151 | 992 | 1160 | 1083 | 37 | 1235 | 1004 | 1198 | 1122 |
| 16 | 1200 | 1000 | 1205 | 1109 | 38 | 1236 | 1000 | 1200 | 1048 |
| 17 | 1129 | 998 | 1214 | 1045 | 39 | 1264 | 1011 | 1198 | 1066 |
| 18 | 1104 | 1003 | 1231 | 1070 | 40 | 1300 | 1006 | 1171 | 1074 |
| 19 | 1110 | 1006 | 1155 | 1074 | 41 | 1289 | 993 | 1149 | 1050 |
| 20 | 1129 | 1012 | 1163 | 1070 | 42 | 1300 | 989 | 1100 | 1052 |
| 21 | 1135 | 943 | 1210 | 1098 | 43 | 1300 | 1000 | 1149 | 1052 |
| 22 | 1155 | 1073 | 1174 | 1102 | | | | | |

TABLE 15

Average Body Weights in Pounds
(Experiment II)

| Animal Number | Days 0 | 75 | 150 | 225 | 300 | |
|---|---|---|---|---|---|---|
| 386 | 1023 | 1026 | 1024 | 1000 | 1069 | |
| 387 | 967 | 948 | 985 | 988 | 985 | |
| 388 | 890 | 915 | 921 | 940 | 835 | * |
| 389 | 1007 | 1014 | 967 | 987 | 949 | |
| 390 | 865 | 862 | 881 | | | |
| 391 | 913 | 923 | 921 | 928 | 718 | ** |
| 392 | 1023 | 1059 | 1048 | 1048 | 998 | * |
| 393 | 902 | 899 | 905 | 979 | 1055 | |
| 394 | 1156 | 1194 | 1185 | 1174 | 1213 | |
| 395 | 1048 | 1019 | 1014 | 1019 | 1085 | |
| 396 | 1001 | 993 | 961 | 963 | 830 | ** |
| 397 | 881 | 912 | 928 | 891 | 897 | |
| 398 | 872 | 869 | 869 | 849 | 847 | |
| 399 | 926 | 876 | 886 | | | |
| 400 | 963 | 983 | 969 | 942 | 987 | |

* Changed to 100 p.p.m.

** Changed to 200 p.p.m.

Animals Continued Through A Second Lactation

| | | | | | |
|---|---|---|---|---|---|
| 386 | 1166 | 1134 | 1140 | 1157 | 1296 |
| 387 | 1014 | 1009 | 1009 | 966 | 994 |
| 394 | 1189 | 1186 | 1182 | 1223 | 1133 |
| 395 | 1101 | 1070 | 1090 | 1047 | 1051 |

TABLE 16

Total Hay and Grain Consumption in Pounds
(Experiment II)

| Animals by treatment | | Days 75 | 150 | 225 | 300 |
|---|---|---|---|---|---|
| Control | | | | | |
| 386 | Hay | 2004.7 | 2335.3 | 2181.6 | 2187.3 |
| | Grain | 768.7 | 742.9 | 659.6 | 520.4 |
| 395 | Hay | 1810.4 | 1928.5 | 1925.6 | 1954.1 |
| | Grain | 1011.2 | 1018.1 | 778.0 | 509.6 |
| 398 | Hay | 1469.2 | 1559.4 | 1484.4 | 1474.9 |
| | Grain | 734.5 | 410.4 | 303.5 | 178.1 |
| Molybdenum 5 p.p.m. | | | | | |
| 390 | Hay | 1769.0 | 1837.1 | | |
| | Grain | 853.3 | 643.5 | | |
| 392 | Hay | 1864.5 | 2240.0 | 2249.3 | |
| | Grain | 893.6 | 712.9 | 540.4 | |
| 399 | Hay | 1474.8 | 1462.6 | | |
| | Grain | 1083.9 | 809.4 | | |
| Molybdenum 10 p.p.m. | | | | | |
| 388 | Hay | 1706.6 | 1770.4 | 1828.7 | |
| | Grain | 694.1 | 528.8 | 483.2 | |
| 391 | Hay | 1594.1 | 1685.6 | 1595.1 | |
| | Grain | 834.6 | 702.3 | 567.1 | |
| 396 | Hay | 1815.3 | 1761.5 | 1791.1 | |
| | Grain | 1181.1 | 799.8 | 163.0 | |
| Molybdenum 20 p.p.m. | | | | | |
| 389 | Hay | 1870.0 | 2037.5 | 1918.6 | 1817.1 |
| | Grain | 904.1 | 843.1 | 619.9 | 402.1 |
| 393 | Hay | 1811.5 | 1798.9 | 1880.4 | 1829.2 |
| | Grain | 728.0 | 656.0 | 459.1 | 299.9 |
| 397 | Hay | 1349.9 | 1499.9 | 1423.3 | 1205.3 |
| | Grain | 840.5 | 634.9 | 394.0 | 317.8 |
| Molybdenum 50 p.p.m. | | | | | |
| 387 | Hay | 1701.1 | 1801.7 | 1779.6 | 1721.8 |
| | Grain | 703.2 | 437.5 | 286.0 | 70.0 |
| 394 | Hay | 1971.9 | 1929.2 | 1925.4 | 2152.0 |
| | Grain | 956.9 | 867.4 | 758.3 | 470.4 |
| 400 | Hay | 1914.7 | 1905.7 | 1596.6 | 1827.1 |
| | | 1283.0 | 1071.3 | 591.3 | 492.5 |

TABLE 16 (cont.)

Total Hay and Grain Consumption in Pounds

(Experiment II)

| Animals by treatment | Days | | | |
|---|---|---|---|---|
| | 75 | 150 | 225 | 300 |
| Molybdenum 100 p.p.m. | | | | |
| 388 Hay | | | | 1283.9 |
| Grain | | | | 257.2 |
| 392 Hay | | | | 1610.7 |
| Grain | | | | 229.8 |
| Molybdenum 200 p.p.m. | | | | |
| 391 Hay | | | | 984.8 |
| Grain | | | | 232.9 |
| 396 Hay | | | | 1298.1 |
| Grain | | | | 200.0 |

## TABLE 17

### Hemoglobins
### (Experiment II)

| Biweekly values | Animal number 386 | 387 | 388 | 389 | 390 | 391 | 392 | 393 | 394 | 395 | 396 | 397 | 398 | 399 | 400 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | mg/100 ml blood | | | | | | | | | | | | | | |
| 1 | 10.97 | 10.85 | 9.76 | 10.97 | 10.91 | 10.91 | 10.73 | 10.91 | 10.06 | 10.36 | 10.24 | 10.12 | 10.18 | 10.48 | 10.82 |
| 2 | 10.61 | 10.51 | 9.76 | 10.32 | 10.00 | 10.79 | 10.24 | 10.61 | 9.45 | 9.21 | 9.39 | 9.94 | 9.33 | 10.00 | 10.54 |
| 3 | 9.52 | 10.18 | 8.97 | 10.30 | 9.21 | 10.00 | 9.64 | 10.42 | 9.21 | 9.94 | 10.00 | 9.45 | 8.79 | 9.39 | 10.00 |
| 4 | 8.91 | 10.00 | 9.39 | 9.76 | 9.76 | 9.94 | 10.48 | 10.91 | 9.09 | 10.48 | 9.58 | 8.42 | 9.03 | 9.03 | 10.00 |
| 5 | 9.32 | 10.06 | 9.03 | 9.15 | 11.03 | 10.18 | 10.79 | 9.82 | 9.52 | 9.39 | 8.79 | 8.12 | 8.97 | 10.00 | 9.94 |
| 6 | 10.36 | 10.36 | 9.94 | 10.79 | 9.58 | 9.27 | 10.12 | 9.82 | 9.21 | 9.33 | 9.15 | 8.97 | 8.79 | 9.09 | 9.94 |
| 7 | 9.15 | 9.82 | 8.91 | 9.82 | 10.00 | 8.91 | 9.82 | 9.76 | 8.24 | 9.82 | 9.82 | 9.09 | 7.88 | 8.79 | 8.79 |
| 8 | 9.82 | 9.88 | 8.91 | 9.52 | 9.58 | 8.97 | 10.12 | 9.76 | 8.76 | 9.76 | 9.82 | 7.21 | 8.67 | 9.52 | 9.09 |
| 9 | 9.09 | 9.82 | 9.09 | 9.45 | 9.09 | 8.67 | 10.06 | 9.52 | 8.73 | 8.42 | 8.18 | 8.73 | 8.79 | 9.70 | 9.82 |
| 10 | 9.03 | 9.82 | 9.21 | 9.76 | 8.61 | 7.88 | 8.91 | 8.79 | 7.58 | 9.15 | 9.09 | 8.67 | 8.18 | 9.94 | 9.09 |
| 11 | 8.79 | 9.27 | 8.00 | 9.09 | 9.52 | 9.70 | 9.09 | 9.64 | 8.73 | 9.39 | 9.27 | 7.94 | 8.00 | | 9.09 |
| 12 | 9.27 | 9.58 | 9.15 | 9.76 | 9.88 | 9.70 | 9.39 | 10.00 | 8.18 | 8.48 | 8.91 | 8.54 | 8.48 | | 8.91 |
| 13 | 9.39 | 9.76 | 9.09 | 9.82 | 9.70 | 10.30 | 9.58 | 9.58 | 8.48 | 8.48 | 8.48 | 8.67 | 8,67 | | 9.70 |
| 14 | 9.09 | 9.70 | 9.15 | 10.00 | | 9.76 | 9.76 | 10.24 | 8.18 | 8.30 | 8.97 | 8.42 | 8.79 | | 8.97 |
| 15 | 9.39 | 9.27 | 9.39 | 10.00 | | 9.64 | 10.00 | 10.61 | 8.48 | 8.54 | 8.91 | 8.06 | 8.73 | | 8.54 |
| 16 | 9.40 | 9.52 | 9.40 | 10.00 | | 9.45 | 9.94 | 10.00 | 8.54 | 8.48 | 9.09 | 8.06 | 8.30 | | 8.48 |
| 17 | 9.64 | 9.76 | 9.52 | 10.42 | | 8.85 | 9.45 | 9.76 | 8.79 | 8.00 | 8.67 | 8.18 | 8.18 | | 9.03 |
| 18 | 9.76 | 9.82 | 9.39 | 10.52 | | 8.67 | 9.15 | 9.21 | 8.00 | 8.88 | 8.48 | 8.00 | 8.12 | | 8.91 |
| 19 | 9.52 | 9.58 | 9.39 | 10.00 | | 8.97 | 8.79 | 9.39 | 8.24 | 8.48 | 7.88 | 8.12 | 8.42 | | 8.91 |
| 20 | 9.21 | 8.73 | 8.54 | 9.64 | | 9.21 | 8.67 | 8.30 | 8.18 | 8.00 | 9.09 | 8.00 | 8.61 | | 9.09 |
| 21 | 8.30 | 8.36 | 8.18 | 9.76 | | 9.60 | 8.71 | 8.85 | 8.30 | 8.85 | 8.06 | 8.61 | 8.92 | | 9.03 |

TABLE 18

Hematocrits

(Experiment II)

| Biweekly values | Animal number | | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | 386 | 387 | 388 | 389 | 390 | 391 | 392 | 393 | 394 | 395 | 396 | 397 | 398 | 399 | 400 |
| | % | | | | | | | | | | | | | | |
| 1 | 33.5 | 34.5 | 31.5 | 32.5 | 35.0 | 38.5 | 35.0 | 37.0 | 30.0 | 34.5 | 33.5 | 30.0 | 28,5 | 34.0 | 35.5 |
| 2 | 30.5 | 34.5 | 30.0 | 31.5 | 31.5 | 33.5 | 35.0 | 33.5 | 30.0 | 31.5 | 30.0 | 29.5 | 28.5 | 34.0 | 35.5 |
| 3 | 29.5 | 32.0 | 30.0 | 31.5 | 32.0 | 31.0 | 32.5 | 31.5 | 28.0 | 29.5 | 29.1 | 29.5 | 27.2 | 33.0 | 31.5 |
| 4 | 27.5 | 30.0 | 29.5 | 31.5 | 31.0 | 30.0 | 32.0 | 32.0 | 30.5 | 32.8 | 29.5 | 31.5 | 29.5 | 30.5 | 31.0 |
| 5 | 32.0 | 34.0 | 32.0 | 33.8 | 35.5 | 32.8 | 35.5 | 32.0 | 29.2 | 30.0 | 28.0 | 27.5 | 27.0 | 31.0 | 30.5 |
| 6 | 29.0 | 31.0 | 29.0 | 32.5 | 30.5 | 31.0 | 33.8 | 32.5 | 27.5 | 31.5 | 30.0 | 28.0 | 24.5 | 29.0 | 31.0 |
| 7 | 31.0 | 33.0 | 30.0 | 31.0 | 32.5 | 30.0 | 33.5 | 31.5 | 27.0 | 30.0 | 30.0 | 25.0 | 27.5 | 32.5 | 31.5 |
| 8 | 28.0 | 32.0 | 29.0 | 31.0 | 31.5 | 28.5 | 32.0 | 29.5 | 25.0 | 29.0 | 27.5 | 27.0 | 29.0 | 32.0 | 31.0 |
| 9 | 28.0 | 32.5 | 28.5 | 31.0 | 32.5 | 28.0 | 30.0 | 32.5 | 27.5 | 29.0 | 29.0 | 27.0 | 28.5 | 31.5 | 30.5 |
| 10 | 31.5 | 34.0 | 29.5 | 33.5 | 33.5 | 30.0 | 33.5 | 33.0 | 28.0 | 30.0 | 29.5 | 25.5 | 26.5 | 32.5 | 29.5 |
| 11 | 31.0 | 32.5 | 30.5 | 34.0 | 33.0 | 30.0 | 34.5 | 33.0 | 26.5 | 29.0 | 29.5 | 26.0 | 26.5 | | 29.0 |
| 12 | 30.5 | 33.5 | 31.5 | 34.0 | 34.0 | 32.5 | 35.0 | 33.0 | 27.0 | 28.5 | 29.5 | 28.0 | 26.0 | | 29.5 |
| 13 | 29.0 | 31.5 | 31.5 | 33.0 | 32.0 | 31.0 | 34.0 | 34.5 | 27.5 | 27.0 | 27.0 | 28.0 | 26.5 | | 29.5 |
| 14 | 29.5 | 30.5 | 32.0 | 32.5 | | 33.5 | 33.0 | 34.0 | 27.5 | 26.5 | 27.5 | 27.0 | 26.5 | | 29.5 |
| 15 | 29.0 | 30.0 | 31.5 | 32.0 | | 33.0 | 33.0 | 34.5 | 27.0 | 26.0 | 28.0 | 25.0 | 26.5 | | 29.5 |
| 16 | 29.5 | 30.5 | 31.0 | 32.5 | | 32.0 | 32.0 | 34.5 | 28.5 | 25.5 | 29.0 | 27.0 | 25.0 | | 28.5 |
| 17 | 30.0 | 30.5 | 30.5 | 34.5 | | 29.5 | 33.0 | 31.5 | 27.0 | 26.0 | 25.0 | 24.5 | 24.5 | | 27.0 |
| 18 | 29.0 | 30.0 | 28.5 | 31.5 | | 28.5 | 30.0 | 31.5 | 26.0 | 26.5 | 26.5 | 23.5 | 27.0 | | 29.5 |
| 19 | 29.5 | 28.5 | 28.0 | 32.0 | | 30.5 | 30.5 | 28.5 | 26.0 | 25.5 | 24.5 | 23.0 | 28.0 | | 30.0 |
| 20 | 29.5 | 26.5 | 25.0 | 31.0 | | 30.0 | 31.5 | 30.0 | 26.0 | 28.0 | 25.0 | 23.5 | 24.5 | | 29.5 |
| 21 | 28.5 | 28.5 | 25.0 | 31.0 | | 27.0 | 29.0 | 29.5 | 26.0 | 28.0 | 26.5 | 24.5 | | | 29.5 |

TABLE 19

Animals Continued Through a Second Lactation
(Experiment II)

| Biweekly values | Animal number 386 | 387 | 394 | 395 | 386 | 387 | 394 | 395 |
|---|---|---|---|---|---|---|---|---|
| | Hemoglobins mg/100 ml blood | | | | Hematocrits % | | | |
| 1 | 9.64 | 9.82 | 9.70 | 9.21 | 30.5 | 31.5 | 31.0 | 29.0 |
| 2 | 10.18 | 10.48 | 8.79 | 9.45 | 30.0 | 32.0 | 30.0 | 30.0 |
| 3 | 10.36 | 12.18 | 8.61 | 9.52 | 28.5 | 33.0 | 25.0 | 31.0 |
| 4 | 10.06 | 11.76 | 8.61 | 9.52 | 29.0 | 33.0 | 26.0 | 30.0 |
| 5 | 9.21 | 10.18 | 8.54 | 9.33 | 29.5 | 32.0 | 25.5 | 30.0 |
| 6 | 9.27 | 10.54 | 8.48 | 8.97 | 29.0 | 31.0 | 25.5 | 30.0 |
| 7 | 9.64 | 9.64 | 8.97 | 8.54 | 29.0 | 32.0 | 26.0 | 30.0 |
| 8 | 9.33 | 9.94 | 8.54 | 8.48 | 29.0 | 32.0 | 27.0 | 30.5 |
| 9 | 9.64 | 9.94 | 8.48 | 8.48 | 29.0 | 32.5 | 28.0 | 30.0 |
| 10 | 9.64 | 9.88 | 9.64 | 9.52 | 30.5 | 31.0 | 28.5 | 31.0 |
| 11 | 9.70 | 9.94 | 9.70 | 9.76 | 30.0 | 33.0 | 29.0 | 30.0 |
| 12 | 9.83 | 10.65 | 9.22 | 9.84 | 30.5 | 33.0 | 29.5 | 30.0 |
| 13 | 9.94 | 10.85 | 9.70 | 9.84 | 30.0 | 34.0 | 29.0 | 30.0 |
| 14 | 10.00 | 10.54 | 9.76 | 9.64 | 30.0 | 35.0 | 29.0 | 30.0 |
| 15 | 10.00 | 10.97 | 9.07 | 9.68 | 31.0 | 33.0 | 29.0 | 30.0 |
| 16 | 10.52 | 10.54 | 9.64 | 9.70 | 32.0 | 30.0 | 29.0 | 30.5 |
| 17 | 10.67 | 10.30 | 9.09 | 10.06 | 33.0 | 28.5 | 28.5 | 30.5 |
| 18 | 10.48 | 9.45 | 9.09 | 9.33 | 32.5 | 30.0 | 28.5 | 29.5 |
| 19 | 10.00 | 9.33 | 9.15 | 9.09 | 32.0 | 30.0 | 29.0 | 29.0 |
| 20 | 9.52 | 8.48 | 9.09 | 9.09 | 29.5 | 28.5 | 29.0 | 28.5 |

TABLE 20

Composition of Milk
(Experiment II)

| Animals by treatment | Milk copper p.p.m. Days 0 | 75 | 150 | 225 | 300 | Milk molybdenum p.p.m. Days 0 | 75 | 150 | 225 | 300 |
|---|---|---|---|---|---|---|---|---|---|---|
| **Sulfate sulfur 0.3%** | | | | | | | | | | |
| 386 | 0.04 | 0.01 | 0.02 | 0.05 | 0.04 | 0.10 | 0.00 | 0.01 | 0.07 | 0.01 |
| 395 | 0.06 | 0.02 | 0.06 | 0.03 | 0.04 | 0.03 | 0.08 | 0.04 | 0.03 | 0.02 |
| 398 | 0.13 | 0.05 | 0.05 | 0.02 | 0.02 | 0.02 | 0.05 | 0.00 | 0.04 | 0.09 |
| **Sulfate sulfur 0.3%, molybdenum 5 p.p.m.** | | | | | | | | | | |
| 390 | 0.28 | 0.01 | 0.02 | | | 0.10 | 0.01 | 0.03 | | |
| 392 | 0.09 | 0.04 | 0.03 | 0.05 | | 0.07 | 0.07 | 0.04 | 0.02 | |
| 399 | 0.00 | 0.01 | 0.11 | | | 0.01 | 0.07 | 0.41 | | |
| **Sulfate sulfur 0.3%, molybdenum 10 p.p.m.** | | | | | | | | | | |
| 388 | 0.12 | 0.01 | 0.04 | 0.06 | | 0.03 | 0.06 | 0.33 | 0.02 | |
| 391 | 0.18 | 0.02 | 0.01 | 0.05 | | 0.12 | 0.05 | 0.04 | 0.08 | |
| 396 | 0.06 | 0.00 | 0.06 | 0.02 | | 0.03 | 0.11 | 0.11 | 0.02 | |
| **Sulfate sulfur 0.3%, molybdenum 20 p.p.m.** | | | | | | | | | | |
| 389 | 0.20 | 0.04 | 0.00 | 0.03 | 0.03 | 0.02 | 0.04 | 0.09 | 0.05 | 0.09 |
| 393 | 0.11 | 0.01 | 0.09 | 0.03 | 0.07 | 0.01 | 0.05 | 0.15 | 0.19 | 0.13 |
| 397 | 0.08 | 0.09 | 0.09 | 0.02 | 0.03 | 0.04 | 0.04 | 0.07 | 0.06 | 0.03 |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m.** | | | | | | | | | | |
| 387 | 0.08 | 0.02 | 0.13 | 0.05 | | 0.03 | 0.14 | 0.17 | 0.20 | |
| 394 | 0.09 | 0.02 | 0.14 | 0.00 | 0.07 | | 0.12 | 0.27 | 0.20 | |
| 400 | 0.11 | 0.14 | 0.09 | 0.03 | 0.04 | 0.03 | 0.06 | 0.14 | 0.25 | 0.18 |

TABLE 20 (Cont.)

Composition of Milk
(Experiment II)

| Animals by treatment | Milk copper p.p.m. Days 0 | 75 | 150 | 225 | 300 | Milk molybdenum p.p.m. Days 0 | 75 | 150 | 225 | 300 |
|---|---|---|---|---|---|---|---|---|---|---|
| **Sulfate sulfur 0.3%, molybdenum 100 p.p.m.** | | | | | | | | | | |
| 388 | | | | | 0.02 | | | | | 0.07 |
| 392 | | | | | 0.00 | | | | | 0.00 |
| **Sulfate sulfur 0.3%, molybdenum 200 p.p.m.** | | | | | | | | | | |
| 391 | | | | | 0.05 | | | | | 0.33 |
| 396 | | | | | 0.06 | | | | | 0.91 |
| **Animals Continued Through A Second Lactation** | | | | | | | | | | |
| **Sulfate sulfur 0.3%** | | | | | | | | | | |
| 386 | 0.01 | 0.00 | 0.05 | | | 0.00 | 0.02 | 0.05 | 0.13 | |
| 395 | 0.12 | 0.04 | 0.00 | 0.08 | 0.03 | 0.06 | 0.04 | 0.01 | 0.07 | 0.12 |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m.** | | | | | | | | | | |
| 387 | 0.02 | | 0.01 | 0.05 | | 0.25 | | 0.04 | 0.07 | |
| 394 | 0.04 | 0.02 | 0.02 | 0.06 | 0.07 | 0.10 | 0.44 | 0.25 | 0.32 | 0.06 |

TABLE 21

Composition of Serum
(Experiment II)

| Animals by treatment | Serum copper p.p.m. Days 0 | 75 | 150 | 225 | 300 | Serum molybdenum p.p.m. Days 0 | 75 | 150 | 225 | 300 |
|---|---|---|---|---|---|---|---|---|---|---|
| **Sulfate sulfur 0.3%** | | | | | | | | | | |
| 386 | 1.1 | 0.9 | 1.0 | 1.0 | 0.7 | 0.00 | 0.03 | 0.02 | 0.03 | 0.29 |
| 395 | 0.8 | 0.7 | 0.7 | 0.6 | 0.9 | 0.01 | 0.00 | 0.03 | 0.00 | 0.42 |
| 398 | 0.7 | 0.6 | 0.6 | 0.6 | 0.52 | 0.04 | 0.05 | 0.00 | 0.00 | 0.00 |
| **Sulfate sulfur 0.3%, molybdenum 5 p.p.m.** | | | | | | | | | | |
| 390 | 1.0 | 0.9 | 0.8 | | | 0.01 | 0.09 | 0.02 | | |
| 392 | 0.8 | 0.9 | 0.8 | 0.6 | | 0.02 | 0.06 | 0.08 | 0.18 | |
| 399 | 0.8 | 0.9 | 0.7 | | | 0.02 | 0.05 | 0.07 | | |
| **Sulfate sulfur 0.3%, molybdenum 10 p.p.m.** | | | | | | | | | | |
| 388 | 0.6 | 0.5 | 0.4 | 0.6 | | 0.02 | 0.26 | 0.05 | 0.22 | |
| 392 | 0.9 | 0.9 | 0.6 | 0.7 | | 0.02 | 0.48 | 0.18 | 0.58 | |
| 399 | 1.2 | 0.8 | 0.9 | 0.6 | | 0.01 | 0.18 | 0.61 | 0.15 | |
| **Sulfate sulfur 0.3%, molybdenum 20 p.p.m.** | | | | | | | | | | |
| 389 | 1.1 | 0.8 | 0.6 | 0.7 | 0.8 | 0.02 | 0.30 | 0.28 | 0.41 | 0.22 |
| 393 | 0.9 | 1.1 | 1.1 | 0.7 | 0.6 | 0.02 | 0.30 | 0.88 | 1.2 | 0.67 |
| 397 | 0.8 | 0.7 | 0.5 | 0.5 | 0.24 | 0.03 | 0.20 | 0.48 | 0.23 | 0.18 |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m.** | | | | | | | | | | |
| 387 | 1.0 | 1.1 | 1.6 | 0.6 | 0.7 | 0.02 | 0.76 | 0.63 | 0.72 | 1.0 |
| 394 | 0.7 | 0.9 | 0.6 | 0.6 | 0.7 | 0.01 | 0.72 | 1.3 | 1.00 | 1.42 |
| 400 | 0.9 | 1.3 | 1.3 | 0.9 | 0.70 | 0.02 | 0.55 | 0.78 | 0.61 | 0.66 |

TABLE 21 (Cont.)

Composition of Serum
(Experiment II)

| Animals by treatment | Serum copper p.p.m. Days 0 | 75 | 150 | 225 | 300 | Serum molybdenum p.p.m. Days 0 | 75 | 150 | 225 | 300 |
|---|---|---|---|---|---|---|---|---|---|---|
| **Sulfate sulfur 0.3%, molybdenum 100 p.p.m.** | | | | | | | | | | |
| 388 | | 1.0 | | | | | 0.78 | | | |
| 392 | | 0.9 | | | | | 1.70 | | | |
| **Sulfate sulfur 0.3%, molybdenum 200 p.p.m.** | | | | | | | | | | |
| 391 | | 0.9 | | | | | 1.3 | | | |
| 396 | | 0.9 | | | | | 2.9 | | | |
| **Animals Continued Through A Second Lactation** | | | | | | | | | | |
| **Sulfate sulfur 0.3%** | | | | | | | | | | |
| 386 | 0.74 | 0.74 | 0.96 | 1.11 | 1.50 | 0.02 | 0.00 | 0.00 | 0.00 | 0.00 |
| 395 | 0.75 | 0.72 | 0.71 | 0.68 | 0.78 | 0.0 | 0.00 | 0.01 | 0.03 | 0.00 |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m.** | | | | | | | | | | |
| 387 | 0.40 | | 0.52 | 0.43 | 0.36 | 0.64 | | 0.16 | 0.18 | |
| 394 | 0.52 | 0.68 | 0.68 | 0.48 | 0.60 | 0.39 | 1.34 | 0.71 | 1.47 | 0.33 |
| **Calves of: Sulfate sulfur 0.3%, molybdenum 50 p.p.m. (at birth)** | | | | | | | | | | |
| 387 | | | 0.00 | | | | | 0.13 | | |
| 394 | | | 0.05 | | | | | 0.46 | | |
| **Sulfate sulfur 0.3% (at birth)** | | | | | | | | | | |
| 386 | | | 0.30 | | | | | 0.05 | | |
| 395 | | | 0.16 | | | | | 0.00 | | |

TABLE 22

Composition of Liver
(Experiment II)

| Animals by treatment | Liver copper p.p.m. Days 0 | 75 | 150 | 225 | 300 | Liver molybdenum p.p.m. Days 0 | 75 | 150 | 225 | 300 |
|---|---|---|---|---|---|---|---|---|---|---|
| **Sulfate sulfur 0.3%** | | | | | | | | | | |
| 386 | 111 | 198 | 158 | 120 | 124 | 4 | 1 | 2 | 40 | 8 |
| 395 | 40 | 70 | 107 | 108 | 75 | 2 | 2 | 9 | 7 | 29 |
| 398 | 13 | 36 | 28 | 26 | 170 | 11 | 1 | 44 | 7 | 2.5 |
| **Sulfate sulfur 0.3%, molybdenum 5 p.p.m.** | | | | | | | | | | |
| 390 | 148 | 100 | 30 | | | 7 | 3 | 6 | | |
| 392 | 140 | 59 | 35 | 5 | | 2 | 4 | 11 | 7 | |
| 399 | 104 | 24 | 8 | | | 0 | 2 | 70 | | |
| **Sulfate sulfur 0.3%, molybdenum 10 p.p.m.** | | | | | | | | | | |
| 388 | 10 | 9 | 11 | 2 | | 10 | 4 | 3 | 7 | |
| 391 | 127 | 69 | 12 | 4 | | 3 | 4 | 5 | 22 | |
| 396 | 1070 | 32 | 31 | 15 | | 2 | 9 | 14 | 7 | |
| **Sulfate sulfur 0.3%, molybdenum 20 p.p.m.** | | | | | | | | | | |
| 389 | 54 | 29 | 12 | 2 | 12 | 6 | 5 | 4 | 15 | 16 |
| 393 | 257 | 179 | 92 | 18 | 9 | 2 | 9 | 24 | 40 | 13 |
| 397 | 71 | 58 | 38 | 16 | 0 | 0 | 11 | 13 | 7 | 9 |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m.** | | | | | | | | | | |
| 387 | 78 | 48 | 32 | 6 | 14 | 7 | 14 | 26 | 30 | 6 |
| 394 | 58 | 11 | 29 | 16 | 24 | 4 | 6 | 56 | 27 | 9 |
| 400 | 86 | 33 | 19 | 19 | 0 | 1 | 16 | 41 | 10 | 14 |

TABLE 22 (Cont.)

Composition of Liver
(Experiment II)

| Animals by treatment | Liver copper p.p.m. Days 0 | 75 | 150 | 225 | 300 | Liver molybdenum p.p.m. Days 0 | 75 | 150 | 225 | 300 |
|---|---|---|---|---|---|---|---|---|---|---|
| **Sulfate sulfur 0.3%, molybdenum 100 p.p.m.** | | | | | | | | | | |
| 388 | | 10 | | | | | 14 | | | |
| 392 | | 24 | | | | | 84 | | | |
| **Sulfate sulfur 0.3%, molybdenum 200 p.p.m.** | | | | | | | | | | |
| 391 | | 22 | | | | | 29 | | | |
| 396 | | 47 | | | | | 36 | | | |
| **Animals Continued Through A Second Lactation** | | | | | | | | | | |
| **Sulfate sulfur 0.3%** | | | | | | | | | | |
| 386 | | 126 | 133 | 83 | 36 | | 8.2 | 6.6 | 5.5 | 2.1 |
| 395 | 44 | 98 | | 106 | 110 | 0 | 4.2 | | 0 | 5.9 |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m.** | | | | | | | | | | |
| 387 | 11 | 86 | 14 | 12 | 8 | 11 | 0.78 | 11 | 13 | 27 |
| 394 | 7.4 | 10 | 20 | 27 | 15 | 23 | 31 | 31 | 35 | 24 |

| Calves of: Sulfate sulfur 0.3%, molybdenum 50 p.p.m. (at birth) | Liver | Spleen | Liver | Spleen |
|---|---|---|---|---|
| 394 | 1.70 | .83 | 0 | 0 |
| 387 | 1.9 | .91 | 0 | 0 |
| **Sulfate sulfur 0.3% (at birth)** | | | | |
| 386 | 118 | .60 | 0 | 0 |
| 395 | 127 | 1.40 | 0 | 0 |

## TABLE 23

### Consumption of Trace Elements as Salts*
### (Experiment III)

| Animals by treatment | $Na_2Mo_4.2H_2O$ | $Na_2HAsO_4.7H_2O$ | $PbSO_4$ | RbCl |
|---|---|---|---|---|
| **Sulfate sulfur 0.3% (outdoors)** | | | | |
| 477 | | | | |
| 482 | | | | |
| 488 | | | | |
| **Molybdenum 50 p.p.m. (outdoors)** | | | | |
| 479 | 609.4362 | | | |
| 481 | 583.9814 | | | |
| 489 | 628.5306 | | | |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m. (outdoors)** | | | | |
| 480 | 662.9193 | | | |
| 483 | 664.2019 | | | |
| 490 | 520.8488 | | | |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m. (indoors)** | | | | |
| 476 | 493.1749 | | | |
| 485 | 557.7292 | | | |
| 487 | 479.3851 | | | |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m., rubidium 100 p.p.m., arsenic 5 p.p.m., and lead 5 p.p.m. (indoors)** | | | | |
| 478 | 595.2067 | 72.1123 | 32.6528 | 613.2326 |
| 484 | 622.1376 | 77.9041 | 35.2567 | 685.4966 |
| 486 | 515.8651 | 65.9668 | 29.8400 | 589.6963 |
| **Animals Carried Through A Second Lactation** | | | | |
| **Sulfate sulfur 0.3%** | | | | |
| 386 | | | | |
| 395 | | | | |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m.** | | | | |
| 387 | 564.4218 | | | |
| 394 | 812.2045 | | | |

*Total for 300 days in grains.

TABLE 24

Body Weight in Pounds
(Experiment III)

| | Animal number | | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Week | 476 | 477 | 478 | 479 | 480 | 481 | 482 | 483 | 484 | 485 | 486 | 487 | 488 | 489 | 490 |
| 1 | 878 | 1011 | 973 | 1041 | 1063 | 1035 | 987 | 1230 | 1049 | 963 | 901 | 893 | 934 | 1109 | 992 |
| 2 | 900 | 977 | 952 | 992 | 1040 | 1000 | 997 | 1200 | 1046 | 937 | 875 | 835 | 912 | 1048 | 965 |
| 3 | 816 | 968 | 935 | 970 | 1017 | 1107 | 996 | 1111 | 1054 | 937 | 865 | 854 | 927 | 1020 | 963 |
| 4 | 857 | 995 | 873 | 967 | 1026 | 1009 | 1010 | 1088 | 1051 | 944 | 854 | 811 | 936 | 974 | 936 |
| 5 | 891 | 1011 | 886 | 976 | 1025 | 1025 | 1034 | 1100 | 1052 | 929 | 852 | 841 | 907 | 992 | 976 |
| 6 | 890 | 1022 | 912 | 1077 | 1044 | 1019 | 1010 | 1098 | 1068 | 967 | 872 | 846 | 919 | 1007 | 974 |
| 7 | 884 | 1007 | 913 | 982 | 1037 | 1031 | 1059 | 1114 | 1059 | 938 | 875 | 822 | 944 | 1016 | 963 |
| 8 | 874 | 1020 | 937 | 1014 | 1043 | 1064 | 1050 | 1100 | 1080 | 952 | 863 | 888 | 920 | 1025 | 974 |
| 9 | 887 | 1026 | 934 | 1022 | 1054 | 1020 | 1089 | 1067 | 1050 | 962 | 873 | 859 | 944 | 1012 | 977 |
| 10 | 904 | 1028 | 932 | 1020 | 1076 | 1053 | 1018 | 1083 | 1053 | 955 | 900 | 835 | 937 | 1033 | 923 |
| 11 | 860 | 1037 | 865 | 930 | 1075 | 1057 | 1062 | 1090 | 1041 | 960 | 881 | 886 | 922 | 1041 | 948 |
| 12 | 895 | 1052 | 957 | 1043 | 1015 | 1052 | 1079 | 1126 | 1048 | 965 | 866 | 862 | 957 | 1043 | 935 |
| 13 | 891 | 1029 | 929 | 947 | 1065 | 1091 | 1052 | 1084 | 1006 | 930 | 886 | 911 | 955 | 1034 | 953 |
| 14 | 903 | 983 | 940 | 1008 | 1059 | 1095 | 1024 | 1047 | 1058 | 985 | 883 | 856 | 963 | 1026 | 948 |
| 15 | 885 | 1047 | 957 | 1024 | 1077 | 1061 | 1072 | 1121 | 1046 | 986 | 899 | 866 | 929 | 1034 | 947 |

TABLE 24 (Cont.)

Body Weight in Pounds
(Experiment III)

| | Animal number | | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Week | 476 | 477 | 478 | 479 | 480 | 481 | 482 | 483 | 484 | 485 | 486 | 487 | 488 | 489 | 490 |
| 16 | 896 | 1032 | 988 | 1038 | 1052 | 1102 | 1059 | 1135 | 1055 | 1000 | 877 | 861 | 972 | 1044 | 920 |
| 17 | 886 | 1008 | 951 | 1026 | 1061 | 1092 | 1079 | 1096 | 1002 | 967 | 856 | 850 | 979 | 1023 | 915 |
| 18 | 874 | 979 | 922 | 1029 | 1096 | 1117 | 1095 | 1074 | 994 | 950 | 840 | 858 | 946 | 1042 | 913 |
| 19 | 886 | 1029 | 974 | 1026 | 1081 | 1104 | 1091 | 1079 | 1016 | 954 | 852 | 847 | 963 | 1054 | 926 |
| 20 | 908 | 987 | 966 | 1015 | 1066 | 1118 | 1105 | 1071 | 1022 | 945 | 872 | 834 | 930 | 1057 | 910 |
| 21 | 900 | 968 | 985 | 1034 | 1058 | 1132 | 1089 | 1047 | 1011 | 958 | 884 | 863 | 1000 | 1053 | 935 |
| 22 | 888 | 932 | 938 | 986 | 1078 | 1109 | 1112 | 1099 | 990 | 954 | 846 | 827 | 985 | 1069 | 939 |
| 23 | 899 | 959 | 925 | 1025 | 1069 | 1118 | 1130 | 1123 | 972 | 947 | 857 | 870 | 975 | 1048 | 889 |
| 24 | 919 | 1008 | 928 | 1016 | 1051 | 1095 | 1130 | 1083 | 980 | 953 | 845 | 856 | 996 | 1042 | 910 |
| 25 | 909 | 1010 | 946 | 1041 | 1055 | 1108 | 1154 | 1103 | 992 | 980 | 865 | 827 | 1010 | 1072 | 920 |
| 26 | 933 | 1028 | 930 | 1022 | 1068 | 1105 | 1136 | 1076 | 980 | 972 | 827 | 830 | 1000 | 1082 | 939 |
| 27 | 931 | 1035 | 931 | 1062 | 1057 | 1118 | 1151 | 1098 | 947 | 948 | 845 | 823 | 1015 | 1034 | 912 |
| 28 | 931 | 1024 | 926 | 1031 | 1071 | 1115 | 1159 | 1066 | 958 | 958 | 865 | 842 | 1032 | 1080 | 900 |
| 29 | 923 | 1019 | 930 | 1064 | 1074 | 1121 | 1155 | 1098 | 1000 | 953 | 874 | 800 | 1032 | 1077 | 893 |
| 30 | 939 | 1015 | 944 | 1022 | 1084 | 1139 | 1188 | 1112 | 952 | 930 | 858 | 855 | 1028 | 1077 | 895 |

TABLE 24 (Cont.)

Body Weight in Pounds
(Experiment III)

| | Animal number | | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Week | 476 | 477 | 478 | 479 | 480 | 481 | 482 | 483 | 484 | 485 | 486 | 487 | 488 | 489 | 490 |
| 31 | 929 | 1030 | 913 | 1057 | 1103 | 1145 | 1184 | 1133 | 977 | 943 | 867 | 829 | 1036 | 1062 | 905 |
| 32 | 926 | 1054 | 930 | 1054 | 1040 | 1111 | 1164 | 1133 | 993 | 962 | 869 | 857 | 1024 | 1105 | 923 |
| 33 | 920 | 1039 | 925 | 1059 | 1092 | 1109 | 1182 | 1163 | 939 | 934 | 849 | 858 | 1029 | 1041 | 922 |
| 34 | 896 | 1059 | 962 | 1070 | 1067 | 1117 | 1173 | 1131 | 984 | 927 | 854 | 860 | 1025 | 1080 | 932 |
| 35 | 904 | 1019 | 943 | 1094 | 1015 | 1126 | 1162 | 1075 | 1015 | 961 | 856 | 847 | 1025 | 1080 | 908 |
| 36 | 930 | 1014 | 912 | 1091 | 1112 | 1152 | 1196 | 1144 | 1020 | 958 | 838 | 829 | 1065 | 1085 | 905 |
| 37 | 915 | 1036 | 922 | 1090 | 1090 | 1188 | 1173 | 1145 | 1000 | 986 | 831 | 883 | 1052 | 1119 | 915 |
| 38 | 928 | 1044 | 955 | 1105 | 1091 | 1167 | 1179 | 1136 | 1022 | 948 | 834 | 792 | 1076 | 1105 | 943 |
| 39 | 886 | 1058 | 963 | 1112 | 1103 | 1183 | 1214 | 1130 | 992 | 935 | 843 | 759 | 1052 | 1122 | 946 |
| 40 | 930 | 1065 | 998 | 1140 | 1092 | 1164 | 1199 | 1142 | 998 | 923 | 872 | 750 | 1067 | 1122 | 951 |
| 41 | 922 | 1046 | 994 | 1111 | 1110 | 1167 | 1220 | 1139 | 1005 | 936 | 842 | 750 | 1070 | 1109 | 988 |
| 42 | 906 | 1096 | 988 | 1107 | 1087 | 1191 | 1203 | 1163 | 993 | 928 | 822 | | 1092 | 1136 | 983 |
| 43 | 878 | 1063 | 992 | 1115 | 1068 | 1189 | 1200 | 1140 | 987 | 921 | 836 | | 1052 | 1140 | 1000 |

**TABLE 25**

**Average Body Weights in Pounds**
**(Experiment III)**

| Animal number | Days | | | | |
|---|---|---|---|---|---|
| | 0 | 75 | 150 | 225 | 300 |
| 476 | 865 | 886 | 899 | 931 | 902 |
| 477 | 985 | 1039 | 962 | 1033 | 1068 |
| 478 | 953 | 963 | 1030 | 929 | 991 |
| 479 | 1001 | 998 | 1012 | 1044 | 1111 |
| 480 | 1040 | 1055 | 1067 | 1076 | 1088 |
| 481 | 1047 | 1054 | 1120 | 1132 | 1182 |
| 482 | 993 | 1038 | 1102 | 1179 | 1208 |
| 483 | 1180 | 1100 | 1072 | 1123 | 1147 |
| 484 | 1050 | 1047 | 1008 | 974 | 995 |
| 485 | 946 | 960 | 952 | 945 | 928 |
| 486 | 880 | 882 | 867 | 864 | 833 |
| 487 | 861 | 861 | 841 | 847 | 750 |
| 488 | 924 | 939 | 972 | 1029 | 1071 |
| 489 | 1059 | 1039 | 1060 | 1081 | 1128 |
| 490 | 973 | 935 | 928 | 908 | 990 |

TABLE 26

Hemoglobins

(Experiment III)

| Biweekly values | Animal number 476 | 477 | 478 | 479 | 480 | 481 | 482 | 483 | 484 | 485 | 486 | 487 | 488 | 489 | 490 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | mg/100 ml blood | | | | | | | | | | | | | | |
| 1 | 10.61 | 11.33 | 10.67 | 9.52 | 10.48 | 9.94 | 9.82 | 9.33 | 10.61 | 9.52 | 9.15 | 10.54 | 11.27 | 10.00 | 10.00 |
| 2 | 9.45 | 11.03 | 10.30 | 8.91 | 9.94 | 9.76 | 9.70 | 9.39 | 10.79 | 9.33 | 9.64 | 10.54 | 11.03 | 10.20 | 10.82 |
| 3 | 8.79 | 10.54 | 9.03 | 8.67 | 10.00 | 9.82 | 9.70 | 9.40 | 10.30 | 9.21 | 9.70 | 10.30 | 10.60 | 10.20 | 11.03 |
| 4 | 8.91 | 11.03 | 9.27 | 8.54 | 10.18 | 10.12 | 9.21 | 9.58 | 10.30 | 9.21 | 9.88 | 10.52 | 10.79 | 10.67 | 11.64 |
| 5 | 8.97 | 11.21 | 9.21 | 8.36 | 10.00 | 9.94 | 10.54 | 9.58 | 10.00 | 9.33 | 9.76 | 10.64 | 10.67 | 10.00 | 11.70 |
| 6 | 9.64 | 11.03 | 9.70 | 8.91 | 10.18 | 10.54 | 11.03 | 10.00 | 10.61 | 9.45 | 9.94 | 10.82 | 10.00 | 10.42 | 11.52 |
| 7 | 9.94 | 11.21 | 9.60 | 9.64 | 10.00 | 11.03 | 10.12 | 9.33 | 10.61 | 9.52 | 9.88 | 10.73 | 9.94 | 10.91 | 11.25 |
| 8 | 10.00 | 10.91 | 10.00 | 8.30 | 9.45 | 11.03 | 10.12 | 9.39 | 10.42 | 10.00 | 9.94 | 10.73 | 9.76 | 11.45 | 11.03 |
| 9 | 9.94 | 10.61 | 10.20 | 7.64 | 9.21 | 11.09 | 10.42 | 9.27 | 10.48 | 10.36 | 10.06 | 10.54 | 10.36 | 11.29 | 11.03 |
| 10 | 10.00 | 10.73 | 10.54 | 8.30 | 9.39 | 10.73 | 10.36 | 9.15 | 10.60 | 10.50 | 10.00 | 10.83 | 10.63 | 11.45 | 11.25 |
| 11 | 10.42 | 10.73 | 10.48 | 8.79 | 9.76 | 10.00 | 10.32 | 9.03 | 10.79 | 10.54 | 9.84 | 10.79 | 10.63 | 11.21 | 11.09 |
| 12 | 10.38 | 10.94 | 10.66 | 8.96 | 9.33 | 10.42 | 10.32 | 8.97 | 10.73 | 10.54 | 9.64 | 9.94 | 10.48 | 10.97 | 10.67 |
| 13 | 10.42 | 10.94 | 10.48 | 8.79 | 9.45 | 10.61 | 10.42 | 8.73 | 11.50 | 11.03 | 9.84 | 10.03 | 10.65 | 10.97 | 9.82 |
| 14 | 10.12 | 11.21 | 10.54 | 8.67 | 9.58 | 10.66 | 10.63 | 9.19 | 11.45 | 10.97 | 9.39 | 10.00 | 10.85 | 11.09 | 9.90 |
| 15 | 10.30 | 11.61 | 10.00 | 8.67 | 9.67 | 10.76 | 10.36 | 9.21 | 11.39 | 11.32 | 8.91 | 10.48 | 9.94 | 10.85 | 9.33 |
| 16 | 10.00 | 11.33 | 11.03 | 8.79 | 10.00 | 10.91 | 10.36 | 8.67 | 10.30 | 10.30 | 9.09 | 8.91 | 9.21 | 9.94 | 9.15 |
| 17 | 9.82 | 11.09 | 11.21 | 8.48 | 10.24 | 10.18 | 10.18 | 9.21 | 9.52 | 10.00 | 8.73 | 7.09 | 9.45 | 8.42 | 9.39 |
| 18 | 9.33 | 11.82 | 10.79 | 8.67 | 10.36 | 10.00 | 9.52 | 8.91 | 9.33 | 9.70 | 8.42 | 6.79 | 9.58 | 9.09 | 9.52 |
| 19 | 8.91 | 11.88 | 10.67 | 8.48 | 10.67 | 10.00 | 9.15 | 8.73 | 9.52 | 9.82 | 8,85 | 6.42 | 9.82 | 9.52 | 9.90 |
| 20 | 8.61 | 13.27 | 10.42 | 8.48 | | 10.18 | 9.45 | 8,85 | 10.00 | 9.52 | 8.73 | | 10.06 | 9.27 | 9.52 |

TABLE 27

Hematocrits
(Experiment III)

| Biweekly values | Animal number 476 | 477 | 478 | 479 | 480 | 481 | 482 | 483 | 484 | 485 | 486 | 487 | 488 | 489 | 490 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | % | | | | | | | | | | | | | | |
| 1 | 32.0 | 31.5 | 34.0 | 29.0 | 33.0 | 30.0 | 30.0 | 29.5 | 34.0 | 32.0 | 32.0 | 36.5 | 36.0 | 32.0 | 34.0 |
| 2 | 28.5 | 33.0 | 32.0 | 30.0 | 32.0 | 29.5 | 30.0 | 28.0 | 34.0 | 31.0 | 30.5 | 31.0 | 33.0 | 33.0 | 32.0 |
| 3 | 29.0 | 34.0 | 28.5 | 27.5 | 31.0 | 30.0 | 30.0 | 30.0 | 35.0 | 30.5 | 30.5 | 32.0 | 33.0 | 33.0 | 33.0 |
| 4 | 27.0 | 36.0 | 30.0 | 28.0 | 32.0 | 30.5 | 29.0 | 30.5 | 34.0 | 30.0 | 31.0 | 32.0 | 33.5 | 33.0 | 34.0 |
| 5 | 28.5 | 35.5 | 30.0 | 24.4 | 31.5 | 30.0 | 29.0 | 30.5 | 34.0 | 30.0 | 31.0 | 32.0 | 33.0 | 33.0 | 35.0 |
| 6 | 30.5 | 35.0 | 31.0 | 25.5 | 31.0 | 30.5 | 29.0 | 30.5 | 34.0 | 30.0 | 31.0 | 32.0 | 33.0 | 33.0 | 36.0 |
| 7 | 31.0 | 33.0 | 32.5 | 24.0 | 31.0 | 30.5 | 29.0 | 30.0 | 35.0 | 30.0 | 31.0 | 32.0 | 32.0 | 33.0 | 36.0 |
| 8 | 32.0 | 33.0 | 33.0 | 25.0 | 30.0 | 30.5 | 29.0 | 30.0 | 36.0 | 32.0 | 31.0 | 32.0 | 33.5 | 34.5 | 36.0 |
| 9 | 33.0 | 34.0 | 31.5 | 25.5 | 30.0 | 30.0 | 30.0 | 30.0 | 36.0 | 33.0 | 31.5 | 32.5 | 33.5 | 35.0 | 36.0 |
| 10 | 32.0 | 34.0 | 31.5 | 25.5 | 30.0 | 31.0 | 32.0 | 28.5 | 35.0 | 34.0 | 32.0 | 32.5 | 33.5 | 34.0 | 36.0 |
| 11 | 33.0 | 35.0 | 31.5 | 26.5 | 28.0 | 31.0 | 32.0 | 28.0 | 36.0 | 34.0 | 32.0 | 32.0 | 33.0 | 35.5 | 35.0 |
| 12 | 33.0 | 35.0 | 32.0 | 27.0 | 28.0 | 31.0 | 33.0 | 27.0 | 37.5 | 35.0 | 31.5 | 32.5 | 33.5 | 34.0 | 34.0 |
| 13 | 33.0 | 35.0 | 32.0 | 28.0 | 29.0 | 31.5 | 34.0 | 27.5 | 36.0 | 32.0 | 31.5 | 30.0 | 33.0 | 34.0 | 33.5 |
| 14 | 34.0 | 36.0 | 32.5 | 26.5 | 29.0 | 32.0 | 34.0 | 28.5 | 35.0 | 32.0 | 30.0 | 29.5 | 32.0 | 33.5 | 32.0 |
| 15 | 34.0 | 36.0 | 32.0 | 26.0 | 29.0 | 30.5 | 34.0 | 27.0 | 33.5 | 31.0 | 28.5 | 29.0 | 31.5 | 34.0 | 29.5 |
| 16 | 32.0 | 37.0 | 32.0 | 26.5 | 30.0 | 31.5 | 33.0 | 26.5 | 32.5 | 33.0 | 28.5 | 29.5 | 30.5 | 30.0 | 29.5 |
| 17 | 31.0 | 37.0 | 31.5 | 25.0 | 31.0 | 31.5 | 31.0 | 29.0 | 32.0 | 34.0 | 28.0 | 27.0 | 30.5 | 26.5 | 29.5 |
| 18 | 29.0 | 38.5 | 34.0 | 26.5 | 33.0 | 31.0 | 33.5 | 28.0 | 31.5 | 33.0 | 27.0 | 21.0 | 31.5 | 28.0 | 28.0 |
| 19 | 28.0 | 40.0 | 34.0 | 26.0 | 33.0 | 30.5 | 33.0 | 28.0 | 32.0 | 32.0 | 27.0 | 20.0 | 32.0 | 30.0 | 28.5 |
| 20 | 27.5 | 41.5 | 33.0 | 25.5 | | 30.5 | 29.0 | 30.0 | 31.5 | 33.0 | 27.5 | | 32.5 | 30.0 | 29.0 |

TABLE 28

Composition of Milk
(Experiment III)

| Animals by treatment | Milk copper p.p.m. Days 0 | 75 | 150 | 225 | 300 | Milk molybdenum p.p.m. Days 0 | 75 | 150 | 225 | 300 |
|---|---|---|---|---|---|---|---|---|---|---|
| **Sulfate sulfur 0.3% (outdoors)** | | | | | | | | | | |
| 477 | 0.08 | 0.04 | 0.01 | 0.00 | 0.08 | --- | 0.02 | 0.01 | 0.01 | 0.05 |
| 482 | 0.06 | 0.02 | 0.00 | 0.07 | 0.07 | 0.03 | 0.03 | 0.01 | 0.24 | 0.02 |
| 488 | 0.08 | 0.02 | 0.00 | 0.05 | 0.06 | 0.03 | 0.00 | 0.00 | 0.03 | 0.06 |
| **Molybdenum 50 p.p.m. (outdoors)** | | | | | | | | | | |
| 479 | 0.06 | 0.02 | 0.00 | 0.00 | --- | 0.02 | 0.60 | 0.79 | 0.55 | 0.88 |
| 481 | 0.12 | 0.01 | 0.00 | 0.04 | 0.04 | 0.04 | 0.84 | 0.54 | 0.52 | 0.03 |
| 489 | 0.07 | 0.02 | 0.00 | 0.02 | 0.01 | 0.00 | 0.86 | 0.33 | 0.30 | --- |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m. (outdoors)** | | | | | | | | | | |
| 480 | 0.13 | 0.01 | 0.00 | 0.00 | 0.07 | 0.03 | 0.24 | 0.06 | 0.16 | 0.16 |
| 483 | 0.08 | 0.01 | 0.00 | 0.01 | 0.03 | 0.03 | 0.16 | 0.14 | 0.11 | 0.25 |
| 490 | 0.08 | 0.00 | 0.00 | 0.02 | --- | 0.24 | 0.10 | 0.12 | 0.40 | --- |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m. (indoors)** | | | | | | | | | | |
| 476 | 0.11 | 0.02 | 0.01 | 0.00 | 0.04 | 0.02 | 0.10 | 0.29 | 0.05 | 0.17 |
| 485 | 0.10 | 0.01 | 0.00 | 0.06 | 0.02 | 0.06 | 0.10 | 0.04 | 0.17 | 0.14 |
| 487 | 0.08 | 0.00 | 0.00 | 0.03 | 0.02 | 0.03 | 0.11 | 0.02 | 0.14 | 0.38 |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m., rubidium 100 p.p.m., arsenic 5 p.p.m., and lead 5 p.p.m. (indoors)** | | | | | | | | | | |
| 478 | 0.11 | 0.01 | 0.00 | 0.00 | 0.06 | 0.02 | 0.33 | 0.18 | 0.22 | 0.05 |
| 484 | 0.12 | 0.01 | 0.00 | 0.02 | 0.03 | 0.03 | 0.46 | 0.13 | 0.24 | 0.51 |
| 486 | 0.06 | 0.00 | 0.00 | 0.05 | 0.09 | 0.02 | 0.05 | 0.01 | 0.10 | 0.15 |

TABLE 29

Composition of Serum
(Experiment III)

| Animals by treatment | Serum copper p.p.m. Days 0 | 75 | 150 | 225 | 300 | Serum molybdenum p.p.m. Days 0 | 75 | 150 | 225 | 300 |
|---|---|---|---|---|---|---|---|---|---|---|
| **Sulfate sulfur 0.3% (outdoors)** | | | | | | | | | | |
| 477 | 0.80 | 0.95 | 0.98 | 0.59 | 0.60 | 0.0 | 0.01 | 0.0 | 0.0 | 0.0 |
| 482 | 0.40 | 0.56 | 0.55 | 0.55 | 0.59 | 0.0 | 0.02 | 0.01 | 0.03 | 0.05 |
| 488 | 1.14 | 0.62 | 0.78 | 0.57 | 0.73 | 0.0 | 0.0 | 0.02 | 0.0 | 0.03 |
| **Molybdenum 50 p.p.m. (outdoors)** | | | | | | | | | | |
| 479 | 0.87 | 0.78 | 0.72 | 0.60 | 0.82 | 0.0 | 2.15 | 1.99 | 1.88 | 2.16 |
| 481 | 1.19 | 0.98 | 1.06 | 1.25 | 0.62 | 0.0 | 4.22 | 0.86 | 1.12 | 1.20 |
| 489 | 0.75 | 1.07 | 0.70 | 0.72 | 0.44 | 0.0 | 0.98 | 0.76 | 0.94 | --- |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m. (outdoors)** | | | | | | | | | | |
| 480 | 0.78 | 1.12 | 0.96 | 0.79 | 0.66 | 0.00 | 1.57 | 0.66 | 0.72 | 0.84 |
| 483 | 0.76 | 0.80 | 0.64 | 0.60 | 0.22 | 0.00 | 0.48 | 0.40 | 0.62 | 0.30 |
| 490 | 0.90 | 0.88 | 0.50 | 0.24 | 0.23 | 0.00 | 0.48 | 0.58 | 0.66 | --- |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m. (indoors)** | | | | | | | | | | |
| 476 | 0.60 | 0.68 | 0.70 | 0.55 | 0.71 | 0.00 | 0.42 | 0.73 | 0.29 | 0.59 |
| 485 | 0.88 | 0.84 | 0.62 | 0.56 | 0.16 | 0.00 | 0.54 | 0.31 | --- | 0.10 |
| 487 | 0.68 | 0.68 | 0.42 | 0.60 | 0.26 | 0.00 | 1.00 | 0.20 | 0.75 | 0.45 |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m., rubidium 100 p.p.m., arsenic 5 p.p.m., and lead 5 p.p.m. (indoors)** | | | | | | | | | | |
| 478 | 0.80 | 0.90 | 0.76 | 0.64 | 0.68 | 0.02 | 1.07 | 1.16 | 1.24 | 0.44 |
| 484 | 0.69 | 1.09 | 0.80 | 0.72 | 0.61 | 0.00 | 1.00 | 0.78 | 0.93 | 1.04 |
| 486 | 0.69 | 0.78 | 0.74 | 0.66 | 0.56 | 0.00 | 0.64 | 0.32 | 0.27 | 0.30 |

TABLE 30

Composition of Liver
(Experiment III)

| Animals by treatment | Liver copper p.p.m. Days 0 | 75 | 150 | 225 | 300 | Liver molybdenum p.p.m. Days 0 | 75 | 150 | 225 | 300 |
|---|---|---|---|---|---|---|---|---|---|---|
| **Sulfate sulfur 0.3% (outdoors)** | | | | | | | | | | |
| 477 | 6.4 | 53 | 75 | 38 | 46 | 2.3 | 2.4 | 6.2 | 20.0 | 6.0 |
| 482 | 0.0 | 23 | 49 | 48 | 14 | 0.0 | 4.3 | 7.0 | 7.8 | 7.8 |
| 488 | 48.0 | 78 | 90 | 65 | 27 | 1.9 | 3.2 | 8.2 | 8.0 | 28.0 |
| **Molybdenum 50 p.p.m. (outdoors)** | | | | | | | | | | |
| 479 | 26.0 | 0 | 9.3 | 0 | 6.8 | 3.3 | 30 | 31 | 27 | 33 |
| 481 | 0.0 | 1.7 | 13.0 | 9.7 | 5.6 | 0.0 | 16 | 29 | 25 | 20 |
| 489 | 123.0 | 59.0 | 30.0 | 16.0 | 85.0 | 0.3 | 25 | 26 | 28 | 28 |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m. (outdoors)** | | | | | | | | | | |
| 480 | 68 | 31 | 25 | 18 | 18 | 0.0 | 22 | 20 | 19 | 29 |
| 483 | 0 | 5.5 | 11 | 17.0 | 9.0 | 0.0 | 12 | 13 | 16 | 8 |
| 490 | 20 | 32 | 12 | 8.0 | 4.9 | 3.0 | 14 | 14 | -- | 24 |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m. (indoors)** | | | | | | | | | | |
| 476 | 19.0 | 1.8 | 25 | 14 | 13.0 | 2.3 | 5.4 | 21 | 13 | 16 |
| 485 | 48.0 | 18.0 | 9.0 | 8.9 | 5.3 | 0.0 | 16 | 16 | 14 | 9.7 |
| 487 | 10.3 | --- | 8.8 | 10 | 5.4 | 3.9 | 14 | 14 | 15 | --- |
| **Sulfate sulfur 0.3%, molybdenum 50 p.p.m., rubidium 100 p.p.m., arsenic 5 p.p.m., and lead 5 p.p.m. (indoors)** | | | | | | | | | | |
| 478 | 0.5 | 14 | 15 | 2.3 | 9.0 | 0.0 | 14 | 18 | 13 | 14 |
| 484 | 44 | 15 | 15 | 13 | 12 | 0.0 | 15 | 16 | 14 | 20 |
| 486 | 67 | 39 | 28 | 27 | 16.8 | 0.0 | 21 | 17 | 25 | 30 |

TABLE 31

Copper-64 Level in Blood
(Experiment IV)

Counts per minute of copper-64 per milliliter of blood*

| Hours after administration of copper-64 | Trial I | | Hours after administration of copper-64 | Trial III | |
|---|---|---|---|---|---|
| | Steer 473 | Steer 474 | | Steer 498 | Steer 499 |
| | | | 0 | 0 | 0 |
| 28 | 346 | 132 | 2 | 115 | 34 |
| 33 | 362 | 186 | 4 | 154 | 50 |
| 38 | 243 | 343 | 6 | 161 | 44 |
| 43 | 324 | 200 | 8 | 206 | 89 |
| 48 | 275 | 275 | 10 | 251 | 109 |
| 55 | 288 | 203 | 12 | 305 | 177 |
| | | | 14 | 353 | 136 |
| | Trial II | | 17 | 411 | 228 |
| | | | 20 | 442 | 268 |
| 0 | 0 | 0 | 23 | 493 | 367 |
| 2 | 0 | 0 | 27 | 532 | 370 |
| 4 | 0 | 51 | 41 | 350 | 321 |
| 6 | 16 | 120 | | | |
| 8 | 38 | 97 | | Trial IV | |
| 10 | 58 | 130 | | | |
| 12 | 90 | 117 | 0 | 0 | 0 |
| 14 | 68 | 105 | 2 | 9 | 1 |
| 17 | 69 | 118 | 4 | 29 | 39 |
| 20 | 102 | 137 | 6 | 54 | 95 |
| 23 | 128 | 204 | 8 | 73 | 59 |
| 27 | 103 | 108 | 10 | 123 | 93 |
| 32 | 73 | 73 | 12 | 138 | 148 |
| 39 | 187 | 227 | 14 | 157 | 136 |
| 44 | 504 | 348 | 17 | 242 | 165 |
| | | | 20 | 259 | 189 |
| | | | 23 | 252 | 155 |
| | | | 26 | 331 | 200 |
| | | | 30 | 424 | 243 |
| | | | 34 | 400 | 268 |
| | | | 38 | 383 | 307 |
| | | | 48 | 470 | 200 |
| | | | 61 | 1188 | 542 |

* Corrected to time when copper-64 level was 5 mc.

TABLE 31 (Cont.)

Copper-64 Level in Blood
(Experiment IV)

Counts per minute of copper-64 per milliliter of blood*

| Hours after administration of copper-64 | Trial V | | Hours after administration of copper-64 | Trial VI | |
|---|---|---|---|---|---|
| | Steer 473 | Steer 474 | | Steer 498 | Steer 499 |
| 0 | 0 | 0 | 0 | 0 | 0 |
| 2 | 7 | 4 | 2 | 16 | 0 |
| 4 | 14 | 14 | 4 | 21 | 31 |
| 6 | 30 | 26 | 6 | 39 | 45 |
| 8 | 35 | 10 | 8 | 66 | 124 |
| 10 | 48 | 58 | 10 | 83 | 80 |
| 12 | 52 | 73 | 12 | 78 | 78 |
| 14 | 87 | 63 | 14 | 58 | 106 |
| 16 | 97 | 34 | 16 | 209 | 163 |
| 18 | 177 | 113 | 18 | 219 | 207 |
| 21 | 188 | 123 | 20 | 273 | 230 |
| 25 | 171 | 138 | 22 | 285 | 236 |
| 30 | 179 | 159 | 24 | 226 | 277 |
| 38 | 324 | 257 | 28 | 288 | 304 |
| 44 | 320 | 400 | 32 | 411 | 453 |
| 50 | 455 | 600 | 44 | 350 | 450 |

* Corrected to time when copper level was 10 mc.

TABLE 32

Results of Statistical Analysis of Data for Experiment I

| | Decreases in levels of copper | | | Increases in levels of molybdenum | | |
|---|---|---|---|---|---|---|
| | Milk | Serum | Liver | Milk | Serum | Liver |
| | Levels of significance | | | | | |
| Average treatment effect | | | | | | |
| 75 days | > 0.05 | >0.05 | >0.05 | 0.01 | 0.05 | > 0.05 |
| 150 days | > 0.05 | >0.05 | >0.05 | 0.01 | 0.01 | 0.01 |
| 225 days | > 0.05 | >0.05 | >0.05 | 0.01 | 0.01 | 0.01 |
| 300 days | > 0.05 | 0.05 | >0.05 | 0.05 | 0.01 | >0.05 |
| Combined average treatment effect | > 0.05 | 0.05 | 0.05 | 0.01 | 0.01 | 0.01 |
| Effect due to time | 0.01 | 0.01 | > 0.05 | 0.01 | 0.05 | >0.05 |
| Interactions of treatment components | > 0.05 | >0.05 | >0.05 | >0.05 | >0.05 | >0.05 |

TABLE 33

Results of Statistical Analysis of Data for Experiment II

| | Decreases in levels of copper | | | Increases in levels of molybdenum | | |
|---|---|---|---|---|---|---|
| | Milk | Serum | Liver | Milk | Serum | Liver |
| | Levels of significance | | | | | |
| Average treatment effect | | | | | | |
| 0 days | >0.05 | >0.05 | >0.05 | >0.05 | >0.05 | >0.05 |
| 75 days | >0.05 | >0.05 | >0.05 | >0.05 | 0.01 | 0.05 |
| 150 days | >0.05 | >0.05 | >0.05 | >0.05 | 0.01 | >0.05 |
| 225 days | >0.05 | >0.05 | >0.05 | 0.05 | 0.05 | >0.05 |
| 300 days | >0.05 | >0.05 | >0.05 | ---- | >0.05 | >0.05 |
| Combined average treatment effect | >0.05 | >0.05 | >0.05 | 0.01 | 0.01 | >0.05 |
| Effect due to time | 0.01 | 0.05 | >0.05 | >0.05 | 0.01 | 0.01 |
| Interactions of treatment components | >0.05 | >0.05 | >0.05 | >0.05 | >0.05 | >0.05 |

TABLE 34

Results of Statistical Analysis of Data for Experiment III

| | Decreases in levels of copper | | | Increases in levels of molybdenum | | |
|---|---|---|---|---|---|---|
| | Milk | Serum | Liver | Milk | Serum | Liver |
| | Levels of significance | | | | | |
| Average treatment effect | | | | | | |
| 0 days | >0.05 | >0.05 | >0.05 | > 0.05 | > 0.05 | >0.05 |
| 75 days | 0.05 | >0.05 | >0.05 | 0.01 | >0.05 | 0.05 |
| 150 days | >0.05 | >0.05 | 0.01 | 0.01 | 0.05 | 0.01 |
| 225 days | > 0.05 | >0.05 | 0.01 | 0.05 | 0.05 | 0.05 |
| 300 days | > 0.05 | >0.05 | >0.05 | >0.05 | >0.05 | >0.05 |
| Combined average treatment effect | >0.05 | 0.05 | 0.01 | 0.01 | 0.01 | 0.01 |
| Effect due to time | 0.01 | 0.01 | >0.05 | 0.01 | 0.01 | 0.01 |
| Interactions of treatment components | >0.05 | >0.05 | >0.05 | >0.05 | >0.05 | >0.05 |

TABLE 35

Results of Statistical Analysis of Data for the Combined Experiments I and II

| | Decreases in levels of copper | | | Increases in levels of molybdenum | | |
|---|---|---|---|---|---|---|
| | Milk | Serum | Liver | Milk | Serum | Liver |
| | Levels of significance | | | | | |
| Average treatment effect | 0.01 | >0.05 | >0.05 | 0.01 | 0.01 | 0.05 |
| Individual treatment components | | | | | | |
| Molybdenum | >0.05 | 0.05 | 0.01 | 0.01 | 0.01 | 0.01 |
| Sulfate | 0.01 | >0.05 | >0.05 | 0.01 | 0.01 | >0.05 |
| Time | 0.01 | 0.01 | >0.05 | 0.01 | 0.05 | >0.05 |
| Interactions of treatment components | | | | | | |
| Molybdenum + sulfate | >0.05 | >0.05 | >0.05 | 0.01 | 0.01 | >0.05 |
| Molybdenum + time | >0.05 | >0.05 | >0.05 | >0.05 | >0.05 | >0.05 |
| Sulfate + time | 0.01 | >0.05 | >0.05 | 0.05 | >0.05 | 0.05 |
| Sulfate + time + molybdenum | >0.05 | >0.05 | >0.05 | >0.05 | >0.05 | >0.05 |

TABLE 35 (cont.)

Results of Statistical Analysis of Data for the Combined Experiments I and II

| | Decreases in levels of copper | | | Increases in levels of molybdenum | | |
|---|---|---|---|---|---|---|
| | Milk | Serum | Liver | Milk | Serum | Liver |
| | Levels of significance | | | | | |
| Effect due to molybdenum | | | | | | |
| 75 days | >0.05 | >0.05 | >0.05 | 0.01 | 0.01 | 0.05 |
| 150 days | >0.05 | >0.05 | >0.05 | 0.01 | 0.01 | >0.05 |
| 225 days | >0.05 | >0.05 | 0.01 | 0.01 | 0.01 | >0.05 |
| 300 days | ---- | >0.05 | >0.05 | >0.05 | 0.01 | >0.05 |
| Effect due to sulfate | | | | | | |
| 75 days | 0.01 | >0.05 | >0.05 | >0.05 | 0.05 | 0.01 |
| 150 days | 0.01 | >0.05 | >0.05 | 0.05 | 0.01 | >0.05 |
| 225 days | 0.01 | >0.05 | >0.05 | 0.01 | 0.01 | >0.05 |
| 300 days | ---- | >0.05 | >0.05 | 0.05 | 0.01 | >0.05 |
| Interactions of molybdenum and sulfate | | | | | | |
| 75 days | >0.05 | >0.05 | >0.05 | 0.05 | 0.05 | >0.05 |
| 150 days | >0.05 | >0.05 | >0.05 | 0.01 | >0.05 | >0.05 |
| 225 days | >0.05 | >0.05 | >0.05 | 0.01 | 0.01 | >0.05 |
| 300 days | ---- | >0.05 | >0.05 | >0.05 | 0.01 | >0.05 |