Diagnosis of Copper Deficiency and Effects of Supplementation in Beef Cows

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ABSTRACT

The effects of feeding supplementary dietary copper to a herd of 400 beef cows, were studied over a two year period. In the first year of the trial, the calves showed clinical signs of copper deficiency. There was improved growth following subcutaneous injection of copper ethylenediamine tetraacetate, and the treated calves had a 2.8% increase in adjusted weaning weights. In the second year of the trial pregnant cows were fed a basal ration of bromegrass silage, barley and minerals over the winter feeding period. The feed was supplemented with copper so that half received 5.5 mg/kg of copper on a dry matter basis and half 40 mg/kg. Calving occurred in the spring and half the calves were treated with injectable copper at birth and again at 12 weeks of age. There was no evidence of copper deficiency in the calves and there was no effect of high level copper supplementation on calf birth weight, or neutrophil candidacidal activity. Susceptibility to diarrhea varied in a complex fashion; morbidity was lowest in calves born to dams fed supplementary copper and highest in calves born to supplemented dams and injected with copper at birth. The cows and calves grazed the same copper deficient pasture over the summer. The average daily gain for calves born to supplemented cows was 0.999 ± 0.010 kg/day ($x \pm$ SEM) which was significantly greater than the 0.972 ± 0.009 kg/day for calves from nonsupplemented dams ($p = 0.044$). The benefit of copper supplementation on 200 day weaning weight was estimated at 4.8 kg. Evidence of copper deficiency was seen when a herd test showed mean serum levels below 9 μmol/L and liver values below 0.09 mmol/kg wet matter.

RÉSUMÉ

Cette expérience s'étalait sur une période de deux ans et elle visait à étudier les effets d'un supplément alimentaire de cuivre, dans un troupeau de 400 vaches à boeuf. Au cours de la première année, les veaux manifestèrent les signes cliniques d'une déficience en cuivre. La croissance de ceux qui reçurent de l'acide éthylènediaminetétracétique de cuivre, par la voie sous-cutanée, s'améliora et leur poids afficha une augmentation d'environ 2,8%, au sevrage. Au cours de l'hiver de la deuxième année, les vaches gravides reçurent, comme ration de base, de l'ensilage de brome, de l'orge et des minéraux; on y ajouta du cuivre, de façon à ce qu'une moitié d'entre elles en reçoive 5,5 mg/kg, sur une base de matière sèche, et l'autre, 40 mg/kg. Les vêlages survinrent au printemps et la moitié des veaux reçurent une injection sous-cutanée d'acide éthylènediaminetétracétique de cuivre, à la naissance, et une autre, à l'âge de 12 semaines. On ne constata aucune évidence d'une déficience en cuivre, chez les veaux, et aucun effet d'un supplément riche en cuivre sur le poids des veaux, à la naissance, ou sur l'activité phagocytaire des neutrophiles, à l'endroit de *Candida* spp. La susceptibilité à l'égard de la diarrhée varia de façon complexe; on enregistra en effet la morbidité la plus faible chez les veaux nés des mères qui avaient reçu un supplément de cuivre, alors que la plus élevée affecta ceux qui, en plus, avaient reçu une injection de cuivre, à la naissance. Les vaches et les veaux passèrent l'été sur le même pâturage, déficient en cuivre. Le gain de poids quotidien moyen des veaux nés des vaches qui avaient reçu un supplément de cuivre fut de $0{,}999 \pm 0{,}010$ kg/jour ($\bar{x} \pm$ EMS), ce qui s'avéra significativement plus élevé que $0{,}972 \pm 0{,}009$ kg/jour, pour ceux des vaches témoins. L'avantage de l'utilisation d'un supplément de cuivre se traduisit par un poids plus élevé d'environ 4,8 kg, lors du sevrage, à l'âge de 200 jours. On constata l'évidence d'une déficience en cuivre, lorsqu' un test du troupeau en révéla < 9 μmol/L de sérum et $< 0{,}09$ mmol/kg de tissu hépatique frais.

INTRODUCTION

Copper deficiency is a widespread problem in western Canada. Slaughterhouse surveys indicate that as many as 29% of cattle have low liver copper levels (1). However, there are few reports of growth responses to copper supplementation in western Canada,

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Funded by Glaxo Canada Limited, Alberta Research Trust Fund and the Saskatchewan Farmlab Program.

Submitted September 19, 1988.

therefore the effects of a low copper status on growth rate are poorly understood. The present study was undertaken to determine whether injectable copper supplementation during the summer months would improve growth rates of beef calves. The effects of maternal copper status on the long-term growth of the offspring was investigated in a second trial. Our work also provides information on the interpretation of serum and liver copper levels for the diagnosis of copper deficiency.

MATERIALS AND METHODS

FARM HISTORY

The study was performed over a 2 yr period on a beef herd in Goodsoil, Saskatchewan. The cows were Charolais-Simmental crossbreeds and were bred to Simmental or Polled Hereford bulls. They were fed at the home ranch over the winter feeding period, calved in early spring and turned on to native pasture in May.

The study was initiated following the identification of a herd problem resulting in the death of 43 of 419 calves. Clinical investigation indicated the calves may have been suffering from copper deficiency as indicated by signs of poor hair coat (82 of 361 calves) and unthriftiness coupled with low liver copper concentrations. Liver samples obtained from six calves at necropsy were low in copper in five cases (0.08, 0.15, 0.30, 0.34, 0.35, 0.618 mmol/kg) and contained normal amounts of molybdenum (8.93, 5.93, 1.0, 12.40, 11.4, 10.71 μmol/kg respectively). The calves also suffered from infections, particularly viral enteritis, septicemia and pneumonia.

TRIAL 1

Prior to the start of the experiment it was decided that for the summer feeding period 256 male and female calves would be pastured on native pasture and 104 female calves on improved pasture. On May 12 all the calves were run through a chute and weighed. Jugular venous blood and liver biopsies (2) were collected from every fourteenth calf; a total of 26 calves were sampled. Every other calf was treated with 50 or a 100 mg of copper as the ethylenediamine tetraacetate (EDTA) salt (Coprin, Glaxo Canada Ltd, Ontario) depending upon whether it was under or over 100 kg in weight. The calves were then moved to their respective summer pastures. The copper treated calves on the improved pasture were readily accessible and they received a second injection of 100 mg of copper EDTA on July 11 (60 days later). All the calves were weighed at weaning in October.

TRIAL 2

This started in November. The cows were randomly allocated into two groups which were penned separately in two 28 hectare paddocks on the same quarter of land. The water source and management of the two groups was identical except for the copper status of the diet. Cows in both groups received a diet of bromegrass silage, barley and minerals formulated to meet or exceed National Research Council (3) requirements for all nutrients except copper. One group received the diet with no supplemental copper (5.5 mg/kg copper on a dry matter basis), the other group received the diet supplemented with additional copper (40 mg/kg copper).

As the cows calved in the following spring, they were moved with their calves to separate paddocks and fed unsupplemented diet. Half the calves were randomly allocated to treatment with 50 mg of copper by subcutaneous injection at one to two days of age. The cows and calves were turned out to pasture on May 15. At this time the calves were approximately 11 wk of age and copper supplemented calves received a further 50 mg or 100 mg of copper by subcutaneous injection.

The calves were weighed at birth, on May 15 and October 17. The calves were weaned after the final weighing. Samples were collected for serum and liver copper analysis on May 15 from every tenth calf. The same cattle were resampled on October 17. The disease status of the calves was monitored daily from birth until May 15. The rancher recorded all treatments for diarrhea, pneumonia and other problems. Calves that died were necropsied by the local veterinarian and abnormal tissues sent to the Western College of Veterinary Medicine for histopathological investigation.

Neutrophil candidacidal activity was measured at 48 to 72 h of age on the first ten calves in each treatment group born in the month of March. The calves were retested at 3 and 6 wk of age.

ASSAYS

Neutrophil candidacidal activity was measured by incubating the neutrophils with candida and differentially staining live and dead candida with methylene blue (4). Serum and liver copper assays and ceruloplasmin estimations were performed as previously described (5).

CALCULATIONS

Average daily gain (ADG, kg/day) was computed from the calf's birth weight (BW, kg), weaning weight (WW, kg) and the number of days from birth to weaning (D, days):

$$ADG = (WW - BW)/D$$

Adjusted weaning weights were supplied by the Federal-Provincial Record of Production (R.O.P) beef improvement program, these values are corrected to a 200 day growing period and for the age of the calf's dam. This program assumes that when the birth weight of the calf is unknown that Charolais and Simmental cows bear male calves that weigh 41.2 and 41.5 kg and female calves that weigh 38.4 and 38.5 kg respectively. If the birth weight is known and the dam is immature (under 5 yr old) adjusted birth weights are calculated; for Charolais dams aged 2, 3, 4 and over 5 yr, 2.6, 1.3, 0.3 and 0 kg for male calves and 2.2, 0.9, 0.1 and 0 kg for female calves respectively are added to the actual birth weights; for Simmental dams the corresponding factors are 2.9, 1.9, 0.8 and 0 kg for male calves and 2.8, 1.4, 0.6 and 0 kg for female calves. Average daily gain (ADG) is then computed from the birth (BWT) and weaning weights (WWT) using the formula:

$$ADG = ((WWT - BWT)/D) + (\text{Age of dam factor}/200)$$

In this formula the actual birth weight is used if known and the adjusted breed average birth weight if birth weight is not known. The age of dam factor adjusts average daily gain for calves nursed by immature dams. For calves born to dams that are 75% or more Charolais aged < 30, 31-42, 43-

54 and > 55 months old the factors for male calves are 30.3, 16.4, 3.9 and 0 kg and for female calves 24.0, 13.2, 5.0 and 0 kg respectively; corresponding factors for calves born to dams that are 75% or more Simmental are 29.0, 14.5, 5.4 and 0 kg for male calves and 21.4, 9.8, 2.1 and 0 kg for female calves. From the average daily gain adjusted 200 day weaning weights are calculated:

Adjusted weaning weight
= (ADG x 200)
+ Adjusted Birth Weight

Values for liver copper concentrations reported in the literature on a dry matter basis were converted to a wet matter basis by dividing by 3.5(6).

STATISTICS

Data were analyzed on a microcomputer after the data base was checked for accuracy against the original work sheets. Statistical calculations were performed using SYSTAT(7,8). The data and the residuals from ANOVA analyses were checked to see if they were normally distributed using normal probability plots. Studentized residuals from the ANOVA were also examined to detect outliers; in no case did the proportion of outliers exceed 3% of the data.

In trial 1 the data were first examined for a possible pasture effect on weight using a two-way ANOVA with factors for sex and pasture. Weaning weights and weight gains were then compared between copper treated and nontreated cows using a one-way ANOVA. The log linear model was used for analysis of categorical data.

In trial 2 ANOVA was performed with treatment effects for copper administration in the feed or by injection, a feed*injection interaction term was also included. Repeated measures models were used when investigating the effects of copper supplementation on neutrophil function, serum and liver copper concentrations over time. There were a large number of missing data in the repeated measures study of liver copper concentrations due to the difficulties in obtaining satisfactory liver biopsies from small calves, so a separate ANOVA was conducted on the October 17 liver copper values. Two types of analysis were performed to investigate the effects of copper supplementation on growth rate. The first analysis used ANOVA to look at treatment effects on average daily gain and adjusted weaning weights. The second used ANCOVA with weaning weight as the dependent variable and incorporated both treatment effects, variables for birth weight and age of calf at weaning (continuous variables), age of dam (categorical variable with four levels corresponding to cows 2, 3, 4 and 5+ years old), sex of calf and breed of calf (Simmental, Hereford or Charolais).

A result is described as being significant if the p value for either the effect or the treatment interaction term was < 0.05. Results are said to be nonsignificant if the p values for the effect and interaction were both >0.10. The p values in the text are for the direct effects unless otherwise stated.

RESULTS

TRIAL 1

On May 12 a sampling of 26 calves revealed serum copper concentrations of 7.5 ± 0.7 μmol/L ($\bar{x} \pm$ SEM) with 88% and 54% of values below 11 and 9 μmol/L respectively. Mean serum ceruloplasmin concentrations were 128 ± 10.2 mg/L. Liver concentrations were 0.08 ± 0.017 mmol/kg; values below 0.4 and 0.09 mmoles/kg were found in 100% and 74% of calves respectively.

There was no effect of pasture on the calves' weight change over the trial period ($p > 0.2$) or on adjusted weaning weight ($p > 0.2$) and the data for both pastures were pooled for studies of the effects of copper treatment. During the trial period 166 copper injected calves gained 177.4 ± 2.1 kg; this gain was marginally greater than the 172.1 ± 2.2 kg gain for 162 noninjected calves ($p = 0.08$). However, the Federal-Provincial R.O.P adjusted weaning weight of supplemented calves was 255.4 ± 2.4 kg which was significantly greater ($p = 0.04$) than the value of 247.1 ± 2.3 kg for nonsupplemented calves.

A total of 32 calves failed to return from pasture at weaning time, 16 were copper treated and 16 had received no copper. Sixteen of the calves that failed to return were observed dead during the trial. These deaths were equally divided between copper treated and nontreated calves.

TRIAL 2

A total of 414 calves were born to 404 cows on trial; 15 of these calves were dead at birth, seven deaths were in the copper fed group and eight in the noncopper group. Fifteen calves missed weighing at the fall round up, the distribution of these missing calves was three from the copper fed, copper injected group, four from the fed but not injected group, six from the nonfed and copper injected group and two from the nonsupplemented group; of the missing calves one in each group had been observed dead by the rancher.

During the course of the experiment there was a significant rise in serum copper ($p < 0.001$) and fall in liver copper concentrations ($p < 0.001$). There was a significant feed*injection interaction effect ($p = 0.04$) on serum copper. The highest serum copper concentrations occurred in injected calves born to unsupplemented dams (Tables I and III). As would be

TABLE I. Serum Copper Content of Calves Treated with Different Amounts of Copper at May 15 Sampling

Concentration of Copper in Dam's Feed during Pregnancy, mg/kg	Injectable Copper to Calf at Birth[a]	Number of Observations	Age, days	Serum Copper Concentration μmol/L
5.5	No	11	80.7 ± 4.1[b]	9.62 ± 0.65
5.5	Yes	8	85.9 ± 6.7	11.23 ± 0.59
40	No	11	83.8 ± 4.0	10.23 ± 0.51
40	Yes	8	79.4 ± 6.0	10.32 ± 0.34

[a]Calves were treated with 50 mg copper (Coprin, Glaxo Canada Ltd)
[b]Mean ± SEM

TABLE II. Liver Copper Content of Calves Treated with Different Amounts of Copper at May 15 Sampling

Concentration of Copper in Dam's Feed during Pregnancy, mg/kg	Injectable Copper to Calf at Birth[a]	Number of Observations	Age, days	Liver Copper Concentration mmol/kg wet matter
5.5	No	7	84.9 ± 5.1[b]	0.191 ± 0.047
5.5	Yes	6	95.1 ± 3.4	0.221 ± 0.041
40	No	9	85.2 ± 4.7	0.287 ± 0.066
40	Yes	6	86.0 ± 7.3	0.501 ± 0.140

[a]Calves were treated with 50 mg copper (Coprin, Glaxo Canada Ltd)
[b]Mean ± SEM

expected, calves born to dams supplemented with copper had significantly higher (p = 0.02) liver copper concentrations, and these stores declined significantly during the summer feeding (Tables II and IV). The repeated measures analysis did not indicate a significant effect of copper injection (n = 25, p = 0.13). Copper concentrations were higher in the livers of injected calves (Tables II and IV) and a significant injection effect (n = 34, p < 0.001) was found if liver copper data for the October 17 sampling only were analyzed. At the May 15 sampling serum copper concentrations were below 11 and 9 mol/L in 69% and 23% of calves respectively. Liver copper content was below 0.4 and 0.09 mmol/kg wet matter in 78% and 0% of calves respectively. At the October 17 sampling liver copper values below 0.4 and 0.09 mmol/kg were found in 100% and 85% of calves respectively.

There was an outbreak of diarrhea in the calves in the period prior to the calves being turned out to pasture on May 15. Coronavirus was isolated from some of the affected calves. The morbidity rates for calves that were unsupplemented, copper-supplemented by injection, supplemented in the feed, and supplemented both by injection and in the feed, were 33%, 32%, 23% and 43%. Significant treatment effects existed with morbidity rate depending on feed*injection interaction effects (p = 0.04). Eight calves died and were necropsied, two had evidence of bovine virus diarrhea infection, three had pulmonary edema, one had evidence of septicemia and no diagnosis was made in two cases.

Copper intake did not significantly affect neutrophil phagocytosis. There was a small but significant rise in phagocytic ability as the calves grew older (p = 0.006); neutrophil phagocytosis was 90.7 ± 0.7%, 89.6 ± 1.0% and 93.0 ± 0.7% at 2.5, 21 and 42 days of life respectively. Copper intake did not significantly affect neutrophil candidacidal activity. However, there was a significant fall (p < 0.001) in candidacidal activity with age of donor calf (Fig. 1).

There was no significant difference in birth weight between calves born to supplemented and nonsupplemented cows. The calves were 234 ± 18.8 days old at weaning. Calves born to supplemented cows gained 27 g more weight daily than calves born to unsupplemented cows and this difference in average daily gain was significant (p = 0.044). Injectable copper administration did not significantly affect growth (Table V). Overall weaning weights were 5.7 kg heavier in calves born to supplemented cows. The ANCOVA which allowed for the possible confounding effects of age of dam, birth weight, sex, age and breed of calf indicated the presence of a significant effect (p < 0.05) of feeding copper to the calf's dam. Overall this equation predicted 43% of the variation in weaning weights, the effects of age of dam, sex, birth weight and age of calf were all significant. Neither the effect of copper injection nor the copper feed*injection interaction effect was significant. The size of the effect of feeding copper to the dam was estimated as a benefit of 5.6 kg in calf weaning weight over that of calves born to nonsupplemented cows; this corresponds to a benefit of 4.8 kg in a 200 day weaning weight. Analysis of the Federal-Provincial R.O.P data indicated that there was a significant effect (p = 0.04) of copper supplementation of the dam on adjusted weaning weight, calves born to supplemented dams had an adjusted weaning weight of 248.9 ± 1.8 kg versus 243.0 ± 2.1 kg for calves born to nonsupplemented dams.

DISCUSSION

In trial 1 the calves showed clinical signs consistent with copper deficiency and there was a growth response to copper supplementation. In trial 2 there was no evidence of copper deficiency in unsupplemented calves; no signs of hair coat changes, lameness, swelling of the fetlock joints (9,10), impaired neutrophil candidacidal activity (11) or growth response to copper supplementation (12-14) were observed. The incidence of diarrhea in the calves did depend on the levels of copper supplementation but the relationship was complex, with the highest incidence of diarrhea in calves receiving copper both orally and by injection. These results may indicate that moderate levels of copper

TABLE III. Serum Copper Content of Calves Treated with Different Amounts of Copper at October 17 Sampling

Concentration of Copper in Dam's Feed during Pregnancy, mg/kg	Injectable Copper[a] to Calf at Birth and 11 Weeks	Number of Observations	Age, days	Serum Copper Concentration µmol/L
5.5	No	11	239.7 ± 4.1[b]	14.71 ± 0.86
5.5	Yes	7	244.6 ± 7.8	17.03 ± 0.70
40	No	11	242.8 ± 4.0	15.16 ± 0.87
40	Yes	9	238.4 ± 6.0	14.60 ± 0.77

[a]Coprin, Glaxo Canada Ltd, given at the rate of 50 mg copper to calves less than 100 kg and 100 mg to heavier calves
[b]Mean ± SEM

TABLE IV. Liver Copper Content of Calves Treated with Different Amounts of Copper at October 17 Sampling

Concentration of Copper in Dam's Feed during Pregnancy, mg/kg	Injectable Copper[a] to Calf at Birth and at 11 Weeks	Number of Observations	Age, days	Liver Copper Concentration mmol/kg wet matter
5.5	No	11	239.8 ± 4.1[b]	0.029 ± 0.006
5.5	Yes	6	240.5 ± 7.8	0.095 ± 0.010
40	No	9	242.0 ± 4.1	0.032 ± 0.006
40	Yes	8	240.6 ± 6.3	0.077 ± 0.018

[a]Coprin, Glaxo Canada Ltd, given at the rate of 50 mg copper to calves less than 100 kg and 100 mg to heavier calves
[b]Mean ± SEM

supplementation are better than the unsupplemented basal diet and that the highest levels used in this study were detrimental.

Veterinarians and ranchers would like to be able to predict when copper supplementation is likely to improve calf performance. Supplementation based on low serum copper concentrations is thought to yield unpredictable results (10) and will depend on the normal values used to interpret the data. It has been suggested that serum copper concentrations below 11 and 8.6 μmol/L be regarded as indicative of marginal and severe copper deficiency (6), while others believe that the respective values are 9 and 3 μmol/L respectively (10,15). The latter set of cutpoints is more consistent with our data since it would classify calves in the second trial as being normal and those in the first trial as being copper deficient. In fact, the finding that in the second trial copper supplemented calves had similar plasma copper concentrations to unsupplemented calves indicates that it may be normal to find a mean plasma copper concentration of 10.28 μmol/L in 11 week old Simmental calves (Table I).

Similar considerations apply to liver copper concentrations. Some (6) regard concentrations below 0.4 mmol/kg wet matter as being indicative of deficiency while others (10,15) use values of 0.09 and 0.02 mmol/kg as indices of marginal and severe deficiency respectively. A cutpoint of 0.09 mmol/kg correctly classifies trial 1 as occurring in a problem year and the May samples from trial 2 as being within the acceptable range.

Support for use of a low set of normal values can also be found in previous western Canadian reports. Four calves with confirmed copper deficiency had plasma copper concentrations of 8 μmol/L (although a subsequent herd test indicated a mean concentration of 10.60 μmol/L) and liver concentrations of 0.033 mmol/kg wet matter(16). Studies with calves fed adequate amounts of copper indicated that mean plasma copper concentrations can be below 11 but above 9 μmol/L (17).

Systematic treatment differences in liver, but not in serum, copper concentrations in trial 2 are consistent with a storage role for the liver (15). The majority of plasma copper is found in ceruloplasmin (15) and levels appear to be maintained until the animal is severely depleted and hepatic ceruloplasmin synthesis inhibited. The fall in liver copper concentrations over the summer feeding period indicates that dietary intake of copper was suboptimal. Plasma concentrations rose during this period indicating that plasma copper concentrations do not necessarily correlate well with liver concentrations; others have found similar results (5,17). This divergence is presumably because serum levels are controlled by the body at the expense of hepatic stores; serum levels fall out of the normal range as liver stores become exhausted.

The efficacy of different methods of copper supplementation is of interest to ranchers, veterinarians and regulatory agencies. Liver copper concentrations at 12 wk of age were higher in calves born to supplemented cows, suggesting that storage of copper occurred *in utero* and provided a reservoir of copper for the calf. It is interesting that copper supplementation of the dams during pregnancy gave benefits to the calves. This difference was expressed as heavier adjusted weaning weights even though the birth weight of calves born to supplemented and unsupplemented

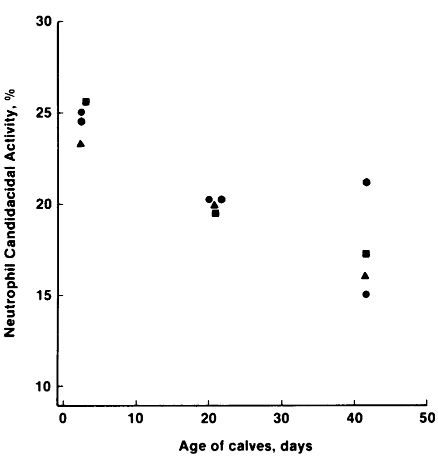

Fig. 1. Graph of mean neutrophil candidacidal activity in four groups of ten calves each. Calves born to supplemented dams and injected with copper are represented by ●, born to supplemented dams and not injected with copper by ■, born to unsupplemented dams and injected with copper by ●, born to unsupplemented dams and not injected with copper by ▲. There were no significant differences between treatment groups but there was a significant decline in candidacidal activity with age.

TABLE V. Birth Weights. Weaning Weights and Adjusted Weaning Weights of Calves According to Copper Supplementation Status

Concentration of Copper in Dam's Diet, mg/kg	5.5	5.5	40	40
Injectable Copper given to Calf	No	Yes	No	Yes
Number of calves	100	93	94	97
Birth weight, kg	42.5 ± 0.6[a]	43.4 ± 0.7	42.3 ± 0.6	43.7 ± 0.6
Weaning weight, kg	269.7 ± 3.3	273.8 ± 4.1	277.6 ± 3.7	276.9 ± 3.7
Average daily gain, kg	0.969 ± 0.012	0.976 ± 0.015	1.000 ± 0.013	0.998 ± 0.014

[a]Mean ± SEM

dams was similar. The lowest incidence of diarrhea was also seen in calves born to supplemented cows. The reasons for these findings are unknown; there have been few studies of the effects of copper status during pregnancy on the growth of the offspring. To some extent the additional hepatic copper stores may have aided growth but this cannot be the whole explanation since calves supplemented with copper by injection at birth did not perform as well as calves who received copper supplementation through the placenta. In sheep it is known that an adequate copper status is necessary for normal myelination in the central nervous system (10,18). It is possible that copper supplementation aided *in utero* development and that this facilitated long-term growth. Alternatively, copper supplementation may have facilitated growth by improving the milk yield of the dam. Beneficial effects of copper supplementation during pregnancy have obvious economic implications but need to be confirmed by other studies before recommendations can be made.

Injectable copper gave a growth response in trial 1. Our study also provides information about the distribution of injected copper. The mass of the liver as a percentage of body weight has been reported to be 2.0% in calves at term, 1.4% in steers at slaughter and 1 to 1.3% in two to three year old dairy cattle (19-21). If it is assumed that the liver of the 12 week old calf weighs 1.7% of body weight, it can be calculated that liver copper in unsupplemented trial 2 calves was about 370 μmol at 12 wk of age (trial 1 calves weighed 114 kg at 12 wk and from Table II liver copper in unsupplemented calves was 191 μmol/kg). Copper injections supply 787 μmol of copper and by July 11 injected calves had received at least 1574 μmol of copper and had significantly higher liver copper concentrations at the October 17 sampling. Only a small amount of this extra copper was present in the liver five months after the final injection because injected calves had only 259 μmol more hepatic copper stores at the October 17 sampling. Hepatic copper stores were below the 0.9 mmol/L "deficient" cut off point in many calves by the end of the trial.

In summary, in beef calves of Simmental parentage, copper deficiency was not seen until mean serum copper concentrations fell below 9 μmol/L and liver copper concentrations fell below 0.09 μmol/g wet matter. Some previous "normal" values may have been set too high to allow correct interpretation of serum and liver copper values in three month old calves. Changes in serum copper concentrations do not necessarily reflect changes in liver stores so liver copper determinations are particularly helpful in determining copper status. The decision to supplement calves can be made by balancing the amount of copper available in the liver stores and the likely availability of copper in the diet. Copper EDTA injection is beneficial in the treatment of calves showing signs of copper deficiency. Providing dams with copper at 40 mg/kg of dry matter of feed during the last trimester of pregnancy had a small, but significant, beneficial effect on growth rate of their offspring, compared to cows fed 5.5 mg/kg.

ACKNOWLEDGMENTS

The help of Mr. Paul Himmelsbach and Mr. Gerhardt Himmelsbach, Dr. Andre Olkowski, Ms. Janet Hehn and Dr. C. Kirk is gratefully acknowledged.

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