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Trace Mineral Supplementation and Ovarian and Luteal Function in Pubertal Heifers

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Cu, Co, Mn and Zn supplemented at high levels do not affect blood progesterone or estradiol concentrations or luteal phase characteristics, but supplemented heifers have fewer large follicles.

Summary

Crossbred heifers ($n=19$) were used to determine the effects of feeding Cu, Co, Mn and Zn in amounts greater than recommended by the NRC on ovarian function. Heifers were fed hay ad libitum and individually fed a control ($n=9$) or trace mineral ($n=10$) supplement for 109 days. Dry-matter intake, average daily gain, day luteal phase began, length of luteal phase, daily mean progesterone production, follicular phase estradiol production and first wave dominant follicle size were not affected by mineral supplementation. However, heifers in the treatment group had fewer large follicles during the early portion of the estrous cycle.

Introduction

Trace minerals are commonly included in rations for developing replacement heifers. Amount of trace minerals supplemented is often determined

by either established NRC requirements, feed tag recommendations or by the producer based on past experience. Recent research suggests reduced reproductive performance of two-year-old cows when trace minerals were fed in amounts above NRC recommendations (1997 Nebraska Beef Report, pp. 15-17) after calving. However, no specific physiological characteristic or mechanism was implicated as the cause of this decrease in reproductive performance. Therefore, the objective of this study was to evaluate effects of feeding greater than recommended amounts of Cu, Co, Mn and Zn on number and size of ovarian follicles during the early part of the estrous cycle, size of the largest ovarian follicle, luteal phase progesterone (P_4) production, length of the luteal phase and estradiol production during the follicular phase of the estrous cycle.

Procedure

Nineteen Angus x MARC II post-pubertal heifers (680 lbs) were randomly assigned to either a control (CON, $n = 9$) or treatment (TRT, $n = 10$) group based on weight and age. Beginning on day zero of the supplementation period, heifers were fed grass hay free choice and individually fed 4.0 lbs of dry-rolled corn and 1.3 lbs of either a control or treatment supplement. This feeding program continued through day 109. The diets were formulated to provide the mineral concentrations listed in Table 1 each day, and the nutrients needed for heifers to gain 1.5 lb/day. Weekly group hay and daily individual

Table 1. Amounts of Cu, Co, Mn and Zn in ration per day^a based on dry-matter intake and feed nutrient analysis.

Nutrient	Ration mg/hd/day		NRC Recommended
	Control	Treatment ^b	
Cu	45	269	84
Co	.92	20	.84
Mn	673	946	168
Zn	120	551	252

^aRation and NRC recommended mineral concentrations based on 18.4 lbs. of dry-matter intake.

^bDaily mineral intake provided from the supplement fed to treatment heifers: 235 mg Cu, 19.4 mg Co, 315 mg Mn, 470 mg Zn.

supplement refusals were monitored to characterize DM intake.

Liver biopsy samples were taken on days -1 and 65 of the experiment. Liver samples were analyzed for Cu, Mn and Zn concentrations (the day before treatments were imposed).

Heifers were synchronized to begin an estrous cycle on approximately day 66 of the experiment by using two injections of prostaglandin given 11 days apart. Beginning on day 66, jugular vein samples were collected twice daily (a.m. and p.m.) for the next 21 days and analyzed for concentration of progesterone (P_4). Length of the luteal phase, was determined during this time (the time in the estrus cycle when progesterone is produced due to the presence of a functional corpus luteum). The first day of the luteal phase was when concentration of P_4 in plasma exceeded .5 ng/ml and the last day of the luteal phase was when concentration of P_4 in plasma fell below

Table 2. Liver biopsy tissue analysis^a.

Group	Control Day			Treatment Day		
	-1 ^b	65	SE	-1 ^b	65	SE
Cu, mg/kg	236	146	124	432	193	113
Mn, mg/kg	7.8	7.5	.4	7.2	7.0	.3
Zn, mg/kg	150 ^c	94 ^d	7	129 ^c	94 ^d	7

^aConcentrations reported on a dry-weight basis.

^bDay -1 indicates the liver tissue samples were taken before any treatments were imposed.

^{c,d}Means within a row with different superscripts are different ($P < .01$).

Table 3. Reproductive characteristics of heifers by treatment group.

	Control	SE	Treatment	SE
Day luteal phase began	5	.5	5.5	.5
Length of luteal phase, days	15.27	.63	14.05	.59
Mean daily luteal phase P ₄ concentration, ng/ml	3.36	.28	2.91	.26
Size of largest follicle, mm	13.4	.8	14.0	.8
Number of large follicles ^a	3.2 ^a	.2	2.6 ^b	.2
Mean sample estradiol concentration, pg/ml	.58	.27	1.05	.26

^{a,b}Means within a row with different superscripts are different ($P < .10$).

.5 ng/ml. The mean daily progesterone production during the luteal phase was also determined.

Ovarian follicular development was monitored using ultrasonography daily from day 66 to day 73 and every other day from day 74 through day 87. To study the follicular phase of the estrous cycle, heifers were once again synchronized using two injections of prostaglandin given 11 days apart so estrus occurred on approximately day 96 of the experiment. On the morning of day 106, all ovarian follicles ≥ 5 mm were ablated via transvaginal follicular aspiration. Beginning approximately 12 hours post-ablation, jugular blood samples were collected every six hours for approximately 72 hours to monitor the amount of estradiol in blood. Heifers were weighed throughout the trial to monitor weight gain.

Results

Average heifer weights at the beginning of the trial were not different between groups and averaged 673 lbs

for heifers in the TRT group and 688 lbs for the CON heifers. Weights at the end of the trial for heifers in the TRT and CON groups were not different and averaged 841 lb and 850 lb, respectively. Mineral supplementation did not affect DM intake or ADG which were 18.37 lb/day, 1.54 lb/day and 18.38 lb/day, 1.49 lb/day for heifers in the CON and TRT groups, respectively.

Liver concentrations of Cu, Mn and Zn were not different between CON and TRT heifers on day -1 or 65 of the experiment (Table 2.). However, liver biopsy samples indicated a numerical decrease in Cu and Mn concentrations in both CON and TRT heifers between day -1 and day 65. In addition, there was a decrease ($P < .01$) in Zn between day -1 and day 65 of the experiment in heifers of both groups (Table 2.). The decrease in liver Cu, Mn and Zn concentrations within both groups is not fully understood. There appears to be large variations in liver mineral concentrations among individual animals as indicated by the standard error associated with the means. As the animal grows, vital

organs (liver, kidneys, spleen, heart) also increase in size. The increase in mass of the organs may have a diluting effect of mineral concentrations in the liver. Because blood volume is a function of body weight, blood volume increases as the animal increases in body size, potentially causing liver concentrations of minerals to decrease.

The beginning day of the luteal phase of the estrous cycle for the CON and TRT groups was not affected by mineral supplementation (Table 3.). Luteal phase length was not affected by mineral supplementation and was $15.3 \pm .6$ days for the CON group and $14.1 \pm .6$ days for the TRT group. Mean daily P₄ production during the luteal phase was also similar for CON and TRT heifers (Table 3.). Furthermore, between days 14 and 16, which is when pregnancy is recognized by the uterus, there was no difference in daily mean P₄ production between the CON ($4.85 \pm .55$ ng/ml) and TRT ($4.26 \pm .53$ ng/ml) heifers.

The size of the largest follicle during the early portion of the estrous cycle was not different between the CON and TRT groups. However, TRT heifers had fewer follicles ($2.6 \pm .2$) greater than 8mm compared to the CON heifers ($3.2 \pm .2$; $P < .10$). Finally, mean sample estradiol concentration in blood was not different between the CON and the TRT groups (Table 3.).

Supplementation of Cu, Co, Mn and Zn at amounts greater than NRC has been shown to decrease female reproductive performance (1997 Nebraska Beef Report, pp. 15-17). Our study indicates supplementing Cu, Co, Mn and Zn at amounts in excess of NRC recommendations does not affect the physiological characteristics at the ovarian level which are important for reproductive success in cattle. However, supplemented heifers had fewer large follicles. How this impacts subsequent reproductive performance is not understood.

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