Influence of repeated trace mineral injections during gestation on beef heifer and subsequent calf performance

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ABSTRACT: Commercial Angus heifers (n = 190; body weight (BW) = 315 ± 49.3 kg) were used to determine the effects of trace mineral injections during gestation on heifer and subsequent calf performance. Heifers received three previous subcutaneous trace mineral (Multimin 90 [**MM**]; n = 93) or sterilized physiological saline (**CON**; n = 97) injections approximately 90 d apart. These treatments were maintained and subsequent injections were given 205, 114, and 44 ± 26 d prepartum. Heifers were provided free-choice inorganic minerals. Heifer BW and body condition scores (BCS) were collected at trial initiation (296 ± 26 d prepartum) and 5- to 10-week intervals thereafter. Liver samples were collected at trial initiation, 5 and 176 ± 3 d postpartum from a subset of cows to determine trace mineral status. Milk production was assessed on 80 cow–calf pairs (40/treatment) at 71 ± 15 d postpartum. Cows were artificially inseminated (AI) 82 d postpartum and then exposed to bulls for 38 d. Data were reported from 174 calves (n = 87 calves/treatment). Calf liver samples were collected 5 and 147 ± 3 d postpartum to determine trace mineral status. Calf weaning BW was collected at 159 ± 26 d postpartum. Calf performance including calving date, birth BW, weaning BW, average daily gain (ADG), and health data were collected. Heifer BW and BCS did not differ ($P \geq 0.72$) throughout the experiment. Multimin heifers tended ($P = 0.08$) to have greater initial liver Se and tended to have decreased ($P = 0.08$) initial liver Zn compared with CON. At calving, MM cows had increased ($P \leq 0.01$) liver Cu and Se. There was no difference ($P \geq 0.47$) in Julian calving date, calving percent, or unassisted births. Calf birth BW was lesser ($P = 0.02$) for MM than CON calves, and MM calves had greater ($P = 0.03$) liver Cu concentrations at birth than CON calves. Despite MM cows having increased ($P < 0.01$) milk production, calf weaning BW and ADG were not different ($P \geq 0.87$). In addition, calf morbidity and mortality were not different ($P \geq 0.43$) between treatments. Calf mineral status was not different ($P \geq 0.57$) at the time of weaning regardless of treatment; however, MM cows had decreased ($P = 0.03$) liver Zn. Multimin cows had decreased ($P = 0.05$) AI pregnancy rates, yet there was no difference ($P = 0.34$) in overall pregnancy rate. Supplementing an injectable trace mineral during heifer development and gestation increased cow milk production and resulted in decreased AI pregnancy rates; however, there was no effect on overall pregnancy rates or preweaning calf health or performance.

Key words: beef calf, beef cow, fetal programming, injectable trace mineral, reproduction

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INTRODUCTION

Fetal programming is complex and can be influenced by numerous factors. The concept of

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trace mineral supplementation potentially altering fetal growth and ultimately long-term health and performance of calves is a novel concept with limited research. Research has primarily focused on organic trace mineral supplementation in late gestation (Gunter et al., 2003; Jacometo et al., 2015). Although trace minerals, such as Cu, Mn, Se, and Zn, may be of particular importance during the last 2 mo of gestation as 75% of fetal growth occurs during this time, they may also be important during early fetal development when differentiation, organogenesis, vascularization, and placental growth occur (Funston et al., 2010). Alterations to maternal nutrition during early gestation may have impacts not only on future growth of the fetus but also on future performance and health of the offspring.

Injectable trace minerals may be advantageous when compared with traditional oral supplement methods in that they provide a targeted delivery of specific amounts of trace minerals to individual animals. This eliminates the variability associated with fluctuation in voluntary intake noted among cattle-provided free-choice mineral (Arthington and Swenson, 2004). Ultimately, research needs to be conducted to determine the role injectable trace mineral supplementation may play in the complex process of fetal development. The injectable trace mineral, Multimin 90 (**MM**; Multimin USA, Fort Collins, CO), is labeled for administration every 90 d in heifers, and the effects of using this injectable trace mineral every 90 d in gestating heifers are yet to be reported. Therefore, the objective of this experiment was to evaluate the effects of repeated trace mineral injections during gestation on beef heifer and subsequent calf performance.

MATERIALS AND METHODS

All experimental procedures were approved by the Institutional Animal Care and Use Committee of the University of Illinois (IACUC #16046) and followed the guidelines recommended in the Guide for the Care and Use of Agricultural Animal in Agricultural Research and Teaching (FASS, 2010).

Animals and Experimental Design

To determine the effects of repeated trace mineral injections on gestating heifer and subsequent calf performance, 190 Angus × Simmental primiparous heifers (315 ± 49.3 kg) were used. The development and reproductive performance of these heifers was previously reported by Stokes et al. (2018). Heifers received three previous subcutaneous trace mineral (MM; Multimin USA) or sterilized physiological saline (**CON**) injections approximately 90 d apart. These treatments were maintained and subsequent injections were given 205, 114, and 44 ± 26 d prepartum (Figure 1). Heifers were artificially inseminated (AI; 296 ± 26 d prepartum; November 30, 2016). All heifers were confirmed pregnant (93 MM and 97 CON heifers) by either AI (43 MM and 46 CON heifers) or clean-up bull (50 MM and 51 CON heifers).

Cattle were stratified by treatment to pastures, housed at the Dixon Springs Agricultural Center in

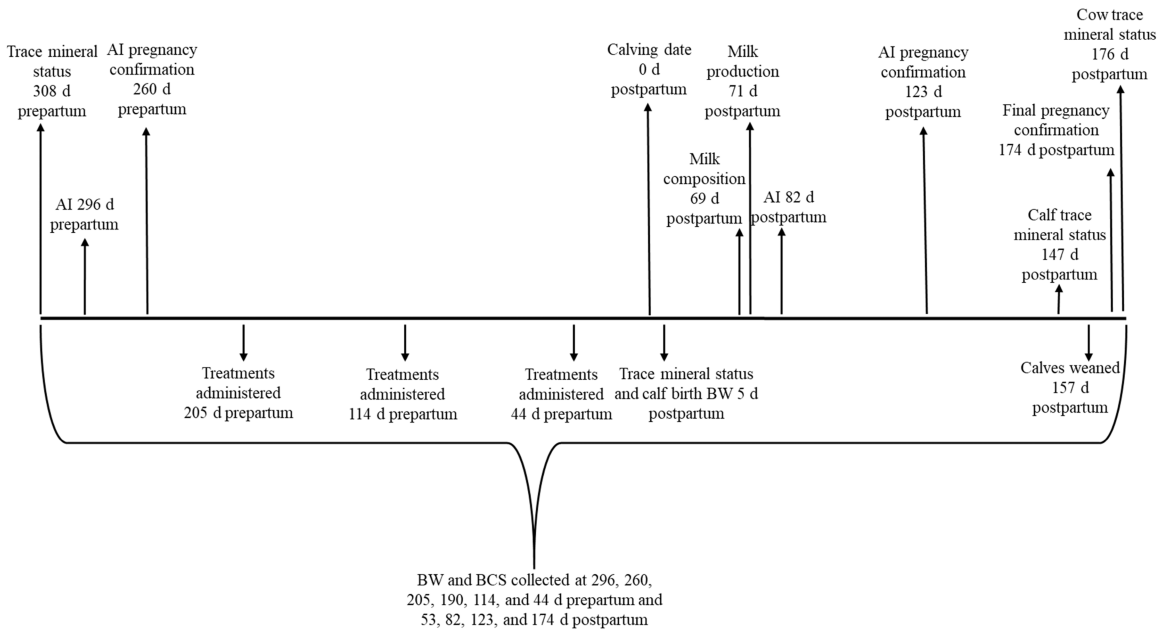

Figure 1. Experimental timeline.

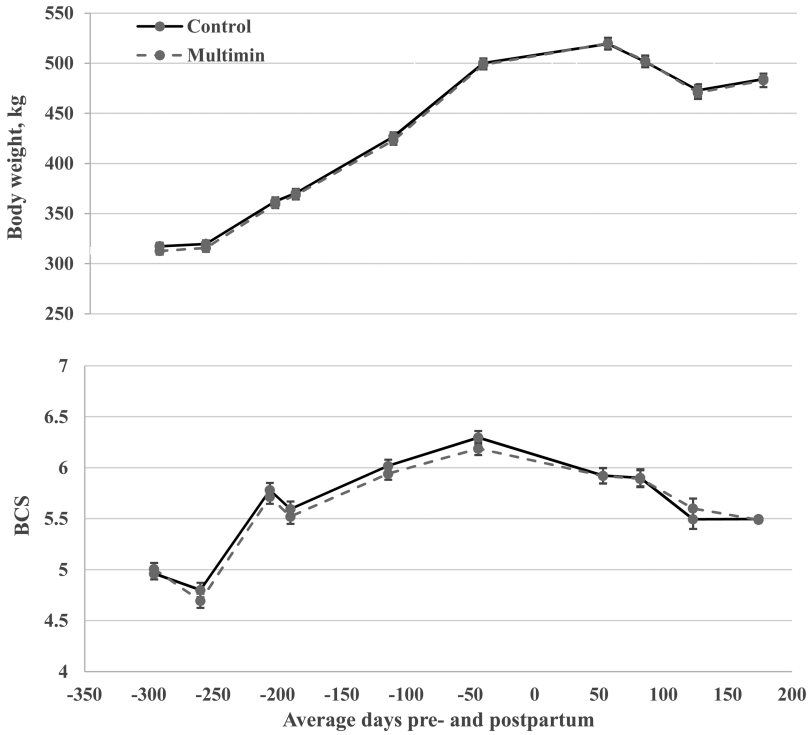

Figure 2. Effect of an injectable trace mineral (Multimin 90) on heifer BW and BCS at trial initiation (296), 260, 205, 190, 114, and 44 ± 26 d prepartum and 53, 82, 123, 174 ± 26 d postpartum. Day 0 represents average calving date. Control cattle received a sterilized saline solution, and Multimin 90 (Multimin) cattle received injectable trace mineral at 205, 114, and 44 ± 26 d prepartum. For BW, treatment by day was not significant ($P = 0.90$), and the main effects of treatment and day were significant at $P = 0.72$ and $P < 0.01$, respectively. For BCS, treatment by day was not significant ($P = 0.49$), and the main effects of treatment and day were significant at $P = 0.55$ and $P < 0.01$, respectively.

Simpson, IL, and grazed endophyte-infected fescue (*Festuca arundinacea*) and red clover pastures (*Trifolium pratense*; spring = 62% NDF, 37% ADF, and 12.3% CP; summer = 62% neutral detergent fiber (**NDF**), 35% acid detergent fiber (**ADF**), and 9.6% crude protein (CP); fall = 69% NDF, 36% ADF, and 7.5% CP). Pasture groups were rotated under the discretion of trained University of Illinois research personnel based on visual appraisal of forage availability. Cattle were supplemented with corn distillers grains (2.7 kg/heifer/d; 43% NDF, 11% ADF, 10.5% fat, and 28.4% CP) from trial initiation until 90 ± 26 d postpartum. At this time, cattle were provided a total mixed ration (**TMR**) for the remainder of the experiment consisting of corn silage, mixed grass hay, corn distillers grains, and soybean hull pellets (54% NDF, 34% ADF, 2.7% fat, and 10.7% CP). In addition, heifers were given access to free-choice inorganic trace minerals (Renaissance Nutrition, Roaring Springs, PA; 0.24% S, 21.37% Ca as calcium carbonate, 2.99% P as monocalcium phosphate, 24.5% salt, 9.35% Na, 5.84% Mg as magnesium oxide, 0.06% K, 2,214 mg/kg Fe as iron oxide, 2,000 mg/kg Mn as manganous oxide, 2,500 mg/kg Zn as zinc oxide, 1,500 mg/kg Cu as copper sulfate, 27 mg/kg Co as cobalt carbonate, 36 mg/kg I, 26 mg/kg Se as sodium selenite, 110,179 IU/kg vitamin A, 3,084 IU/kg vitamin D, and 545 IU/kg vitamin E). Mineral consumption was measured for two periods throughout the experiment, with the first period representing gestating heifers and the second period representing cow-calf pairs. Gestating heifers consumed 47.7 g/heifer/d of free-choice mineral and cow-calf pairs consumed 54.6 g/pair/d. Four cows (two CON and two MM) were removed from the trial due to death or poor performance. All analysis included cow performance data until the date they were removed from study.

Sample Collection and Analytical Procedures

Cattle body weights (BW) and body condition scores [BCS; emaciated = 1; obese = 9; as described by Wagner et al. [1988]) were collected at trial initiation (296 d prepartum), 260, 205, 190, 114, and 44 ± 26 d prepartum (Figure 2), and 53, 82, 123, 174 ± 26 d postpartum. Previously mineral status–determined (Stokes et al., 2018) heifers (38) and their subsequent calves were used for additional sampling. Liver and blood samples were collected from these heifers 8 d before the start of the experiment (308 ± 3 d prepartum), and 5 and 176 ± 3 d postpartum and from their calves 5 ± 3 d after

calving for trace mineral determination. At the time of calving, an additional 30 cow (15/treatment) and their AI sired bull calves were selected for additional blood and liver biopsies. Blood and liver biopsies were collected from calves 5 ± 3 d and 147 ± 3 d postpartum. Liver biopsy samples were collected using the method of Engle and Spears (2000) with the modification that all heifers were given an intradermal 5 mL of Lidocaine Injectable-2% (MWI Animal Health, Boise, ID) at the site of the biopsy. Following the biopsy, samples were transported to the laboratory on ice and were frozen at –20°C for subsequent trace mineral analysis. Blood was collected at the time of biopsy via jugular venipuncture into trace element serum vacuum tubes (6.0 mL; Becton, Dickinson and Company, Franklin Lakes, NJ). Blood samples were centrifuged at 1,300 × *g* for 20 min at 4°C and plasma was stored at –20°C for subsequent trace mineral analysis. Milk samples were collected at 69 ± 3 d postpartum for trace mineral analysis. These samples were collected using a method previously described by Clements et al. (2017) with the modification that cows were administered 1 mL/cow oxytocin intramuscularly (MWI Animal Health) to stimulate milk letdown. Cows were then hand milked to obtain a 50-mL sample. Following collection, samples were stored on ice and transported to the laboratory where they were centrifuged at 218 × *g* for 10 min at 4°C. The skim portion was removed and stored at 2°C for 24 h until shipped for analysis. Blood, liver, and milk samples were shipped to Michigan State University Diagnostic Center for Population and Animal Health (East Lansing, MI), and concentrations of Cu, Mn, Se, and Zn were analyzed using an Agilent 7500ce Inductively Coupled Plasma Mass Spectrometer (Agilent Technologies, Inc., Santa Clara, CA) via procedures described previously (Wahlen et al., 2005).

Milk production was assessed on 80 cow–calf pairs (40/treatment) at 71 ± 15 d postpartum via the weigh-suckle-weigh technique as described by Beal et al. (1990), with age and sex of calf equally represented across treatments. Cows were enrolled in a 7-d CO-Synch + controlled internal drug release (**CIDR**) procedure (Johnson et al., 2013) 73 ± 26 d postpartum and AI 82 ± 26 d postpartum. Sire and AI technician were stratified across treatments. Ten days following AI, cows were placed with six bulls (three bulls/pasture) that had previously passed a breeding soundness exam for a 38-d breeding season. First-service AI conception rates and overall pregnancy rates were determined at 123 and 174 ± 26 d postpartum by a trained technician via ultrasonography (Aloka 500 instrument; Hitachi Aloka Medical America, Inc., Wallingford, CT; 7.5 MHz general purpose transducer array).

Calf BW was collected using a hand scale within 48 h of birth. Bull calves were surgically castrated at birth. All calves were weighed at weaning (159 ± 26 d postpartum) and calf overall average daily gain (ADG) was calculated from birth to weaning. Before weaning, calves were vaccinated with Bovishield Gold FP5 VL5 HB (Zoetis, Florham Park, NJ), Covexin 8 (Merck Animal Health, Madison, NJ), and Pulmo-Guard MpB (AgriLabs, St. Joseph, MO). Calf health was monitored throughout the experiment by trained university farm personnel. Data were reported from 174 calves (87 calves/treatment; CON = 41 heifers and 46 steers; MM = 34 heifers and 53 steers). Twenty-three calves (11 CON and 12 MM) were removed throughout the experiment due to death or chronic illness. All analysis included calf performance data until the date they were removed from the study.

For nutrient composition analysis, feed and forage samples were collected monthly and composited and dried at 55°C for a minimum of 3 d and ground through a 1-mm screen using a Wiley Mill (Arthur H. Thomas, Philadelphia, PA). Ground samples were analyzed for CP (TruMac; LECO Corporation, St. Joseph, MI), NDF and ADF using an Ankom 200 Fiber Analyzer (Ankom Technology, Macedon, NY), and for crude fat using an Ankom XT10 fat extractor (Ankom Technology).

Statistical Analysis

Cow and calf BW, BCS, calf Julian birth date, calf ADG, milk production, and plasma, liver, and milk mineral were analyzed using the MIXED procedures of SAS (SAS Inst. Inc., Cary, NC). The model included the fixed effects of treatment and pasture. Calf parameters included the fixed effects of sire and sex. Cow BW and BCS were analyzed as repeated measures with the fixed effects of treatment, pasture, day, and the interaction between treatment and day. For all repeated variables, the unstructured covariance structure was used, as it provided the smallest Akaike information criterion. Day was the repeated effect for all repeated measures. All binary data, including calving percent, percent of unassisted births, calf morbidity and mortality, and AI and overall pregnancy rates, were analyzed using the GLIMMIX procedure of SAS. The model included the fixed effect of treatment and pasture for all binary variables. Technician and

AI sire were not significant for AI pregnancy rates and thus were removed from the model. Animal served as the experimental unit for all analyses. Treatment effects were considered significant at $P \leq 0.05$, and tendencies were noted at $0.05 < P \leq 0.10$. Means reported in tables are least squares means ± SEM.

RESULTS AND DISCUSSION

During development, Stokes et al. (2018) reported that these heifers had no difference in BW and BCS despite the increased Cu and Se status of MM-treated heifers. As CON cows were considered marginally Se deficient according to Kincaid (2000) at trial initiation, the authors' hypothesized potential differences in performance parameters should have been noted. However, BW and BCS were not different ($P \geq 0.49$; Figure 2) throughout the experiment between MM and CON cows. In other work by Stokes et al. (2017), in one of three experiments there was no difference in BW and BCS between heifers receiving an injection of MM or sterilized saline. However, in the other two experiments, one noted an increased BCS for control cattle at the time of breeding, and the other reported an increased BCS for MM-treated cattle at the time of breeding. Gadberry and Baldridge (2013) also noted no difference in BW or BCS when Angus cows were administered two dose of an injectable trace mineral, one before calving and another before breeding. Conversely, Mundell et al. (2012) reported a greater BCS increase in cows between calving and AI when treated with an injectable trace mineral 105 d before calving and again 30 d before fixed-time AI. Drawing comparisons across these experiments is challenging, as time and frequency of injectable trace mineral administration is variable, and mineral status of cattle is often not reported.

Previously, heifers enrolled in this study had received similar treatments of either an injectable trace mineral or saline at approximately 221, 319, and 401 d of age (Stokes et al., 2018). In this experiment, initial liver Cu and Mn were not different ($P \geq 0.11$; Table 1) between treatments; however, liver Se tended ($P = 0.08$) to be greater for MM cows than CON cows. Multimin-supplemented cows also tended ($P = 0.08$) to have decreased liver Zn and had decreased ($P = 0.04$) plasma Zn. Similar results were noted in these heifers during development, with MM-treated heifers having decreased liver Zn before breeding (Stokes et al., 2018). Blood measures, such as plasma Zn, may provide little information regarding trace mineral status as circulating concentrations of trace minerals are often regulated by homeostatic mechanisms until reserves become substantially depleted (Miller, 1975). In addition, adequate markers of Zn status have yet to be established as both plasma and liver concentrations can vary based on immune status and age (Kincaid, 2000). Despite these initial differences in Zn concentrations, there was no difference ($P \geq 0.81$) in liver or plasma Zn at the time of calving. There was also no difference ($P \geq 0.45$) in plasma or liver Mn at the time of calving. It is also important to note that throughout the experiment, cow Mn and Zn status remained well within adequate levels as defined by Kincaid (2000).

Plasma and liver Cu and liver Se concentrations were greater ($P \leq 0.01$) for MM cows than CON cows, and plasma Se concentrations tended ($P = 0.08$) to be greater for MM cows than CON cows. At the time of calving, liver Cu concentrations of MM cows had decreased by 28%, and CON cows had decreased liver Cu concentrations by almost 80% compared to initial status. This change in liver Cu status resulted in CON cattle being classified as deficient according to Kincaid (2000). Deficiencies in Cu can result in a variety of clinical symptoms including infertility, anemia, and suppression of immune function (Underwood and Suttle, 1999). Though dramatic, this decrease in Cu status at calving was expected as status of the cow is affected by the demand for Cu from the fetus. Small (1996) used 26 Hereford-cross multiparous cows and primparous heifers to investigate the effect parturition had on serum Cu concentration and noted that Cu concentrations were lesser (Serum Cu = 0.47 mg/ L) at parturition than at 7 d before or after calving (Serum Cu = 0.58 and 0.66 mg/L, respectively). Xin et al. (1993) used 18 multiparous Holstein cows supplemented with three levels of dietary Cu, 5.5 mg/kg of Cu, 10 mg/kg of Cu, and 20 mg/kg of Cu, and assessed the changes of Cu concentrations in the blood and liver from 8 wks prepartum to 8 wks postpartum. Liver Cu concentrations declined continuously in these cattle with the least concentration noted at parturition, with a 49% decrease in liver Cu from cattle supplemented only 5.5 mg/kg of Cu daily. In addition, as with the cows used in this experiment, supplementing additional Cu at either 10 or 20 mg/kg seemed to mitigate the drastic decrease in cow liver Cu concentrations.

At birth (5 ± 3 d of age) calf plasma Cu, Mn, Se, and Zn were not different ($P \geq 0.17$; Table 2) between treatments. In addition, liver Mn and Zn

Table 1. Influence of an injectable trace mineral supplementation on dam mineral status

	Treatment[1]			
Item	Control	MM	SEM	*P* value
Plasma mineral				
Initial[2]	*n* = 16	*n* = 22		
Cu, mg/L	0.75	0.84	0.047	0.19
Mn, µg/L	1.75	1.63	0.126	0.52
Se, µg/L	68.4	75.8	1.74	0.52
Zn, mg/L	1.04	0.90	0.045	0.04
Calving[3]	*n* = 31	*n* = 37		
Cu, mg/L	0.55	0.86	0.037	<0.01
Mn, µg/L	3.51	2.71	0.738	0.45
Se, µg/L	57.6	62.0	1.75	0.08
Zn, mg/L	0.79	0.77	0.033	0.83
Weaning[4]	*n* = 16	*n* = 22		
Cu, mg/L	0.83	0.80	0.033	0.47
Mn, µg/L	3.82	3.89	0.441	0.90
Se, µg/L	92.7	87.4	1.96	0.07
Zn, mg/L	1.11	0.99	0.036	0.03
Liver mineral, mg/kg				
Initial[2]	*n* = 16	*n* = 22		
Cu	132.3	175.3	18.18	0.11
Mn	9.98	10.03	0.258	0.91
Se	1.06	1.55	0.190	0.08
Zn	111.0	103.6	2.81	0.08
Calving[3]	*n* = 31	*n* = 37		
Cu	25.7	126.9	10.14	<0.01
Mn	9.73	9.71	0.298	0.96
Se	0.98	1.42	0.048	<0.01
Zn	120.5	123.5	8.58	0.81
Weaning[4]	*n* = 16	*n* = 22		
Cu	180.9	216.8	23.94	0.31
Mn	11.56	11.66	0.389	0.86
Se	1.63	1.60	0.082	0.81
Zn	127.3	112.9	4.43	0.03

[1]Control cattle received a sterilized saline solution, and multimin (MM) cattle received injectable trace mineral at 205, 114, and 44 ± 26 d prepartum.

[2]308 ± 3 d prepartum; samples were collected from 22 MM and 16 CON heifers.

[3]5 ± 3 d postpartum; samples were collected from 34 MM and 30 CON cows.

[4]176 ± 3 d postpartum; samples were collected from 20 MM and 13 CON cows.

were not different ($P \geq 0.78$) in calves at the time of birth. As dam Mn status was considered adequate and not different throughout the experiment, the authors hypothesized that little to no difference would be noted in calf Mn status. Though limited research has been conducted regarding Mn trace mineral supplementation and fetal development, Hansen et al. (2006) did report that calves born to heifers consuming a diet of 16.6 mg of Mn/kg of diet had decreased birth BW and exhibited varying signs of Mn deficiency. This occurred even though dams from both treatments had similar whole-blood Mn concentrations and similar serum cholesterol concentrations. These data suggest that gestating heifers require additional Mn to overcome fetal deficiencies. Cattle in this study were given access to free-choice mineral, containing Mn, and this was likely sufficient to overcome any deficiencies as severe as those reported by Hansen et al. (2006).

Foreshadowed by the difference in maternal Cu and Se status, calves from MM-supplemented dams also had increased ($P \leq 0.03$) liver Cu and Se concentrations at birth. In addition, calves from both treatments had over two times the liver Cu concentration than that of their dams. These data suggest that the calf may see an even greater benefit from Cu supplementation during late gestation as the calf appears to receive precedence over the dam for Cu accumulation and storage. Although the accumulation of fetal liver Cu has been well established in

Table 2. Influence of maternal injectable trace mineral supplementation on calf mineral status

	Treatment[1]			
Item	Control	MM	SEM	*P* value
Plasma mineral				
Birth[2]	*n* = 28	*n* = 33		
Cu, mg/L	0.60	0.61	0.035	0.92
Mn, µg/L	1.63	1.98	0.174	0.17
Se, µg/L	48.2	50.2	2.89	0.63
Zn, mg/L	0.91	0.90	0.065	0.91
Liver mineral, mg/kg				
Birth[2]	*n* =28	*n* = 33		
Cu	182.6	302.2	21.28	<0.01
Mn	9.09	9.30	0.524	0.78
Se	1.13	1.38	0.079	0.03
Zn	274.1	269.7	29.67	0.92
Weaning[3]	*n* = 15	*n* = 15		
Cu	108.2	103.8	15.77	0.85
Mn	8.77	9.02	0.698	0.66
Se	1.22	1.41	0.223	0.57
Zn	125.1	123.0	5.99	0.81

[1]Control dams received a sterilized saline solution, and multimin (MM) dams received injectable trace mineral at 205, 114, and 44 ± 26 d prepartum.

[2]5 ± 3 d postpartum; samples were collected from 33 MM and 27 CON calves.

[3]147 ± 3 d postpartum; samples were collected from 15 MM and 14 CON calves.

the literature (Gooneratne and Christensen, 1988; Graham et al., 1994), it is perhaps more important to note that calf liver Cu and Se status mimicked that of their dams. This increase in calf liver Cu and Se status may be critical for proper immune function and inflammatory response as these trace elements are a key component of an animal's health and productivity (Berry et al., 2000; Arthington et al., 2014; Genther-Schroeder and Hansen, 2015).

Final trace mineral status was determined approximately 176 d postpartum, and at this point cows had not been supplemented with an injectable trace mineral for 220 d. Likely due to the amount of time since supplementation, plasma Cu and Mn and liver Cu, Mn, and Se were not different ($P \geq 0.31$) between treatments. Plasma and liver Zn were lesser ($P = 0.03$) in MM cows than CON cows. This is consistent with previous data from these heifers, that even when supplemented, MM-treated heifers had decreased liver Zn concentrations compared to controls at the time of breeding (Stokes et al., 2018). This persistent difference in liver Zn, even after supplementation, suggests that this differences in status may be specific to this group of cattle. Interestingly, MM cows tended to have decreased ($P = 0.07$) plasma Se concentrations compared to controls at the time of weaning. Though there tended to be differences in plasma Se, both treatment groups did have adequate Se status. Pogge et al. (2012) demonstrated that within 15 d of administration, an injectable trace mineral is an effective way to improve liver trace mineral status of cattle, particularly Cu and Se. Although administration of an injectable trace mineral did also increase initial plasma Zn and Mn concentrations, by 24 h after injection both plasma minerals had returned to similar values as that of the control (Pogge et al., 2012). Though little research has been conducted regarding trace mineral status of cattle following the withdrawal or removal from an injectable trace mineral supplementation program, it is likely that differences in mineral status would be ablated 220 d after supplementation. Stockdale and Gill (2011) supplemented dairy cows with either 20, 30, 40, or 60 mg of Se from Se yeast for 6 wks and monitored blood and milk Se concentrations for 21 wks following the withdrawal from supplementation. Blood and milk Se concentrations were markedly increased by week 6 of supplementation; however by 21 wks after supplementation Se concentrations were not different. Stockdale and Gill (2011) stopped assessing Se status at this point and so it is unclear if they would have seen similar results at 25 wk after supplementation, with supplemented cows having decreased plasma Se concentrations. Despite these differences in cow mineral status at the time of weaning, calf liver mineral concentrations were not different

($P \geq 0.57$) regardless of maternal treatment. This lack of difference in calf weaning mineral status was not overly surprising as treatments were only administered to dams, and calves would have been relying on stores of trace minerals built up during gestation and milk mineral content.

Cow milk production and milk mineral composition data were collected at 71 ± 15 d postpartum and 69 ± 3 d postpartum, respectively. Dams supplemented with MM had 1.57 kg/d greater ($P < 0.01$; Table 3) 24 h milk production compared to their CON counterparts (6.13 and 4.56 kg/d for MM and CON, respectively). Supplementing injectable trace mineral increased liver Cu and Se stores of dams postpartum. These trace minerals are required for numerous biochemical processes and are key components of structural proteins (Suttle, 2010). The increased stores of trace minerals postpartum may have allowed cattle to better maintain peak lactation as observed in this experiment. In a meta-analysis, Rabiee et al. (2010) reported that organic trace minerals supplemented to dairy cows increased milk production by 0.93 kg/d. Contrastingly, Machado et al. (2013) administered Holstein dairy cows a trace mineral injection (5 mL containing 300 mg of Zn, 50 mg of Mn, 25 mg of Se, and 75 mg of copper) at 230 and 260 d of gestation and again 35 d postpartum and noted no differences in milk production. However, due to the variability in breed of cattle and type of mineral supplementation, drawing comparisons across these experiments becomes challenging. Despite differences in milk production, milk Mn and Se concentrations were not different ($P \geq 0.65$) between treatments. There was a tendency for increased ($P = 0.08$) milk Zn concentration in MM-treated cows than CON cows. Milk Cu concentrations were below detectable limits; however, this is commonly noted as milk Cu concentrations are typically as low as 0.1–0.2 mg/L (Lonnerdal et al., 1981).

Calving percent, Julian calving date, and percent unassisted births were not different ($P \geq 0.47$) between CON and MM-supplemented dams. However, calf birth BW was lesser ($P = 0.02$;

Table 3. Influence of an injectable trace mineral on cow calving, milk production, milk mineral composition, and subsequent reproduction

	Treatment[1]			
Item	Control	MM	SEM	*P* value
Calving	*n* = 87	*n* = 87		
Calving, %	91	94	—	0.47
Calving date, Julian d	266	264	2.8	0.61
Unassisted birth, %	98	97	—	0.65
Milk production,[2] kg/d	4.56	6.13	0.346	<0.01
Milk composition[3]	*n* = 31	*n* = 37		
Cu,[4] mg/L	—	—	—	—
Mn, µg/L	23.2	24.1	1.56	0.65
Se, µg/L	18.4	18.5	0.55	0.93
Zn, mg/L	4.49	5.02	0.22	0.08
Artificial insemination pregnancy rate, %	67	53	—	0.05
Overall pregnancy rate, %	96	93	—	0.34

[1]Control dams received a sterilized saline solution, and multimin (MM) dams received injectable trace mineral at 205, 114, and 44 ± 26 d prepartum.

[2]71 ± 15 d postpartum; 40 cows/treatment.

[3]69 ± 3 d postpartum.

[4]Copper milk concentrations were below detectable limits.

Table 4. Influence of maternal injectable trace mineral supplementation on calf performance and health

	Treatment[1]			
Item	Control	MM	SEM	*P* value
Birth BW, kg	30.5	28.7	0.55	0.02
Weaning BW, kg	164.6	163.9	2.78	0.87
ADG, kg	0.81	0.81	0.015	0.90
Morbidity, %	9.4	6.0	—	0.43
Mortality, %	10.5	11.7	—	0.80

[1]Control dams received a sterilized saline solution, and multimin (MM) dams received injectable trace mineral at 205, 114, and 44 ± 26 d prepartum.

Table 4) for calves from MM-supplemented dams than their CON counterparts. Even though calf BW gain has been shown to be driven by milk production (Clutter and Nielsen, 1987) and MM dams had greater milk production, calf weaning BW and ADG were not different ($P \geq 0.87$) between treatments. Dams and calves were comingled across treatments to minimize difference due to pasture variation, and this may have allowed calves to cross-nurse between treatments, potentially explaining the lack of difference noted in calf ADG. There were also no differences ($P \geq 0.43$) between treatments in calf morbidity and mortality. Arthington et al. (2014) reported that beef calves administered an injectable trace mineral had an increased mineral status, greater humoral response to a novel antigen, and a heightened acute phase protein response when subjected to transportation stress, suggesting direct administration of an injectable trace mineral may improve calf health. Although no differences in health parameters were noted in this study, the incidence of morbidity was lower than that of previously reported years (Clements et al., 2017; morbidity = 27%), and if the health of these calves in this experiment had been more challenged perhaps differences would have been noted.

Although the effects of maternal macronutrient restriction have been extensively reviewed (Wu et al., 2006; Funston et al, 2010), the effects of maternal trace mineral supplementation on subsequent calf health and performance are minimally studied. Nutritional deficiencies in Cu, Mn, Se, and Zn impair performance and immune defense parameters, and although the exact mechanism these trace mineral work remains unclear, it is likely these trace minerals work in concert through specific mechanisms to execute a coordinated response within the animal. To the best of our knowledge, no other experiments have assessed the effect of injectable trace minerals on subsequent calf performance. However, other authors have assessed the impact of various organic and inorganic trace mineral supplementation strategies on subsequent calf health and performance (Muehlenbein et al., 2001; Gunter et al., 2003; Jacometo et al., 2015). Minimal differences have been reported with maternal organic trace mineral supplementation and subsequent calf performance (Muehlenbein et al., 2001; Gunter et al., 2003). However, Jacometo et al. (2015) did report that maternal trace mineral supplementation resulted in changes in calf gene expression that could alter the neonatal immune response. Ultimately, more research is needed to understand the complex role trace minerals may be playing in utero and how they may ultimately affect the long-term health and productivity of cattle.

The final round of treatments was administered approximately 108 d before breeding. Interestingly, MM cows had decreased ($P = 0.05$) AI pregnancy rates (53%) compared to CON cows (67%). However, there was no difference ($P = 0.34$) in overall pregnancy rates. This is in contrast to the primiparous AI pregnancy rate for these females, which was not different between treatments (Stokes et al., 2018). In other work, Stokes et al. (2017) noted increased AI pregnancy rates in Simmental × Angus heifers administered an injectable trace mineral 30 d before breeding. Mundell et al. (2012) also reported increased AI conception for cows receiving an injectable trace mineral administered 105 d before calving and again 30 d before fixed-time AI. Although the cattle used in this experiment would have received a similar injection before calving, treatments were not readministered before breeding as reported by Mundell et al. (2012). This difference in time of treatment administration combined with the use of different-timed AI protocols may help explain why variable results were noted across experiment. Vanegas et al. (2004) reported a significant decrease in first-service conception rate when administering dairy cows two doses of an injectable trace mineral, 60 d apart, prior to breeding. Liver samples for mineral analysis were not collected in these experiments to help explain these results.

Cow BCS was similar between treatments at the time milk production was assessed (CON = 5.9 and MM = 5.9) and at the time of breeding (CON = 5.9 and MM = 5.9). In addition, BCS was not changing at this time, suggesting that cattle nutritional needs were being adequately met. However, the difference in AI pregnancy rate, noted in this experiment, could have been driven by the differences noted in milk production, with MM cows having increased 24-h milk production. Both suckling behavior and milk yield can affect the activity of the hypothalamus and ovaries, ultimately extending the anestrous period after calving and inhibiting follicular development (Montiel and Ahuja, 2005). Likely, all cows were exhibiting estrus later into the breeding season and would have then been bull bred, potentially explaining the lack of differences in overall pregnancy rate. Ultimately though, estrus and suckling behavior were not assessed in this experiment, and these data may have helped explain differences noted in cow reproductive performance.

Repeated trace mineral injections during gestation resulted in an increased Cu status of both dam and calf at birth. In addition, cows supplemented

with an injectable trace mineral had increased milk production, which may have contributed to decreased AI pregnancy rates. However, overall pregnancy rates were not different regardless of treatment. Despite calves from dams treated with an injectable trace mineral had a decreased birth BW, there was no effect on calf preweaning health or performance. These data suggest that repeated trace mineral injections during gestation may increase trace mineral status and milk production; however, this resulted in no improvement in beef calf health and performance. Additional research will be required to determine how these repeated trace mineral injections during gestation may impact the health of calves when stressed or challenged, and how this change in gestational supplementation may impact long-term calf performance.

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