

Injectable trace-mineral supplementation improves sperm motility and morphology of young beef bulls¹

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ABSTRACT

This experiment evaluated effects of supplemental s.c. trace-mineral injections on growth and breeding soundness of bull calves. Weaned bulls (n = 488; initial BW = 308 ± 45 kg, initial age = 203 ± 17 d) of 2 breeds (Angus and Charolais) and originating from 13 ranches in the Great Plains were transported to a common confinement facility and assigned randomly to 2 treatments: (1) s.c. injections of trace mineral (TM) containing 15 mg/mL Cu, 5 mg/mL Se, 10 mg/mL Mn, and 60 mg/mL Zn or (2) s.c. injections of physiological saline (control). Treatments were administered at arrival (d -2 or -1; 1 mL per 45 kg of BW) and on d 90 \pm 1 (1 mL per 68 kg of BW). On d 0, bulls were stratified by treatment, breed, and ranch of origin and assigned randomly to 8 pens in which they were fed a growing diet for 225 d. The diet was formulated to promote a 1.5-kg ADG at a DMI of 2.6% of BW and to meet or exceed NRC (2000) requirements for Ca, Co, Cu, I, Mg, Mn, Na, P, K, Se, and Zn. Initial BW were measured and pretreatment blood plasma samples were collected on d-2 or -1. Breeding soundness examinations (BSE) were conducted and BW were measured at 10 and 12 mo of age (d 90 \pm 1 and d 150 \pm 1, respectively). Scrotal circumference was measured and semen samples were collected via electro-ejaculation. Motility and morphology of sperm were evaluated via light microscopy. Scrotal circumference, BW, and ADG did not differ (P > 0.16)between treatments at any time. Proportions of controland TM-treated bulls achieving minimal satisfactory BSE classifications did not differ at 10 mo of age (P = 0.98; 50 $\pm 3.8\%$ for both TM and control) or at 12 mo of age (P = 0.43; 89 and 86 \pm 2.2% for TM and control, respectively). Conversely, improved (P = 0.05) sperm motility was detected in TM-treated bulls compared with control-treated bulls at 12 mo of age; moreover, TM-treated bulls had greater (P < 0.05) improvements in sperm morphology and motility between 10 and 12 mo of age than controltreated bulls. Among bulls that failed BSE at 10 mo of age, more TM-treated bulls tended (P = 0.10) to pass BSE at 12 mo of age than control-treated bulls (98 and 94 \pm 1.6% for TM and control, respectively). Under the conditions of this experiment, sperm motility and morphology at 12 mo of age were improved in bulls treated with injectable TM at 7 and 10 mo of age compared with bulls treated with saline.

Key words: breeding soundness, sperm motility, sperm morphology, trace minerals

INTRODUCTION

Costs associated with developing young beef bulls to sexual maturity motivate breeders to minimize the number of animals that are culled for inadequate reproductive fitness before sale. To minimize risk of reproductive failure, many breeders develop young bulls in confinement during the postweaning period to ensure adequate growth and timely puberty. Bull-development diets fed in confinement are generally formulated to meet or exceed NRC (2000) or NASEM (2016) recommendations for trace minerals; however, variation in DMI between bulls may limit mineral intake, and gut-level antagonisms may limit intestinal mineral absorption.

Injectable supplemental trace minerals may be used to bypass gut-level antagonisms and to overcome poor intestinal absorption and variation in trace-mineral intake. Minerals of particular significance to sexual development of bulls include Cu, Mn, Se, and Zn. They have integral roles in either spermatogenesis (Underwood and Somers, 1969), sperm motility and morphology (Brown and Burk, 1973; Swarup and Sekhon, 1976; Hunter, 1977; Parillo et al., 2014), testicular hypertrophy (Miller and Miller, 1962), tissue repair (Machado et al., 2014), or steroid hormone synthesis (Hurley and Doane, 1989; Kumar et al., 2006).

Because of the relatively high value of breeding bulls compared with nonbreeding bulls, small improvements in breeding soundness achieved through injectable trace-mineral supplementation may result in greater net revenue for seedstock producers. Injectable trace-mineral supplements may have particular value for seedstock producers because trace-mineral status can be elevated coincident with predicted onset of puberty or the timing of a breeding soundness examination (**BSE**). The process of complete sper-

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matogenesis (spermatogonia to mature sperm) in beef and dairy bulls averages 61 d (Amann, 1962; Johnson et al., 1994). Treatment with an injectable trace-mineral source approximately 61 d before a scheduled BSE may result in a greater number of bulls with satisfactory breeding soundness. Therefore, the objective of this experiment was to evaluate the performance and breeding soundness of weaned bull calves that were developed in confinement and subject to 2 s.c. injections of either trace minerals or physiological saline.

MATERIALS AND METHODS

Animal care practices used in our experiment were reviewed and approved by the Kansas State University Animal Care and Use Committee (protocol no. 3426).

Weaned, fall-born bull calves (n = 488; initial BW = 308 ± 45 kg; initial age = 203 ± 17 d) of 2 breeds (Angus and Charolais) originating from 13 ranches in Kansas, Oklahoma, and Colorado were used in this experiment. To initiate the experiment, bull calves from all ranches were shipped to a private confinement facility near Randolph, KS. Each participating ranch was required to deliver both Angus and Charolais bulls to the central confinement facility to be used in our experiment—307 Angus bulls and 181 Charolais bulls were included in our analyses.

Bulls from individual ranches of origin arrived at the confinement facility over a 2-d time period (i.e., d - 2 or -1; deliveries were staggered such that all bulls originating from a single ranch could be processed as a group upon arrival. Each bull was identified with a unique visual ear tag and a radio-frequency transponder button (half-duplex RFID, Allflex USA Inc., Fort Worth, TX). Pretreatment blood samples (approximately 10 mL) were collected via caudal vessel puncture. Serum mineral concentrations were analyzed for Co, Cu, Fe, Mn, Mo, Se, and Zn concentrations via inductively coupled plasma spectrometry (Varian ICP, Santa Clara, CA) at the Diagnostic Center for Population and Animal Health, Michigan State University. Bulls were then weighed and assigned randomly to 2 treatments: (1) supplemental s.c. trace-mineral injections (TM; Multimin 90, Multimin North America Inc., Fort Collins, CO; Table 1) or (2) s.c. injections of physiological saline (control). Treatments were administered at initial processing (1 mL per 45 kg of BW) and on d 90 \pm 1 (1 mL per 68 kg of BW).

Following initial processing, bulls were temporarily penned (i.e., d -2 and -1) with their respective ranch-oforigin group until all processing activities were complete. The following d (i.e., d 0), bulls were stratified by treatment, breed, and ranch of origin and assigned randomly to 8 pens. All pens contained bulls from each ranch of origin and breed; treatment allocation to each pen was approximately equal. Pens (minimum area $\geq 200 \text{ m}^2$ per bull; linear bunk space = 0.46 m per bull) afforded ad libitum access to water via concrete tanks. A growing diet (Table 2) formulated to promote a 1.5-kg ADG at a DMI of 2.6% of

BW was fed for 225 d. Bunks were evaluated each morning at 0630 h, and feed was delivered once daily at 0700 h. Bunks were managed using a slick-bunk management method to minimize feed refusals. If all feed delivered to a pen was consumed, delivery at the next feeding was increased to approximately 102% of the previous delivery until intake was stable at approximately 2.6% of aggregate pen BW. Diet samples were collected from bunks weekly and frozen at -20° C. Samples were composited by weight at the conclusion of the experiment and submitted to a commercial laboratory (SDK Laboratories, Hutchinson, KS) for estimation of TDN and analysis of DM, CP, Ca, Mg, P, S, Co, Cu, Fe, I, Mn, Se, and Zn (Table 2). Diet NE values were calculated from TDN using equations proposed by the NRC (2000). Bulls were again weighed individually on d 90 \pm 1 and d 150 \pm 1 beginning at 0600 h and before daily feed delivery.

Breeding soundness examinations were conducted on d 90 \pm 1 and d 150 \pm 1 at approximately 10 mo (301 \pm 17 d) and 12 mo of age, respectively. One veterinarian measured scrotal circumference and collected semen samples using a programmable electro-ejaculator. Semen samples were assessed visually for motility and morphology via light microscopy by one experienced technician immediately following collection. Bulls with white blood cells (**WBC**) in ejaculate were presumed to have vesiculitis and were treated using a macrolide antibiotic (Draxxin; Zoetis Inc., Kalamazoo, MI) according to label directions under the supervision of a veterinarian. Breeding soundness classifications of satisfactory or unsatisfactory were assigned as specified by the Society for Theriogenology (Chenoweth et al., 1993). Briefly, bulls were considered to be unsatisfactory breeders when one or more of the following criteria was true: scrotal circumference <30 cm; sperm-cell motility <30% motile cells; sperm-cell morphology < 70% normal cells; or presence of WBC in ejaculate. Bulls that passed the BSE on d 150 were marketed via live auction on d 225 at 14 to 15 mo of age.

Bulls were monitored daily during the experiment for symptoms of respiratory disease, infectious keratoconjunctivitis, and interdigital infection. Bulls with clinical signs

Table 1. Composition of injectable trace-mineral solution administered to weaned bull calves at 7 and 10 mo of age	
ltem	Multimin 90,1 mg/mL
Cu ²	15
Mn ²	10
Se ³	5
Zn²	60
	n America (Fort Collins, CO). Implex with disodium EDTA. Iodium selenite.

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