New Concepts in Trace Mineral Supplementation of Grazing Cattle
Hydroxy Sources, Injectable Sources and Pasture Application

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Introduction

The trace mineral nutrition of grazing cattle is complicated by several factors among which are the impacts of trace mineral antagonists in grazed forage and the reliance on predictable, uniform intake of free-choice mineral supplements. Numerous options are available to assist in the management of trace mineral nutrition of grazing cattle. In recent years, significant research efforts have been focused on new technologies, which have revealed insight toward their utilization in trace mineral supplementation programs. This article will focus on three of these technologies, (1) hydroxy trace minerals, (2) injectable trace minerals, and (3) pasture application of Se.

Hydroxy Trace Minerals

One of the newest technologies to impact the trace mineral nutrition of livestock is the creation of hydroxy trace mineral sources of Cu, Zn, and Mn. These specific crystalline inorganic mineral sources are formed by covalent bonds within a crystalline matrix. This covalent bond structure differs from the ionic bonds present in common sulfate-based minerals and is more similar to the covalent bonds present in organic trace mineral sources. Whereas organic trace minerals are covalently bound to a carbon-containing ligand, hydroxy trace minerals are covalently bound to an OH group. One of the most functional characteristics of hydroxy trace minerals is their lack of solubility at neutral pH ranges, such as the rumen of healthy cattle. Dissolution of the metal occurs at lower pH values, which are common in the lower gastrointestinal tract. In addition to these nutritional characteristics, the crystalline matrix of the hydroxy trace minerals allows for exceptional handling characteristics. They are non-hydroscopic and free of dust leading to handling and mixing advantages absent in most other inorganic and organic trace mineral sources. Within the blended formulation, hydroxy trace minerals are highly stable, particularly when compared to sulfate counterparts. This stability aids in the reduction of oxidative loss of fat-soluble vitamins. Lu et al. (2010) reported a 52% reduction in vitamin E loss when broiler feeds were supplemented with 200 mg/kg of hydroxy Cu vs. Cu sulfate (Figure 1). Hydroxy trace minerals have also been suggested to have greater bioavailability compared to sulfate counterparts (Spears et al., 2004) and due to their lower solubility, they may avoid certain trace mineral antagonisms in the rumen (i.e. Cu x S x Mo; Arthington and Spears, 2007). Additional to these functional characteristics, hydroxy trace minerals are also highly concentrated allowing for greater flexibility with formulation space. For example, a

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mineral supplement containing 4,000 mg/kg Zn would require only 0.73% of the formulation space for hydroxy Zn inclusion (IntelliBond [Micronutrients]; 55% Zn), but would require 2.7% of the formulation space for organic Zn inclusion (i.e. Bioplex [Alltech] Zn; 15% Zn).

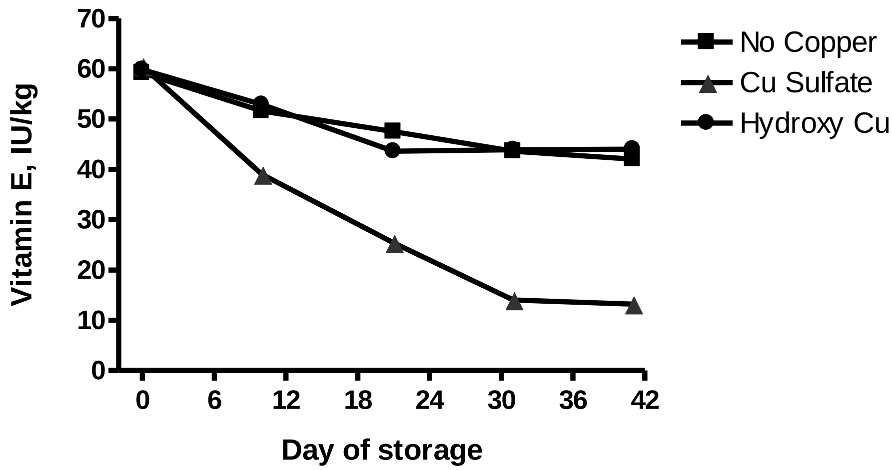

Figure 1. Vitamin E stability of broiler feeds supplemented with no Cu or 250 mg/kg of Cu from sulfate and hydroxy sources. Adapted from Lu et al. (2010). Vitamin E concentrations are less ($P < 0.05$) in Cu sulfate-supplemented feeds on each sampling day compared to the other two treatments. Average standard deviation of the mean = 3.30, 3.60, and 3.85 for No Cu, Cu sulfate, and hydroxy Cu, respectively.

Limit-Creep Feeding and the Effects of Trace Mineral Source on Voluntary Intake by Cattle

We have had a long-term interest in nutritional management applications that will optimize the trace mineral status in beef calves prior to weaning. Weaning is one of the most stressful events that a calf will encounter throughout its lifetime and trace mineral loss is a consequence of that stress. Normal calf management practices such as castration and vaccination also contribute to stress and trace mineral loss. Therefore, optimizing the trace mineral nutrition of calves, prior to weaning will help to ensure adequate trace mineral status following recovery from the stress of weaning. One area of investigation is the use of “limit-fed” creep supplements. The concept of “limit-fed” is essential in this application. Many studies have confirmed that the efficiency of added gain among creep-fed calves is poor, in fact, the poorest of all phases of the beef production system. Therefore, we sought to use limited creep feeding as a system for delivering trace minerals to pre-weaned calves. In our first study, we discovered that calves had a strong aversion to consumption of mineral-fortified creep feed, which did not exist in calves consuming the same supplement without mineral fortification (Figure 2; Moriel and Arthington, 2013). We hypothesized that the sulfate sources of minerals, particularly Cu and Zn, were disassociating in the calves’ mouths, causing a taste aversion, such as a person might experience with a metallic taste experience. This hypothesis is supported by the highly soluble nature of Cu- and Zn sulfate. Visual observation of the calves' reactions as they attempted to consume the supplements also supported our hypothesis.

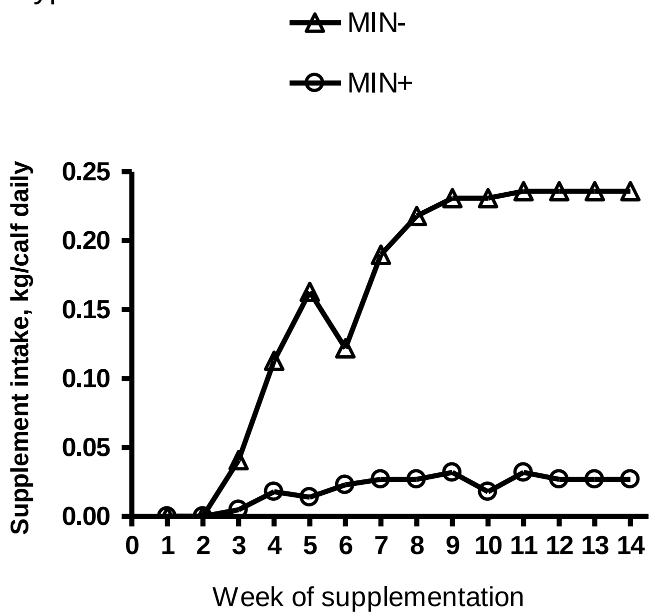

Figure 2. Voluntary intake (as-fed) of limit creep feed supplements with (MIN+; sulfate sources of Cu, Zn, and Mn) or without (MIN-) mineral fortification (pooled SEM = 0.008). Calves limited to a maximum of 230 g/calf daily. Average daily intake over the entire supplementation period was greater ($P < 0.001$) for MIN- vs. MIN+ (0.16 vs. 0.02 kg/calf daily; SEM = 0.006). Figure adapted from Moriel and Arthington (2013).

To test our hypothesis, we designed a study to evaluate the preference for intake of three experimental supplements, each containing the same base ingredient formulation, but differing by source of Cu, Zn, and Mn. This was achieved in 4 individual studies. These studies involved 8 pens of early-weaned calves (2 calves/pen) with an average age of 120 days and an average body weight of 115 kg. Each pen was provided free-choice access to concentrate and grass hay. On each study day at 1000 h, all feed was withdrawn from the pens and calves were offered three different mineral fortified supplements, for a 4-hour period. The supplements were provided in three separate feeding containers. The supplements differed by the source of Cu, Zn, and Mn, which were hydroxy- (IntelliBond; Micronutrients, Inc.), organic- (Bioplex; Alltech, Inc.), and sulfate-sources. The supplements were created using a base mixture containing 52, 46, and 2% cottonseed meal, ground corn, and salt fortified with 2,000, 750, and 3,000 mg/kg of only Zn (Experiment 1), only Cu (Experiment 2), and only Mn (Experiment 3), respectively. The last evaluation (Experiment 4) contained the same base supplement mixture fortified with a mixture of Zn, Cu, and Mn. Preferential intake was measured over 7- (Experiments 1, 2, and 3) and 14-d (Experiment 4) evaluation periods. Results are expressed as preferential intake as a % of the amount of supplement offered. These results reveal a lesser preferential intake of supplements fortified with organic sources of Cu and Zn compared to supplements fortified with hydroxy and sulfate sources of these elements (Figure 3).

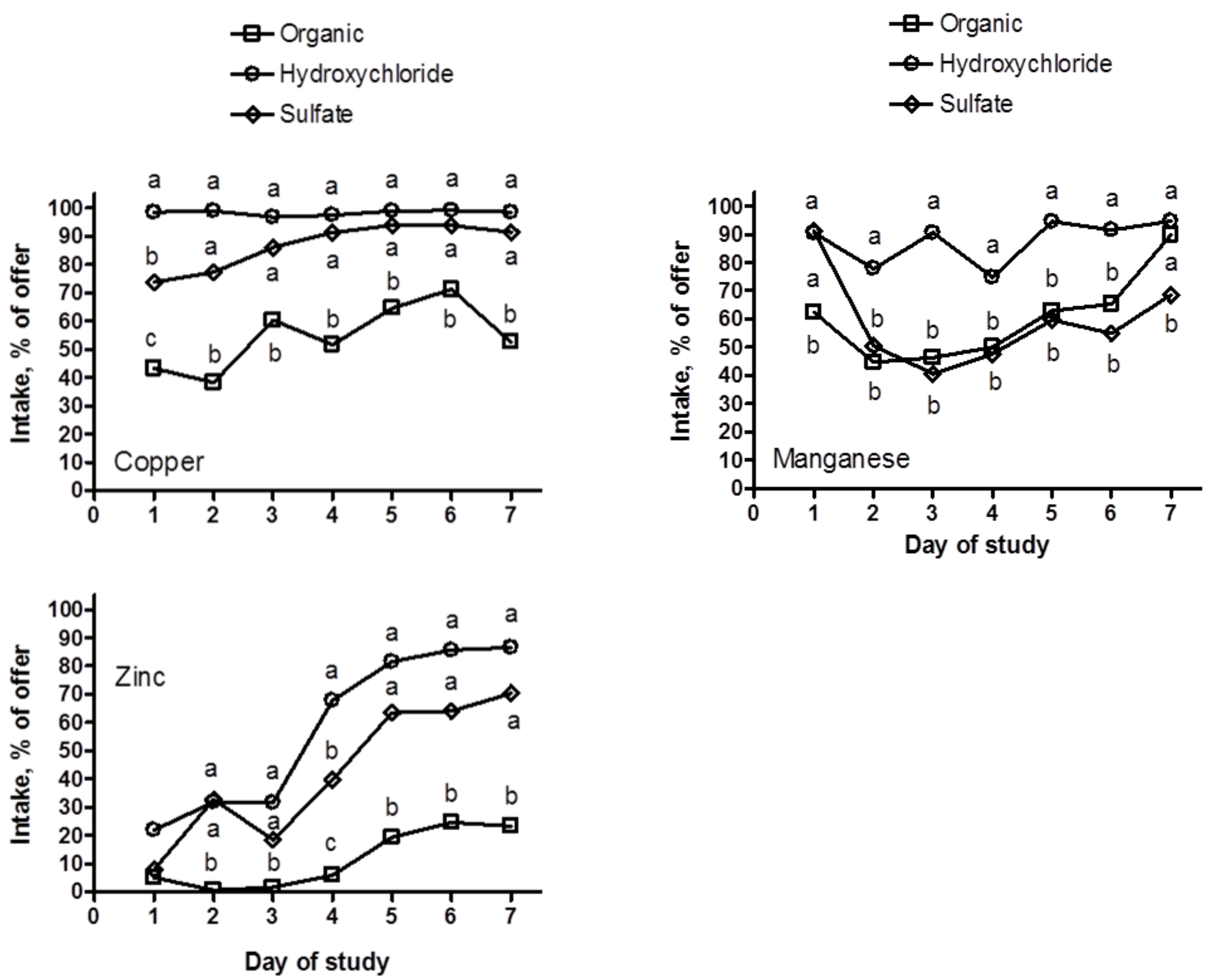

Figure 3. Preferential intake of supplements individually fortified with Cu (750 mg/kg), Zn (2,000 mg/kg), and Mn (3,000 mg/kg) from organic (Bioplex; Alltech, Inc.), hydroxy chloride (IntelliBond; Micronutrients, Inc.), or sulfate sources. Pooled SEM = 7.71, 7.90, and 7.34 for Cu, Zn, and Mn, respectively. Means with unlike superscripts within day differ ($P < 0.05$).

In the case of Mn, the preferential intake of the hydroxy source was greater than both organic and sulfate sources for all days, except 1 and 7. When all three trace minerals were combined together, preferential supplement intake differences were dramatically different with calves almost exclusively selecting the supplement fortified with the mixture of hydroxy Cu, Zn, and Mn (Figure 4).

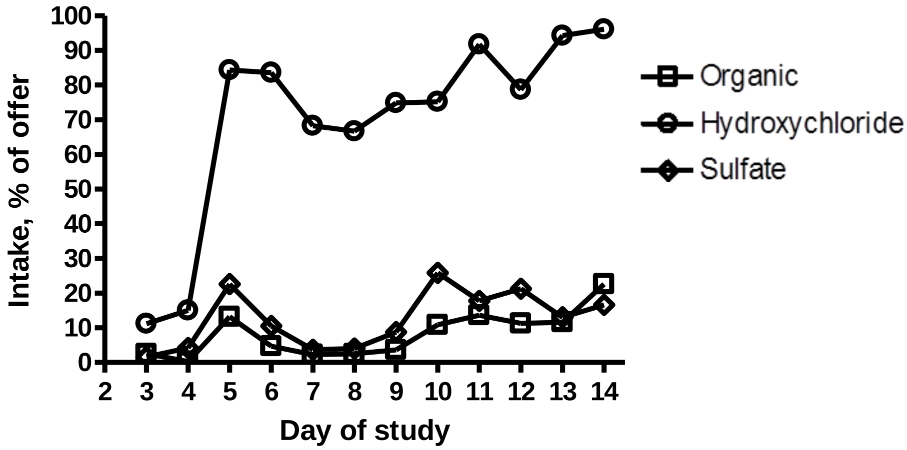

Figure 4. Preferential intake of supplements fortified with Cu (750 mg/kg), Zn (2,000 mg/kg), and Mn (3,000 mg/kg) from organic (Bioplex; Alltech, Inc.), hydroxy chloride (IntelliBond; Micronutrients, Inc.), or sulfate sources. Pooled SEM = 7.72.

With this knowledge, we designed the next limit-fed creep feeding study with three treatment formulations; (1) mineral fortification with hydroxy sources of Cu, Zn, and Mn, (2) mineral fortification with sulfate sources of Cu, Zn, and Mn, and (3) no mineral fortification (Table 1). In this study, voluntary intake increased over the 13-week supplementation period ($P < 0.001$), but there was no treatment x time interaction for voluntary creep feed intake ($P = 0.33$). Nonetheless, over the entire supplementation period, calves provided mineral-fortified creep with hydroxy sources of Cu, Zn, and Mn tended to consume more ($P = 0.10$) of the limit-creep feed offered than calves provided sulfate sources of these elements (7.4 vs. 4.9 kg; SEM = 0.97; Figure 5).

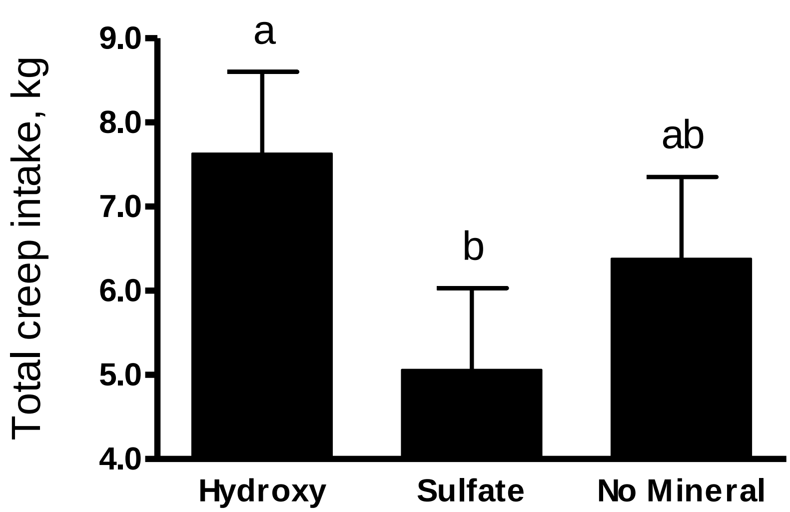

Figure 5. Total creep intake/calf over an 89-d study. Total creep intake tended ($P = 0.10$) to be greater over the entire 89-day supplementation period for calves provided mineral-fortified creep with hydroxy sources of Cu, Zn, and Mn compared to calves provided sulfate sources of these elements (7.4 vs. 4.9 kg; SEM = 0.97). Means with unlike superscripts differ ($P < 0.05$).

Table 1. Ingredient composition of limit-fed creep supplements[1]

Item	Hydroxy	Sulfate	No mineral
	-------------------------	%	-------------------------
Soybean meal	73.75	73.75	73.75
Alfalfa meal	10.00	10.00	10.00
Wheat middlings	5.08	4.87	6.40
Molasses, dried	5.00	5.00	5.00
Ca carbonate	2.50	2.50	2.50
Salt	1.25	1.25	1.25
Fat, liquid	1.00	1.00	1.00
Ca propionate	0.10	0.10	0.10
Zn sulfate	0	0.63	0
Mn oxide	0	0.50	0
Cu sulfate	0	0.31	0
Na selenite (1% suppl.)	0.08	0.08	0
Ethyleneiamine dihydroiodide (EDDI)	0.01	0.01	0
Co carbonate	0.002	0.002	0
IntelliBond[2] M	0.68	0	0
IntelliBond[2] Z	0.41	0	0
IntelliBond[2] C	0.13	0	0

[1] Diets formulated to provide 750, 2,000 and 3,000 mg/kg of Cu, Zn, and Mn, respectively. Creep supplements provided in cow exclusion areas in amounts not to exceed 230 g/calf daily. [2] Micronutrients Inc.

Liver tissue collected for biopsy from calves at the time of weaning revealed greater concentrations of Co, Cu, and Se among calves consuming mineral-fortified creep feed, irrespective of source, compared to calves consuming creep feed without mineral fortification ($P \leq 0.004$; Table 2). Although differences in liver concentrations of Cu and Co were detected among treatments, all were within normal ranges for cattle. However, calves not provided mineral-fortified creep feed were highly deficient in Se (average = 0.16 mg/kg DM), whereas calves provided mineral-fortified creep feeds were only marginally deficient (0.52 mg/kg DM).

These results provide meaningful insights to the management of trace mineral nutrition of pre-weaned calves. Research efforts in 2015 will further focus on the applications of hydroxy Cu, Zn, and Mn in both limit-fed creep feeding applications and free-choice, salt-based trace mineral supplementation systems.

Table 2. Effect of mineral fortification of limit creep feed using sulfate or hydroxy sources of Cu, Zn, and Mn on liver trace mineral concentrations of weaned calves[1]

Item	Hydroxy[2]	Sulfate[2]	No mineral	No creep	SEM	Hydroxy vs. sulfate	Mineral vs. no mineral	Creep vs. no creep
	-------------- mg/kg (DM basis) -------------							
Co	0.36	0.13	0.08	0.04	0.037	0.001	< 0.001	0.001
Cu	241	179	114	98	26.9	0.13	0.001	0.007
Fe	204	223	259	267	80.2	0.86	0.46	0.56
Mn	8.8	9.0	6.1	7.7	1.43	0.92	0.15	0.84
Mo	3.5	2.8	2.9	2.5	0.64	0.45	0.45	0.32
Se	0.60	0.43	0.18	0.14	0.120	0.31	0.004	0.02
Zn	172	171	172	153	17.5	0.99	0.54	0.21

[1]Calves were provided limit-creep feed, 3 times weekly, in amounts not exceeding 230 g/calf daily, except for the No creep treatment. Liver biopsy samples were collected at weaning following an 89 day period of limit-creep supplementation.
[2]Creep feed contained 750, 2,000 and 3,000 mg/kg of Cu, Zn, and Mn, respectively, from IntelliBond (hydroxy-sources), and Cu- and Zn-sulfate and manganous oxide (sulfate).

Injectable Trace Minerals

Injectable trace minerals (**ITM**) have been available for many years, but the technology, targeted application, and scientific assessment of efficacy has more recently been a subject of attention. An advantage of ITM, compared with traditional oral supplementation methods is the targeted delivery of a known amount of trace minerals to individual animals. This removes the variability associated with annual fluctuations in voluntary intake, which is common among cattle provided free-choice mineral formulations (Arthington and Swenson, 2004). In addition, ITM can be used within production environments that might experience difficulty managing the routine delivery of free-choice mineral mixes, such as extensive rangeland systems, seasonal grazing of mountain meadows, and seasonally flooded pastures. Further, the contribution of wildlife to the overall consumption and disappearance of free-choice mineral mixes also can cause complications in these production environments and add further value to the use of ITM. Our interest in ITM investigation originated from research findings from colleagues at other Universities which included increased mineral status (Pogge et al., 2012),increased feed efficiency (Clark et al., 2006), reduced treatments for illness (Berry et al., 2000), and reduced morbidity treatment costs (Richeson and Kegley, 2011) in stressed feeder calves. Our specific aim was to assess measures of mineral status, performance, and immune competence in beef calves receiving ITM or a control injection of sterile saline.

In the first experiment (Arthington and Havenga, 2012), we evaluated a single 7 mL subcutaneous injection of ITM (MultiMin®) containing 15, 40, and 10 mg/mL of Cu, Zn, and Mn, respectively as disodium EDTA chelates, and 5 mg/mL of Se as Na selenite or 7 mL sterile saline (Control). These treatments were administered to weaned steer calves concurrently with a single dose of a commercially-available modified live vaccine (Arthington and Havenga, 2012). All calves enrolled in the study were determined to be seronegative for the key viral pathogens targeted by the vaccine (BHV-1, BVDV-1, and BVDV-2). As a response variable, we measured serum neutralizing antibody titers following vaccination. On the day of vaccination and treatment administration, serum concentrations of Cu, Zn, Mn, and Se were similar among all steers and all values were within the sufficient range for cattle, suggesting that there were no pre-existing mineral deficiencies among the group of steers utilized in this study. By d 14 after treatment administration, steers receiving the saline control treatment experienced a decrease in serum Zn and Se concentrations and on that sampling day were less than steers receiving ITM. Neutralizing antibody concentrations to BVD-1 and 2 and BHV-1 (the primary causative pathogen for infectious bovine rhinotracheitis - IBR) increased in all steers following vaccination. Antibody titers against BHV-1 were greatest for steers receiving ITM vs. Control on day 14, 30, and 60 post-vaccination (Figure 6). Additionally, there were no visible signs of injection site inflammation which were sometimes common in earlier ITM preparations, particularly Cu-containing injectable supplements (Boila et al., 1984; Chirase et al., 1994).

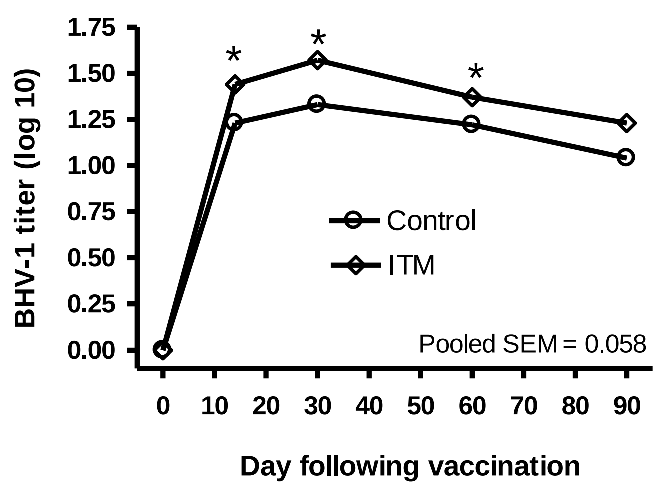

Figure 6. Bovine herpesvirus-1 (BHV-1) serum titers of calves provided a 7-mL injection of trace minerals (ITM) or 7 mL of sterile saline (Control). Seronegative calves were vaccinated on d 0. * = Values within the day and between treatments differ (P < 0.05). Data adapted from Arthington and Havenga (2012).

In the next experiment, 34 yearling heifers were randomly assigned to receive 4, 2.5 mL injections of ITM or sterile saline (Control) on d 0, 51, 83, and 127 of the study (Arthington et al., 2014). The ITM product used in Experiment 2 contained 15, 60, and 10 mg/mL of Cu, Zn, and Mn, respectively as disodium EDTA chelates, and 5 mg/mL of

Se, as Na selenite (MultiMin 90®; Multimin USA). The heifers grazed winter, stockpiled limpograss pastures and were provided free-choice, stock salt with no added trace minerals. On day 51, at the time of the second injection, all heifers were challenged with a 10-mL injection of a 25% porcine red blood cell solution to represent a novel exposure to a pathogen. The production of antibodies against the porcine red blood cells (via hemagglutination procedures) was found greater for heifers receiving ITM, compared to Control (Figure 7). Heifers receiving ITM had a 21% greater ADG compared to Control heifers (0.69 vs. 0.57 lb/d). In addition, by the end of the evaluation, heifers receiving ITM had greater liver concentrations of Se compared to control heifers (0.88 vs. 0.48 mg/kg; DM basis).

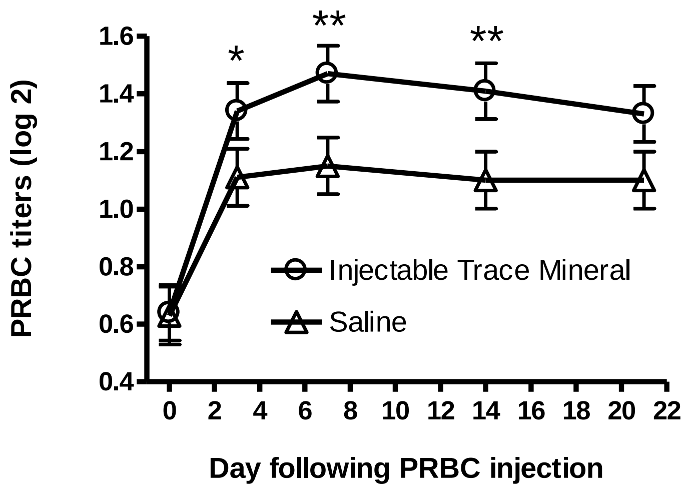

Figure 7. Effect of injectable trace minerals (ITM) on humoral immune response to porcine red blood cell (PRBC) injection. Heifers received ITM or sterile saline (2.5 mL) on d 0 and 51 and humoral immune response to PRBC was evaluated on d 51 following the second treatment administration. Treatment means differ (**P < 0.03) on d 7 and 14 and tend to differ (* P > 0.10) on d 3. Data adapted from Arthington et al. (2014).

Collectively, these findings suggest that the trace mineral status of cattle can be increased by administration of ITM. Additionally, antibody production to vaccine appears to be heightened in calves receiving ITM. These responses appear to be evident even in calves exhibiting adequate trace mineral status. It is unclear; therefore, if these observed increases in antibody titers are responses to increased trace mineral status or a priming response to the immune system. Nonetheless, this heightened immune response may be an important contributing factor to the improved measures of health and performance reported by other investigators in previous studies.

Pasture Application of Selenium

Selenium is an essential trace element for all categories of livestock. In grazing cattle, Se nutrition is complicated by the regional differences in soil Se abundance causing variation in plant Se content. Of the trace elements commonly found to be deficient in forage (i.e. Se, Cu, Co, Zn, and sometimes Mn), Se is the only trace mineral that is sometimes found in toxic concentrations in forages grown in specific regions of the US. The range between adequate and toxic concentrations is narrower for Se compared to other essential trace minerals; however, in most regions of the country, Se-deficient forage is much more common than cases of Se excess. In a survey of 253 cow/calf operations in 18 US states, over 18% were classified as marginally or severely Se deficient by blood Se parameters (Dargatz and Ross, 1996). Among the states analyzed, those located in the southeast region had the greatest percentage of operations classified as marginally or severely Se deficient (35.8%). A complicating factor impacting Se supplementation is the FDA control over maximum Se fortification of free-choice cattle mineral supplements, which limits Se supplementation to a level not exceeding 3 mg/head daily (21CFR573.920 rev. April 1, 2014). In almost all situations, this upper limit is sufficient to supply adequate Se nutrition to grazing cattle; however, this assumes a consistent intake at the target level for which the free-choice supplement was formulated. Unfortunately, we know that there are significant and often dramatic fluctuations in free-choice intake of salt-based mineral supplements. During periods of reduced voluntary intake, the potential occurrence of Se deficiency becomes a concern. This is further accentuated in scenarios involving high-S diets (i.e. > 0.30 % S; DM basis), which is a major antagonist impacting Se metabolism.

One potential method for addressing Se nutrition in grazing cattle is the implementation of pasture Se applications with the intent of increasing plant Se content and thus the Se status of cattle grazing these forages. In Florida, spraying bermudagrass with Na selenate at Se application ranges of 120 to 480 g/ha resulted in substantial increases in forage Se content by 2 wk after application, decreasing rapidly by 12 wk post-application (Table 3; Valle et al., 1993).

Table 3. Average forage Se concentrations (mg/kg; DM basis) at different weeks after spraying with Na selenate[1]

	Weeks after spraying Na selenate				
Se application rate, g/ha	2	4	6	12	18
0	1.4[a]	0.9[a]	1.5[a]	0.5[a]	0.4[a]
24	2.9[a]	2.7[a]	2.3[a]	0.7[b]	0.8[b]
120	12.8[a]	6.3[b]	4.8[bc]	0.5[c]	0.6[b]
240	26.1[a]	15.5[b]	11.9[b]	0.8[c]	1.0[b]
480	51.5[a]	28.2[b]	25.7[b]	0.7[c]	0.7[b]

[1]Data adapted from Valle et al. (1993). Means are based on 4 replicates per treatment.
[2]Means with unlike superscripts within each row differ ($P < 0.05$).

Selenium from selenate sources appears to be much more available for plant uptake compared to selenite sources (Archer, 1983). Feeding forages grown on Se-fertilized hay fields impacts both Se status and performance of grazing cattle. In one study (Hall et al., 2013), weaned Angus-type calves were fed Se-fertilized alfalfa hay over a 7-week period. Alfalfa hay was grown on fields receiving applications of Na selenate in amounts providing 0, 23, 45, or 90 g Se/ha. These application rates resulted in a linear (R^2 = 0.997) response for Se application rate and subsequent Se content of alfalfa hay harvested 40 d after Se application (Figure 8; Inset A). In addition, calves consuming these hay treatments (approximately 2.5% BW daily) experienced a linear (R^2 = 0.979) increase in whole blood Se concentrations as Se application rate (and Se content of hay) increased (Figure 8; Inset B).

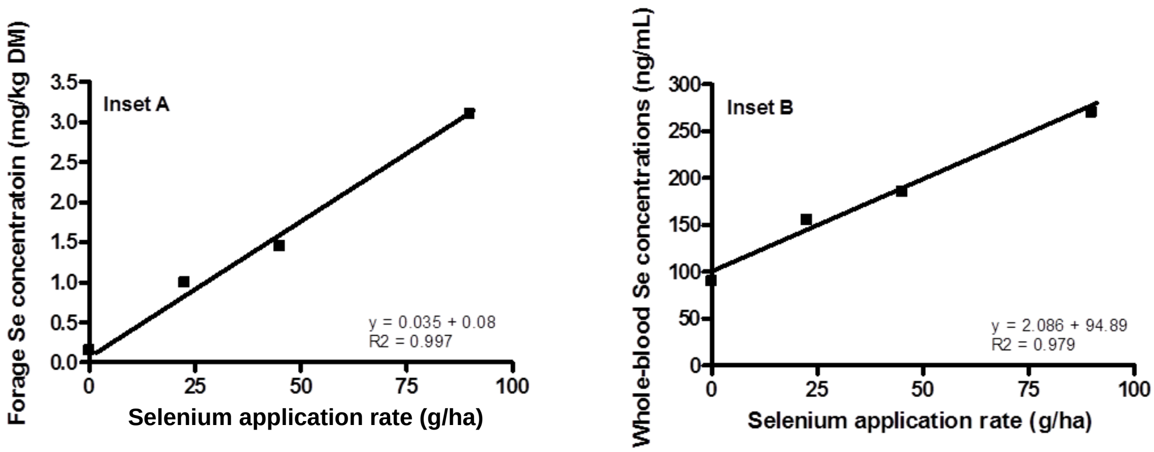

Figure 8. Effects of Na selenate application to alfalfa hay fields on subsequent forage Se content (A) and Se status (B) of calves consuming the hay. Data adapted from Hall et al. (2013).

In a recent study at the UF/IFAS, Range Cattle REC, we produced a high-Se hay crop by spraying a Jiggs bermudagrass hayfield with Na selenate at a rate of 257 g Se/ha. Selenium content of hay, harvested 8 wk after Na selenate application, was greater for Se-treated vs. control pastures (7.73 ± 1.81 vs. 0.07 ± 0.04 mg/kg DM; $P < 0.001$). In a subsequent study, this hay crop was fed to weaned calves and Se status was evaluated over a 42-d study. Calves were stratified by initial BW and randomly assigned to treatments including high-Se hay, low-Se hay + supplemental Na selenite, or No supplemental Se (n = 14, 14, and 4 calves, respectively). Calves were housed in drylot pens (2 calves/pen; 7, 7, and 2 pens per treatment). A pair-feeding design was utilized, whereas each pen of high-Se hay calves was paired to a pen of Na selenite - supplemented calves. Calves assigned to the high-Se hay treatment were provided ground, high-Se hay for a 4 h period each morning. Pen DMI was calculated and total daily Se intake/pen was estimated. Each Na selenite paired pen was then provided the same daily amount of Se via Na selenite hand-mixed into a limit-fed grain supplement. Therefore, each pen of calves receiving high-Se hay had a paired partner pen of calves receiving the same amount of Se via Na selenite. Liver Se concentrations remained unchanged for the negative control calves receiving no supplemental Se over the 42-d feeding period, but they were increased ($P < 0.001$) in calves receiving both high-Se hay and Na selenite treatments. Calves receiving high-Se hay had greater ($P < 0.05$) liver Se concentrations on d 21 and 42 than calves receiving Na selenite (Figure 9).

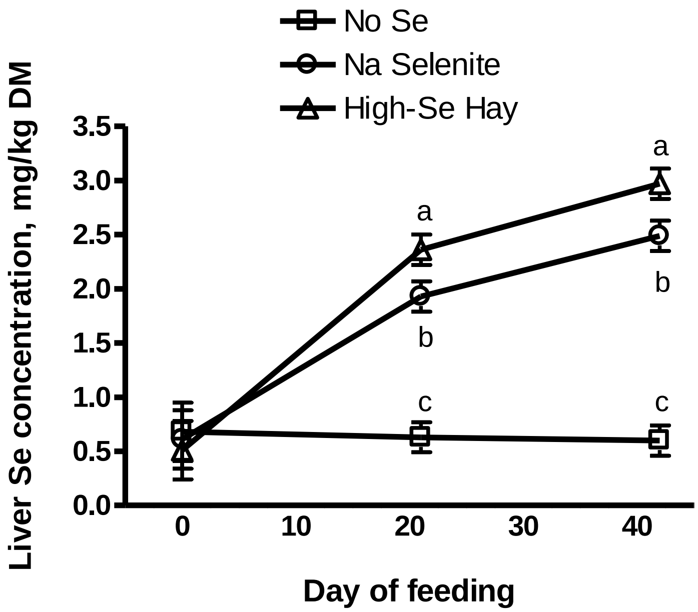

Figure 9. Liver Se concentrations among calves offered high-Se hay or a Na selenite supplement. Basal diet contained 0.6 mg Se daily (No Se treatment). Calves fed high-Se hay and the Na selenite supplement were pair fed to control overall daily Se intake (average 2.8 mg Se/d). [a,b,c] Means differ within day; $P < 0.05$.

Interestingly, this difference was attributed only to the paired pens consuming < 3 mg Se daily (Figure 10). From these initial data, we hypothesize that there is a differential availability of Se in forage vs. inorganic sources dependent upon the total daily intake with a critical point of approximately 3 mg/d in beef calves. We are currently examining these data further in both periparturient cows and calves.

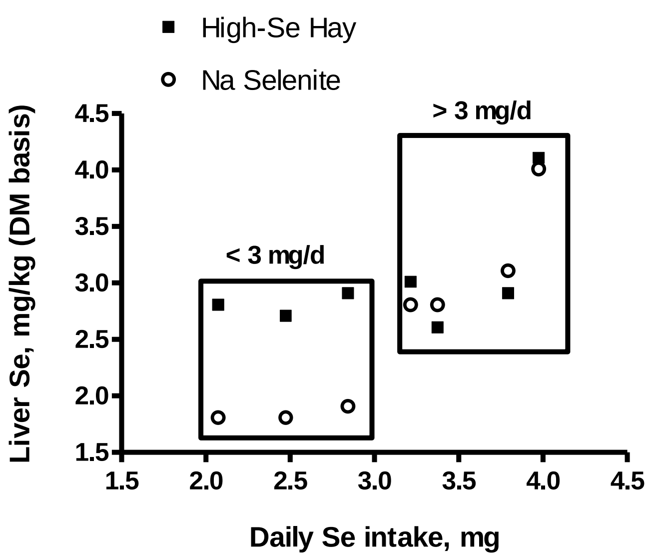

Figure 10. Liver Se concentrations (d 42) among pair-fed calves calves. X-axis denotes average daily Se intake (mg/d) among each pair-fed calf group.

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SESSION NOTES