

The Effect of Trace Mineral Source and Concentration on Mineral Solubility in the Rumen and Diet Digestibility

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Summary and Implications

Supplementing the trace minerals copper (Cu), manganese (Mn), and zinc (Zn) in the metal hydroxyl form does not impact dry matter (DM) digestibility, while sulfate-bound trace minerals decreased DM digestibility. Metal hydroxy trace minerals solubilize in the pH found in the abomasum, enabling absorption, suggesting that these minerals solubilize only in the abomasum and will not inhibit digestion. Trace minerals present in the basal diet prior to supplementation appear to be adequate for proper function of rumen microbes.

Introduction

Rumen microorganisms require trace minerals for proper function. However most research has shown that microorganism requirements for Cu, Mn and Zn are minor, and much lower than those typically provided by ruminant diets. However, *in vitro* data also suggest that relatively small concentrations of Cu, Mn and Zn can impact fiber digestion. The previously mentioned research suggests that rumen microbes can maintain proper function using trace minerals found in the basal diet, without supplementation. However, supplementation may create a rumen environment with trace mineral concentrations that are in excess of microbe needs, perhaps impacting their ability to function. The solubility of trace minerals can differ depending on the source, and it is likely that only ruminally soluble minerals will be available to the microbes for use. Amino acid bound Zn sources are more soluble in the rumen than inorganic sources while tetrabasic or metal-hydroxy bound sources are less soluble in the pH range typically found in the rumen. IntellibondC (Cu), IntellibondM (Mn), and IntellibondZ (Zn) are metal hydroxy-bound mineral sources (Micronutrients, Inc) that should be less soluble in the rumen, and then solubilize at a lower pH, as found in the abomasum and early small intestine, where absorption of these trace minerals occurs. The low rumen solubility of these sources may prevent negative impacts of trace mineral supplementation on rumen microbe function while still remaining available to the animal for absorption. The objective of this experiment was to compare the impact of sulfate and metal hydroxy trace mineral sources at different inclusion rates on dry matter digestibility, neutral detergent fiber digestibility, and solubility of Cu, Mn and Zn in the rumen and abomasum of cattle on diets formulated for a lactating dairy cow.

Materials and Methods

Five ruminally-fistulated steers were used in a 5 x 5 Latin square experimental design. Steers were fed a corn-silage based diet twice daily, formulated to meet the needs of a high-producing lactating dairy cow and allowed to consume feed on an *ad libitum* basis. There were five dietary treatments: Control (CON): no supplemental Cu, Mn or Zn; Low Sulfate (LS): 5 ppm Cu from CuSO₄, 15 ppm Mn from MnSO₄, and 30 ppm Zn from ZnSO₄; Low Metal-hydroxy (LMH): 5 ppm Cu from tribasic copper chloride (IntellibondC), 15 ppm Mn from tribasic manganese chloride (IntellibondM), and 30 ppm Zn from tribasic zinc chloride (IntellibondZ); High Sulfate (HS): 25 ppm Cu from CuSO₄, 60 ppm Mn from MnSO₄, and 120 ppm Zn from ZnSO₄; and Low Metal-hydroxy (HMH): 25 ppm Cu from tribasic copper chloride (IntellibondC), 60 ppm Mn from tribasic manganese chloride (IntellibondM), and 120 ppm Zn from tribasic zinc chloride (IntellibondZ). Steers were adapted to a common diet, similar to the final diet, for 21 d prior to the beginning of the first period. Each period was 12 days long, to allow for 10 d to adapt to the period diet, followed by insertion of Dacron bags on d 11.

Dry matter (DM) disappearance was measured using Dacron bags with small pores. A total mixed ration (TMR) sample from the CON treatment was dried and ground through a 2 mm screen in a Wiley mill, and approximately 4 g of TMR was added to each bag and the bags were heat-sealed. All bags (a total of 24) were added at 0 h, and 6 bags were removed at each of 4 timepoints, after 6 h, 12 h, 24 h, and 36 h of digestion. Once removed, bags were immediately immersed in ice water to stop fermentation, and then transported back to the laboratory on ice and frozen at -20°C. Bags were washed, dried, then weighed to determine DM disappearance. Thirty-six hours post insertion, rumen contents were sampled via a suction strainer. Rumen fluid samples were separated into soluble and insoluble mineral fractions by centrifugation, and the soluble portion was analyzed for trace mineral content. The insoluble mineral fraction (pellet) was resuspended with 3 mL of the soluble fraction and subjected to simulated abomasal digestion with hydrochloric acid and pepsin at a pH of approximately 2.5, at 102°F for one hour. After digestion, soluble and insoluble portions were separated by centrifugation, and the abomasal soluble fraction was analyzed for trace mineral content. Neutral detergent fiber analysis of substrate remaining in Dacron bags at each time point was completed using an ANKOM fiber analyzer. Total mixed ration samples and residual feeds were dried, ground and composited by period and analyzed for trace mineral content.

Dry matter and NDF disappearance and rates, and DM intakes were analyzed using the MIXED procedure of SAS including the random effect of period and steer, fixed effects of treatment and the repeated effect of hour (SAS Institute Inc., Cary, NC). Rumen fluid trace mineral solubilities were also analyzed using the MIXED procedure of SAS including the random effects of period and steer, and the fixed effect of treatment. Individual degree of contrasts included: CON vs. all trace mineral supplemented treatments, Metal hydroxy bound trace minerals vs. sulfate bound trace minerals, CON vs. Sulfate bound trace mineral treatments, and CON vs. Metal Hydroxy bound trace mineral treatments.

Results and Discussion

Dry matter intake. There were no differences in dry matter intake due to treatment ($P = 0.66$; Table 1). No contrasts either between CON and supplemental treatments, or between trace mineral sources impacted dry matter intake ($P > 0.15$). Daily trace mineral intakes were not different between LS and LMH, or HS and HMH treatments for Cu, Mn or Zn intakes ($P > 0.30$).

Dry matter disappearance. Dry matter disappearance of the CON treatment tended to be greater than all other supplemental trace mineral treatments ($P = 0.06$; Table 1). The addition of sulfate trace minerals decreased dry matter disappearance when compared to CON ($P = 0.03$), but the addition of metal-hydroxy trace minerals did not impact DM disappearance compared with the CON ($P = 0.18$). There was no difference between the sulfate and metal-hydroxy trace mineral supplemented treatments in dry matter disappearance ($P = 0.32$). Dry matter disappearance results suggest that the addition of supplemental trace minerals from sulfate sources decreases DM disappearance. However there was no difference in overall dry matter disappearance from metal-hydroxy trace minerals when compared to controls. This result implies that the differences between CON and trace mineral supplemented treatments were mediated through the differences between sulfate-supplemented treatments and the control treatments. Sulfate-bound trace minerals negatively impact dry matter disappearance while metal hydroxy trace minerals do not interfere with dry matter disappearance.

Neutral detergent fiber. Neutral detergent fiber percent disappearance was not impacted by treatment ($P = 0.70$; Table 1). There was no difference between sulfate bound and metal hydroxy bound trace minerals on NDF disappearance ($P = 0.90$). The overall rate of NDF disappearance was not impacted by treatment ($P = 0.93$). The lack of differences due to treatment in the overall NDF disappearance indicates that trace minerals do not change total NDF disappearance. However, differences in the rate of disappearance over time were affected by treatment. Trace minerals supplemented at high concentrations may change the rate at which rumen microbes digest fiber; as between hours 6 and 12 the NDF disappearance rate for HS

and HMH was lower than other treatments ($P < 0.10$). Previous work has suggested that some trace minerals, including Zn, may bind to the exterior of fiber particles, or to the exterior of microbes, inhibiting bacterial attachment and changing fiber digestion. These results may indicate that some of the trace minerals may bind to NDF particles, changing the efficiency of bacterial digestion, especially when supplemented at high concentrations.

Ruminal and abomasal solubility. Metal-hydroxy bound trace minerals are not soluble in the high pH of the rumen, but become soluble in the low pH of the abomasum. This is reflected in the results of the Cu and Mn analysis of the rumen fluid. The CON diet did not contain any supplemental Cu, Mn and Zn, and as such had the lowest concentration of both Cu and Mn. In both rumen soluble Cu and Mn, the LMH treatments were not different from CON treatment, despite the LMH diet containing supplemental Cu and Mn. Rumen soluble Cu concentrations for HMH were not different from LS and were lower than HS, again supporting the previous research stating that metal-hydroxy minerals are not soluble at the pH found in the rumen. Rumen soluble Mn concentrations for HMH were higher than LS, unlike Cu, but it was less than HS, so they have lower solubility than sulfate bound trace minerals.

Rumen soluble Zn followed a different trend than Cu and Mn. While there was no difference between CON, LS, LMH or HS, HMH Zn concentration was greater than the other treatments, suggesting that at high trace mineral concentrations metal-hydroxy bound Zn may be more soluble. These results were unexpected as metal hydroxy bound Zn was expected to be less soluble in the rumen than sulfate trace minerals.

The insoluble pelleted portion of the rumen fluid sample after ultra-centrifugation was resuspended with the supernatant, and digested with acid to evaluate the solubility of trace mineral sources in the acidic environment of the abomasum. There were no differences due to treatment in abomasal soluble Cu. However, the higher dietary concentration treatments, HS and HMH had greater Cu concentrations than other treatments, and were not different, suggesting that both sulfate bound and metal hydroxy bound trace minerals are equally soluble in the abomasum.

Abomasal Mn CON concentrations were lowest, while there was no difference between LS and LMH, in contrast to the rumen soluble Mn, where LMH was lower than LS. This suggests that in the pH range of the abomasum, Mn from the LMH becomes as soluble as Mn from LS. There was a difference between the HS and the HMH abomasal soluble Mn concentrations, which was similar to the rumen soluble concentration.

Soluble Zn concentrations in the abomasum were lowest in the CON treatment. Concentrations were greater for the low trace mineral supplemented dietary treatments, but not different between LS and LMH. Both high supplemented treatments had the greatest Zn concentrations,

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though HMH Zn concentrations remained higher than HS, as previously shown in the rumen.

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Table 1. Effect of trace mineral concentration and source on fiber digestion and trace mineral solubility.

	Treatments					SEM	Trt	Contrasts ¹
	CON	LS	LMH	HS	HMH			
DMI, lbs	29.1	29.5	28.6	29.3	28.8	3.17	0.66	
DM digestibility, %	65.60	64.01	64.53	63.50	64.36	1.409	0.29	A†, C*
NDF digestibility, %	39.05	37.55	37.72	37.07	36.60	1.939	0.70	
Rumen soluble								
Cu	0.11	0.19	0.14	0.30	0.18	0.015	0.0002	A**, B**, C**, D*
Mn	1.53	2.75	1.80	5.47	4.52	0.178	<0.0001	A**, B**, C**, D**
Zn	0.52	0.57	0.67	0.68	1.02	0.070	0.004	A*, B*, D**
Abomasal Soluble								
Cu	0.06	0.06	0.06	0.08	0.08	0.116	0.38	
Mn	1.88	3.16	2.86	5.91	4.80	0.203	<0.0001	A**, B**, C**, D**
Zn	2.85	3.80	3.88	6.26	7.86	0.321	<0.001	A**, B*, C**, D**

†P<0.10, *P<0.05, **P<0.001,

¹Contrasts: A: CON vs. all trace mineral supplemented treatments, B: Metal hydroxy bound trace minerals vs. sulfate bound trace minerals, C: CON vs. Sulfate bound trace mineral treatments, and D: CON vs. Metal Hydroxy bound trace mineral treatments