

# The effect of supplemental inorganic and organic forms of copper and zinc on digestibility and growth in yearling geldings in training

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## Introduction

A mineral proteinate is a chelated mineral complex formed by reacting a mineral salt with a specifically prepared mixture of amino acids and small peptides. The chelate that results from the binding of the mineral and amino acid carries no electrical charge, and remains stable in the presence of pH changes in the gastrointestinal tract. Because of this electrical neutrality, the mineral in organic complex form is theorized to be more bioavailable to the animal, and utilize a different absorptive mechanism than a mineral ion. Numerous studies in rats, poultry, pigs, and ruminants have shown increased bioavailability of the mineral proteinate as compared to the inorganic form. However, there are few controlled studies testing the effects of supplemental inorganic and organic forms of trace minerals on digestibility, daily balance, or bone density in the equine.

## Objectives

The objectives of this trial were to determine the effects of supplemental inorganic and organic forms of copper (Cu) and zinc (Zn) on digestibility, daily balance and bone density in yearling geldings in training.

## Materials and methods

Ten yearling stock-type geldings were stratified by weight and randomly assigned to one of three diets: control (no added Cu or Zn), inorganic Cu/Zn or organic Cu/Zn. The trial consisted of four 30-day experimental periods with 72-hr collection periods at the end of each experimental period. Total urine and fecal collections were taken during the 72-hr collection periods. Blood samples and radiographs were taken the first morning of each collection period. Beginning on day 50, all horses were placed on an exercise regime consisting of 15 min non-circular longeing at the trot. Horses were fed individually at 12-hr intervals, provided fresh water *ad libitum*, and allowed 2 to 4 hrs to consume their respective ration before being turned out each day. Radiographs were scanned at the nutrient foramen with a Bio-Rad Model 620 Densitometer to determine bone density. A logarithmic regression was formed using the thickness of steps on an aluminum penetrometer to determine the radiographic bone aluminum equivalence (RBAE) from the maximal optical density readings of both cortices for each view of the metacarpal.

Diets were formulated to meet or exceed NRC (1989) requirements and to reflect mineral supplementation common to the equine industry for yearlings in training at moderate growth. Concentrates were fed with matua-alfalfa hay in a 65:35 grain to hay ratio. The concentrate diets were formulated using a base concentrate of corn, soybean meal, cottonseed hulls, and dehydrated alfalfa meal. The control diet contained no supplemental Cu or Zn. The inorganic mineral treatment contained approximately 50 ppm Cu and 200 ppm Zn from added Cu sulfate and Zn oxide. The organic mineral supplement consisted of the same total added amounts of Cu and Zn but was formulated by replacing 45% of the Cu and Zn with Bioplex™ Cu and Zn (Alltech, Inc.). Concentrate formulation and analyses of concentrate and hay are shown in Tables 1 and 2.

**Table 1. Concentrate formulation.**

Ingredient, unit	Control	Inorganic	Organic
Ground shelled corn, %	61.0	61.0	61.0
Soybean meal (44% CP), %	15.0	15.0	15.0
Cottonseed hulls, %	10.0	10.0	10.0
Dehydrated alfalfa, %	6.5	6.5	6.5
Soybean oil, %	3.0	3.0	3.0
Liquid molasses, %	2.0	2.0	2.0
Trace mineralized malt, %	1.0	1.0	1.0
Limestone, %	1.0	1.0	1.0
Dicalcium phosphate, %	0.5	0.5	0.5
Copper sulfate, g/ton	--	144.3	79.4
Zinc oxide, g/ton	--	202.1	111.1
Bioplex Copper <sup>a</sup> , g/ton	--	--	164.0
Bioplex Zinc <sup>a</sup> , g/ton	--	--	437.0

<sup>a</sup>Alltech Inc., Nicholasville, KY

**Table 2. Concentrate and Matua/alfalfa hay analyses (dry matter basis).**

	Concentrate			Matua/alfalfa
	Control	Inorganic	Organic	
Moisture, %	7.0	7.1	7.1	14.0
CP, %	14.2	13.6	13.8	13.5
ADF, %	14.0	13.9	13.0	31.2
Ash, %	1.3	3.1	3.3	13.1
Ether extract, %	6.5	6.9	5.6	2.6
Calcium, %	0.7	0.7	0.7	0.8
Phosphorus, %	0.4	0.4	0.3	0.2
Copper, ppm	23.4	51.2	61.0	35.1
Zinc, ppm	66.6	189.9	216.6	29.5

Statistical analyses were conducted using repeated measures within the General Linear Models procedure, with treatment and time as main effects; time as the repeated variable; and all interactions (SAS, 2000). Due to the death of two geldings (unrelated to treatment), one from the inorganic and one from the organic treatment group, least square means were calculated. Non-orthogonal contrasts were used to determine differences between time means (day 30 vs days 60, 90, and 120). Orthogonal contrasts were used to compare treatment means (control vs the mean of the inorganic and organic treatments, and inorganic vs organic).

## Results

Horses consuming diets containing organic minerals had greater mean apparent daily Cu retention ( $P < 0.001$ ; Figure 1), retention as a percent of intake ( $P < 0.001$ ; Figure 2), and apparent Cu digestibility ( $P < 0.001$ ; Figure 3) as compared to horses consuming Cu sulfate. Similarly, horses consuming the organic minerals had a greater mean apparent daily Zn retention ( $P < 0.001$ ; Figure 4) as compared to horses consuming the inorganic form. There was no difference between Cu sources on mean serum Cu concentration ( $P < 0.792$ , Table 3). There was no difference between Zn sources on mean serum Zn concentration ( $P < 0.634$ ; Table 4). No differences due to treatment were detected in lateral, medial, palmar, or dorsal RBAE at any time during the trial.

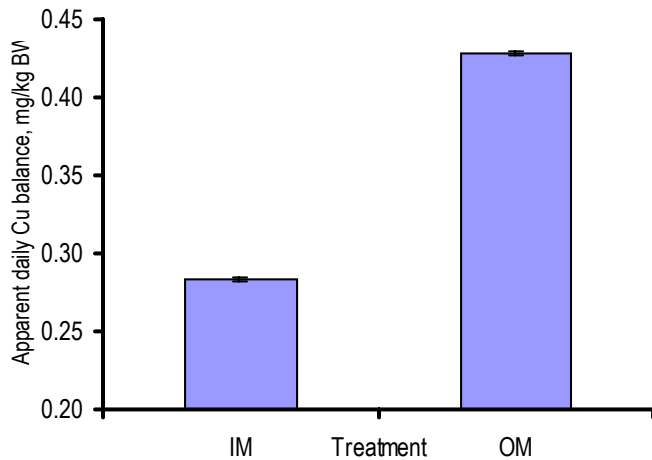


Figure 1. The effect of treatment (inorganic vs organic) on mean apparent daily Cu retention over 120 days ( $P < 0.001$ ).

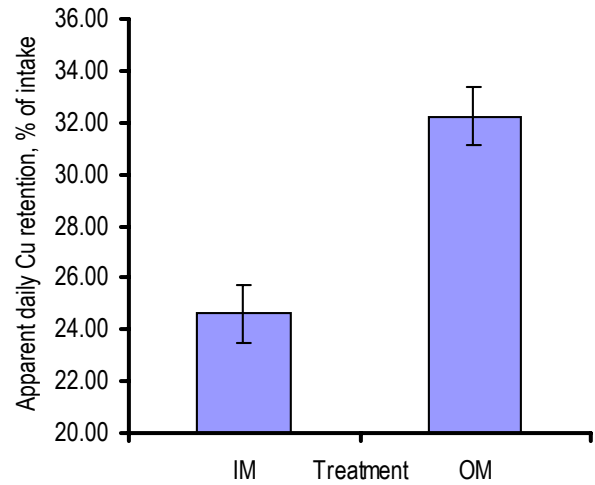


Figure 2. The effect of treatment (IM vs OM) on mean apparent daily Cu retention as a percent of intake over 120 d ( $P < 0.001$ ).

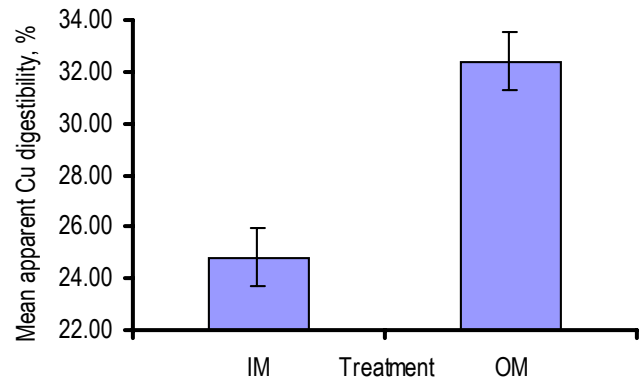


Figure 3. The effect of treatment (inorganic vs organic) on mean apparent Cu digestibility over 120 days ( $P < 0.001$ ).

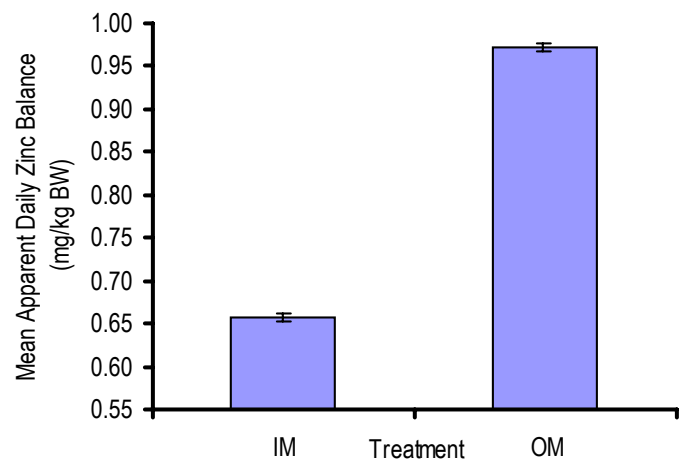


Figure 4. The effect of treatment (inorganic vs organic) on mean apparent daily Zn retention over 120 days ( $P < 0.001$ ).

**Table 3. Mean daily Cu intake, fecal and urinary excretion; apparent daily retention, retention as a percent of intake, and retention as a percent of absorption; apparent digestibility; and serum concentration at 30, 60, 90 and 120 days.**

	Time (days)			
	30	60	90	120
<i>Cu Intake, mg/kg BW</i>				
Control	0.69	0.70	0.69	0.69
Inorganic	1.15	1.16	1.15	1.15
Organic	1.35	1.32	1.32	1.32
<i>Fecal Cu, mg/kg BW</i>				
Control	0.36	0.33	0.42	0.42
Inorganic	0.72	0.88	1.00	0.88
Organic	0.79	0.89	1.04	0.87
<i>Urine Cu, ug/kg BW</i>				
Control	1.58	1.71	1.94	2.02
Inorganic	1.72	2.73	2.27	1.92
Organic	1.67	2.17	2.20	2.25
<i>Cu retention, mg/kg BW</i>				
Control	0.34	0.36	0.27	0.27
Inorganic	0.43	0.27	0.16	0.27
Organic	0.56	0.43	0.28	0.45
<i>Cu retention, % of Cu intake</i>				
Control	48.50	52.00	38.41	38.66
Inorganic	37.67	23.54	13.42	23.79
Organic	41.52	32.43	20.89	34.06
<i>Ca retention, % of Cu absorbed</i>				
Control	99.52	99.51	99.24	99.24
Inorganic	99.59	99.02	98.22	99.29
Organic	99.70	99.49	99.20	99.50
<i>Cu digestibility, %</i>				
Control	48.73	52.24	38.69	38.96
Inorganic	37.83	23.78	13.62	23.96
Organic	41.64	32.60	21.06	34.23
<i>Serum Cu, ppm</i>				
Control	1.31	1.26	1.08	1.10
Inorganic	1.12	1.34	1.10	1.09
Organic	1.18	1.41	1.04	1.10

**Table 4. Mean daily Zn intake, fecal and urinary excretion; apparent daily retention, retention as a percent of intake, and retention as a percent of absorption; apparent digestibility; and serum Zn concentration for all treatments at 30, 60, 90 and 120 days.**

	Time (days)			
	30	60	90	120
<i>Zn intake, mg/kg BW</i>				
Control	1.36	1.37	1.35	1.35
Inorganic	3.42	3.40	3.39	3.38
Organic	3.87	3.83	3.38	3.85
<i>Fecal Zn, mg/kg BW</i>				
Control	1.10	1.10	1.13	1.16
Inorganic	2.74	2.73	2.80	2.67
Organic	2.87	2.95	3.06	2.59
<i>Urine Zn, ug/kg BW</i>				
Control	9.06	7.59	5.37	6.23
Inorganic	5.84	8.13	6.10	4.62
Organic	7.60	8.64	6.36	5.31
<i>Zn Balance, mg/kg BW</i>				
Control	0.25	0.26	0.21	0.18
Inorganic	0.67	0.66	0.58	0.71
Organic	0.99	0.87	0.78	1.25
<i>Zn Balance as % Intake, %</i>				
Control	18.26	19.16	15.73	13.06
Inorganic	19.62	19.44	17.18	20.89
Organic	25.66	22.76	20.23	32.39
<i>Zn retention, % of absorbed</i>				
Control	95.84	96.38	97.24	96.08
Inorganic	98.98	98.41	98.73	99.33
Organic	99.24	99.02	99.19	99.58
<i>Zn digestibility, %</i>				
Control	18.93	19.72	16.13	13.53
Inorganic	19.85	19.68	17.36	21.03
Organic	25.85	22.98	20.40	32.53
<i>Serum Zn, ppm</i>				
Control	0.96	0.39	0.82	0.63
Inorganic	0.94	0.47	0.84	0.64
Organic	0.87	0.45	0.86	0.67

## Discussion

The concentrate diets fed the inorganic (IM) and organic mineral (OM) treatment groups were formulated to contain equivalent amounts of Cu and Zn; however upon analysis the OM concentrate contained 61 ppm Cu and 216 ppm Zn, while the IM concentrate contained 61 ppm Cu and 190 ppm Zn. Due to the resulting differences in Cu and Zn intake between treatments, it is important to consider the retention data with the balance as a percent of intake data to compare the supplemental forms of the minerals. It is also interesting to note that the basal concentrate mixture contained 23 ppm Cu and 66 ppm Zn using standard ingredients, which supply well above NRC requirements for these minerals (10 ppm Cu, 40 ppm Zn).

Apparent daily mineral retention data can sometimes be difficult to interpret in feeding trials. Oftentimes, as mineral intake increases above daily requirements, retention also increases. A cursory glance at the balance data from this trial might lead one to conclude that this is the cause of the significantly higher daily Cu and Zn balance in horses given the OM diet (Figures 1 and 4), as intake of both minerals was higher for these horses. However, this is not the case with Cu, as Cu retention as a percent of daily Cu intake (Figure 6) was also significantly higher for those consuming OM compared to those consuming IM.

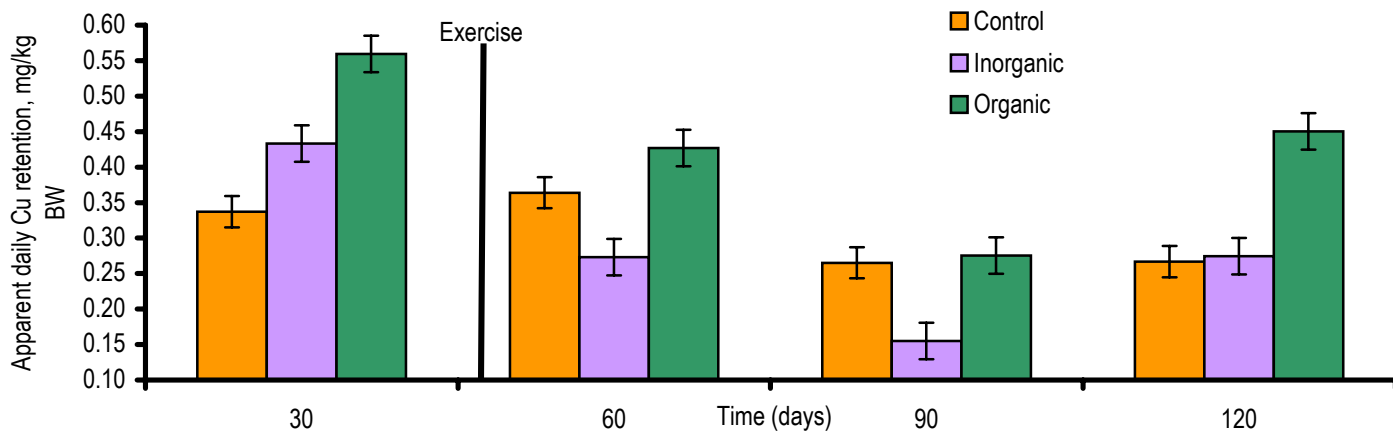


Figure 5. The effect of sampling time and treatment on mean apparent daily Cu retention (interaction:  $P < 0.001$ ).

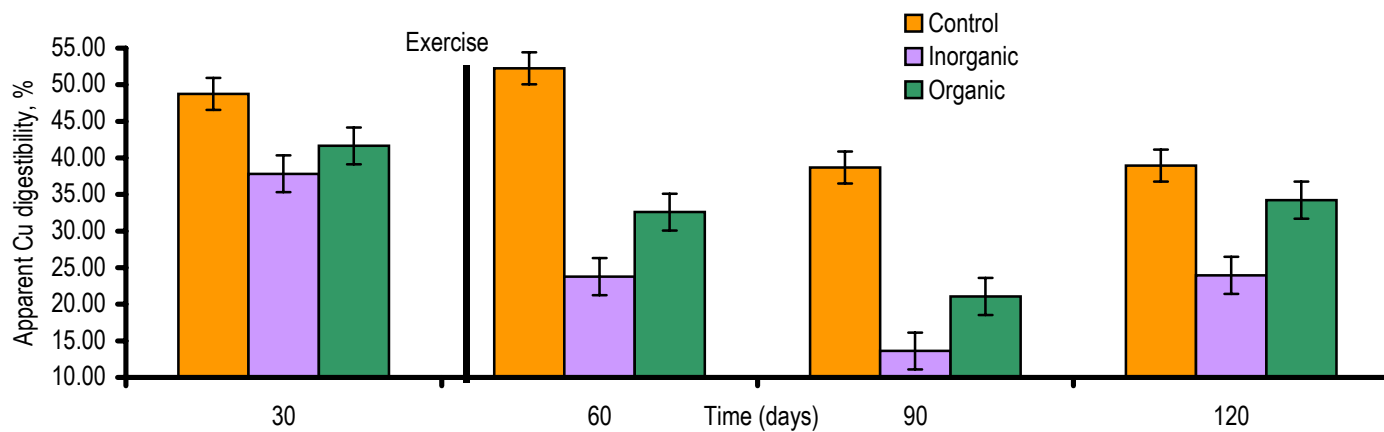


Figure 6. The effect of sampling time and treatment on mean apparent Cu digestibility (interaction:  $P < 0.007$ ).

As can be seen in Figure 3, apparent daily Cu retention for those horses consuming both IM and OM appear to follow a very similar trend. Retention in both groups decreased at day 60 and again at day 90, before increasing at day 120. This decrease and subsequent increase could be explained by the beginning of the exercise regimen on day 50 causing an increased need for Cu by various enzymes and/or the increased bone remodeling that is taking place. Another possible explanation for this trend is an immune response to disease and a subsequent depletion of body stores of Cu. Although all horses were vaccinated for *Streptococcus equi* (strangles) upon arrival at the WTAMU Horse Center, all horses became symptomatic of strangles before the beginning of the trial, and continued to suffer mild symptoms through approximately day 100.

The digestibility data in Figures 6 and 3 provide further evidence for the effectiveness of the organically chelated Cu and Zn used in this study. As a general rule, mineral digestibility percentages tend to decrease as intake increases above the daily requirement. This theory holds true with regard to horses consuming the control as compared to both IM and OM treatment diets (Figure 6). However, even though horses consuming OM were

consuming more Cu than horses on the IM diet (1.32 vs. 1.15 mg/kg BW, respectively), Cu digestibility in the OM treatment group was higher at every time point measured (Figure 6), and was significantly higher overall (Figure 3).

No differences due to treatment were detected for Zn retention as a percent of intake or apparent Zn digestibility. However, horses on the OM diet had significantly higher apparent daily Zn retention than horses consuming the IM diet. This increased daily balance could be explained by a slightly higher Zn intake by horses consuming OM; however, as observed in the Cu data, this increase could be due to horses more efficiently retaining the organically chelated Zn used in this study.

Results from this trial indicate that yearling geldings supplemented with the organically chelated forms of Cu and Zn (Bioplex™ Cu and Zn) had significantly higher Cu digestibilities, daily retention, retention as a percent of intake, and significantly higher apparent daily Zn retention compared to those supplemented with the inorganic forms of these minerals. More research is necessary to ascertain the effects of organically chelated Zn on Zn digestibility in horses.