

**Research Article** 

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# The effect of copper complex of methionine compared with copper sulfate in growing pigs

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Abstract: The aim of this study was to compare 2 copper products and examine their influence on hematological parameters, body weight, and content of copper and iron in the liver and spleen of pigs. In this experiment, 60 day-old pigs, bw $\approx$ 10 kg, (Camborough hybrid) were fed 1 of 3 treatments for 20 days: 20 ppm copper from copper methionine (Cu MET), 40 ppm copper from Cu MET, and 20 ppm Cu from copper sulfate (Cu SUL). Control group did not receive any copper supplementation. Our data show that a daily dose of 20 ppm Cu MET supplemented to the ration of pigs exerts positive effects on blood hemoglobin on day 10 and erythrocyte levels on day 20, and copper and iron storage in the liver and spleen of pigs as compared with the control. Growth response from added Cu MET showed some improvements over pigs fed the control diet; however, no significant difference was found between these groups.

Key words: Pig, copper source, blood parameters, performance

## Introduction

Trace elements play a very vital role in animal feeding. Trace elements are part of cell structure, component of soft tissues, and they are needed for regulating many biological processes in the organism (1). These elements are found in most feed ingredients, but their quantity and bioavailability vary significantly (2). The trace elements are usually supplemented in feed ration in small quantities in forms of sulfates, chlorides, carbonates, and oxides (3). Previous investigations have shown that the bioavailability of trace elements can be increased by connecting them with organic substances, usually a mixture of amino acids and low molecular peptides (ligands) that leads to organic minerals (chelates) (4).

Iron, copper, and zinc are specific organic minerals supplemented in pig rations (3). Organic minerals are used in prophylaxis and metaphylaxis of copper deficiency in newborn and growing pigs, in relation with signs of anemia, hypotrophic syndrome, diarrhea, etc. Their implementation in appropriate dosages in rations leads to a decreased level of diseases in pigs.

Copper is an essential trace mineral in pig nutrition and it acts as a growth promoter in growingfinishing pigs when supplemented at high dietary concentrations. Copper plays different roles in pig metabolism, but 2 antioxidant enzymes, "ceruloplasmin" and "Cu-Zn superoxide dismutase", are thought to be the most important molecules that copper participates in actively.

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The ceruloplasmin taking part in absorption and transport of Fe and Cu-Zn superoxide dismutase helps protecting the cells of toxic effect of chemicals linked to the catalytic copper ions (5).

When the supply of copper in feed is insufficient, a deficiency symptom of anemia can be observed related to decreased hemoglobin and erythrocyte count under respective physiological limits (6,7).

The copper is supplemented in form of inorganic salts to feed pig rations to improve their growth (1). However, recent studies have shown that organic copper sources are more effective as a growth promoter and create adequate concentration in the body to overcome the copper insufficiency (8-14). On the other hand, some comparative investigations have shown that organic copper sources supplemented to pig ratios have demonstrated similar effect like inorganic copper sources. Apgar and Kornegay (15) reported that the absorption and retention of copper lysate were similar in comparison to copper sulfate.

There is lack of information on usage of organic copper sources in pig's rations and their effectiveness on growth promotion in Bulgaria. Therefore, the aim of the present study was to investigate the new synthesized Bulgarian copper chelate called copper methionine (Cu MET) on performance, copper and iron levels of liver and spleen, and hematological parameters.

#### Materials and methods

Twenty four growing pigs of the hybrid "Camborough" at 60 days of age with 10 kg average body weight were included in this study. The pigs were divided into 4 groups with 6 pigs per group (n = 6). Similar conditions applied for all animals including feeding and rearing. Within the experimental period (20 days) the pigs were fed 0.5 kg feed daily (meal). The basal ration meets all nutrient requirements for swine.

The pigs in group 1 received 20 ppm copper from Cu MET; group 2 received 40 ppm from Cu MET; group 3 received 20 ppm copper from copper sulfate, and group 4 was control (no copper supplementation).

The general health status was evaluated throughout the experimental period. Blood for

hematological examinations (hemoglobin, erythrocyte, and hematocrit levels) was collected from sinus ophtalmicus of each pig into tubes containing EDTA on day 10 and 20. The body weight was also determined. The hematological parameters were evaluated by an automatic analyzer (Cellscan 13, Spain). The livers and spleens were taken after the end of the examination to estimate the copper and iron storage. They were euthanized by a mixture of 10 mg/kg bw ketamine and 2.5 mg/kg bw diazepam.

The Cu and Fe quantity was evaluated by ICP (Varian, Vista-PRO). The samples preliminary were dry mineralized in a muffle oven (NABERTHERM, Germany) at 550 °C.

During the care and use of experimental animals, the law for protection and humane attitude to animals was observed (Ordinance 15/03.02.2006, Bulgaria).

For the statistical analysis of the results, one-way analysis of variance (ANOVA) was performed. The results considered statistically significant at  $P \leq 0.05$ .

## Results

The data about the body weight and hematological parameters of pigs treated with Cu MET and Cu SUL are presented at Table 1. The results showed that pigs received 20 ppm Cu MET in feed (group 1) had the highest body weight on day 10 and day 20 after treatment in comparison with the other groups. At the end of the study, mean body weight of the animals by group was  $16.83 \pm 0.53$  kg (group 1, 20 ppm C MET),  $15.00 \pm 0.70$  kg (group 2, 40 ppm C MET),  $14.41 \pm 0.37$  kg (group 3, 20 ppm Cu SUL), and  $12.53 \pm 0.49$  kg (control group). However, the differences between experimental and control animals were not statistically significant (P > 0.05), (Table 1).

The hematological parameters (Hb, Er, and Ht) of pigs before treatment in all experimental groups were within physiological limits (Table 1).

The HB levels were significantly higher (P < 0.05) for pigs fed the diets supplemented with 20 and 40 ppm copper from Cu MET comparing with the control group.

			Table 1. Tł	he effect of C	u source on h	Table 1. The effect of Cu source on hematological parameters and body weight in pigs from 60 to 80 days of age.	arameters ai	nd body weig	ht in pigs fron	n 60 to 80 day	s of age.		
							Parameters	ers					
C	-		Starting	Starting values			10 <sup>th</sup> day	ĥ			20 <sup>th</sup> day	y	
Groups	of pigs (n)	Hb (g/L)	Er (T/L)	Ht (U/L)	Body weight (kg)	Hb (g/L)	Er (T/L)	Ht (1/L)	Body weight (kg)	Hb (g/L)	Er (T/L)	Ht (I/L)	Body weight (kg)
Group 1	9	$107.6 \pm 4.38$	$6.53 \pm 0.31$	$35.08 \pm 1.42$	$10.23 \pm 0.25$	111.33a ± 3.46	$6.72 \pm 0.38$	$34.75 \pm 2.29$	$13.95 \pm 0.66$	$115.25 \pm 3.53$	7.08b ± 0.60	$35.66 \pm 1.50$	$16.83 \pm 0.53$
Group 2	9	$108.1 \pm 2.31$	$6.41 \pm 0.51$	$34.85\pm1.40$	$10.06\pm0.40$	110.05a ± 1.56	$6.68\pm0.81$	$34.71 \pm 2.31$	$12.16 \pm 0.51$	$111.83 \pm 1.29$	$6.78b\pm0.38$	$34.15b \pm 0.91$	$15.00 \pm 0.70$
Group 3	9	$106.8\pm3.03$	$6.46 \pm 0.45$	$34.86 \pm 1.18$	$9.85 \pm 0.54$	$108.33 \pm 8.70$	$6.49 \pm 0.46$	$35.01 \pm 0.70$	$12.25 \pm 1.08$	$110.55 \pm 1.64$	$6.44 \pm 0.34$	$35.25 \pm 0.74$	$14.41 \pm 0.37$
Group 4	9	$105.6\pm6.01$	$6.40\pm0.77$	$36.5 \pm 3.27$	$9.66 \pm 0.81$	$105.91 \pm 4.11$	$6.18\pm0.74$	$36.31 \pm 1.72$	$11.33\pm0.75$	$106.06 \pm 0.32$	$6.21\pm0.15$	$36.43\pm1.73$	$12.53 \pm 0.49$
$^{a}$ P < 0.05													
$^{\rm b}$ P < 0.01													
$^{c}$ P < 0.001													
Statistical	significance	Statistical significance toward control group	group										
Group 1-	treated with	Group 1- treated with Cu MET at 20 ppm concentration;	ppm concentrat	tion;									
Group 2-	treated with	Group 2- treated with Cu MET at 40 ppm concentration;	ppm concentrat	tion;									

Group 3- treated with Cu SUL at 20 ppm concentration;

Group 4- control group;

Plasma Er concentrations on day 20 for pigs fed 20 ppm Cu MET (group 1) and 40 ppm Cu MET (group 2) were significantly different (P<0.01) different comparing to the control group, with experimental means  $\pm$  SE (T / l) of 7.08  $\pm$  0.60, 6.78  $\pm$  0.38, 6.21  $\pm$  0.15, respectively. Plasma Ht concentration on day 20 decreased significantly (P < 0.01) in group 2 that fed 40 ppm Cu diet from Cu MET comparing with the control group.

The data about the quantity of trace elements copper and iron in the liver and spleen in pigs are presented in Table 2.

According to our results, there were higher values of iron and copper in the livers and spleens of pigs that received 20 ppm Cu MET supplementation (group 1). Thus, on day 20, the quantity of iron in the liver in the control group was  $125.3 \pm 7.7$  ppm and of copper was  $15.6 \pm 1.3$  ppm; in pigs of the group 3 (20 ppm Cu SUL) these values were, respectively,  $131.4 \pm 8.9$  ppm iron (P > 0.1) and 29.7 ± 3.6 ppm copper (P > 0.1); in group 2 (40 ppm Cu MET) the iron content was  $134.8 \pm 10.4$  (P > 0.1) and the copper content was  $30.9 \pm 4.8$ ppm (P > 0.25); in group 1 (pigs treated by 20 ppm Cu MET) these values were highest:  $137.3 \pm 12.3$  ppm iron (P < 0.1) and  $32.00 \pm 5.2$  ppm copper (P < 0.01).

The similar tendency was observed for iron and copper content in spleens of pigs in all experimental groups (Table 2).

## Discussion

The obtained results about hematological parameters in pigs treated with 20 ppm Cu MET are in accordance with data of Coffey et al. (9), Smits and Henwan (12), and Veum et al. (13) who used organic copper in their experiments (copper-lysine complex) in comparison to copper sulfate supplemented in ration of pigs as in the present study. Coffey et al. (9) evaluated the copper-lysine complex effectiveness in comparison with copper sulfate in growing pigs and established a better growth rate (11.5%, P < 0.05) in pigs treated with organic copper, which is an outcome that they attribute to high utilization of copper from organic sources, supported by higher copper levels in the liver and spleen. Contrary to Coffey et al. (9), none of the Cu sources were able to improve or impede pig growth criteria when fed from 60 to 81 days of age.

The general evaluation from the conducted comparative experiments of treatment by Cu MET and Cu SUL in growing pigs is that the dosage of 20 ppm Cu MET exerts better influence on hematological parameters as well as on copper storage in the liver and spleen in comparison with inorganic copper form (Cu SUL).

	Numbers of pigs (n)	T R A C E E L E M E N T S ( ppm )				
Groups		COPPER		IRON		
		Livers x ± Sx	Spleens x ± Sx	Livers x ± Sx	Spleens x ± Sx	
Group 1	6	$32.00 \pm 5.2$	$14.2 \pm 2.3$	37.3 ± 12.3	88.6 ± 9.2	
Group 2	6	$30.9 \pm 4.8$	$13.4 \pm 1.9$	$134.8\pm10.4$	$85.9\pm8.4$	
Group 3	6	$29.7 \pm 3.6$	$12.8 \pm 1.1$	$131.4 \pm 8.9$	$83.5\pm7.6$	
Group 4	6	$15.6 \pm 1.3$	$7.3\pm0.9$	$125.3\pm7.7$	$74.7\pm6.3$	

Table 2. The effect of Cu source on Cu content of liver and spleen of pigs from 60 to 80 days of age.

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