

# Copper balance, bone mineralization and the growth performance of turkeys fed diet with two types of Cu supplements

**D. Mikulski<sup>1,3</sup>, J. Jankowski<sup>1</sup>, Z. Zduńczyk<sup>2</sup>, M. Wróblewska<sup>2</sup>  
and M. Mikulska<sup>1</sup>**

*<sup>1</sup>Warmia and Mazury University, Department of Poultry Science  
Oczapowskiego 5, 10-718 Olsztyn, Poland*

*<sup>2</sup>Institute of Animal Reproduction and Food Research, Polish Academy of Sciences  
Tuwima 10, 10-747 Olsztyn, Poland*

(Received 6 October 2008; revised version 21 April 2009; accepted 6 November 2009)

## ABSTRACT

The aim of this study was to examine the effect supplementation of diet with 20 mg Cu/kg from copper sulphate (Cu-SUL) and/or copper amino acid chelate (Cu-AA) on the copper balance, bone mineralization status and growth performance of turkeys. A total of 840 one-day-old heavy Large White BIG-6 turkey males were randomly assigned to 4 groups: Cu-0, Cu-SUL, Cu-SUL/Cu-AA and Cu-AA. The results of the present study confirm the Cu-requirements for growing/fattening turkeys given by scientific bodies (e.g., NRC, INRA, GfE). The Cu unsupplemented basal diet contained 11.3 mg Cu/kg is enough for turkeys and further Cu-supplementations are not necessary. Further supplementations from inorganic and/or organic sources did not significantly improve turkeys performance and health. Plasma superoxide dismutase activity, physico-chemical properties of bone and the concentrations of Ca and P in the tibial bone were generally not affected by dietary treatment. Further Cu supplementation from inorganic and/or organic sources increase the Cu-excretion (from 808 to 1915-2164 mg per animal) and may cause additional environmental problems.

KEY WORDS: copper, chelate, bone mineralization, performance, turkeys

---

<sup>3</sup> Corresponding author: e-mail: [dariusz.mikulski@uwm.edu.pl](mailto:dariusz.mikulski@uwm.edu.pl)

## INTRODUCTION

Copper is an essential mineral required for proper bone growth and development as well as enzyme function. In a number of enzymes and proteins, the role of Cu has been defined for many physiological functions related mostly to a catalytic agent in the active sites of cuproenzymes (McDowell, 1992). Copper supplementation is very important to maintain normal functions of the cardiovascular system in animals (Savage et al., 1966).

Copper sulphate was included as a Cu source as it is the typical form used by the poultry industry. Inorganic Cu in the form of Cu-sulphate pentahydrate ( $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$ ) is most commonly used as a dietary supplement for animals due to cost and commercial availability. Additional Cu sources have become available recently, and the potential for commercial use as feed additives has expanded. Many authors share the opinion that the excretion of trace elements can be reduced by dietary inclusion of low doses of readily available minerals. Several researchers (Hemken et al., 1993; Du et al., 1996) confirm that metal chelates of amino acids and peptides can enhance the bioavailability of trace elements. An important function of chelated and complexed metals is the extent to which organic ligands remain associated with the metal ligand under physiological conditions (Guo et al., 2001). Some commercially available chelated and complexed Cu compounds are purported to protect the mineral from becoming degraded or complexed with other compounds until the product reaches the section of the gastrointestinal tract where absorption occurs. The target site for these Cu products is the small intestine, and more specifically the duodenum which is the major site of absorption of minerals in chickens (Hurwitz and Bar, 1965).

In studies conducted by Guo et al. (2001) the bioavailability of several organic copper products for chickens was significantly greater than that of copper sulphate. When Cu sulphate was assigned a value of 100% as the standard, linear regression slope ratios of  $\log_{10}$  liver Cu concentration regressed on added dietary Cu concentration gave estimated relative bioavailability values of 124, 122 and 111% for Cu lysine complex, Cu amino acid chelate and Cu proteinate, respectively. The bioavailability estimates for Cu lysine complex and Cu amino acid chelate were greater ( $P \leq 0.05$ ) than that for Cu sulphate (Guo et al., 2001). Aoyagi and Baker (1993) reported that the relative bioavailability of Cu in a Lysine-Cu complex was 126% compared with 100% for that in copper sulphate.

NRC (1994) requirements of Cu for broiler chickens and turkeys is only 8 mg/kg. According to European Commission Health (SCAN, 2003) the copper content of diets for animals should not exceed 35 mg/kg feed, including up to 20 mg Cu/kg feed from cupric chelate of amino acid hydrate. Information on the effectiveness of organic copper sources in turkey diets is scant. Therefore, the aim

of this study was to examine the effect of practical wheat/soyabean meal-based diets supplemented with 20 mg Cu/kg feed from copper amino acid chelate on the copper balance, bone mineralization status and growth performance of turkeys. In this study copper sulphate was used as a reference point for comparing the effectiveness of Cu amino acid chelate.

## MATERIAL AND METHODS

### *Animals and diets*

The experiment was conducted according to the guidelines of the Local Animal Experimentation Ethics Committee. A total of 840 one-day-old heavy Large White BIG-6 turkey males, sexed at the local commercial hatchery, were used. The initial body weight (BW) of day-old poults was 61 g. Rearing was conducted to 126 days of age, according to the technology recommended by the British United Turkeys Company (B.U.T., 2002).

Each of the experimental diets consisted of a wheat-soyabean meal based basal diet formulated using least-cost linear programming software to meet the nutrient requirements of turkeys, according to B.U.T. (2002) recommendations. The composition and calculated nutritive value of basal mixtures are presented in Table 1. The birds were fed *ad libitum*, according to 4-step program, i.e. starter diet (1-28 days), grower 1 diet (29-56 days), grower 2 diet (57- 84 days) and finisher diet (85 to 126 days). Starter diets were offered as crumbles and the other diets - as 3 mm pellets. The diets contained no growth promoters or coccidiostatics. Pelleted feed was supplied by a local commercial animal feed mill. Fresh feed and water were provided daily and were available *ad libitum*.

Each of the 4 dietary treatments consisted of 7 replicate pens with 30 birds in each pen. The treatment diets were as follows: 1. basal diet (Cu-0, no Cu source), 2. basal diet with 20 ppm Cu from  $\text{CuSO}_4$  (Cu-SUL), 3. basal diet with a blend of 10 ppm Cu from  $\text{CuSO}_4$  and 10 ppm Cu from copper amino acid chelate (Cu-SUL/AA), 4. basal diet with 20 ppm Cu from copper amino acid chelate (Cu-AA). The diets were supplemented with elemental Cu as  $\text{CuSO}_4$  ( $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$ ; Merck Ltd.) or as Cu-amino acid chelate (as Albion Copper Amino Acid Chelate supplied by Albion Laborat. Inc., France). Tested products were incorporated in the vitamin-mineral premixes which was supplied by the Polsanders Co. (Pruszcz Gdański, Poland).

Table 1. Composition (g/kg<sup>-1</sup>) and calculated nutrient content of the basal diets fed to turkeys from 1 to 126 days of age

Item	Starter 1	Grower 1	Grower 2	Finisher
	1 to 28 d	29 to 56 d	57 to 84 d	85 to 126 d
<i>Ingredient</i>				
wheat	243.2	279.0	370.4	459.0
maize	200.0	200.0	200.0	200.0
potato protein	50.0	30.0	-	-
soyabean meal (46%)	423.0	403.0	341.0	253.0
soyabean oil	33.0	20.0	21.0	24.0
animal fat	-	25.0	30.0	30.0
limestone	14.7	11.7	9.8	8.6
monocalcium phosphate	19.8	15.2	11.4	9.7
salt	2.6	2.1	1.5	1.5
NaHCO <sub>3</sub>	1.5	1.5	1.5	1.5
DL-methionine 99	1.6	1.3	1.0	0.8
L-lysine HCl	0.6	1.2	2.3	1.1
L-threonine	-	-	0.1	0.8
vitamin-mineral premix <sup>1</sup>	10.0	10.0	10.0	10.0
<i>Calculated analyses</i>				
ME, kcal/kg	2800	2900	3000	3100
crude protein, %	28.00	26.01	22.02	19.00
lysine, %	1.61	1.51	1.30	1.00
Met + Cys, %	1.05	0.95	0.80	0.70
Ca, %	1.20	1.00	0.85	0.75
available P, %	0.60	0.50	0.42	0.38
Na, %	0.17	0.15	0.12	0.12

<sup>1</sup>supplied per kg of feed, IU: vit A 15 000, vit D<sub>3</sub> 4 500; mg: vit. E 50, vit. K<sub>3</sub> 2.5, vit. B<sub>1</sub> 3.5, vit. B<sub>2</sub> 10, vit. B<sub>6</sub> 6, vit. B<sub>12</sub> 0.03, folic acid 2, biotin 0.36, nicotinic acid 75, pantothenic acid 21, choline 600, Mn 150 from magnesium oxide, Zn 90 from zinc oxide, Fe 60 from ferrous sulphate, J 1.0 from ethylene diamine dihydroiodide, Se 0.3 from sodium selenite, Co 0.2; g: Ca 1.72

### *Animals management*

The poults were vaccinated against turkey rhinotracheitis (TRT) by spray application at 1 d of age and were allocated at random to 28 pens with straw as bedding material. Stocking density was approximately 50 kg BW/m<sup>2</sup> of usable floor space in all pens. Visual health inspection of all birds was performed on a daily basis. Brooder rings for poults (till 10 days of age) and additional heat sources (till 28 days of age) were installed in the pens. Heating was provided by a central heating system and electric heaters (red light). The brooder unit's temperature was set at 35°C and then altered as needed to suit bird comfort. Room temperature was set at 28°C on the day of placement, and was subsequently reduced by 2°C per week. Temperature and humidity were recorded on a daily basis at 8 a.m. and 3 p.m. The lighting programme in the room was as follows: 23 h light at about 100 lux till 3 days of age and 14 h light at 5-6

lux from day 4 until the end of the experiment. Relative humidity was about 65 to 70%. Air changes were 0.4-0.5 m<sup>3</sup>/h/kg BW from 2 to 7 weeks of age and 7-8 m<sup>3</sup>/h/kg BW from 8 weeks of age.

Body weight and feed consumption were recorded on 28, 56, 84 and 126 day of age. Culling and mortality rates (specifying the cause of death and reason for rejection) were monitored on a regular basis. The cases and incidence of aortic rupture were recorded systematically.

#### *Cu balance, superoxide dismutase activity, bone mineralization status*

Copper balance was determined from day 25 to 29. At 21 days of age, ten birds were selected randomly of each treatment (5 replications, each of 2 birds) and transferred to individual cages for balance trials. Battery cages were thermostatically controlled, and turkeys had free access to water from nipple drinkers. After a three-day adaptation period (25th day of age), the birds were fasted for 14 h, next they received a starter diet (administered in the growth performance test) *ad libitum* for 5 days, and then they were fasted again for 14 h. During the proper experimental period (5 days) feed intake was monitored on a regular basis, and excreta were collected. After removal of the non-faecal material, the excreta were stored in plastic bags at -20°C. After the collection period the moisture content of the excreta was determined by drying in a ventilated drying oven (48 h at 60°C). Samples of feed and dried excreta were ground through a 0.5 mm screen and were then analysed for DM and Cu content. Dry matter content was determined by oven drying at 100°C for 16 h. In order to determine copper content, samples of feed and dried excreta were mineralized in a mixture of nitric acid and perchloric acid (3:1). Mineralization was carried out in an electric aluminium heating block with selectable temperatures (VELP DK 20, VELP Scientifica, Italy), for 4 to 6 h, over a temperature range of 120 to 200°C. The obtained colourless mineralizate was transferred to 50 cm<sup>3</sup> measuring flasks, which were filled up to the mark with deionized water. Copper retention was determined by accounting for the differences between total Cu in the excreta and feed and adjusted to a DM basis. Cu intake was counted based at total feed intake by 2 birds for 5 days and described as average daily Cu intake per 1 bird. Cu excreted was counted based at total excreta voided by 2 birds for 5 days and described as average daily Cu excreted per 1 bird. Calculations were made for each replication separately. At the completion of the balance trial the birds were returned to the pens.

On day 56 and 126, blood samples were taken from 7 turkeys of each treatment, and next the birds were sacrificed. Superoxide dismutase (EC 1.1.5.1; SOD) activities in erythrocyte lysates were assayed using kits from Randox Laboratories Ltd. (Crumlin, UK) according to the method described by Woolliams et al. (1983).

The Randox method uses xanthine and xanthine oxidase to generate superoxide radicals that react with 2-(4-iodophenyl)-3-(4-nitrophenol)-5-phenyltetrazolium (I.N.T.) to form a red formazan dye. The SOD activity is then calculated from the degree of inhibition of this reaction compared to a standard curve of SOD.

On day 56 and 126, the left femoral and tibia bones were excised from all 28 birds, weighed (following the separation of muscles and cartilage), and their volume was measured in distilled water. Bone weight (w) and volume (v) provided a basis for calculating bone specific gravity (w:v). Tibias were next dried for 16 h at 100°C, defatted by refluxing with diethyl ether for 6 h, dried in a 100°C oven for 16 h, and analysed for ash content, total Cu, Ca and P. In order to determine ash content, samples were ashed for 6 h in an electric furnace at 300°C, and then at 530-540°C until white ash was obtained. The ash was dissolved in a 5 M solution of nitric acid (Suprapur, Merck, Germany). Analytical samples were prepared simultaneously, and reference material SRM 1400 (Bone Ash) was analysed. The obtained mineralizates (non-diluted or diluted) were assayed for the concentrations of Cu and Ca by flame atomic absorption spectrometry (acetylene-air flame), as described by Whiteside and Miner (1984), using a Unicam 939 Solar atomic absorption spectrometer (Cambridge, UK) equipped with an Optimus data station, background correction (deuterium discharge lamp) and cathode lamps. Prior to Ca determination, a 10% aqueous solution of lanthanum chloride was added to all solutions, to ensure the final La<sup>+3</sup> concentration of 1% (Whiteside and Miner, 1984). Total P was determined colorimetrically with ammonium molybdate, sodium sulphate and hydroquinone. Conversion of phosphates into phosphoromolybdates in the sulphuric acid environment, with ammonium molybdate, followed by their reduction to phosphoromolybdenum blue with sodium sulphate and hydroquinone. Absorbance was measured using a VIS 6000 spectrophotometer (Krüss–Optronic, Germany) at a wavelength of 610 nm.

### *Statistical analysis*

All data were analysed statistically by a one-way ANOVA and Duncan's multiple range test, using Statistica 6.0PL software. Replicate means served as experimental units for the statistical analysis. Treatment effects were considered to be significant at  $P \leq 0.05$  or trend at  $P < 0.1$ .

## RESULTS

*Cu balance.* The Cu content of feed offered to turkeys in group Cu-0 eached 11.35 mg/kg, while in Cu-supplemented diets it was by 14.6-14.8 mg/kg (Cu-SUL

and Cu-SUL/AA) to 20.3 mg/kg (Cu-AA) higher (Table 2). Feed consumption and faeces excreted during the balance test was comparable in all treatments. As expected, Cu intake in Cu-supplemented groups was significantly ( $P \leq 0.05$ ) higher than in group Cu-0 turkeys. In group Cu-AA turkeys, Cu intake was significantly ( $P \leq 0.05$ ) higher than in the other groups. Copper retention in Cu-0, Cu-SUL and Cu-SUL/AA turkeys was at a similar level (29.1 to 30.1%). The Cu retention coefficient of Cu-AA turkeys was significantly ( $P \leq 0.05$ ) higher than in birds administered Cu-SUL, Cu-SUL/AA and Cu-0. The Cu unsupplemented basal diet contained 11.3 mg Cu/kg and further Cu supplementation from inorganic and/or organic sources significantly increased ( $P \leq 0.05$ ) Cu-excretion (from 808 to 1915-2164 mg per bird).

Table 2. The effects of copper source on Cu balance in male turkeys from 25 to 29 d of age<sup>1</sup>

Item	Cu source <sup>2</sup>				SEM
	Cu-0	Cu-SUL	Cu-SUL/AA	Cu-AA	
<i>Feed</i>					
Cu content in feed, µg/g	11.35	26.20	26.00	31.68	-
feed intake, g/day per bird	101.3	107.9	105.4	104.0	1.1
Cu intake, µg	1150 <sup>A</sup>	2827 <sup>B</sup>	2741 <sup>B</sup>	3294 <sup>C</sup>	187
<i>Faeces, after drying</i>					
Cu content, µg/g	28.30	66.00	65.17	75.78	-
faeces excreted, g/day/bird	28.6	30.4	29.4	28.6	0.4
Cu excreted, µg	808 <sup>A</sup>	2004 <sup>B</sup>	1915 <sup>B</sup>	2164 <sup>C</sup>	124
Apparent Cu retention, % of Cu intake	29.6 <sup>A</sup>	29.1 <sup>A</sup>	30.1 <sup>A</sup>	34.3 <sup>B</sup>	0.8

<sup>1</sup> data represent mean values of five replicate; <sup>2</sup> experimental diets: Cu-0 - wheat based feed without copper supplements and experimental diets containing 20 mg/kg of copper from CuSO<sub>4</sub> (Cu-SUL) or copper amino acid chelate (Cu-AA), or both source (10 mg + 10 mg; Cu-SUL/AA) means within the same line with no common superscripts differ significantly: <sup>A-B</sup>  $P \leq 0.05$  as a result of a Duncan means comparison

*Turkey growth, feed conversion and mortality.* The response to birds to dietary copper varied widely (Table 3). At 28 days of age, the body weight of turkeys fed a diet with Cu-SUL or Cu-AA was significantly ( $P \leq 0.05$ ) higher than the body weight of control group turkeys (Cu-0) and those fed a diet with Cu-SUL/AA. At 56 days of age, only the body weight of birds given a diet with Cu-AA was significantly ( $P \leq 0.05$ ) higher than Cu-0 and Cu-SUL/AA turkeys. On day 84, similarly as on day 28, the body weight of group Cu-AA turkeys was significantly ( $P \leq 0.05$ ) higher than the body weight of turkeys in groups Cu-SUL/AA and Cu-0. On day 84, the body weight of turkeys in group Cu-SUL was significantly ( $P \leq 0.05$ ) higher than in Cu-0, but tended ( $P < 0.1$ ) to lower than in group Cu-AA. At the end of rearing (day 126), the body weights of turkeys in particular treatments were similar.

Table 3. Body weight (BW), feed conversion ratio (FCR) and mortality of turkeys raised to 18 weeks of age<sup>1</sup>

Item	Cu source <sup>2</sup>				SEM
	Cu-0	Cu-SUL	Cu-SUL/AA	Cu-AA	
<i>Body weight, kg/bird</i>					
at 28	1.136 <sup>A</sup>	1.174 <sup>B</sup>	1.137 <sup>A</sup>	1.178 <sup>B</sup>	0.004
at 56	4.830 <sup>A</sup>	4.875 <sup>AB</sup>	4.821 <sup>A</sup>	4.917 <sup>B</sup>	0.014
at 84	9.585 <sup>A</sup>	9.741 <sup>BCa</sup>	9.661 <sup>AC</sup>	9.864 <sup>Bb</sup>	0.027
at 126	17.142	17.126	17.165	17.142	0.051
<i>FCR, kg/kg</i>					
1 to 28 d	1.447 <sup>b</sup>	1.414 <sup>a</sup>	1.416 <sup>a</sup>	1.421 <sup>ab</sup>	0.006
1 to 56 d	1.772	1.753	1.762	1.744	0.005
1 to 84 d	2.121 <sup>B</sup>	2.053 <sup>A</sup>	2.043 <sup>A</sup>	2.013 <sup>A</sup>	0.012
1 to 126 d	2.632	2.603	2.575	2.597	0.012
<i>Mortality, %</i>					
1 to 28 d	1.4	-	0.46	-	
1 to 126 d	2.79	2.30	4.17	2.76	

<sup>1</sup>data represent mean values of seven replicate pens of 30 turkeys each; <sup>2</sup>as in Table 2 means within the same line with no common superscripts differ: <sup>a-b</sup>-  $P < 0.10$ ; <sup>A-B</sup>-  $P \leq 0.05$

Over a period of 1 to 28 days, a tendency towards a lower ( $P < 0.1$ ) feed conversion ratio (FCR) was noted in groups Cu-SUL and Cu-SUL/AA, in comparison with the control group. During a period of 1 to 56 days, FCR were generally not affected by dietary treatment. Throughout a period of 1 to 84 days, FCR was significantly ( $P \leq 0.05$ ) lower in turkeys fed a Cu-supplemented diet than in group Cu-0. The FCR estimated for the entire rearing period (1-126 days) was at a comparable level in all treatments, irrespective of the copper source in the diet.

The health condition of turkeys was generally good over the experimental period (Table 3). Until day 28, the mortality rate in treatments varied from 0.46 to 1.4%, and throughout the rearing period, i.e. 126 days, it ranged from 2.3 to 4.2%. Between 14 and 18 weeks of age, mortality due to aortic rupture was diagnosed in 5 turkeys, including 1 bird in group Cu-SUL and 2 birds in groups Cu-AA and Cu-SUL/AA. There was no relationship between the causes of mortality and the copper source in feed.

*Plasma superoxide dismutase activity.* Average activity of superoxide dismutase in blood plasma of turkeys of different groups was in small degree differentiate and difference between dietary treatments was statistically non-significant (Figure 1).

*Bone mineral content.* Femur relative weight and specific gravity were unaffected ( $P > 0.1$ ) by dietary treatments on days 56 and 126 (Figure 2). Tibia specific gravity, ash content and the concentrations of major mineral elements (Ca and P) were also unaffected ( $P > 0.1$ ) by dietary treatments (Table 4). At the age of 56 days, a tendency towards a higher tibia relative weight and tibia Cu

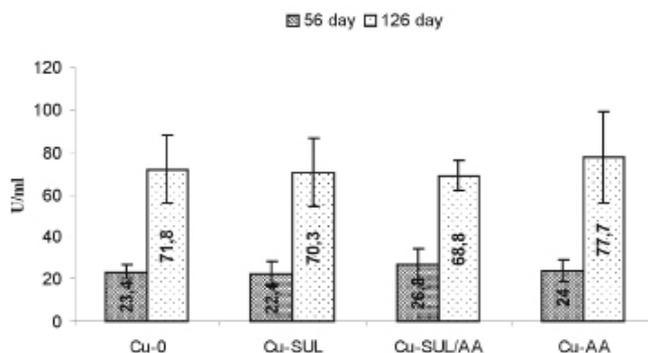


Figure 1. Superoxide dismutase (SOD) activity in blood plasma of turkeys at 56 days and 126 days of age, U/ml<sup>1</sup>; <sup>1</sup> data represent mean values of seven turkeys per treatment

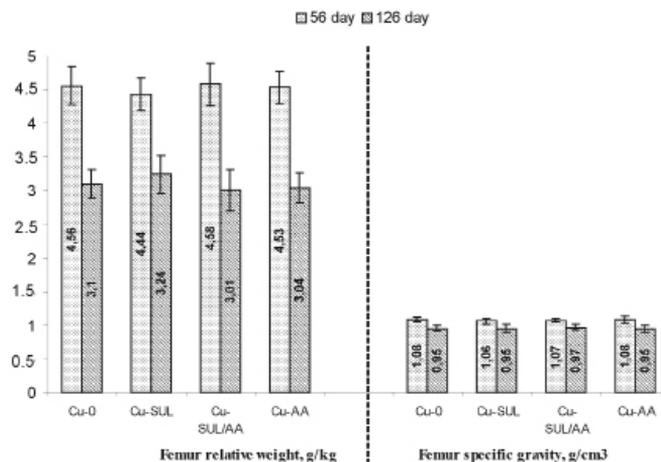


Figure 2. Femur relative weight and specific gravity of turkeys at 56 and 126 days of age<sup>1</sup>; <sup>1</sup>data represent mean values of seven turkeys per treatment

concentration ( $P < 0.1$ ) was noted in turkeys fed a Cu-AA supplemented diet, compared to Cu-SUL and Cu-0 treatment. Such a rising trend (by 25%;  $P < 0.1$ ) in tibia Cu concentration was also reported in group Cu-AA turkeys on day 126, compared with turkeys fed inorganic Cu, i.e.  $\text{CuSO}_4$  (Cu-SUL). The addition of 20 ppm copper in the form of Cu-SUL to diets for turkeys had no effect on tibia Cu concentration. At the age of 126 days, dietary supplementation with Cu-SUL/AA or Cu-AA significantly increased ( $P \leq 0.05$ ) the concentration of Cu in tibia bone, compared to the control treatment.

Table 4. Characteristics of tibia bone of turkeys at 56 and 126 days of age<sup>1</sup>

Item	Cu source <sup>2</sup>				SEM
	Cu-0	Cu-SUL	Cu-SUL/AA	Cu-AA	
<i>56 days</i>					
tibia relative weight, g/kg BW	5.80 <sup>a</sup>	5.81 <sup>a</sup>	5.96 <sup>ab</sup>	6.26 <sup>b</sup>	0.088
tibia specific gravity, g/cm <sup>3</sup>	1.17	1.18	1.18	1.19	0.006
tibia ash, %	45.47	45.88	46.54	46.56	0.322
Cu content, mg/kg DM	1.05 <sup>a</sup>	1.07 <sup>a</sup>	1.12	1.22 <sup>b</sup>	0.032
Ca content, mg/kg DM	184.1	187.9	190.0	193.0	2.176
P content, mg/kg DM	86.39	87.11	88.50	88.82	0.990
<i>126 days</i>					
tibia relative weight, g/kg BW	4.28	4.40	4.01	4.39	0.076
tibia specific gravity, g/cm <sup>3</sup>	1.01	1.02	0.99	1.01	0.005
tibia ash, %	63.00	63.29	63.39	62.97	0.163
Cu content, mg/kg DM	0.257 <sup>A</sup>	0.317 <sup>a</sup>	0.341 <sup>B</sup>	0.395 <sup>Bb</sup>	0.016
Ca content, mg/kg DM	241.5	245.1	240.2	238.1	1.191
P content, mg/kg DM	113.6	115.8	115.1	113.8	0.974

<sup>1</sup> data represent mean values of seven turkeys per treatment; <sup>2</sup> as in Table 2  
means within the same line with no common superscripts differ: <sup>a,b</sup>- P<0.10; <sup>A,B</sup>- P≤0.05

## DISCUSSION

In earlier experiments of other authors (Baker et al., 1991; Mondal et al., 2007) dietary Cu in organic form, including Cu-lysine, Cu-methionine and Cu-proteinate, was absorbed to the same or to a higher degree than Cu in the form of cupric sulphate. Paik et al. (1999) reported that Cu from a Cu-methionine complex was better absorbed and accumulated to a higher degree in the breast muscles and to a lower degree in the liver of chickens than Cu from copper sulphate. In our study Cu unsupplemented basal diet contained 11.3 mg Cu/kg was enough for turkeys. The increase of Cu content in diet to 26 mg/kg (Cu-SUL and Cu-SUL/AA) had no effect on apparent Cu retention in turkeys. Further Cu supplementation from inorganic and/or organic sources increased the Cu-excretion (from 808 to 1915-2164 mg per bird) and may cause additional environmental problems.

Studies with broiler chickens indicated that copper in organic form was more effective in stimulating growth at lower concentrations than copper sulphate (Pesti and Bakali, 1996; Ewing et al., 1998). In this experiment the Cu unsupplemented basal diet contained 11.3 mg Cu/kg was enough for turkeys and confirm the Cu-requirements for growing/fattening turkeys given by scientific bodies (e.g., NRC, INRA, GfE). Further Cu-supplementations from inorganic and/or organic sources did not significantly improve turkeys performance and health. Other authors (Guo et al., 2001; Banks et al., 2004a) reported also that copper source did not affect broiler performance.

The concentration of Cu in body tissues of mammals varies greatly with species and age. It is high in the livers and kidneys of cattle and sheep, and low in the same organs of swine and chickens (Doyle and Spaulding, 1978). Bones are tissues of intermediate Cu concentration and, like the liver and kidney, are particularly responsive to changes in Cu intake (Underwood, 1977). In the present study, tibia Cu concentration was found an increasing trend ( $P < 0.1$ ) in 56-day- and 126-day-old turkeys fed a Cu-AA chelate supplemented diet, compared to birds given a diet with Cu-sulphate. In an experiment performed by Banks et al. (2004b), bone mineralization results indicated that chickens fed diet with Cu-lysinate had the greatest ( $P \leq 0.05$ ) tibia and toe ash percentages and weights as compared with birds fed Cu-citrate or Cu-sulphate diets. Our results show that the tibia major mineral concentration (Ca and P) were unaffected ( $P > 0.1$ ) by dietary treatments both at days 56 and 126 of the experiment. Similarly, Mondal et al. (2007) reported no impact of dietary copper source on major mineral balance and plasma concentrations of such elements as Ca, P and Mg in broilers.

## CONCLUSIONS

In conclusion, the results of the present study confirm recommendations of scientific bodies concerning Cu-requirements of growing/fattening turkeys (e.g., NRC, INRA, GfE). The Cu unsupplemented basal diet contained 11.3 mg Cu/kg and further Cu-supplementations from inorganic and/or organic sources did not significantly improve turkeys performance and health. Plasma superoxide dismutase activity, physico-chemical properties of bone and the concentrations of Ca and P in the tibia bone were generally not affected by the copper source. There is no need for additional supplementation of inorganic or organic copper. Further Cu supplementation increase the Cu-excretion (from 808 to 1915-2164 mg per animal) and may cause additional environmental problems.

## REFERENCES

- Aoyagi S., Baker D.H., 1993. Bioavailability of copper in analytical-grade and feed-grade inorganic copper sources when fed to provide copper at levels below the chick's requirement. *Poultry Sci.* 72, 1075-1083
- Baker D.H., Odle J., Funk M.A., Wieland T.M., 1991. Research note: bioavailability of copper in cupric oxide, cuprous oxide and in copper lysine complex. *Poultry Sci.* 70, 177-179
- Banks K.M., Thompson K.L., Jaynes P., Applegate T.J., 2004a. The effects of copper on the efficacy of phytase, growth, and phosphorus retention in broiler chicks. *Poultry Sci.* 83, 1335-1341

- Banks K.M., Thompson K.L., Rush J.K., Applegate T.J., 2004b. Effects of copper source on phosphorus retention in broiler chicks and laying hens. *Poultry Sci.* 83, 990-996
- B.U.T. Commercial Performance Goals, 2002. British United Turkeys Ltd. 5th Edition. Waren Hall Broughton, Chester (England). Available at: <http://www.aviagen.com/home.aspx?siteId=8>
- Doyle J.J., Spaulding J.E., 1978. Toxic and essential elements in meat. *J. Anim. Sci.* 47, 398-419
- Du Z., Hemken R.W., Jackson J.A., Trammell D.S., 1996. Utilization of copper in copper proteinate, copper lysine, and cupric sulfate using the rat as an experimental model. *J. Anim. Sci.* 74, 1657-1663
- Ewing H.P., Pesti G.M., Bakallii R.I., Menten J.F., 1998. Studies on the feeding of cupric sulfate pentahydrate, cupric citrate, and copper oxychloride to broiler chickens. *Poultry Sci.* 77, 445-448
- Guo R., Henry P.R., Holwerda R.A., Cao J., Littell R.C., Miles R.D., Ammerman C.B., 2001. Chemical characteristics and relative bioavailability of supplemental organic copper sources for poultry. *J. Anim. Sci.* 79, 1132-1141
- Hemken R.W., Clark T.W., Du Z., 1993. Copper: its role in animal nutrition. In: T.P. Lyons (Editor). *Biotechnology in the Feed Industry*. Alltech Technical Publications. Nicholasville, KY (USA), pp. 35-39
- Hurwitz S., Bar A., 1965. Absorption of calcium and phosphorus along the gastrointestinal tract of the laying fowl as influenced by dietary calcium and egg shell formation. *J. Nutr.* 86, 433-438
- McDowell L.R., 1992. Copper and molybdenum. In: *Minerals in Animal and Human Nutrition*. Academic Press Inc. San Diego, CA, pp. 176-204
- Mondal M.K., Das T.K., Biswas P., Samanta C.C., Bairagi B., 2007. Influence of dietary inorganic and organic copper salt and level of soybean oil on plasma lipids, metabolites and mineral balance of broiler chickens. *Anim. Feed Sci. Tech.* 139, 212-233
- NRC, 1994. *Nutritional Requirements of Poultry*. 9th revised Edition. National Academy Press. Washington, DC
- Paik I.K., Seo S.H., Um J.S., Chang M.B., Lee B.H., 1999. Effects of supplementary copper-chelate on the performance and cholesterol level in plasma and breast muscle of broiler chickens. *Asian-Austr. J. Anim. Sci.* 12, 794-798
- Pesti G.M., Bakallii R.I., 1996. Studies on the feeding of cupric sulfate pentahydrate and cupric citrate to broiler chickens. *Poultry Sci.* 75, 1086-1091
- Savage J.E., Bird D.W., Reynolds G., O'Dell B.L., 1966. Comparison of copper deficiency and lathyrism in turkey poults. *J. Nutr.* 88, 15-25
- SCAN, 2003. Opinion of the Scientific Committee for Animal Nutrition on the Use of Copper in Feedingstuffs. [http://ec.europa.eu/food/fs/sc/scan/index\\_en.html](http://ec.europa.eu/food/fs/sc/scan/index_en.html)
- Underwood E.J., 1977. Copper. In: *Trace Elements in Human and Animal Nutrition*. 4th Edition. Academic Press. New York, NY
- Whiteside P., Miner B., 1984. *Pye Unicam Atomic Absorption Data Book*. Pye Unicam LTD. Cambridge (England)
- Woolliams J.A., Wiener G., Anderson P.H., McMurray C.H., 1983. Variation in the activities of glutathione peroxidase and superoxide dismutase in the concentration of copper in the blood in various breed crosses of sheep. *Res. Vet. Sci.* 34, 253-256