ISSN 1682-8356
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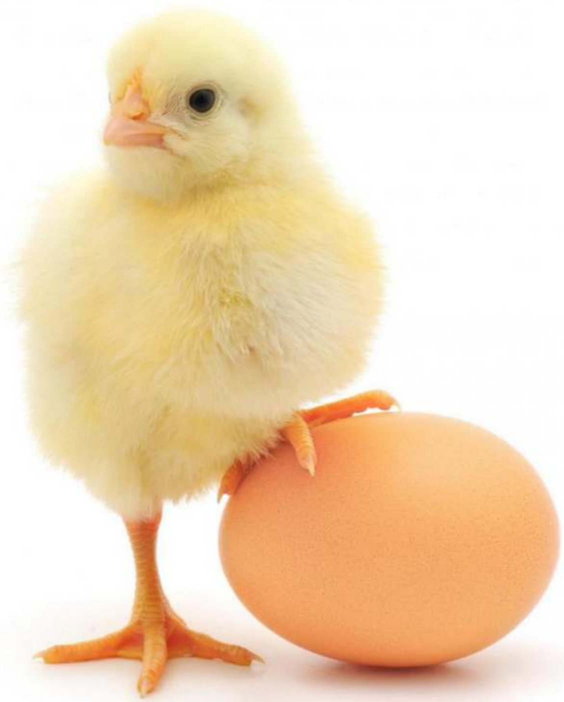

INTERNATIONAL JOURNAL OF POULTRY SCIENCE

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International Journal of Poultry Science 9 (8): 726-730, 2010
ISSN 1682-8356

Effects of Dietary Copper Source and Level on Growth, Organ Weights and Carcass Characteristics of Cherry Valley Meat Ducks

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Abstract: Tri-basic copper chloride (TBCC®, 58% copper; Micronutrients, Indianapolis, Indiana, USA) and copper sulfate pentahydrate (25% copper) were evaluated as dietary supplements at substantially higher than requirement levels for improving growth and carcass characteristics of ducks. Newly hatched Cherry Valley meat-strain ducklings (1,280) were assigned randomly to 4 treatment groups and fed a basal diet supplemented with 1 of the following: 1) 10 mg copper/kg diet from TBCC® (control; standard diets); 2) 150 mg copper/kg from copper sulfate pentahydrate; 3) 150 mg copper/kg from TBCC®; or 4) 0 mg added copper/kg (8.9 and 7.2 mg copper/kg in starter and grower by analysis) but with antibiotic growth promoter (40 mg zinc bacitracin and 40 mg garlicin/kg). Feed/gain ratios of high TBCC® and of antibiotic-fed ducks from 21-42 and 0-42 d were improved ($p = 0.045$; $p = 0.029$) vs. control ducks, with high copper sulfate pentahydrate results intermediate. The high TBCC® group had lower ($p = 0.045$) mortality % 21-42 d than the high copper sulfate pentahydrate group, with control or antibiotic-fed group results intermediate. For the entire trial (0-42d), feed/gain ratios of high TBCC® or antibiotic-fed groups were significantly improved ($p = 0.029$) compared to control group, with the high copper sulfate pentahydrate group intermediate. The high TBCC® (150 mg copper/kg of feed) significantly lowered feed/gain ratio of meat ducks compared with control (10 mg copper from TBCC®/kg of feed) during the starter, grower and entire trial periods and reduced mortality % compared with high copper sulfate pentahydrate (150 mg copper from copper sulfate pentahydrate/kg of feed) during the grower period.

Key words: Copper, duck, performance, carcass, organ weights, tri-basic copper chloride

INTRODUCTION

Copper (Cu) is an essential trace mineral for poultry which functions in numerous physiological processes primarily as a constituent of several enzyme systems. The Cu is involved in mitochondrial oxidative phosphorylation, free radical detoxification, neurotransmitter synthesis and denaturation, pigment formation, connective tissue synthesis and Iron (Fe) metabolism (Underwood and Suttle, 1999; Crisponi *et al*., 2010). The recommended Cu requirement for ducks is 8 mg/kg diet (Leeson and Summers, 1997), the same as for broilers (National Research Council, 1994); however, prophylactic high dosages, up to 250 mg/kg of Cu, have traditionally been added to broiler diets due to its antimicrobial and growth promoting effects (Pesti and Bakalli, 1996; Banks *et al*., 2004; Zhang *et al*., 2009b).

There is considerable interest in alternatives to antibiotics that can produce similar effects in terms of intestinal microflora regulation, as well as giving optimal animal performance. Besides being an essential nutrient, Cu has received considerable attention due to its antimicrobial properties and promotion of performance in animals when it has been fed at prophylactic levels (Zhou *et al*., 1994; Ibrahim *et al*., 2008; Pang *et al*., 2009).

Different sources or forms of Cu have different Cu bioavailabilities and effects on animals. For example, organic Cu and nanosized Cu have been shown to be more bioavailable than Cu sulfate pentahydrate (Du *et al*., 1996; Creech *et al*., 2004; Gonzales-Eguia *et al*., 2009), the most commonly used Cu source for supplementation in poultry diet is copper sulfate pentahydrate ($CuSO_4 \cdot 5H_2O$) due to its lower cost and commercial availability.

Tri-basic Cu chloride [$Cu_2(OH)_3Cl$; TBCC®) is a relatively new Cu product in China that is not soluble in water but soluble in acidic solutions (Pang and Applegate, 2007). The TBCC® is reported to be a safer Cu source, due to its lower toxicity, with higher Cu bioavailability in chicks than Cu sulfate (Miles *et al*., 1998; Luo *et al*., 2005). The TBCC® has been successfully evaluated in diets of laying hens (Banks *et al*., 2004; Liu *et al*., 2005) and broiler chickens (Miles *et al*., 1998; Luo *et al*., 2005; Pang *et al*., 2009). Although TBCC® has been proven beneficial for many animal species, it has apparently not been scientifically evaluated as a source of dietary Cu for ducks.

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Therefore, the purpose of this research was to evaluate and compare the effects of TBCC® relative to copper sulfate pentahydrate, with each at high levels (providing 150 mg Cu/kg of feed) of dietary supplementation, on performance, organ weights and carcass parameters of Cherry Valley meat ducks versus the premix copper supplementation of 10 mg per kg of feed typical of the application level in many Chinese feed mills. In an additional treatment, diets were unsupplemented with copper but contained an antibiotic (zinc bacitracin plus garlicin) growth promoter.

MATERIALS AND METHODS

Experimental design and dietary treatments: The experimental designs and procedures were approved by the Committee for Animal Care and Use of Nanjing Agricultural University following the requirements of the Regulations for the Administration of Affairs Concerning Experimental Animals of China as published by The State Science and Technology Commission in 1988.

A total of 1,280 newly hatched Cherry Valley meat-strain ducks were randomly allocated to four treatments for a 42-d feeding trial, including starter phase (0-21 d) and grower phase (21-42 d). Each treatment had 8 replicate pens with 40 straight-run ducklings per pen. Ducks were fed basal diets supplemented with either: 1) 10 mg of copper (Cu)/kg of diet from TBCC® (CON); 2) 150 mg of Cu/kg of diet from CuSO4 (high CuSO4; 3) 150 mg of Cu/kg of diet from TBCC® (high TBCC®); or 4) 0 mg added Cu/kg but with antibiotic growth promoter (AGP; 40 mg zinc bacitracin/kg and 40 mg garlicin/kg of diet). Tri-basic copper chloride ($Cu_2(OH)_3Cl$), commonly known as TBCC® (58% Cu), was provided by Micronutrients, Division of Heritage Technologies, LLC, Indianapolis, Indiana.

The starter and grower basal diets were obtained from a commercial feed company in Xuzhou City, Jiangsu Province, China. The formulas and calculated nutrient contents are shown in Table 1.

The diets were analyzed to contain 17.9, 142.6 and 165.6, 8.9 mg Cu/kg in starter and 15.7, 140.0, 158.8 and 7.2 mg Cu/kg in grower, respectively. The AGP starter and grower diets with 8.9 and 7.2 mg Cu/kg by analysis, but without added Cu, approximately met the estimated Cu requirement of 8 mg/kg of diet for ducks (Leeson and Summers, 1997).

Ducks were allowed *ad libitum* access to feed and water and were housed in an environmentally controlled room. The body weight, feed intake and mortality of ducks were recorded by pen. Outside temperatures during the trial often exceeded 33-35°C.

Live performance: Average body weight of ducks was determined at 0 d (the beginning of the trial), 21 d, and 42 d (the end of the experiment). Feed was withdrawn for 12 h with water being provided *ad libitum* before weighing ducks and feed at 21 d and 42 d.

Table 1: Formulas and calculated nutrient levels for basal starter and grower duck diets

Ingredients	Starter (0-21 d) (%)	Grower (21-42 d) (%)
Corn	41.00	32.00
Soybean meal	23.20	6.00
Cottonseed meal	5.00	8.00
Wheat middling and red dog	10.00	18.00
Rice bran	7.00	20.00
Corn germ meal	5.00	8.00
Corn gluten meal	2.60	1.60
Soybean oil	1.50	2.50
Limestone	1.26	1.46
Dicalcium phosphate	1.80	0.83
L-Lysine•HCl	0.20	0.24
dl-Methionine	0.14	0.07
Vitamin and trace mineral premix[1]	1.00	1.00
Salt	0.30	0.30
Total	100.00	100.00
Calculated nutrient levels		
Metabolizable energy (kcal/kg)	2,870.00	2,920.00
Crude protein (%)	21.40	16.88
Calcium (%)	1.02	0.84
Available phosphorus (%)	0.42	0.30
Lysine (%)	1.15	0.89
Methionine + Cysteine (%)	0.80	0.64

[1]Premix provided per kg of diet: Transretinyl acetate, 20 mg (vitamin A 10,000 IU); cholecalciferol, 6 mg (vitamin D3 3,000 IU); all-rac-α-tocopherol acetate (vitamin E), 30 mg; menadione, 1.3 mg; thiamine, 2.2 mg; riboflavin, 10 mg; nicotinamide, 50 mg; choline chloride, 600 mg; calcium pantothenate, 12 mg; pyridoxine•HCl, 4 mg; biotin, 0.04 mg; folic acid, 1.5 mg; vitamin B12 (cobalamin), 0.012 mg; Fe (from ferrous sulfate), 100 mg; Mn (from manganese sulfate), 80 mg; Zn (from zinc oxide), 60 mg; I (from calcium iodate), 1 mg; Se (from sodium selenite), 0.3 mg

The feed/gain ratio was calculated. Average body weight, average daily gain, and feed/gain ratio were used to assess the live performance of ducks by dietary treatment.

Organ weights and carcass characteristics: Organ weights and carcass parameters were measured by slaughter and processing. At 42 d, 8 ducks per treatment were weighed after feed deprivation for 12 h, then feather-scalded and blood removed by exsanquination to obtain dressed weight. The dressed (bled-defeathered) yield was defined as the percentage of dressed weight relative to live body weight. The eviscerated weight was defined as the weight of eviscerated carcass (blood, heart, abdominal fat, etc. removed; with head, neck, and kidney kept), and the eviscerated yield was eviscerated weight relative to dressed weight. The relative weights of liver, spleen, thymus and bursa were calculated as organ weight (g)/body weight (kg), which is equivalent to % of body weight.

Statistical analysis: Data were analyzed by one-way ANOVA with 4 treatments and 8 replicate pens each. Least Significant Difference (LSD) procedure was used to determine whether means were significantly different between treatments. Significance (p-value) was evaluated at the 0.05 level. The means and total Standard Errors (SEM) are presented. All the statistical analyses were performed using SPSS statistical software (version 13.0 for windows, SPSS).

RESULTS

The live performance responses of ducks to the different copper (Cu) sources and levels are shown in Table 2. Compared with CON, the feed/gain ratios of ducks fed high TBCC® or the AGP diets from 22-42 d were significantly improved (p = 0.045), with high $CuSO_4$ results intermediate. The 22-42 d mortality in the high TBCC® group was significantly lower (p = 0.049) than in the high $CuSO_4$ group, whereas CON and AGP results were intermediate. For the entire trial (0-42 d), feed/gain ratios of the high TBCC® and the AGP groups were significantly improved (p = 0.029) compared to CON group, with the high $CuSO_4$ and the AGP groups intermediate.

The relative weights (% of body weight) of liver, thymus, spleen and bursa of ducks at the end of the starter (21 d) and grower (42 d) periods were not affected by dietary treatments (p>0.05, Table 3). At 42 d, ducks from the AGP group (0 mg added Cu/kg diet) had lower breast muscle yield compared with CON or high $CuSO_4$ treatment results (p = 0.043), however, breast muscle yield was not different among CON, high $CuSO_4$ and high TBCC® treatments (p>0.05).

DISCUSSION

Previous researchers have reported that dietary copper (Cu) enhances performance in poultry when fed at prophylactic dosages over the minimum nutritional requirement (Pesti and Bakalli, 1996; Zhang *et al*., 2009a). The feed intakes and body weights of 21-day-old chickens fed 300 or 450 mg Cu/kg diets from TBCC® were increased compared to 300 or 450 mg Cu/kg levels from Cu sulfate but were similar at dietary concentrations of 150 mg/kg (Miles *et al*., 1998). Luo *et al*. (2005) demonstrated that the average daily gain and average daily feed intake of broilers were not significantly but higher than chicks consuming Cu sulfate with the dose of supplemental 150 or 300 mg Cu/kg diets, whereas chickens fed TBCC® had significantly higher average daily gain or average daily feed intake than from Cu sulfate at 450 mg Cu/kg dose (p<0.05). However, there were also reports which showed that the effects on performance of birds given TBCC® did not differ significantly from that of Cu sulfate at high dose (Banks *et al*., 2004; Pang and Applegate, 2007; Zhang *et al*., 2009b). Actual results in a given situation may be related to the type of poultry, age, doses, feed and microbial environment which affect the antimicrobial effects of Cu or antibiotics (Arias and Koutsos, 2006).

Ducks fed diets with the combination antibiotic growth promoter (AGP group) had the lowest breast muscle yield at 42 d. No significant effects on carcass characteristics of ducks were observed among the different Cu treatments. This was consistent with results observed in previously reported studies. Arias and Koutsos (2006) found that carcass weights of 45-day-old chickens fed 188 mg/kg Cu from TBCC® were heavier than when chickens were fed control diets containing 8 mg/kg Cu. Zhang *et al*. (2009b) reported that the eviscerated yield of broilers fed diets containing 50 mg Cu/kg from TBCC® was higher than that of broilers fed diets with 50 mg added Cu/kg from Cu sulfate.

Pharmacological levels of Cu may be beneficial for altering intestinal microbiota, increasing digestibility of other nutrients and/or improving gastrointestinal

Table 2: Effects of dietary copper (Cu) source and level on live performance of ducks[1]

	CON	High $CuSO_4$	HighTBCC®	AGP	SEM	p-value
Starter (0-21 d)						
Body wt (g; 21 d)[2]	1,021.00^a	1,031.00^a	1,032.00^a	1,032.00^a	2.81	0.450
ADG (g/bird/day)[2]	45.95^a	46.41^a	46.46^a	46.49^a	0.13	0.449
Feed/gain ratio (g/g)[2]	1.70^a	1.72^a	1.69^a	1.70^a	0.01	0.330
Mortality (%)	1.79^a	1.25^a	1.07^a	2.14^a	0.48	0.870
Grower (22-42 d)						
Body wt (g; 42 d)	2,914.00^a	2,931.00^a	2,942.00^a	2,914.00^a	8.43	0.595
ADG (g/bird/day)	94.66^a	95.01^a	95.50^a	94.08^a	0.38	0.631
Feed/gain ratio (g/g)	2.45^a	2.44^{ab}	2.37^b	2.38^b	0.01	0.045
Mortality (%)	1.25^{ab}	1.61^a	0.00^b	0.18^{ab}	0.29	0.049
Whole period (0-42 d)						
ADG (g/bird/day)	69.71^a	70.12^a	70.38^a	69.70^a	0.21	0.594
Feed/gain ratio (g/g)	2.21^a	2.20^{ab}	2.15^c	2.16^{bc}	0.01	0.029
Mortality (%)	3.04^a	2.86^a	1.07^a	2.32^a	0.57	0.636

$^{a-c}$Means with different superscript letters in a row differ significantly at p-value indicated.

[1]A total of 1,280 ducklings were allocated to 4 treatments with 8 replicate pens and 40 ducklings per pen. CON indicates basal diets + 10 mg Cu from TBCC®/kg. High $CuSO_4$ indicates basal diets + 150 mg Cu from $CuSO_4$/kg. High TBCC® indicates basal diets + 150 mg Cu/kg from TBCC®. AGP indicates basal diets (0 mg added Cu/kg) + zinc bacitracin and garlicin at 40 mg/kg each.

[2]Body wt is body weight. ADG is average daily gain

Table 3: Effects of dietary copper (Cu) source and level on organ relative weights (% of body weight) and carcass characteristics of ducks at 21 and 42 d of age[1]

	CON	High $CuSO_4$	High TBCC®	AGP	SEM	p-value
End of starter period (21 d)						
Liver (% of body weight)	30.18[a]	31.57[a]	29.83[a]	30.09[a]	0.61	0.795
Thymus (% of body weight)	4.31[a]	4.50[a]	4.50[a]	4.22[a]	0.15	0.906
Spleen (% of body weight)	1.07[a]	1.06[a]	1.01[a]	1.08[a]	0.04	0.946
Bursa (% of body weight)	1.41[a]	1.46[a]	1.38[a]	1.29[a]	0.03	0.355
End of grower period (42 d)						
Liver (% of body weight)	20.32[a]	21.79[a]	21.19[a]	21.65[a]	0.42	0.639
Thymus (% of body weight)	3.09[a]	3.13[a]	3.54[a]	3.41[a]	0.15	0.712
Spleen (% of body weight)	0.61[a]	0.68[a]	0.70[a]	0.71[a]	0.03	0.605
Bursa (% of body weight)	0.90[a]	0.94[a]	0.92[a]	0.88[a]	0.03	0.958
Dressed yield (% of body wt)[2]	88.60[a]	89.02[a]	88.94[a]	88.92[a]	0.12	0.674
Eviscerated yield (% DY)[2]	77.31[a]	77.50[a]	77.36[a]	76.79[a]	0.15	0.416
Breast muscle yield (% DY)	10.81[a]	10.87[a]	10.23[ab]	9.76[b]	0.18	0.043

[a-c]Means with different superscript letters in a column differ significantly at p-value indicated.
[1]CON indicates basal diets + 10 mg Cu from TBCC®/kg. $CuSO_4$ indicates basal diets + 150 mg Cu from $CuSO_4$/kg. TBCC indicates basal diets + 150 mg Cu/kg from TBCC®. AGP indicates basal diets (0 mg added Cu/kg) + zinc bacitracin and garlicin at 40 mg/kg each. [2]Dressed yield % of body weight (abbreviated % DY) is weight with blood and feathers removed (after scalding) divided by body weight, then x 100. Eviscerated yield % DY is weight with blood, feathers, heart, adominal fat, lungs and so on removed but with head, neck and kidneys kept divided by dressed (bled-defeathered) weight, then x 100

physiology. Pang *et al.* (2009) found that supplementation with TBCC® at 187.5 mg Cu/kg dosage significantly increased the similarity coefficients of microbiota (by the molecular technique of DGGE) in the ileal mucosa compared with a similar level of Cu from Cu sulfate which suggested that TBCC® may alter the microbial community associated with the ileal mucosa in broiler chickens yet did not significantly influence live performance or ileal phosphorus digestibility in their trial. Arias and Koutsos (2006) reported that chicks fed TBCC® or positive control (antibiotic) diets had better crypt depth in the ileum compared with those fed control or Cu sulfate, and a greater quantity of duodenum intraepithelial lymphocytes compared with the negative control.

The effects of TBCC® on poultry may be due to particular chemical characteristics of Cu source. The TBCC® is almost totally insoluble in water compared with solubility of > 99% for Cu sulfate, yet the TBCC® is highly (100%) soluble in weak acid (0.4% HCl, 2% citric acid, or neutral ammonium citrate). This characteristic should result in complete solubility of TBCC® in an animal's gut. The TBCC® has low hygroscopicity and very low chemical reactivity which is a benefit in vitamin and trace mineral premixes or in complete feeds.

In conclusion, high TBCC® (150 mg copper/kg of feed) significantly lowered feed/gain ratio of meat ducks compared with control (10 mg copper from TBCC®/kg of feed) during the starter, grower, and entire trial periods and reduced mortality % compared with high copper sulfate pentahydrate (150 mg copper from copper sulfate pentahydrate/kg of feed) during the grower period. It was demonstrated that TBCC® is a safe and effective dietary copper source when added to ducks diets at a high Cu dose (150 mg/kg feed).

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