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Potential Effects of Different Dietary Copper Sources to Improve Productive Performance, Plasma Biochemical Parameters and Oxidative Response Activities of Broiler Chickens

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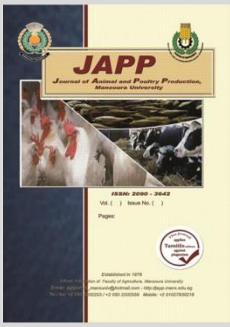

ABSTRACT

This investigation was established to assess the effect of dietary different copper sources and levels supplementation on growth performance, plasma biochemical parameters and antioxidant activities of broiler chickens. Four hundred and twenty-one-day-old chicks (Ross 308) were randomly allocated into 7 experimental treatment (n= 60 birds per each).At the end of the experiment, results obtained revealed that birds fed basal diet with inorganic or organic Cu had significantly ($P < 0.05$) better live body weight, weight gain and feed conversion ratio, while feed intake was reduced comparison with control group. Chicks supplemented with organic CuNo3 significantly ($P < 0.05$) improved growth performance s compared with the other groups. Considerable reductions in plasma total cholesterol and triglyceride, high density lipoprotein cholesterol (HDL-C), low density lipoprotein cholesterol (LDL-C) were detected in the group that fed diet with 125 mg / kg diet Cu in comparison with other diets. Plasma glutathione (GPx), superoxide dismutase (SOD) and total antioxidant activity (T-AOC) were significantly enhanced with Cu-supplemented diets, while malondialdehyde (MDA) was significantly declined in the experimental groups compared with the control group. In conclusion, Cu supplemented at level of 125 mg/ kg diet either copper nitrate or copper sulfate improved the productive performance, profile of lipid metabolism and oxidative response enzymes of broiler chickens. Broilers fed organic form of copper (copper nitrite) had better growth and immune response than those fed inorganic form of copper (copper sulfate) sulfate.

***Keywords**:* copper source, performance, plasma lipid profile, antioxidant system, broilers.

INTRODUCTION

Poultry sector contributes a critical role in reducing starvation, poverty, as well as encouraging economic progress in developing countries, since meats and eggs are extremely valuable in providing income and high-quality animal protein (Herrero *et al.*, 2013, Smith *et al.*, 2013, El-Attrouny *et al.*, 2021) . Furthermore, white meat is conceding a good source of essential vitamins and minerals and is rich with unsaturated fatty acids like; Omega 3 and Omega 6 (Farrell, 2013; Brenes and Roura 2010 and Ahmed *et al.*, 2015).

Addition of microelements in poultry feed is a crucial issue and have effectiveness for health and growth performance. All animals and birds depend on microelements for growth and various metabolic activities because they serve as catalysts or components which used in in the enzymatic systems of numerous cells. (Świątkiewicz *et al.*, 2014, Khalil *et al.*, 2019, Alagawany *et al.*, 2021). The accessibility of minerals from traditional inorganic sources, such as oxides, sulphates, or carbonates, as well as from feed materials derived from plants, is relatively low, whereas the requirements of contemporary, high-producing lines of laying hens and broiler chickens for microelements are very high. These details, in addition to the most recent understanding of the significance of microelements in immune response (Saripinar-Aksu *et al.*, 2012, Świątkiewicz *et al.*, 2014, Mateos *et al.*, 2020).

Copper (Cu), is a vital microelement in poultry diets and is essential to preserve the proper physiological functions of the body in chickens, Cu is worked as a growth promoter and/or an antimicrobial agent (Świątkiewicz *et al.*, 2014, Sharif *et al.*, 2021). Cu generates the active centers of various enzymes including; superoxide dismutase, bilirubin oxidase, and lysis oxidase, which are crucial to the body's normal operation. (Makarski and Zadura 2006, Lutsenko *et al.*, 2007, Festa *et al.*, 2011). The main site of nutritional absorption of Cu stretches from the stomach to the distal small intestine Wapnir (1998). Cu is regularly added to chicken diets at high concentrations, frequently in amounts higher than what the birds actually need Świątkiewicz *et al.*, (2014), as a potential option an antibiotic growth promoter.

When Cu is given to broilers at significantly higher therapeutic levels, i.e. 125 to 250 ppm, it improves growth performance and feed efficiency (Samanta *et al.*, 2011). However, too much copper in the diet can also be harmful. Also, Cu has a significant impact in lowering broiler meat and plasma cholesterol levels. (Kim *et al.*, 1992). Therefore, the purpose of the current study was to ascertain the impact of dietary different copper sources with different levels on growth performance, feed utilization, plasma biochemical parameters and oxidative response activities of Ross 308 broiler chicken.

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MATERIALS AND METHODS

Ethics approval and Experimental Location

The Poultry Research Farm, Faculty of Agriculture at Benha University, Egypt served as the study's location. The Institutional Animal Care and Use Committee (IACUC) at Benha University authorized all experimental protocols. During the period from 21 May to 26 June 2021.

Experimental animals and design:

A total number of 420 Ross-308 one-day broiler chicks with nearly live body weight were randomly assigned into three groups each of 120 chicks in three replicates (20 chicks per each) with 10 birds / m^2 stocking density. Chicks of the first group were fed the starter and grower diets (considered as a control group). Birds of the second group were fed the starter and grower basal diets supplemented with copper nitrate [organic copper 31% copper] in form powder of Indian origin, produced by a company HARYANA) at a level of (75, 100, 125 mg/kg diet). While, chicks of the 3rd group were fed starter and grower basal diets supplemented with copper sulfate [Inorganic copper 39% copper] at a level of 75, 100, 125 mg/kg diet.

Chicks of all experimental treatments were kept under similar management, hygienic and environmental conation in separate pens until the end of the experiment. Wood shaving was used at 10 cm depth as a litter. Artificial lighting was provided over 24 hours daily during the first 5 days of age, then from 6 to 35 days of age 23 hours florescent light :1-hour dark was applied. Floor brooder with gas heaters were used to provide chicks with heat needed for brooding. Brooding temperature was maintaining at 35 ^{O}C during the first 5 days of chicks age then decreased by 2 ^{O}C weekly until the end of brooding period.

Water and respective feed were available at any time. The experimental diets were formulated according to NRC (1994) to meet standards nutritional requirements of chicken during the starter (1–21 days) and grower periods (22-35 days). Table 1 displays the composition and computed nutritional content of the experimental diets.

Table 1. The ingredients and nutrients composition of the basal starter and grower diets (DM basis).

Ingredients (%)	Starter (1-21 days)	Gower (22-35 days)
Corn	58.00	63.00
Soybean (44% CP)[a]	28.20	24.90
Gluten (60% CP)[b]	7.00	6.0
Dicalcium phosphate	1.80	1.20
Limestone	1.30	1.30
Soybean oil	2.00	2.00
Salt	0.30	0.30
L-Lysine HCl	0.20	0.20
DL-Methionine	0.20	0.10
Premix[c]	1.00	1.00
Total	100	100
Calculated composition		
ME (kcal/kg)[d]	3012.26	3068.97
Crude protein (%)	21.99	20.28
Calcium (%)	0.98	0.84
Total phosphor (%)	0.72	0.59
Methionine (%)	0.59	0.47
Methionine C cysteine (%)	0.95	0.80
Lysine (%)	1.14	1.06

[a,b]Crude protein. [c]Provided the following per kilogram of diet: 13,000 IU of vitamin A; 1,300 IU of vitamin D; 65 IU of vitamin E; 3.4 mg of menadione; 37 mg of pantothenic acid; 6.6 mg of riboflavin; 3.7 mg of folic acid; 39 mg of niacin; 1.0 mg of thiamine; 4.3 mg of vitamin B6; 0.23 mg biotin; 0.075 mg of vitamin B12: 43 mg of choline chloride. 170 mg of zinc; 140 mg of iron; 34 mg of manganese; 16 mg of copper; 0.29 mg of iodine; 0.29 mg of selenium. [d]Metabolizable energy.

Data collection

Parameters estimate and data collection

Growth performance: Chicks of all groups were weighed individually in the start and the end of the experimental period (5 weeks). The average feed intake (AFI), weight gain (ABWG) were recorded throughout the trial period (1-5 wks. of age) and feed conversion rate (FCR) were calculated according to the following formulas; FCR = AFI /ABWG.

Blood parameters

Blood samples (n = 5 per group) were extracted from the bird's wing at the ending of the experiment (5 weeks old), and they were put in heparinized tubes. Blood plasma was produced after centrifugation at 3500 rpm for 10 min at 4°C, and it was stored in a deep freezer at about -20°C until it was time for chemical analysis. Colorimetric method by means of commercial kits were employed for determination of plasma Protein fractions (total protein, albumin, globulin and A/G ratio), Liver function tests (aspartate aminotransferase (AST), alanine aminotransferase (ALT)), Kidney function tests (creatinine and uric acid), Lipid profile (triglycerides, total cholesterol, low density lipoproteins (LDL) and high density lipoproteins (HDL) and Measurements of antioxidant capacities in plasma total antioxidant capacity (T-AOC), glutathione and peroxidase (GPX), superoxide dismutase (SOD).

Statistical analysis:

Two-way ANOVA was used to examine the data, using the General Linear Models (GLM) method of (SAS, 2004). A factorial design (2×3) was used to assess the interaction between the main effect's elements (dietary copper source and amount). Duncan's multiple range test was applied to evaluate differences among treatments, where significant differences (Duncan 1955). According to the following liner model:

$$X_{ijk} = \mu + S_i + L_j + (SL)_{ij} + e_{ijk}$$

Whereas: **Xijk = the observation of traits for ijkth birds; μ = the overall mean.; Si = the effect of the ith copper sources; Lj = the effect of the jth copper levels.; (SL)ij = the fixed effect of the interaction between the ith sources and the jth levels.; eij = random error assumed to be independently and randomly distributed.**

RESULTS AND DISCUSSION

Growth performance

Results obtained in Table (2) showed significant *(P<0.05)* variations in body weight (BW) and average weight gain (AWG) for chicks fed experimental diets due to the effect of copper source. No significant (P > 0.05) variations were found in feed intake (FI) and feed conversion ratio (FCR). The highest averages of BW and AWG were recorded in chicks fed diet supplemented with copper nitrate. Compared with group supplanted with copper sulfate and control, FI and FCR were improved in chicks fed diet supplemented with copper nitrate.

Regardless to the effect of the copper levels, the levels used had significant effect on BW, BWG, FI and FCR. BW and BWG increased with increasing the level of copper supplementation. While, the rate of fed intake decreased and feed conversion ratio was improved by increasing the level of copper supplementation .It is clearly observed that broilers chicks fed diet supplemented with 125 mg/kg diet either copper sulfate or copper nitrate significantly increased BW, BWG and improved FCR followed by those fed diet supplemented 100 mg/kg diet, when compared with those fed 75 mg/kg diet and control group.

The results obtained may be attributed to Cu's growth-promoting properties are linked to the growth hormone axis (Yang *et al.,* 2011) and hypothalamic appetite regulating genes (Zhu *et al.,* 2011). Moreover, Cu has been demonstrated to promote mitogen activity and increase the quantity of expression of growth hormone (GH) mRNA in the pituitary glands. On the other hand, GH levels is known to be influenced by a variety of circumstances. Cu has the ability to effect GH gene expression either directly on pituitary cells or indirectly on factors that influence GH gene expression have been reported by (Prashanth *et al.,* 2015). The higher growth performance and feed utilization in this study may be due to copper supplementation effects with different forms, which could be decreased number of pathogenic bacteria in the intestine (Xia *et al.,* 2004). Jegede *et al.,* (2011) clarified that Cu proteinate is superior to CuSO4 in terms of growth support, as evidenced by the significantly higher final live weight, body weight gain, and FCR improved seen in broilers fed copper proteinate.

Table 2. Live bodyweight, weight gain, average feed intake and feed conversion as affected by dietary supplementation of copper sources and levels

sources	Levels (mg/kg diet)	BW (g) at day 1	BW (g) at 35 day	BWG (g) at 1-35 day	FI (g/bird) at 1-35 day	FCR (g/bird) at 1-35 day
Copper sources	Cu nitrate	44.2±0.34	2042.7±18.9^{ab}	1998.5±18.9^{ab}	3184.9±11.1	1.59±0.02
	Cu sulfate	43.6±0.35	1999.8±19.4^{b}	1956.1±19.4^{b}	3189.8±11.1	1.62±0.02
Copper levels	0	43.43±0.64	1915.5±35.4^{b}	1872.1±33.9^{b}	3370.1±11.3^{a}	1.85±0.006^{a}
	75	43.72±0.35	1940.4±19.4^{b}	1896.99±23.0^{b}	3225.0±13.8^{b}	1.68±0.008^{b}
	100	44.16±0.35	2052.2±19.4^{a}	2007.9±22.4^{a}	3194.2±13.8^{b}	1.59±0.008^{c}
	125	43.98±0.35	2069.2±19.4^{a}	2025.2±22.4^{a}	3150.5±13.8^{c}	1.55±0.008^{d}
Control	0	43.43±0.65	1862.5±34.7^{b}	1819.4±34.7^{b}	3375.1±11.7^{a}	1.85±0.007^{a}
Cu nitrate	75	44.11±0.60	2007.0±32.5^{a}	1962.9±32.1^{a}	3228.0±15.1^{b}	1.64±0.007^{c}
	100	44.03±0.64	2060.3±32.5^{a}	2016.3±32.6^{a}	3193.2±15.1^{bc}	1.58±0.007^{d}
	125	44.47±0.60	2062.0±32.5^{a}	2017.5±32.6^{a}	3133.5±15.1^{d}	1.55±0.007^{e}
Cu sulfate	75	42.93±0.61	1915.5±32.5^{b}	1872.1±34.7^{b}	3214.7±15.1^{b}	1.72±0.007^{b}
	100	44.44±0.61	2044.0±32.5^{a}	1999.6±32.6^{a}	3197.5±15.1^{bc}	1.59±0.007^{d}
	125	43.56±0.61	2076.4±32.5^{a}	2032.8±32.6^{a}	3157.2±15.1^{cd}	1.56±0.007^{e}
			Two-way ANOVA			
Source	-	P=0.5704	P=0.0455	P=0.0312	P=0.7449	P=0.3133
level	-	P=0.7126	P=0.0041	P=0.0001	P=0.0001	P=0.0001
Source x level	-	p= 0.7251	P=0.0501	P=0.0001	P=0.0001	P=0.0001

[a,b,c]Means with different superscript in the same column are significantly different at (P<0.05).

Moreover, broiler chicks fed diets supplemented with different sources and levels of Cu displayed decreased feed consumption and improved FCR when compared with the control group, with significant differences shown by those fed different levels of Cu. Furthermore, the best level of copper sources (copper nitrate or sulfate) was 125 mg/kg diet. No significant (P > 0.05) effects were found in AFI and FCR due to copper sources (Table 2).The results obtained of AFI are consistent with those reported by Idowu *et al.* (2006), who reported that increasing Cu level significantly decreased feed intake. El-Husseiny *et al.* (2012) recorded that organic trace mineral (Cu) supplementation significantly (P < 0.001) boosted FI in the second week of birds age. However, Organic Trace Mineral (OTM) supplementation lowered FI during the third week of the experiment. Santos *et al.* (2020) noticed that, birds which treated with 150 ppm Cu had significantly lower feed intake compared to birds control. On the other hand, Abdallah *et al.* (2009) noticed that chicks fed meals containing 100 percent organic minerals; Zn, Cu, Mn, and Fe) had the better FCR than those fed diets supplemented with inorganic minerals. Gheisari *et al.* (2010) conceived that when broiler diets were supplemented with 50% of their Cu requirements as OTM, FCR were enhanced compared to adding 100% of those mineral recommendations. El-Husseiny *et al.* (2012) indicated that organic trace mineral (Cu) supplementation significantly (P < 0001) enhanced FCR in the second and third weeks of age. The 50 % Cu organic (ORG) diet improved FCR considerably (P < 0.001) in the 4th and 5th weeks of the experimental period, respectively.

Blood biochemical parameters

Protein fractions

Inspecting of data obtained Table 3 revealed no significant (P > 0.05) variations were found in plasma total protein fractions parameters, except plasma levels of globulin due to copper sources applied (P>0.05). Treating birds with CuSo4 increased plasma level of total protein (TP), albumen (AL) and globulin (GL) and A/G ratio as compared with birds fed diet supplemented with CuNO3. On the other hand, levels of dietary Cu applied showed highly significant (P < 0.001) effect on plasma levels of total proteins, albumen, globulin and A/G ratio (Table. 3). Birds fed diet supplemented with 125 mg/kg diet recorded the higher values of TP, AL, GL and A/G ratio followed by those fed diets supplemented with 100 mg/kg diet as compared with different levels applied and control. This indicated that plasma protein increased with increasing Cu level . Also, specify that Cu levels applied showed a magnitude of change in plasma level of protein fractions. Results obtained showed significantly increased in plasma total protein, albumin, A/G ratio and globulin according to the interaction between CuSo4 and CuNo3 × 125 mg/kg diet, respectively.

These improvements in plasma might be due to the antibacterial properties of Cu, which reduces pathogens and boosts good bacteria. (Waldroup *et al.,* 2003, Pang *et al.,* 2009). The outcomes were consistent with those established by Jegede *et al.* (2012), who discovered that serum total protein, albumin, globulin, and creatinine levels were not significantly impacted by the Cu source.

Table 3. plasma protein fractions (total protein, albumin, globulin and A/ G ratio) as affected bydietary supplementation of copper sources and levels

Sources	Levels (mg/kg diet)	Plasma (g / dl)			
		Total Protein	Albumin	Globulin	A/G ratio
Copper sources	Cu nitrate	7.15±0.10	4.26±0.03	2.89±0.07	1.63±0.04
	Cu sulfate	6.88±0.10	4.20±0.03	2.68±0.07	1.59±0.04
Copper level	0	$6.12±0.05^{d}$	$3.84±0.03^{d}$	$2.28±0.05^{c}$	$1.72±0.03^{a}$
	75	$6.65±0.05^{c}$	$4.12±0.03^{c}$	$2.54±0.05^{b}$	$1.68±0.03^{a}$
	100	$7.06±0.05^{b}$	$4.22±0.03^{b}$	$2.84±0.05^{a}$	$1.50±0.03^{b}$
	125	$7.34±0.05^{a}$	$4.35±0.03^{a}$	$2.99±0.05^{a}$	$1.65±0.03^{a}$
Control	0	$6.12±0.03^{g}$	$3.84±7.6^{e}$	$2.28±0.29^{f}$	$1.72±0.03^{a}$
Cu nitrate	75	$6.75±0.03^{e}$	$4.22±7.6^{c}$	$2.54±0.29^{e}$	$1.74±0.29^{a}$
	100	$7.24±0.03^{b}$	$4.22±7.6^{c}$	$3.02±0.29^{b}$	$1.40±0.29^{c}$
	125	7.46±0.03a	$4.34±7.6^{b}$	$3.12±0.29^{a}$	$1.75±0.29^{a}$
Cu sulfate	75	$6.54±0.03^{f}$	$4.02±7.6^{d}$	$2.54±0.29^{e}$	$1.74±0.29^{a}$
	100	$6.88±0.03^{d}$	$4.22±7.6^{c}$	$2.66±0.29^{d}$	$1.60±0.29^{b}$
	125	$7.22±0.03^{c}$	$4.36±7.6^{a}$	$2.86±0.29^{c}$	$1.56±0.29^{b}$
Two-way ANOVA					
Source	-	P=0.073	P=0.2776	P=0.0576	P=0.5539
level	-	P=0.0001	P=0.0001	P=0.0001	P=0.0098
Source x level	-	p= 0.0001	P=0.0001	P=0.027	P=0.0001

[a,b,c]Means with different superscript in the same column are significantly different at (P<0.05).

According to Attia *et al.* (2011), there was a strong association between the interaction impact of Cu sources and its levels for plasma total protein and globulin concentration, demonstrating that breeders fed 120 ppm of OCu had significantly higher values than those fed the same dose of ICu. El-Hady and Mohamed (2019) noticed that Cu supplementation dramatically raised globulin levels while, having no discernible effects on total protein or albumin. On the other hand, Attia *et al.* (2011) and Kumar *et al.* (2013) who observed that in chickens fed diet supplemented with 100–400 mg/kg diet CuSO4, could increase the plasma total protein.

Liver and kidney functions

The results of the blood analysis are considered to be within the normal range for a healthy chicken. Cu sources had no significant effects on blood constituents AST, ALT, uric acid and creatinine (Table 4) .Significant ($P < 0.05$) differences were found in biochemical blood values between chicks fed diets supplemented with Cu at different levels. The highest values of AST, ALT, uric acid and creatinine were observed in control group without Cu supplementation while, the lowest values were recorded in the groups fed diet supplemented with 125 mg/kg diet Cu nitrate and sulfate.

Broilers fed diets supplemented with inorganic CuSo4 and organic CuNO3 at different levels had significantly lower values of AST, ALT, uric acid and creatinine than the control group, with the lowest biochemical blood values observed in CuNo3 which was supplemented Cu 125 mg/kg diet compared to the other treated groups (Table 4). Other treatments applied showed lower biochemical blood levels compared to the control group. The supplementation of Cu reduced the activity of the enzyme AST and ALT levels compared with their activity in control group, which appears to emphasize the condensation of the essential amino acids necessary for the synthesis of proteins. These results were in agreement with the findings of Tayeb and Qader (2012); Jegede *et al.* (2012); Kwiecien *et al.* (2014) they reported significant differences ($P < 0.05$) in blood parameters between treatment groups supplemented with Cu and control group. Attia *et al.* (2011) and Kumar *et al.*, (2013) they observed that in the chickens fed diet supplemented with 100–400 mg/kg diet CuSO4 could be improved plasma albumin.

Table 4. Liver enzymes (AST & ALT) and kidney functions (uric acid & creatinine) as affected by dietary supplementation of copper sources and levels

sources	Levels (mg/kg diet)	Plasma (U/L)		Plasma (mg/dl)	
		AST	ALT	Uric acid	Creatinine
Copper sources	Cu nitrate	$26.40±0.34^{ab}$	26.40±0.26	18.0±0.32	0.79±0.02
	Cu sulfate	$27.41±0.34^{a}$	27.41±0.26	18.61±0.32	0.84±0.02
Copper levels	0	$32.20±0.58^{a}$	$19.88±0.04^{a}$	$20.14±0.58^{a}$	$1.01±0.04^{a}$
	75	$27.05±0.33^{b}$	$17.49±0.02^{b}$	$18.56±0.33^{b}$	$0.85±0.02^{b}$
	100	$25.85±0.33^{bc}$	$16.48±0.02^{b}$	$17.88±0.33^{bc}$	$0.77±0.02^{bc}$
	125	$24.74±0.33^{c}$	$15.30±0.02^{c}$	$17.28±0.33^{c}$	$0.69±0.02^{c}$
Control	0	$32.20±0.42^{a}$	$19.88±0.23^{a}$	$20.14±0.55^{a}$	$1.01±0.04^{a}$
Cu nitrate	75	$27.50±0.42^{b}$	$17.86±0.23^{c}$	$18.58±0.55^{abc}$	$0.88±0.04^{abc}$
	100	$26.52±0.42^{cd}$	$16.92±0.23^{d}$	$18.00±0.55^{bcd}$	$0.78±0.04^{cd}$
	125	$25.20±0.42^{de}$	$15.38±0.23^{gh}$	$17.42±0.55^{bcd}$	$0.71±0.04^{de}$
Cu sulfate	75	$28.96±0.42^{b}$	$18.78±0.23^{b}$	$19.18±0.55^{ab}$	$0.92±0.04^{ab}$
	100	$27.16±0.42^{cd}$	$17.54±0.23^{cd}$	$18.58±0.55^{abc}$	$0.83±0.04^{bcd}$
	125	$26.12±0.42^{de}$	$16.48±0.23^{df}$	$18.08±0.55^{bcd}$	$0.77±0.04^{cd}$
Two-way ANOVA					
Source	-	P=0.0455	P=0.1206	P=0.7449	P=0.3133
level	-	P=0.0041	P=0.0001	P=0.0001	P=0.0001
Source x level	-	P=0.0761	P=0.0001	P=0.0001	P=0.0001

[a, b,c]Means with different superscript in the same column are significantly different at (P<0.05).

Lipid profile

Plasma lipid profile; triglycerides, total cholesterol, LDL-C and HDL-C were significantly ($P< 0.05$) influenced by addition of Cu forms in chickens diet (Table 5). Birds fed diet supplemented with CuNO3 significantly ($P <0.05$) decreased lipid profile than those fed diet supplemented with CuSo4. The addition of CuNo3 significantly ($P < 0.05$) decreased the level of plasma total cholesterol, triglycerides, LDL and HDL than those of the control group.

The hypocholesterolemia effect of copper observed in this study is well documented. Since the addition of Cu to the feed ingredients decreased the level of total cholesterol compared to the control treatment. Significant (P < 0.05) differences in lipid profile values between chicks fed diets supplemented with Cu at different levels were found. The lowest values of triglycerides, cholesterol, and HDL-C were observed in the groups fed of Cu at a level of 125 mg/kg diet, While the highest values were recorded in control. Addition of Cu with high level resulted in reducing lipid profile of birds. While, the levels of triglycerides, cholesterol, LDL, and HDL in birds fed diet supplemented with 125 mg/kg diet Cu were significantly (P< 0.05) lower than those at other levels. Plasma cholesterol was reduced with increasing the level of Cu supplementation, indicating that feeding broilers with high Cu concentrations may lower their plasma cholesterol levels. These findings could be explained by the fact of t Cu can controls cholesterol biosynthesis by lowering hepatic glutathione levels and altering the ratio of oxidized glutathione, which reduced the activity of 3-hydroxyl-3-methylglutaryl Co- (HMG-CoA) reductase (Skrivan *et al.*, 2000; Mondal *et al.*, 2007; Jegede *et al.*, 2011). The current findings were in line with those of Ševčíková *et al.*, (2003), who demonstrated that adding Cu-glycine chelate to the feed considerably reduced the cholesterol concentration 24.9% compared to the control group. Additionally, Saripinar-Aksu *et al.* (2012) found that chickens fed diets supplemented with organic forms of Zn, Cu, and Mn had lower cholesterol and LDL-C levels and higher HDL-C levels in their plasma. (Bakalli *et al.*, 1995) y observed that a high dietary copper level (250 mg/kg diet) to broiler chicks decreased blood reduced glutathione concentration and subsequently reduced plasma total cholesterol.

Table 5. Plasma lipid profile (Triglycerides, Total cholesterol, LDL and HDL) as affected by dietary supplementation of copper sources and levels

Sources	Levels (mg/kg diet)	Plasma (g/dl) Triglycerides	Total cholesterol	LDL	HDL
Copper sources	Cu nitrate	101.7±2.46^{b}	146.8±3.31^{b}	64.4±2.3^{a}	82.4±2.0^{b}
	Cu sulfate	109.5±2.46^{a}	154.6±3.31^{a}	59.4±2.3^{b}	95.3±2.0^{a}
Copper level	0	122.6±2.45^{a}	182.3±2.34^{a}	66.6±2.03^{a}	115.8±4.1^{a}
	75	111.1±1.73^{b}	159.4±1.65^{b}	64.8±1.43^{a}	94.7±2.9^{b}
	100	105.9±1.73^{b}	149.4±1.65^{c}	63.4±1.43^{a}	86.2±2.9^{b}
	125	99.9±1.73^{c}	143.3±1.65^{d}	57.6±1.43^{b}	85.8±2.9^{b}
Control	0	122.6±0.10^{a}	182.3±0.14^{a}	66.6±3.6^{a}	115.8±2.8^{a}
Cu nitrate	75	108.2±0.10^{d}	156.0±0.14^{b}	64.2±3.6^{d}	91.8±2.8^{e}
	100	101.1±0.10^{f}	145.1±0.14^{f}	68.2±3.6^{a}	77.2±2.8^{g}
	125	95.7±0.10^{g}	139.3±0.14^{g}	61.0±3.6^{e}	78.4±2.8^{f}
Cu sulfate	75	114.1±0.10^{b}	162.9±0.14^{b}	65.4±3.6^{c}	97.6±2.8^{b}
	100	110.8±0.10^{c}	153.7±0.14^{d}	58.6±3.6^{f}	95.2±2.8^{c}
	125	104.2±0.10^{e}	147.3±0.14^{e}	54.2±3.6^{g}	93.20±2.8^{d}
Two-way ANOVA					
Source	-	P=0.0001	P=0.0001	P=0.0001	P=0.0001
level	-	P=0.0001	P=0.0001	P=0.0001	P=0.0001
Source x level	-	p= 0.0001	P=0.0001	P=0.0001	P=0.0001

a,b,c Means with different superscript in the same column are significantly different at (P<0.05).

Birds fed diet supplemented with CuNo3 at a level of 125 mg/kg diet had significantly (P < 0.05) lower plasma triglycerides, cholesterol and HDL-C levels compared with those fed CuSO4 supplemented diets at different levels. Diets supplemented with different Cu levels and forms showed significantly (P < 0.05) lower in lipid profile compared to the control group. This finding contradicts with Cromwell *et al.* (1989), who claimed that the sulphate source of copper produced higher cholesterol levels than other sources. Similarly, Pesti and Bakalli (1996) and Konjufca *et al.* (1997) showed that broiler hens' cholesterol levels were reduced in diets supplemented with copper pentahydrate or copper citrate. Additionally, according to Paik et al. (1999), dietary doses of copper chelate decreased total plasma cholesterol. This may be because copper regulates cholesterol production by lowering hepatic glutathione levels (Kim *et al.*, 1992). Triglycerides-lowering effect of Cu supplementation could be reduced the impact of the activity of fatty acid synthetase (FAS) which in turn resulted in a reduction in fatty acid synthesis from Acetyl COA and finally decreasing triglycerides biosynthesis.

Antioxidants response

Results in Table (6) showed that the antioxidant response against different oxidative stressors was significantly improved by Cu supplementation. In this concern, total antioxidant capacity (T-AOC), plasma glutathione (GP_X) and superoxide dismutase (SOD) were significantly (P < 0.05) increased in chickens fed diet supplemented with copper nitrate, while malondialdehyde (MDA) activity was significantly decreased. The improvement in oxidative response in this study was clearly recorded for broilers fed diet supplemented with Cu at a level 100 or 125 mg/kg diet. Idowu *et al.* (2011) reported that supplementation of dietary copper with 250 mg/kg diet caused a high lipid peroxidation and low activity in glutathione, SOD and catalase in the liver and erythrocytes of broiler chickens.

Broiler chickens fed diet supplemented with 125 mg/kg diet from either copper nitrate or copper sulfate, respectively significantly, (P < 0.05) increased plasma T-AOC, GPX and SOD, when compared with different levels applied and control group, which showed the highs value of plasma MDA activity.

The reduction observed of MDA levels in birds fed Cu supplemented diets is consistent with the findings of Kumar *et al.* (2013), who found that plasma MDA of broiler chickens was reduced when fed diets supplemented with copper sulphate pentahydrate. Although the ability of free copper ions exaggerates the elaboration of reactive oxygen species (Gaetke and Chow 2003), however, the lipid peroxidation (MDA) significantly reduced in quails fed up to 150 mg/kg diet.

The present results revealed significant increases in plasma T-AOC, GP_X and SOD, of broiler chicks fed diet supplemented with Cu compared with control group (Table 6). This result in agreement with that obtained by Bakalli *et al.* (1995) and Konjufca *et al.* (1997). Also, Robbins and Baker (1980) sustained that Cu, has a strong oxidizing ability associated with the presence of free SH-groups, which may affect and /or limit the availability of GSH because the difficulty of Cu-SH bond to be dissociated easily. Supplementation of copper with form of organic or inorganic source decreased the activity of MDA, which as an indicator of cell membranes injury form free radicals oxidative stress, because the higher levels of MDA may be a diagnostic criteria for the antioxidant defense system damage due to oxidative stress (Farombi *et al.*, 2004 and Kato *et al.*, 2007).

Table 6. Plasma T-AOC, GPX, SOD and MDA as affected by dietary supplementation of copper sources and levels

Sources	**Levels (mg/kg diet)**	**Plasma**			
		T-AOC (Mm/ L)	**GPX (U/ mg)**	**SOD (U/ mg)**	**MDA (nmol/ mL)**
Copper sources	Cu nitrate	0.83±0.03^{a}	289.9±5.2^{ab}	4.80±0.17	3.78±0.15^{b}
	Cu sulfate	0.78±0.03^{b}	281.3±5.2^{b}	4.60±0.17	4.02±0.15^{a}
Copper level	0	0.63±0.04^{d}	251.6±8.0^{c}	3.84±0.29^{c}	4.86±0.25^{a}
	75	0.74±0.02^{c}	276.1±4.6^{b}	4.52±0.16^{b}	4.16±0.14^{b}
	100	0.83±0.02^{b}	290.0±4.6^{b}	4.80±0.16^{ab}	3.77±0.14^{bc}
	125	0.97±0.02^{a}	306.3±4.6^{a}	5.16±0.16^{a}	3.32±0.14^{c}
Control	0	0.63±0.03^{e}	251.6±7.6^{g}	3.84±0.29^{e}	4.86±0.29^{a}
Cu nitrate	75	0.73±0.03^{cd}	276.0±7.6^{e}	4.56±0.29^{c}	4.18±0.29^{bc}
	100	0.82±0.03^{b}	288.4±7.6^{b}	4.76±0.29^{b}	3.82±0.29^{d}
	125	0.95±0.03^{a}	305.4±7.6^{a}	5.10±0.29^{a}	3.36±0.29^{g}
Cu sulfate	75	0.69±0.03^{de}	268.4±7.6^{d}	٤,٣٢±0.29^{d}	4.40±0.29^{b}
	100	0.76±0.03^{c}	282.6±7.6^{c}	4.٦٤±0.29^{bc}	3.98±0.29^{c}
	125	0.88±0.03^{a}	293.0±7.6^{b}	4.86±0.29^{a}	3.70±0.29^{e}
Two-way ANOVA					
Source	-	P=0.0037	P=0.036	P=0.172	P=0.042
level	-	P=0.0001	P=0.0001	P=0.0016	P=0.0001
Source x level	-	p= 0.0001	P=0.0001	P=0.027	P=0.0001

[a,b,c]Means with different superscript in the same column are significantly different at (P<0.05).

CONCLUSION

Based on the findings, it can be stated that broiler chickens blood biochemical parameters and growth performance are both positively impacted by excess copper. Ross 308 chickens ideal copper requirements were 125 mg/kg of feed; regardless of copper source, poultry farmers are advised to use copper as a growth booster in addition to lowering plasma cholesterol levels and enhancing antioxidant defense.

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التأثيرات المحتملة للتغذية علي مصادر النحاس المختلفة لتحسين الأداء الإنتاجي ، ومقايس بلازما الدم وأنشطة الاستجابة التأكسدية لدجاج التسمين

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الملخص

اجريت التجربة لدراسة تأثير مصادر ومستويات النحاس الغذائية على أداء النمو وبعض مقايس الدم والأنشطة المضادة للأكسدة لدجاج التسمين. تم إستخدام عدد ٤٢٠ كتكتوت تسمين (روص ٣٠٨)عمر يوم قسمت عشوائياً إلى ٧ معاملات تجريبية (٦٠ طائر / معاملة) .سجلت الطيور المغذاة على عليقة مضاف إليها النحاس بمصادرة المختلفة أعلي وزن الجسم ومعدل الزيادة اليومية فى وزن الجسم وكفاءة التحويل الغذائى بينما إنخفض معدل إستهلاك الغذاء مقارنة بمجموعة الكنترول. سجلت المعاملة المغذاة علي نترات النحاس العضوى أعلي اداء لمعظم الصفات المدروسة مقارنة بالمجموعات الأخرى. إنخفضض مستويات الدهون الكلية ، والدهون الثلاثية ، الكوليستيرول الكلى بالبلازما و الدهون الثلاثية مرتفع و منخفض الكثافة فى الكتاكيت المغذاة على ١٢٥ ملليجرام نحاس / كجم مقارنة بالمجموعات الاخري. تحسنت حالة مضادات التأكسد للطيور من واقع زيادة مستوي إنزيم سوبر اكسيد ديسميوتاز ، جلوتاثيون بيروكسيديز والقدرة الكلية المضادة للاكسدة بينما إنخفض إنزيم مالونديالديهيد في المجاميع المغذاة علي النحاس مقارنة بمجموعة الكنترول. وقد خلصت نتائج الدراسة إلي ان إضافة النحاس للعليقة بمعدل ١٢٥ ملليجرام / كجم عليقة بمصادرة المختلفة, كان له تأثير إيجابي ومعنوي علي الآداء الإنتاجى ونظام الدفاع لمضادات الأكسدة وانخفض محتوى الكوليستيرول فى كتاكيت التسمين. كتاكيت التسمين المغذاة على نترات النحاس العضوى حسنت اداء النمو والإستجابة المناعية مقارنة بالمغذاة علي كبريتات النحاس الغير عضوية.

الكلمات الدالة : مصادر النحاس, الآداء, ملف الدهون فى البلازما, نظام مضادات الأكسدة , كتاكيت التسمين.