

The Effect of Adding Different Levels of Copper Sulfate And vitamin C to the Diet on Some Cellular and Biochemical Parameters of Broilers

¹Tarek I. Majeed ; ²Qutaiba J. Gheni

^{1,2} Animal Production Dept. Collage of Agriculture, University of Basrah ,Iraq
qutiba.chemi@uobasrah.edu.iq

Abstract

The current study was conducted on the poultry field in University of Basrah - college of Agriculture - Production Department for the period from 5/9/2017 to 11/10/2017 during a (5 weeks) experiment to investigate the effect of adding different concentrations (0, 100, 200 and 400 mg/kg feed) of copper sulphate and vitamin C concentration (500 mg /kg feed) to the basic diet provided to the birds. In some of the physiological characters of broilers blood. In the study two hundred and eighty eight day-old Ross 308 farnels broiler chicks were assigned equally into eight dietary treatments for (35) days , with three replicates of 12 chicks each, according to the complete randomized CRD design, the chicks were given copper sulfate and vitamin C from the first day to the end of the experiment 35 days, Treatments were as T1 control treatment without additions, T2 (500 mg/kg) of vitamin C, T3 (100 mg/kg) of copper sulphate, T4 (100 mg/kg) of copper sulfate + (500 mg/kg) of vitamin C, T5 (200 mg/kg) of copper sulphate, T6 (200 mg/kg) of copper sulphate + (500 mg/kg) of vitamin C, T7 (400 mg/kg) of copper sulphate, T8 (400 mg/kg) of copper sulfate + (500 mg/kg) of Vitamin C. The results indicated Significant increase ($p \leq 0.05$) in the PCV (Packed Cell Volume) and the concentration of HB Hemoglobin in addition treatments to non-addition treatments ,

There was a significant superiority ($p \leq 0.05$) in the concentration of total protein and globulin in plasma blood of birds added treatments compared to blood plasma birds of non - additive treatments. While there was a significant decrease in the level of cholesterol in the blood of birds by increasing the concentration of copper sulphate for diet treatments compared to the control treatment, significant increase ($p \leq 0.05$) in the concentration of copper in plasma blood and birds faeces by increasing the concentration of copper sulphate in the diet for the adding copper treatments compared with birds of non-addition treatments.

Keywords: Copper Sulfate , vitamin C, cellular, biochemical, Broilers. Corresponding author Email

1. Introduction

Fodder additives are any substance or group of substances that are pure or loaded, added to the feed material or the mixture in small quantities to meet a specification of requirements. Wenk.(2000), Feed additives are added to bird diets to improve their nutritional value, enhance bird performance by increasing their growth rate, improve feed conversion efficiency, and reduce mortality by reducing disease infection. Onu. (2010).

Although the needs of poultry are low, copper sulphate (CuSO_4) is very important, because it enters into many vital and metabolic processes in the body, it acts as a catalyst for growth, Many researchers have noted that adding levels of 125 to 250 mg copper sulfate has led to a positive response to body weight and feed conversion efficiency. Arias and Koutsos, 2006, also, copper can be added to the diet of birds under various forms as a stimulant for growth, However, the most commonly used form is sulfate. Mondal et al., (2007) With the growing public interest in bacterial resistance to antibiotics in the poultry industry attracted attention to alternatives that have antimicrobial properties, While at the same time maintaining the health of the digestive system so emerged the process of using modern methods

of unconventional in the treatment of birds , Such as the use of a biologic booster (probiotic) and the use of certain enzymes as well as the use of certain hormones and minerals. In 2008, the US Environmental Protection Agency (EPA) approved the use of dozens of useful natural alternative therapies in a list (GRAS(General Recognized As Safe, It is a list of safe materials that can be used in the areas of human nutrition and feeding of farm animals.Karpanen et al., (2012). Copper has received great attention because of its antimicrobial properties, which also improve production performance at the same time.

Vitamin C is a water-soluble vitamin, Vitamin C increases the resistance of birds to certain bacterial and viral diseases and thus reduces mortality rates. Padayatty et al., (2003). It also reduces immunosuppression by increasing the hormone corticosterone and also increases the cellular immune response. Chand et al., (2014). The objective of this study is to study the effect of the addition of copper sulphate at different levels on some physiological characteristics of the blood of broilers.

2. Materials and Methods

This study was carried out in the field of poultry broiler that belong to the Department of Animal Production / Agriculture College/ University of Basra, for the period from 5/9/2017 until 11/10/2017. A total of 288 birds of one-day old famels Ross broiler which were provided from aprivate Hatcheries from Baghdad province. birds were housed in (24) cages.Water and feed were provided libitum and birds reared under strict hygienic and standard management conditions.

Copper sulfata and vitamin C mixed with the food and added as:

T1 = control treatment without additions.

T2 = addition (500 mg/kg) of vitamin C.

T3= addition (100 mg/kg) of copper sulphate.

T4 = (100 mg/kg) of copper sulfate + (500 mg/kg) of vitamin C. T5 = (200 mg/kg) of copper sulphate.

T6 = (200 mg/kg) of copper sulphate + (500 mg/kg) of vitamin C. T7 = (400 mg/kg) of copper sulphate.

T8= (400 mg/kg) of copper sulfate + (500 mg/kg) of Vitamin C.

Blood samples were withdrawn at the age of 35 days were collected (5 ml) of blood from birds and thigh vein and divided into two parts , The first was in the EDTA test tubes and the second was placed in tubes free of anticoagulation to prepare the plasma so as to conduct complete blood tests which include , Hemoglobin,PCV , also other measurements on plasma tests the concentration of total protein, albumin protein, globulin protein, total cholesterol, and copper concentration in blood plasma. The experimental data were analyzed using CRD SPSS 2012 Version 19.

Contents		grower diet)02-53(Days
Metabolizable energy(kcal / kg)	2332	0933
Crude protein)%(20.92	23.32
Calcium)%(9	3.32
Available phosphorus)%(3.29	3.22
Copper mg / kg feed	93.223	29.212

Chemical analysis of forage materials

3. Results and discussion

Table (1) shows the effect of the addition of copper sulfate with different concentrations and vitamin C to the diet fed to birds on PCV and hemoglobin Hb levels of birds at 35 days, The results of the statistical analysis indicate an increase in the volume of PCV blood cells and the concentration of Hb for all copper sulfate addition treatments compared to control treatment T1 and tratment T2 , It was also observed that there was a

significant effect ($p \leq 0.05$) to add copper sulfate to these traits at the age of (35) days , where control treatment T1 recorded The lowest mean value of PCV 30.33% and the concentration of Hb was 9.99 g/ml compared to the other study treatments. We also note the superiority of treatment T7, which did not differ significantly from the treatment T8 for PCV and both recorded (37.66)% , While it was observed a significant increase ($p \leq 0.05$) in the treatment of T8 blood concentration of hemoglobin on the rest of the treatments as recorded (11.76) g/ml ,These results agreed with Mousa,(2014) Which pointed to the superiority of treatment that was added to it (250 mg/kg feed) of copper sulfate on the control treatment in the rate of PCV also pointed to the superiority of the treatment of adding copper sulfate with (350 mg/kg feed) On the addition of garlic powder concentrations (0.75% and 1.5%) and this study agreed with the results of Salwa.D (2016), The reason of superiority for addition copper sulfate treatments in PCV values and concentration of HB on control treatment It may be attributed to the role of copper, which has a significant and major effect in stimulating the formation of red blood cells, By facilitating the absorption of iron from the gastrointestinal tract and release it from its stores within the body to the endometrial tissue and from the visceral tissue of the liver to the blood plasma Mroczek-Sosnowska et al., (2013).

In the case of copper deficiency, iron cannot enter the process of manufacturing hemoglobin blood but deposited in the liver Sharma et al., (2009). Copper is also highly correlated with alpha- globin, and ceruloplasmin, and when the deficiency of these proteins indicates anemia Hefnawy and El-khaiat, (2015)

Table (1) Effect of addition of copper sulfate with different concentrations and vitamin C on PCV and hemoglobin HB blood ratio (gm / 100 ml) at 35 days for meat broilers.

Trait studied Treatments	%PCV	Hemoglobin concentration g/100 ml
T1	03.33 ^c ± 5.41	9.99 ^d ± 0.12
T2	31.33 ^c ± 3.00	10.19 ^d ± 0.16
T3	34.66 ^b ± 0.88	53.41 ^c ± 0.06
T4	34.33 ^b ± 0.66	53.56 ^c ± 0.04
T5	34.66 ^b ± 1.15	10.97 ^b ± 0.09
T6	34.33 ^b ± 0.88	11.08 ^b ± 0.15
T7	37.66 ^a ± 0.88	11.41 ^a ± 0.04
T8	37.66 ^a ± 1.20	11.76 ^a ± 0.05
Significant	*	*

* Significant different letters vertically represent significant differences at the level of ($P \leq 0.05$).

Table (2) shows the effect of the addition of copper sulfate with different concentrations and vitamin C to the diet fed to birds on the total protein concentration in plasma at 35 days, It is also noted from Table (2) that there is a significant superiority ($p \leq 0.05$) for the addition of copper sulphate treatments compared with the non-addition of copper sulphate (T1 and T2) for the concentration of total protein in plasma blood. The T7 and T6 treatments were superior at the rest of the treatments (4.40 and 4.13 g / 100 ml respectively), whereas T1 (control) is the lowest performance of this characteristic recorded (2.76) g/100 ml, also there were no significant differences between T5 and T8 (3.92 and 3.95) g/100 ml respectively, These results are agreed with Waleed, (2015), The results of this study did not agree with Rahman et al., (2001). The moral improvement in the total protein concentration of birds added to their diet copper sulfate may be attributed to the increased metabolic rate and construction rates in their body, Which correspond with high rates of live body weight for added copper sulfate treatments Mroczek-Sosnowska et al., (2017). The reason may be attributed in the superiority of add (200 and 400 mg/kg) copper sulfate in the total protein concentration rate in the plasma to the copper active role in increasing the secretion of growth hormone from the pituitary gland Yang et al., (2011). Which

affects the metabolism of proteins as lead to an increase amino acids osmosis and increase concentration within cells and this, in turn, increases the proportion of proteins manufacturing process Sturkie, (2012), As well as the important role of copper on antioxidant enzymes Bonham et al., (2002). This provides protection against Catabolism reactions in the body through its active role in capturing free radicals and inhibiting protein degradation Suttle, (2010). It may also be to add vitamin C key role in the moral improvement in the total protein concentration because of its positive effect on the antioxidant Ezzat, (2011). The significant improvement in total protein concentration was accompanied by a significant improvement ($p \leq 0.05$) in the concentration of globulin and albumin proteins. Table (2) showed a significant effect of adding copper sulphate with different concentrations and vitamin concentration on the concentration of globulin proteins at (35) days. While no significant effect ($p \leq 0.05$) was observed to add copper sulfate and vitamin C at the same age in albumin concentration, As shown in the analysis of variance there is a significant improvement in albumin concentration at the age of (35) days, The addition of copper sulfate was superior at the non-addition treatments for the concentration of globulin protein, which significantly exceeded ($p \leq 0.05$) the treatment of T7 on the rest of the treatments and recorded (2.50) g/ml, The control treatment T1 recorded the lowest concentration of globulin protein (0.96) g/ml, While the concentration of albumin protein did not increase by increasing the level of copper sulphate to any significant differences between the treatments as there were no significant differences between the treatment of T3 and T7 and the results of this study with Kumar et al., (2013) and did not agree with Rahman et al., (2001), Who compared the use of copper sulfate at the level of (350 mg/kg) and control treatment without addition. The significant increase ($p \leq 0.05$) in the concentration of globulin protein and the improvement in the concentration of albumin protein may be attributed as clarify Bonham et al., (2002) to the role of copper sulfate in stimulating the function of the immune system by increasing the level of immunoglobulin (IgA, IgM) as well as the effect of copper sulfate in stimulating the production of thyroxine. As the transfer of this hormone needs to globulin, which stimulated the production of globulin Sturkie, (2012), in addition to the role of copper in the production of alpha-globulin.

Table (2) Addition of copper sulfate with different concentrations and vitamin C on total protein , globulin and Albumin concentrations in blood plasma at 35 days for broilers.

Trait studied Treatments	Total protein plasma g/100ml	Glubulin g/ml	Albumin g/ml
T1	2.76 ^c ± 0.50	0.96 ^c ± 0.47	1.70 ± 0.04
T2	3.35 ^{bc} ± 0.28	1.76 ^b ± 0.21	1.80 ± 0.14
T3	3.62 ^b ± 0.05	1.80 ^{ab} ± 0.10	1.82 ± 0.10
T4	3.73 ^{ab} ± 0.06	1.99 ^{ab} ± 0.14	1.76 ± 0.12
T5	3.92 ^{ab} ± 0.12	2.22 ^{ab} ± 0.26	1.76 ± 0.20
T6	4.13 ^a ± 0.13	1.93 ^{ab} ± 0.11	2.15 ± 0.21
T7	4.40 ^a ± 0.14	2.50 ^a ± 0.25	1.92 ± 0.16
T8	3.95 ^{ab} ± 0.12	1.84 ^{ab} ± 0.11	2.23 ± 0.08
Significant	*	*	N.S

* Significant different letters vertically represent significant differences at the level of (P≤ 0.05).

N.S: None Significant

The addition of copper sulfate and vitamin C has a clear effect on the cholesterol level in the blood plasma as explained the results table (4). The results of the previous table showed a significant effect (p≤0.05) to add copper sulfate to the level of cholesterol in plasma blood, the results confirmed the existence of a significant decrease in cholesterol level in the addition of copper sulfate treatments compared with the treatment of T1 and T2, also observed significant increase (p≤0.05) in the control treatment (T1) in the level of cholesterol in plasma blood at age (28-35) days and recorded (128.33) g/100 ml, As for the addition of copper sulphate, it can be observed that with the increased concentration of copper sulfate we notice a decrease in the level of cholesterol and the lowest T7 treatment of this attribute and reached (77.00) g / 100 ml at the age of (35) days, The results of the current study were agreed with (Scott et al., 2018), The significant decrease in blood plasma cholesterol level in copper sulfate supplementation would be attributed to the addition of copper sulfate to an improvement in fat metabolism as indicated by Bakalli et al., (1995), Who pointed out that the addition of copper sulfate in concentration (250 mg/kg feed) has led to lower cholesterol levels and triglycerides in blood plasma and breast meat of broilers at the age of (42) days. Also, there is a number of researchers mention the effective role of copper sulfate in reducing fat and cholesterol in the blood plasma of broilers (Skriwan et al., 2002), Where they agreed that the reason for the low level of cholesterol in the blood plasma of broilers due to the effect of copper sulfate in reducing concentrations of protein Glutathione GSH in liver cells, leading to minimize or reduce the stimulation of the production of the enzyme HMG-CoA, which plays an important role in cholesterol manufacture in the liver (70%) of the total cholesterol in the body. Morais et al. (2001) showed no significant differences in the significant decrease in cholesterol when using different levels of

copper sulfate (50, 100, 150 and 200 mg/kg feed) between males and females, While pointed Idowu et al., (2011) to exceed the treatment of addiction (250 mg/kg feed) copper sulphate on the treatment(250 mg copper sulfate + 100 mg vitamin C) as well of treatment (100 mg/kg feed of vitamin C) at the age of (35) days in lowering the level of cholesterol in blood plasma and abdominal fat or abdominal fat.

Table (3) Effect of addition of copper sulfate with different concentrations and vitamin C at the level of cholesterol in blood plasma at 35 days for broilers.

Trait studied Treatments	Cholesterol level in blood plasma g/100 ml
T1	128.33 ^a ± 5.36
T2	124.00 ^{ab} ± 3.78
T3	118.00 ^b ± 3.46
T4	99.33 ^c ± 1.76
T5	91.00 ^{cd} ± 2.88
T6	95.66 ^c ± 4.37
T7	77.00 ^e ± 1.52
T8	88.66 ^{de} ± 2.18
Significant	*

* Significant different letters vertically represent significant differences at the level of (P ≤ 0.05).

The concentration of copper in the blood plasma of birds was significantly affected (p < 0.05) when the level of copper sulfate was increased in the bird feed, Table (4) shows the effect of the addition of copper sulfate with different concentrations and vitamin C on the concentration of copper in blood plasma in different treatments at age 35 days, It excelled significantly (p < 0.05) treatment T7 on the rest of the treatments amounted to (8.08 mg/L), while the lowest concentration in the treatment of T2 reached (0.27) mg/L, The results of the above table show that the main source of copper in blood plasma is the level of copper sulfate added to the diet and these results agree with (Payvastegan et al., 2013) where they linked the level of absorption and the bioavailability of the levels of some metals such as copper and zinc in plasma blood and body and the idea that these minerals when added or exist in small quantities or a few have a better chance of absorption in the digestive tract than others, Because these small quantities contain a relatively larger surface area in which metals can be exposed to enzymes or proteins for binding, transport, and metabolism (Pang and Applegate, 2007). These results also agreed also with (Hamdi et al., 2018) who compared the addition of copper sulfate levels (15, 150 and 300 mg/kg feed) and found that the increase was highly significant and linear concentrations of copper in both liver and blood plasma, The increase was consistent with the increased

level of copper sulfate added to the diet, which was due to the high melting of copper sulfate. As noted above, the effect of vitamin C is evident in the detraction of absorption copper sulfate in the additional treatments ofcopper sulfate and vitamin C, which confirms the role of vitamin C preventive against high levels of copper sulfate and these results agreed with them (Kakkar et al., 2017)

Table (4) Effect of addition of copper sulfate with different concentrations and vitamin C on copper concentration in plasma blood at 35 days for broilers.

Trait studied Treatments	Concentrate copper in blood plasma mg/100 ml
T1	3.40 ^f ± 3.050
T2	0.27 ^f ± 0.096
T3	1.08 ^e ± 0.074
T4	0.94 ^e ± 0.092
T5	2.75 ^c ± 0.083
T6	2.00 ^d ± 0.077
T7	8.08 ^a ± 0.050
T8	6.41 ^b ± 0.033
Significant	*

* Significant different letters vertically represent significant differences at the level of (P≤ 0.05).

4. conclusion

In general, we can conclude from this study that the addition of copper sulfate to broiler had a significant effect on some physiological characteristics of blood. There was also no significant damage to birds when using copper sulfate at a concentration of 400 mg/kg feed.

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