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EFFECT OF DIETARY NATURAL FEED ADDITIVES TO MINIMIZE NEGATIVE ROLE OF PEROXIDE HYDROGEN IN BROILER

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ABSTRACT : This study was conducted in poultry farm belong to Animal Resources and Fisheries Research center, Ministry of Sciences and Technology, from 21/11/2018 to 2/1/2019 for 42 day. The aim of study was to find out effect of Green tea and Organic Selenium, as antioxidant and their effects on immunity response and oxidation parameters of broiler. One Hundred – Ninety Eight of males broiler (Ross 308) chicks with average initial body weight of 42 gm./ chick were used in this study. Chicks were randomly assigned to six treatments, each treatment was sub-divided into three replicates (11 chicks per replicate). The treatments were as follows: T_1 basal diet free from any addition (Negative control), T_2 included addition of 0.5% H_2O_2 to drinking water at the age 10 days (Positive control), T_3 basal diet + 1.5% green tea to feed, T_4 basal diet + 1 mg/kg Organic Selenium to feed, T_5 basal diet supplemented with 1.5% green tea + H_2O_2 (0.50%), T_6 basal diet supplemented with 1 mg/kg Organic Selenium + H_2O_2 (0.50%). The results revealed significant differences between treatments in (HI) titer against Newcastle and Gumboro diseases. However, T_2 (Negative control) recorded lowest values with significant difference with other treatments. Also, no significant differences were observed by adding green tea and organic selenium on relative weights of Bursa, while relative weights of spleen and thymus recorded significant differences between treatments and T_5 showed significant increase as compared with other treatments, while Highly significant (P<0.01) observed for T_6 in relative weight of thymus and recorded 0.370. Results included significant differences between treatments in Creatinine and Uric acid. T_2 showed a significant increase in Malondialdehyde (MDA), Peroxide Value (P.V) and Free Fatty Acids (FFA) in liver tissue as compared with other treatments. On other hand, feed additives treatments showed improvement in liver tissues content of Glutathione peroxidase and catalase as compared with T_2 (Negative control). As well as, T_1 (positive control) has highly significant (P<0.01) in Glutathione peroxidase and catalase enzymes.

Key words : Green tea, organic selenium, immunity, oxidation status, broiler.

INTRODUCTION

Nutrition is a very important link in chain of operations that must be conducted in order to reach the optimal production process, especially poultry production eggs or meat (Zuidhof *et al*, 2015). The adoption of modern strategies in the poultry industry tends to increase the efficiency of food conversion, which is an inevitable option on a commercial scale, but it causes decrease on health status of bird (Das *et al*, 2011). Recent research has focused on enhancing the immune ability of broiler meat, as it is important to increase immune capacity (Khan *et al*, 2016). High intensity of poultry production requires fast growing strains and rations of high energy density, so poultry rations are often supplemented with oils. Feeding diets with added fat to poultry can confer several economic advantages by providing increased energy levels at a lower cost and is becoming common practice (Mousa *et al*, 2018). Fats added to the diet for fast growing broilers are generally rich in polyunsaturated fatty acid (PUFA) (Al-Rawi *et al*, 2019). An increase in the degree of un saturation of carcass fat of broiler, due to dietary unsaturated fat supplements decreases the carcass lipid stability. Lipid oxidation is a major cause of quality deterioration in meat and meat products and can give rise to rancidity and formation of undesirable odors and flavors, which affect the functional, sensory, and nutritive value of the meat products (Amaral *et al*, 2018). Under such condition, antioxidant supplementation even is effective in stabilizing these tissues from oxidation reactions. The high stocking densities of broiler are highly susceptible to infectious agents either as a result of reduced immune potential (Qaid *et al*, 2016) or as a result of deteriorating environmental hygiene (Umar *et al*, 2017).

One of several approaches to increase responsiveness in high intensity production is to supplement rations with micronutrients (Mousa and AL-Rawi, 2015). It has also been proven by conclusive scientific evidence that vegetable kingdom possesses a large number of by-products that are distinguished by its ability to treat incurable diseases in humans and animals, especially birds through their biological efficacy and physiological effects (Alloui *et al*, 2014). Among these plants is (Green tea) rich in nutrients and phenolic compounds which are the body's most important defenses against free radicals (Vishnoi *et al*, 2018). Green tea also has many advantages, including that it contains Polyphenols which had antioxidant properties (Bao and Peng, 2016). Catechins has many health benefits in addition to being an antioxidant, as it is an anti-mutagenic (Santhy and Geetha, 2013) anti-carcinogenic (Najeeb *et al*, 2016) and anti-inflammatory (Khan, 2014). Vishnoi *et al* (2018) reported that green tea extract contains polyphenols flavonoids so it is effective in inhibiting free radicals. Catechin organizes the inner lining of blood vessels by activating nitric oxide, and that catechins work to inhibit the action of Nuclear factor- kappa B (NF-κB), which works to stick white blood cells (WBC) together or on the walls of blood vessels, as well as prevent platelet aggregation hematology. These factors collectively work to prevent blood clotting (Mosawy *et al*, 2016). Catechines are the flavanol monomers and the most important catechins in green tea are epigallocatechin-3-gallate and epicatechin-3-gallate, which are antioxidant compounds that are predominantly and active, meaning that they have higher antioxidation effective there are also compounds with anti-oxidant propagation that are both epicatechin and epigallocatechin, and there are also compounds with antioxidant efficacy that are both epicatechin and epigallocatechin and epigallocatechin-3-gallate (EGCG), which are known to be the most biologically active (Chu *et al*, 2017). Fiecke *et al* (2019) showed that effect of (EGCG) which is attributed to most of the benefits of green tea as an antioxidant in body (*in vivo*) depends on the degree or level of oxidative stress and the presence of other antioxidants in feeds.

Selenium is one of the essential elements that human and animal body need for its important role in functioning of antioxidant system in body as it is considered one of the essential components of the glutathione peroxidase enzyme (GSH-Px) found in most tissues of body (Surai *et al*, 2018). There are two types of selenium, the first represents the inorganic selenium, which is in the form of Selenate (SeO_4) and Selenite (SeO) and other type is organic selenium (Upton *et al*, 2009). In previous studies, Cichoski *et al* (2012) reported that selenium metabolism in body is closely related to activity of the enzyme Glutathione peroxidase that eliminates hydroxyl peroxide in cellular structures. Therefore, the study aimed to know the effect of adding green tea powder and organic selenium to diets and their effect on the productive performance of broilers. The objective of this study was to determine the effect of inclusion green tea and organic selenium and effects on antioxidant status of broiler.

MATERIALS AND METHODS

The study was conducted in the poultry field belong to Agricultural Research Department in the Ministry of Science and Technology in Baghdad, Al-Zaafaraniya and continued for 42 days. One Hundred-ninety eight of males broiler (Ross 308) chicks were used with average weight 42 gm were randomly distributed to six treatments with 3 replicates (11 chicks/replicate) in a closed farm containing ground cages with an area of $1m^2$. Use a continuous lighting program 24 hours/day from the first day of the experiment to the end. The chicks were fed three phases rations, starter from (1- 14 days), Grower (15-28) days and the finisher (29-42) days (Table 1). The treatments were as follows: The first treatment T_1 (basal diet) without any additives, T_2 (basal diet + H_2O_2 0.50%) to drink water, T_3 supplemented with 1.5% green tea to feed, T_4 supplemented with Organic Selenium at 1 mg/kg feed, T_5 basal diet with adding 1.5% green tea + H_2O_2 0.50%, T_6 basal diet with adding 1 mg/kg Organic Selenium + H_2O_2 0.50%. The required quantity of green tea was purchased from local markets, and it was purified, ground and placed in plastic bags for use. Likewise, organic selenium was purchased from the local market by the Uruk Company in the Al-Sinak/Baghdad region, where the required quantity of Zinpro products was provided, and the material is available in powder and metal (selenium) form carried on the amino acid methionine, so the selenium concentration is 50% according to recommendations the Producing company.

Determination of immune response

- The immune response was evaluated by measuring the humeral immunity using heamagglutination inhibition (HI) test against Newcastle disease (Beard, 1989).
- The results of HI titer of the serum samples were recorded and were given titer reference number (TRN) according to Kaleta and Siegmann (1971) and then they were subjected to data analysis to calculate the geometric mean HI antibody titer.
- The relative weights of spleen, thymus and bursa of fabricuius were determined (immune organs).

Table 1 : Ingredient and the chemical composition calculated of experimental diets.

Ingredients	Starter(1-14) day	Grower(15-28) day
Wheat	60.32	62.93
Soybeans (48%)	29.00	25.70
Protein [1] %	5.00	5.00
Vegetable oil	3.40	4.50
Limestone	1.10	1.00
Dicalcium phosphate [2]	0.70	0.50
DL-methionine	0.17	0.17
L-lysine	0.21	0.10
Salt	0.10	0.10
Total	100	100
Chemical analysis, Calculated [3]		
Crude protein %	23.00	21.90
ME kcal/kg	3001	3100
Protein: energy ratio	130	142
Ether extract	5.40	6.58
Crude fiber	3.10	3.00
Methionine	0.64	0.62
Methionine + cysteine	1.01	0.95
Lysine	1.40	1.22
Calcium	0.95	0.88
Available phosphorus	0.47	0.46

[1]: The imported protein center from the Dutch company Al-Wafi and the container used 40% crude protein, 2107 kcal energy represented/kg feed, 5% calcium, methionine 3.7%, methionine + cysteine 4.12%, lysine 3.85%, free phosphorus 4.68% calcium 5.6% tryptophan 0.42 %, Threonine 1.70%, sodium 2.50% and chloride 4.20%,

[2]: Contains 24% calcium and 18% phosphorus,

[3] : chemical composition according to the analysis of feed material contained in NRC (1994).

Measurements of some blood constituents

- serum uric acid according to (Caraway,1963), total serum creatinine (Folin, 1934).

Antioxidant Measurement

- Catalase (CAT) activity was determined according to Cohen *et al* (1970).
- Glutathione peroxidase (GSH-PX) was determined according to Godin *et al* (1988).
- Malone dialdehyde (MDA) compound which is a byproduct of lipid oxidation process has been estimated according to the standard method described by Ohkawa *et al* (1979).

At the end of the trial, from the slaughtered birds' liver was removed and frozen immediately at (–20°C), then thawed and ground. The fat extracted from the liver according to the method of Folch *et al* (1957). The extracted fat was used for determined the peroxide value according to A.O.C.S. (1966).

Statistical analysis

The results of the study were analyzed using Complete Randomized Design (CRD) to investigate the effect of different treatments in the studied traits as well as the Duncan Multivariate test (Duncan, 1955) was used to examine the differences between the averages at the mean level of 0.05 and 0.01 by using the Analysis Statistical System (SAS).

RESULTS AND DISCUSSION

It is noted from Fig. 1 there were significant differences in titer against Newcastle and titer against Gumboro disease where results of statistical analysis showed treatment T_2 (H_2O_2 0.50%) recorded lowest values and this is due to the oxidative stress by addition hydrogen peroxide, while green tea and organic selenium treatments recorded a significant improvement in titers against Newcastle and Gumboro titers and this may be due to the role of catechines and role of organic selenium in cells protecting from oxidative stress by increasing internal antioxidant enzymes that in turn worked to maintain balance of antioxidants (Lee *et al*, 2009). The improvement in antibody titers may refers to catechines in green tea and organic selenium which is necessary for mitochondria and microsomes of liver against oxidative stress (Tan *et al*, 2010). The immunostimulant effect of green tea and organic selenium is mainly probably due to its antioxidant effect (polyphenols and catechins) could modify the overall balance of antioxidants in cell, resulting in different pools of antioxidants in cells that carry different immunoregulatory properties (Chao-Lin *et al*, 2014). Catechines and some active components in green tea had the ability to enhance the immune system in human (Haque and Ansari, 2014) and poultry against coccidiosis (Jang *et al*, 2007). In present study, green tea and organic selenium improved titer to Newcastle disease vaccine (NDV) and that may be could useful as a method to increase vaccination effectiveness and improve the immunity response. In summary, the present study demonstrated that dietary green tea and organic selenium enhances the antibody response to Newcastle and Gumboro diseases vaccination. Green tea and organic selenium supplementation may have different effects on cellular free radicals, antioxidant balance events and activation states of immune cells. Our results were in agreement with Farahat *et al* (2016), who mentioned that specific antibody titre against Newcastle disease virus vaccines was significantly increased at 28 and 35 d of age in broilers fed diets supplemented with green tea and concluded inclusion of green tea has possess antioxidant and immunostimulant characteristics for broilers. In contrast, result of this study were disagreement with Torki

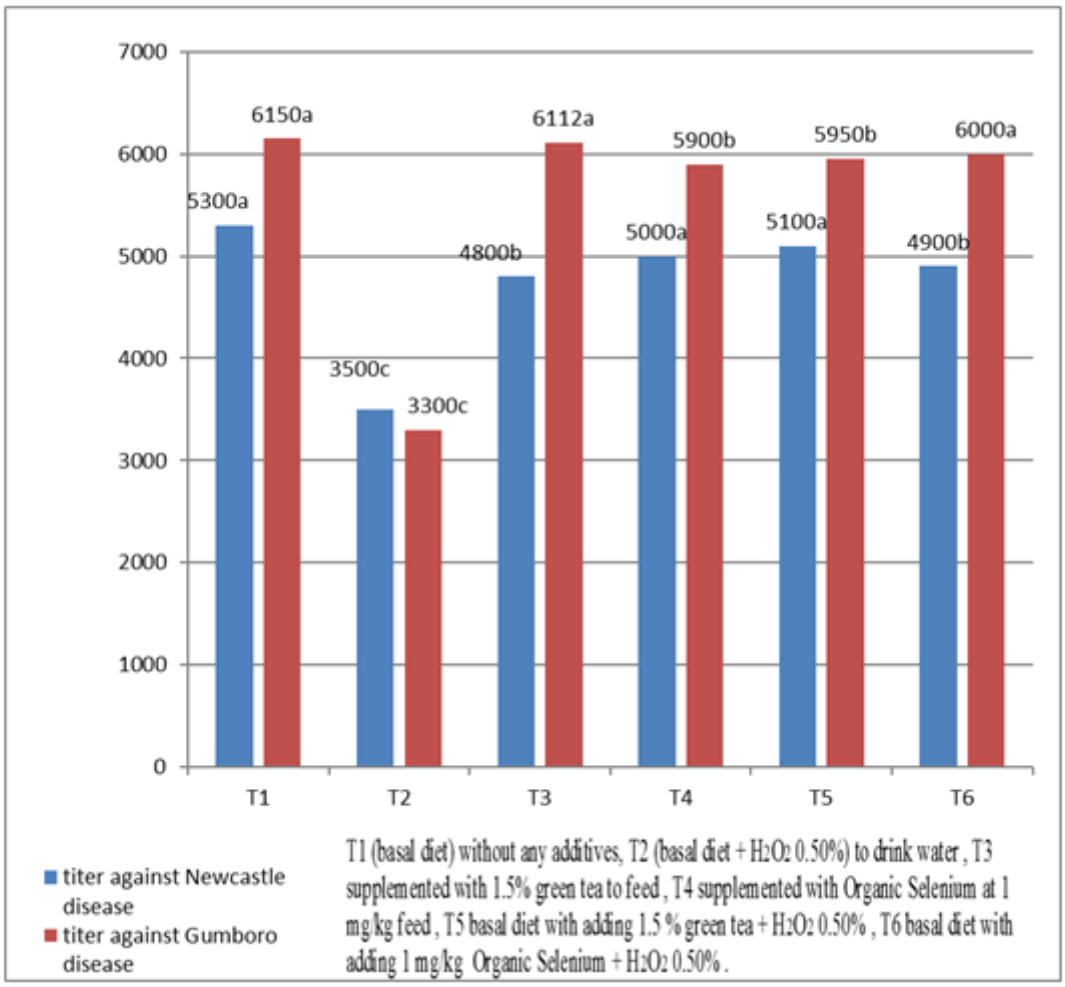

Fig. 1 : Effect of dietary natural feed additives on titers against Newcastle and Gumboro disease in broiler chickens.

et al (2015) who showed that titer to Newcastle disease vaccine (NDV) was no significantly (P≤0.05) in broiler receiving 1 mg selenium/kg diet.

Table 2 showed relative weights of lymphoid tissues (spleen, thymus and bursa of fabricuius) which have a considerable role in bird's immunity (Jones and Bark, 1979). The effect of green tea and organic selenium supplementation on lymphoid organs are presented in Table 2. Relative weight of bursa of fabricuius was not affected by supplementation with green tea and organic selenium. Thymus, bursa of Fabricius and spleen are major lymphoid organs in birds. During the immune challenge, immune cells and lymphocytes interact with injected antigens in most tissues. It is clearly accepted that immune tissue may responsible of the immune system functionality (Latif *et al*, 2014). Regarding the weight of lymphoid organs, little changes in its relative weight were observed in our study. This finding was consistent with previous results of Habibian *et al* (2014), who indicated that dietary selenium had no positive effect on relative weight of bursa under heat stress. Also, Khalaji *et al* (2011) did not show any positive effect by green tea added to broiler diets with level 300 mg/kg on the immune response. Furthermore, El-Deek *et al* (2012) reported in his study no significant differences in humoral immune response of broiler fed diets with 1500 and 3000 mg/kg of green tea against New castle Disease virus vaccine. However, spleen and thymus of birds fed diets supplemented with green tea and organic selenium were affected (Table 2). Tatum *et al* (2000) reported that selenium had the ability to inhibit toxic in his study to investigate adding selenium and copper to broiler diets. Moreover, Xu *et al* (2003) reported that dietary Selenium and green tea had a synergestic effect on the lymphoid organ weights in broiler under normal environmental conditions. The significant differences between treatments is probably due to first; the increased cellular multiplication by green tea and selenium, rather than to the improvement in antioxidants in cell and decrease the adverse environment (Suzuki, 2005). Second; high levels of (polyphenols in green tea) and organic selenium also cause an increase in phagocytosis, during the first few hours after antigen stimulant (Suzuki *et al*, 2007). Third; catechines and organic selenium stimulates the helper

Table 2 : Effect of dietary natural feed additives on relative weight of lymphoid organs in broiler chickens.

Item	Experimental treatments						SEM	*P-value*
	T_1	T_2	T_3	T_4	T_5	T_6		
Spleen	0.109 c	0.7601 b	0.182 c	0.108 c	0.920 a	0.104 c	0.02	0.677
Bursa of fabricuius	0.185 a	0.177 ab	0.170 ab	0.10 c	0.192 a	0.168 b	0.064	NS
Thymus	0.354 ab	0.289 c	0.330 b	0.340 b	0.356 ab	0.370 a	0.261	0.399

a,b,c Different superscripts within same row indicate significantly different (P<0.01).
**T_1 (basal diet) without any additives, T_2 (basal diet + H_2O_2 0.50%) to drink water, T_3 supplemented with 1.5% green tea to feed, T_4 supplemented with Organic Selenium at 1 mg/kg feed, T_5 basal diet with adding 1.5% green tea + H_2O_2 0.50%, T_6 basal diet with adding 1 mg/kg Organic selenium + H_2O_2 0.50%.
NS: Non significant.

Table 3 : Effect of dietary natural feed additives on blood plasma creatinine and uric acid in broiler chickens.

Item	Experimental treatments						SEM	*P-value*
	T_1	T_2	T_3	T_4	T_5	T_6		
Creatinine (mg/dL)	0.2 b	0.1 c	0.35 a	0.36 a	0.22 b	0.2 b	0.011	0.014
Uric acid (mg/dL)	3.6 ab	4.2 a	2.5 b	2.2 b	3.3 ab	2.7 b	0.031	0.011

a,b,c Different superscripts within same row indicate significantly different (P<0.01).
**T_1 (basal diet) without any additives, T_2 (basal diet + H_2O_2 0.50%) to drink water, T_3 supplemented with 1.5% green tea to feed, T_4 supplemented with Organic Selenium at 1 mg/kg feed, T_5 basal diet with adding 1.5% green tea + H_2O_2 0.50%, T_6 basal diet with adding 1 mg/kg Organic Selenium + H_2O_2 0.50%.

Table 4 : Effect of dietary natural feed additives on relative expression of antioxidant enzymes and oxidation status in broiler chickens.

Item	Experimental treatments						SEM	*P-value*
	T_1	T_2	T_3	T_4	T_5	T_6		
MDA	35 d	65.8 a	54.4 b	45.9 c	52.5 b	50.4 b	2.36	0.001
FFA%	0.725 b	1.75 a	0.466 d	0.546 c	0.560 c	0.516 cd	0.097	0.0049
PV	2.86 b	4.26 a	1.56 cd	1.24 d	1.98 c	1.58 cd	0.351	0.0357
Glutathione	230 a	111 d	212 c	220 b	225 ab	220 b	77.8	1.231
Catalase	250.3 a	170.5 b	240.2 ab	245.3 ab	247.4 a	250.0 a	1.166	2.226

*a,b,c Different superscripts within same row indicate significantly different (P<0.01).
**T_1 (basal diet) without any additives, T_2 (basal diet + H_2O_2 0.50%) to drink water, T_3 supplemented with 1.5% green tea to feed, T_4 supplemented with Organic Selenium at 1 mg/kg feed, T_5 basal diet with adding 1.5% green tea + H_2O_2 0.50%, T_6 basal diet with adding 1 mg/kg organic selenium + H_2O_2 0.50%.

activity and cooperation between T-cells and B- cells in immunolo globulin production (Khan *et al*, 2016).

Data in Table 3 summarized effect of feeding diets supplemented with different levels of green tea and organic selenium on Creatinine and uric acid, significant effects were detected attributable by adding green tea and organic selenium. Adding H_2O_2 0.5% to drinking water in (T_2) recorded depressed significantly in creatinine with lowest value (0.1 mg/dl), feed additives treatments were recorded significant differences in creatinine values as compared with T_2. Furthermore, a significant difference (P<0.01) recorded for T_3 (1.5% green tea) and T_4 (Organic Selenium at 1 mg/kg feed) as compared to T_1 (positive control). In contrast, Dalia *et al* (2017) reported that inclusion Selenium as inorganic and organic form to chicken rations decreased the serum creatinine level compared to control group. Data in Table 3 indicated significant differences in uric acid values for study treatments, birds in T_2 (adding H_2O_2 0.5%) were significantly higher compared to the other treatments. The Results of the present study revealed that there is no significant difference (P<0.01) in values of uric acid between feed additives treatments and T_1 (control). Result of Shirai *et al* (2013) was disagreement with our results, significant effects on uric acid of male mice fed diets supplemented with 0.25% green tea extract. The findings of this study were consistent with previous studies which reported that Serum uric acid concentrations decreased (P<0.01) by adding selenium 1mg/kg or combined with copper 200 mg/kg feed (Torki *et al*, 2015). On other hand, Saraeia *et al* (2016) had no effect of adding fish oil and green tea powder to broiler diets on creatinine values.

Table 4 shows the values of the enzymatic antioxidant

(catalase) at 42 days of age, a significant improvement was observed in values of catalase enzyme for T_4, T_5 and T_6, which achieved 250.3, 247.4 and 250.0 respectively at (P≤0.01) as compared with second treatment T_2, while treatments did not differ with T_1 and T_3 treatments, while T_2 recorded lowest value 170.5. This indicates show the ability of additives to get rid of free radicals produced by hydrogen peroxide. The increase in value of catalase enzyme in second treatment may be caused by oxidative stress resulting from addition hydrogen peroxide with absence of non-enzymatic antioxidants or feed additives such as green tea or organic selenium and thus role of enzyme defense is clear through its high concentration to resist oxidation factors resulting by addition H_2O_2 and the continues of oxidative stress leads to a deterioration in health and productivity status, also it leads to high levels of MDA. Cells can face some of oxidative stress degrees and overcome it by increase manufacturing of antioxidants catalase and Glutathione peroxidase, but increase oxidative stress leads to continued oxidation and chain reaction of free radicals, which leads to oxidative damage.

Table 4 showed effect of feed additives on oxidation status values (MDA, PV, FFA), where are by-products from process of oxidation and peroxidation, as it represents best indicator or evidence for measuring oxidation in blood and tissue because it is more stable than hydrogen peroxides and Peroxide Value (Ayala *et al*, 2014). The control treatment recorded a significant decrease at (P≤0.01) in MDA values compared with treatments at age (42 days) and this due to not subjecting it to hydrogen peroxide challenge. As shown in Table 4 T_4 recorded a significant improvement when compared to T_2, T_3, T_5 and T_6 which achieved 65.8, 54.4, 52.5 and 50.4, respectively. Control treatment (T_1) recorded lowest values 35 at (P≤0.01). No significant differences between third, fifth and sixth treatments, these results demonstrated ability of green tea and organic selenium to scavenge free radicals , catechines and its different isomers play an anti-oxidant role by reducing free radicals through interaction with these radicals, also decrease reaction of hydrogen peroxide with lipids which causes liver toxicity (Abozid *et al*, 2018). Higher values of MDA in second treatment may be due to hydrogen peroxide and it role in oxidative stress and increase production of Reactive Oxygen Species (ROS) that attacks poly unsaturated fatty acids (PUFA) in cell membranes. This is consistent exist in Table 4, where an increase in MDA values that make a change in permeability of cellular membranes and lead to increase damage of liver cells (Muriel, 2009). In natural state, there is a balance between free radicals formed during biological processes and antioxidants production and this imbalance occurs in this equilibrium, such as a pathological condition or environmental pollution, such as the addition of H_2O_2 causes an oxidative stress, which foundation or substance occurs, causes an imbalance between the production of free radicals and ability of body to defense them or detoxification by antioxidants. One of reasons for increase in malondialdehyde levels (MDA) in T_2 (without antioxidants), Poly unsaturated fatty acids (PUFA) in diets and that increase free radicals formed by addition of H_2O_2 which could increase lipid peroxide and cause oxidative damage.

Data on Glutathione Peroxidase at 42 days of age in broiler chickens fed inclusion of organic Selenium and green tea to diets summarized in Table 4. The results show a significant improvement for T_4 (1 mg/kg Organic Selenium) , which achieved 230 compared to T_2 (H_2O_2 0.5% with drinking water), T_3 (green tea 1.5%), T_1 (control) and T_6 (1 mg/kg Organic Selenium + H_2O_2 0.50%) respectively. It is also noted that no significant differences appeared between treatments T_4 and T_5. Likewise, no significant differences were observed between T_5(1.5 % green tea + H_2O_2 0.50%), T_6 (1 mg/kg Organic Selenium + H_2O_2 0.50%) and T_1, which recorded a significant improvement (P≤0.01) in values of Glutathione Peroxidase (GHS-Px) 225, 220 and 240, respectively compared to T_3 which recorded 212 and T2 which recorded the lowest values 111. The decrease in values of Glutathione peroxidase in T_2 (H_2O_2 0.50%) may be due to fact that continuity with addition of hydrogen peroxide worked to obtain excessive oxidation with expense of body's production of antioxidant enzymes. It is an imbalance between the production of free radicals and ability of the body to produce antioxidant enzymes needed to stop chain of oxidative reactions or remove the toxins from it (Cheng *et al*, 2016). With regard to rise of antioxidant enzymes in additive treatments, these results are consistent with Yanishlieva-Maslarova *et al* (2001) in a study aimed to investigating effect of *Eimeria tenella*, a parasite responsible for acetic disease, on criteria related to oxidative stress in broilers, which indicated that a rise in values of antioxidant enzymes due to Ecological Oxidative Balance (EOB), which is weak oxidation balance in internal environment of chicks after oxidative stress occurred after experimental infection of birds. The significant differences in values of CAT, GSH-PX, MDA can be explained by role of green tea and organic selenium in reducing the occurrence of oxidative processes as well as its role in reducing negative effects of oxidation in the body. For this body need second

line of defense which was weak, antioxidant enzymes, so its concentration in addition treatments did not rise while its concentration in T_2 increased (Ighodaro and Akinloye, 2018). The researchers attributed the preventing role of green tea to flavonoids and their effects as an antioxidant and concludes that green tea has a preventive ability to protect liver in addition to its role as an antioxidant that will be useful when liver exposure to toxins. Also, it is noted an improvement in values of GHS-PX, which works to remove secondary products from metabolizes a group of hormones and some chemicals and an increase in glutathione improves condition of liver (Honda *et al*, 2017). Results were consistent with Haug *et al* (2007), which indicated that organic selenium is an effective antioxidant that works to inhibit oxidation of long-chain Poly unsaturated fatty acids (PUFA) in cell membranes and increase works to change level of antioxidant enzymes that works to inhibit activity of free radicals resulting from auto-oxidation.

CONCLUSION

Results of present study suggest that inclusion green tea at 1.5% and organic selenium at 1mg/kg feed in powder form to broiler chicken diets had affected on creatinine and uric acid in blood plasma, while in fact, diets supplementation with green tea and organic selenium was clear beneficial effects on antibody production against Newcastle and gumboro diseases and relative weights of lymphoid orgns which responsible of immunity. Nevertheless, results of addition natural feed additives were similar to the observed in previous studies without serious adverse effect on physiological performance and makes broilers with less fat.In conclusion, we can stated, that green tea and organic selenium may be good alternatives additives to antibiotic growth promoters in broiler chickens fattening.

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