

THE EFFECT OF DIETARY SUPPLEMENTATION OF ORGANIC CHROMIUM, ORGANIC SELENIUM AND VITAMIN E ON PHYSIOLOGICAL RESPONSES IN BROILERS UNDER NATURAL SUMMER CONDITIONS

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SUMMARY

An experiment was conducted to investigate the effects of increasing dietary levels of organic chromium, organic selenium and vitamin E for deleterious impacts of heat stress on broilers. A total of 120 1-d old commercial broiler chicks (Cobb) were obtained from a local hatchery. Upon arrival the chicks were divided into four groups with six replications, each of five chicks. The first group was the control group, while the second group was fed the basal diet supplemented with organic chromium (0.8 mg/kg diet), the third group was fed the basal diet supplemented with organic selenium (0.3 mg/kg diet) and the fourth group was fed the basal diet supplemented with vitamin E (200 IU/kg diet). Body weight (BW); body weight gain (BWG); feed intake (FI) and feed conversion ratio (FC) were significantly ($P \leq 0.05$) improved in treatment groups compared to the control at 21 and 42 days of the age. Red blood cell count (RBC), hematocrit (HT), hemoglobin (Hb), hemoglobin mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), hemoglobin mean corpuscular volume (MCV) and heterophils lymphocytes ratios (H/L ratio) were significantly ($P \leq 0.05$) different in vitamin E; organic selenium and organic chromium compared to the control at 21 and 42 days of the age, but RBC count at 21 days of age were not significant. Moreover, it has been noticed a significant ($P \leq 0.05$) increase occurrence in values of glutathione peroxidase (GPX), Super-oxide dismutase (SOD) and catalase (CAT) but the level of malondialdehyde (MDA) was significantly ($P \leq 0.05$) decreased by supplementing broiler diets with vitamin E; organic selenium and organic chromium, respectively compared to the control. Supplementing broiler diets with vitamin E; organic selenium and organic chromium improved ($P \leq 0.05$) the carcass characteristics, respectively compared to the control. However, these improvements were insignificant in abdominal fats, thymus, bursa and spleen. The present results indicate that the supplementation of diets with vitamin E; organic selenium and organic chromium, especially vitamin E, is necessary to overcome the deleterious effects of heat stress on broilers.

Keywords: *Broiler chicks, performance, vitamin E; organic selenium, organic chromium, blood parameters, antioxidant, glutathione peroxidase, super-oxide dismutase (SOD), catalase (CAT) and malondialdehyde (MDA).*

INTRODUCTION

Homeostasis is constantly challenged by intrinsic and extrinsic stressors (Lin *et al.*, 2006). Heat stress is of major concern for poultry industry, especially in the hot regions. The important traits governing productivity (growth performance, immune suppression and high mortality rate, etc.) are adversely affected by heat stress (Mujahid *et al.*, 2005, 2007 and Niu *et al.*, 2009). When the temperature exceeds 30 °C, signs of heat stress are likely to appear (Yardibi and Turkay, 2008). Biochemical and physiological events associated with hyperthermia can potentially promote reactive oxygen species formation which results in the disturbance of balance between the oxidation and anti oxidants defense systems, causing lipid peroxidation (LPO), in cell membranes, free radical peroxidation and oxidative injury in biological molecules, DNA and proteins. (Ando *et al.*, 1997; Lin *et al.*, 2006; Mujahid *et al.*, 2006 and 2008 and Aslam *et al.*, 2010). Further more, heat stress increases mineral and vitamin mobilization from tissues and their excretion, thus may exacerbate a marginal vitamin and mineral deficiency or an increased mineral and vitamin requirement. Several methods are available to alleviate the negative effects of heat stress, mostly focused on dietary manipulation. The major effects of heat stress are decreased feed intake and

lower weight gain (El Moniary, 1991, Cahaner and Leenstra, 1992, El -Moniary *et al.*, 1993 and Geraert *et al.*, 1996).

High temperature is enough to cause increased body temperature also change circulating leucocyte component in broilers and increased in H/L ratio (Altan *et al.*, 2000). Heat stress not only adversely affects production performance but also inhibits immune function (Mashaly *et al.*, 2004).

Cells generate small amounts of free radicals or reactive oxygen species (ROS) during their normal metabolism, Mujahid *et al.* (2006) reported that acute heat stress resulted in increased levels of ROS in mitochondria. Excessive levels of ROS result in the disturbance of balance between the oxidation and antioxidant defense systems.

There is an abundance of literature on possible techniques to alleviate the adverse effects of heat stress in broiler chickens. One of the practical approaches that have yielded promising results is altering birds' abilities to cope with high ambient temperatures. Converging evidence suggests that stressful experiences during the neonatal stage can have considerable impact on various facets of an animal's physiology and behavior.

Dietary chromium supplementation has been reported to have a positive effect on growth rate and feed efficiency of growing poultry under stress conditions (Sahin *et al.*, 2001 and Lien *et al.*, 1999). Stress increases chromium mobilization from tissues and its excretion and also depresses in humans and animals including poultry (Borel *et al.*, 1984 and Pardue *et al.*, 1985). Stress thus may exacerbate a marginal chromium deficiency or an increased chromium requirement, implying that both chromium. Selenium supplementation especially at 0.2 and 0.3 mg/ kg diet significantly ($P \leq 0.05$) increased live body weight of chicks (El-Sheikh *et al.*, 2010 and Zhou and Wang, 2011). In contrast, Patton *et al.* (2002) suggested that adding selenium yeast at 0.1, 0.2 or 0.3 ppm, was not significantly ($P < 0.05$) affected feed intake. Similar results were observed by (Spears *et al.*, 2003; Jiakui and Xiaolong 2004; Utterback *et al.*, 2005 and Ševčíková *et al.*, 2006).

Vit.E were discovered and its role as an antioxidants was further characterized (Wolf, 2005). The Vit.E that is integrated into cellular membranes exerts its antioxidant effects by intercepting peroxyl radicals more rapidly than can polyunsaturated fatty acids (Burton and Traber, 1990). Lin *et al.* (2005) found that cockerels receiving supplements of more than 40 mg/kg vitamin E had higher body weight gain.

Therefore, the objective of the present study is to elucidate the effect of increasing dietary supplements levels of organic chromium, organic selenium and Vitamin E on growth performance, some antioxidant and heat acclimation markers, hematological parameters and the carcass characteristics of broilers under heat stress conditions. Finally, results from this study could provide a fundamental knowledge for using antioxidants to reduce oxidative stress and improve the productive performance and some Physiological parameters in broiler production under hot climates of summer season in Egypt.

MATERIALS AND METHODS

A total of 120 1-d old commercial broiler chicks (Cobb) were obtained from a local hatchery. Upon arrival, the chicks were divided into four groups with six replications, each of five chicks. The first group was the control group, while the second group was fed the basal diet supplemented with 0.8 mg/kg diet organic Chromium, the third group was fed basal diet supplemented with 0.3 mg/kg diet organic selenium, the fourth group was fed basal diet supplemented with 200 IU/kg diet vitamin E. At 42-d of age, all groups were exposed to $41 \pm 1^\circ\text{C}$ for 1h.

The experimental diets and their calculated analysis are shown in Table (1). Feed and water were offered *ad libitum* and artificial light was provided for 23 hours daily allover the experimental period, which lasted for 6 weeks. Chicks of the four treatments were kept under similar hygienic and environmental conditions and vaccinated against common diseases.

Measurements:

Chicks were weighed at 1, 21 and 42 days of age (DOA). Body weight (BW) was recorded to the nearest 0.1g. The average body weight gain (BWG) was calculated by subtracting the average of initial body weight of the birds in a certain stage from the final one in the same stage. Feed intake was recorded to the nearest 0.1g and then the feed conversion ratio (FCR) was calculated as gram feed to gram body weight gain.

Blood samples were collected from birds at 21 and 42 DOA during their exsanguination into Waserman plastic tubes and Hemoglobin (Hb), hematocrit (HT), red blood cell count (RBS), hemoglobin mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), hemoglobin mean corpuscular volume (MCV) and heterophils to lymphocytes ratios (H/L ratio).

Hemoglobin (Hb) values as g/100ml. of whole blood were determined by cyanomethemoglobin method using commercial kit purchased from Spectrum Diagnostics Cairo, Egypt, as described by Tietz (1990). Red blood cell count (RBS_c) were performed by the method described by Natt and Herrick (1952), The blood samples were pipetted in heparinized microhematocrite tubes, centrifuged at 3000 r.p.m for 15 minutes and the hematocrite (Ht) percent was recorded, hemoglobin mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), hemoglobin mean corpuscular volume (MCV), were calculated depended on Hb, RBS_c and Ht. Blood smears were also done, stained with Wright's stain procedure and used to calculate the number of lymphocytes (L) and heterophils (H) in 100 white blood cells, then the H/L ratio was calculated.

After that the clotted blood was centrifuged at the speed of 4000 r.p.m for 15 minutes using laboratory centrifuge (SMIC, YJ03, Shanghai, China). Serum were decanted into (1.5 ml) Ependorf tubes determine Glutathione peroxidase enzyme (GPX) activity in plasma was determined using commercial kit according to Bell *et al.* (1986), superoxide dismutase (SOD) levels in plasma was measured by the method of Marklund and Marklund (1974), catalase (CAT) activity was determined by the method of Cohen *et al.* (1970) and Malondialdehyde (MDA) production was determined by the method described by Placer *et al.* (1966).

At the end of period (42 day of age), three birds from each treatment group were randomly taken, weighed and slaughtered. Feathers were manually removed and eviscerated, weights of Carcass, thigh, breast, liver, heart, gizzard and abdominal fat were also recorded to the nearest 1 gram and thymus, bursa and spleen were weighted and recorded to the nearest 0.01 gram.

Data were subjected to one way analysis of variance using the General Linear Model procedures (SAS, 1996), Differences among means were detected by using Duncan's multiple range test (Duncan, 1955).

Table (1). Composition and calculated analysis of the experimental diets.

Ingredient	Starter %, 1-21 D	Grower %, 22-42 D
Yellow corn	55.35	60.45
Soybean meal (44%)	27.5	25
Corn gluten meal (62%)	8.9	6
Mono-Ca phosphate	1.65	1.6
Limestone	1.7	1.65
Vegetable oil	3.7	4.15
Salt	0.45	0.4
Methionine	0.15	0.15
Lysine	0.3	0.3
Vitamin and mineral mixture	0.3	0.3
<u>Total</u>	100	100
<u>Calculated analysis:</u>		
ME kcal/kg	3143	3190
Crude protein %	22.77	20.31
Lysine %	1.27	1.19
Methionine %	0.55	0.51
Methionine+ Cystine %	0.93	0.85
Calcium %	1.00	0.97
Av.Phosphorus %	0.47	0.47

*: Vitamin and mineral mixture per kg of diet: Each kilogram of diet contains = VA, 12000 I.U., D3, 2500 I.U., E, 10mg., B1, 2mg., B2, 5mg., B6, 4mg., B12, 10µg., Niacin, 25mg., Pantothenic acid, 10mg., Biotin, 50µg., Folic acid, 1000µg., and Coline chloride, 255mg. Selenium, 300µg., Copper, 10mg., Iodine, 1.0mg., K, 2.0mg., Iron, 33mg., Manganese, 60mg.and, 60mg. Zinc.

RESULTS AND DISCUSSION

Productive performance:

Effect of different treatments on BW, BWG, F.I and F.C traits of broilers chickens were illustrated in Table 2, It appeared that BW at 21 and 42 day of age were Significantly increased (P<0.05) from vitamin E, organic selenium and organic chromium respectively compared with control. A similar trend was also observed for BWG values at the same periods.

It is generally observed that feed intake was significantly ($P<0.05$) increased in vitamin E, organic selenium and organic chromium compared with control at periods from 1 to 21, 21 to 42 and 1 to 42 days of age, the same trend in improvements was observed when we calculated the feed conversion ratio at these periods, which the improvement in feed conversion ratio were significantly ($P<0.05$) at 21 to 42 and 1 to 42 days of age, but at 1 to 21 were insignificant.

Table (2). Effect of dietary supplements by organic chromium, organic selenium and Vitamin E on productive performance of broiler chicks at different ages.

Trait	Treatment				Sign.
	Overall mean \pm SD				
	Control	Chromium	Selenium	Vit. E	
B.W, 1 D.O.A	41.21 \pm 0.617	40.86 \pm 0.254	40.80 \pm 0.488	41.14 \pm 0.129	NS
B.W, 21 D.O.A	611.00 ^c \pm 21.86	633.33 ^{bc} \pm 25.81	650.00 ^{ab} \pm 37.41	675.00 ^a \pm 35.07	*
B.W, 42 D.O.A	1730.17 ^b \pm 62.05	1846.83 ^a \pm 22.67	1867.50 ^a \pm 59.11	1885.67 ^a \pm 57.97	*
B.W.G, 1-21	569.79 ^c \pm 21.34	592.48 ^{bc} \pm 25.57	609.20 ^{ab} \pm 36.97	633.86 ^a \pm 34.95	*
B.W.G, 21-42	1119.17 ^b \pm 42.50	1213.50 ^a \pm 9.13	1217.50 ^a \pm 26.21	1210.67 ^a \pm 43.39	*
B.W.G, 1-42	1688.96 ^b \pm 61.46	1805.98 ^a \pm 22.43	1826.70 ^a \pm 58.65	1844.52 ^a \pm 57.87	*
F.I, 1-21	794.33 ^c \pm 12.14	814.33 ^b \pm 5.24	820.16 ^b \pm 8.06	833.50 ^a \pm 3.39	*
F.I, 21-42	2553.50 ^b \pm 1.04	2586.33 ^{ab} \pm 1.21	2580.83 ^{ab} \pm 4.83	2615.50 ^a \pm 82.01	**
F.I, 1-42	3347.83 ^b \pm 13.13	3400.67 ^{ab} \pm 6.15	3401.00 ^{ab} \pm 12.83	3449.00 ^a \pm 84.62	*
F.C, 1-21	1.315 \pm 0.032	1.350 \pm 0.052	1.376 \pm 0.072	1.395 \pm 0.069	NS
F.C, 21-42	2.28 ^a \pm 0.087	2.13 ^b \pm 0.016	2.12 ^b \pm 0.041	2.16 ^b \pm 0.047	*
F.C, 1-42	1.9 ^a \pm 0.065	1.88 ^b \pm 0.020	1.86 ^b \pm 0.053	1.87 ^b \pm 0.016	*

^{a, b, c} Means within a row with different superscripts are significantly different ($P\leq 0.05$).

NS = non-significant, * = $P\leq 0.05$; ** = $P\leq 0.01$.

It is suggested that supplemental treatments could improve heat tolerance and alleviate performance reduction associated with stress conditions. These findings are in agreement with those reported by Sahin and Kucuk (2001), who indicated that the synthesis of vitamin E is reduced during heat stress, we used it in poultry diets because of their anti-stress effects, their increased on body weight and feed intake during heat stress, also Ferket and Qureshi (1992) showed that the supplementation of vitamin E on broilers diets submitted to heat stress improved weight gain and feed conversion ratio. Hegazy and Adachi (2000) observed a significant improvement in weight gain and feed conversion ratio of birds were fed dietary supplementation of organic Se. (Edens, 2001; Spears *et al.*, 2003 and Niu *et al.*, 2009) who found a significant influence on body weight and feed intake by dietary supplementation of organic Se. (Sahin *et al.* 2008; Chun *et al.*, 2009; Attia *et al.*, 2010; Zhou and Wang 2011; and Yang *et al.*, 2012) revealed that, a linear increase in feed intake and improvement in feed efficiency was found in Se-supplemented groups were reared under heat stress conditions.

It is hypothesized that supplementation of Cr to the diet improves nutrient utilization such as carbohydrates and proteins by the role of Cr in increasing the sensitiveness of insulin receptors to insulin hormone and consequently increases the absorption of glucose by cells and considering that trivalent Cr is needed for proper metabolism of glucose in the body and the amount of Cr affects the ability of insulin in maintaining blood sugar levels (NRC, 1997) therefore by increasing levels of chromium in the diet, the metabolism of nutrients, including carbohydrates completely done and insulin hormone can also maintain blood sugar at longer time intervals, the chromium role in improving amino acid uptake by tissues and muscle cells and increases in protein retention in the body, this finding are illustrated the improvement on BW, BWG, FI and FCR by organic Cr supplementation which agreement with (Lien *et al.*, 1999; Contreras *et al.*, 2000; GURSOY, 2000; and Sahin *et al.*, 2002, 2005).

Hematological parameters:

Data presented in table 3 clarify the effect of supplements of organic chromium, organic selenium and Vitamin E on red blood cell count (RBS), hematocrit (HT), hemoglobin (Hb), hemoglobin mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), hemoglobin mean corpuscular volume (MCV) and heterophils lymphocytes ratios (H/L ratio), present data show significantly ($p<0.05$) improvement on the Hematological parameters were studied by the supplemented dietary than the control, but RBS count at 21 day of age were not significant. Free radicals and peroxides play a significant role in physiological phenomena and in the pathogenesis of various diseases and are thought to participate in ageing, damaging oxidative tissues and increasing stress (Guemouri *et al.*, 1991). The biological effects of these highly reactive substances are controlled *in vivo* by an endogenous antioxidant system, consisting of vitamin E and selenium, which improvement the blood parameter

Vitamin E works to protect phospholipids of cellular membranes and sub cellular, by preventing the oxidation of fatty acids with unsaturated bonds. This antioxidant effects of vitamin E in efficient high concentrations of oxygen and therefore is concentrated in red blood cells and membranes Dowd and Zheng (1995) and Muller (2010). Our results are in accordance with Shlig (2009) obtained his results from selenium and Vitamin E. Determination of some blood parameters has a substantial merit in understanding metabolic changes in heat-stressed poultry fed with dietary vitamin E and selenium.

The results from the present study show that, hemoglobin increased significantly and hematocrit tended to increase in broilers fed supplemental Cr. Wilson (1971) reported that hematology may be used to diagnose both quantitative and morphologic physiological alterations that might be associated to heat stress, such as changes in hematocrit and hemoglobin. According to Kubena *et al.* (1972) exposure of chickens to high temperatures causes a decrease in blood hematocrit and hemoglobin values. Toghiani *et al.* (2006) reported that Cr supplementation increased hematocrit of stressed broiler. Research on Cr and its hematological effect in broiler are very limited.

Table (3). Effect of dietary supplements by organic chromium, organic selenium and Vitamin E on Hematological parameters of broiler chicks at different ages.

Age (days)	Trait	Treatment				Sig n.
		Overall mean ± SD				
		Control	Chromium	Selenium	Vit. E	
(at 21 D.O.A)	Hb	8.56 ^b ±0.404	10.90 ^b ±0.100	10.80 ^b ±0.200	11.40 ^a ±0.200	*
	HT	30.0 ^b ±2.00	33.3 ^a ±1.15	33.6 ^a ±1.52	35.0 ^a ±1.00	**
	RBS _c ×10 ⁶	3.20±0.200	2.83±0.152	2.93±0.416	3.14±0.020	NS
	MCH	26.78 ^a ±0.47	38.54 ^a ±2.20	37.33 ^a ±5.44	36.30 ^a ±0.55	*
	MCHc	28.60 ^b ±1.56	32.72 ^a ±0.90	32.11 ^a ±1.41	32.58 ^a ±0.82	*
	MCV	93.87 ^b ±6.25	117.84 ^a ±7.01	116.02 ^a ±13.91	111.47 ^a ±3.57	**
(at 42 D.O.A)	H/L ratio	0.63 ^a ±0.010	0.54 ^b ±0.026	0.53 ^b ±0.010	0.51 ^b ±0.010	**
	Hb	9.36 ^b ±0.416	13.06 ^a ±0.493	12.96 ^a ±0.416	12.90 ^a ±0.20	*
	HT	26.33 ^b ±0.57	29.33 ^a ±1.15	29.66 ^a ±1.52	31.00 ^a ±1.00	*
	RBS _c ×10 ⁶	3.10 ^c ±0.10	3.80 ^b ±0.20	4.03 ^{ab} ±0.15	4.16 ^a ±0.02	*
	MCH	30.21 ^c ±0.93	34.411 ^a ±1.04	32.16 ^b ±1.24	31.01 ^{bc} ±0.42	*
	MCHc	35.56 ^b ±1.19	44.61 ^a ±2.91	43.83 ^a ±3.59	41.63 ^a ±1.18	*
	MCV	85.00 ^a ±3.25	77.42 ^b ±6.79	73.69 ^b ±6.09	74.52 ^b ±2.59	**
	H/L ratio	0.65 ^a ±0.010	0.50 ^b ±0.026	0.49 ^b ±0.010	0.48 ^b ±0.010	*

^{a-b} Means within a column with different superscripts are significantly different ($P \leq 0.05$).

NS = non-significant, * = $P \leq 0.05$; ** = $P \leq 0.01$

Antioxidant Status:

Results determined of the present study (Table 4) showed that significantly ($p < 0.05$) higher activities of Glutathione peroxidase (GPX), superoxide dismutase (SOD) and catalase (CAT) enzymes at 42 DOA from vitamin E, organic selenium and organic chromium compared with control. However, All supplemented groups showed significantly ($p < 0.05$) lowered malondialdehyde (MDA) level than control at the same age. Glutathione peroxidase GPX activity was highest in Se-fed broilers (Mahmoud and Edens, 2003). Edens *et al.* (2001) reported that Se-fed broilers catalyzed a more rapid oxidation of GSH to GSSH because of higher glutathione peroxidase activity, These effects were similar to the present study The results of increasing glutathione peroxidase activity in plasma by organic Se in the present experiment are in agreement with the previous reports by Jiang *et al.* (2009) who reported that the addition of 0.15mg/kg or 0.225 mg/kg Se-Met significantly elevated GPX activities in broiler. Also, Previous work has also shown that Se supplementation increases the activity of GPX in the tissues (Surai., 2000) , which in close agreement with our results . It is well documented that, selenium is an important component of the antioxidant enzyme GPx and it's actively involved in the antioxidant defense systems (Chantiratikul *et al.*, 2011).

Se is an essential component of Se-dependent glutathione peroxidase enzyme, which reduces peroxide and protects cells against the damaging effects of oxidation (Reddy *et al.*, 2009). (Jianhua *et al.*, 2000, and Payne and Southern, 2005) recorded that dietary Se supplementation increased the plasma GPx activity in the broiler chickens. Khajali *et al.* (2010) found that the inclusion of organic Se source (selenomethionine) significantly elevated plasma GPx activity when measured at 42 days of bird's age, which can be regarded as an improvement of antioxidant status. In addition to the role of Se as antioxidant, methionine moiety can be converted to cysteine which in turn, converts to GSH. Both

cysteine and GSH can function as direct scavengers of reactive oxygen species (ROS). GSH and cysteine can also protect proteins from irreversible oxidative damage through interactions between these thiols and proteins and the formation of mixed disulfides, such as glutathiolated protein (Mallis *et al.*, 2002).

Organic Selenium is actively absorbed by the same transport pathways as methionine and later incorporated (non-specifically) into tissues in place of methionine (Wolffram, 1999). Vitamin E functions as the primary chain-breaking antioxidant in the avian body, by scavenging free radicals and inhibiting the propagation of lipid oxidation (Jensen *et al.*, 1995; Surai, 2006). Selenium plays an integral supportive role to Vit.E as a component of several glutathione peroxidases (GSH-Px), which are responsible for the cellular removal of the precursors of free radicals (i.e., hydro-peroxides) (Surai, 2006; Puvača and Stanačev, 2011).

In addition, Se is involved in the recycling of Vit.E through the seleno-enzyme thioredoxin reductase, which recycles ascorbic acid and, in turn, promotes the recycling of Vit.E (Surai, 2002; Surai, 2006; Skřivan *et al.*, 2008). In fact, it has been shown that the action of these Se-dependent enzymes often display a “sparing” effect on the Vit.E stores in poultry by reducing the demand on Vit.E for the inhibition of lipid peroxidation, ultimately rendering more Vit.E available to the animal (Surai, 2000; Surai, 2006; Skřivan *et al.*, 2008).

The results of SOD activity is in accordance with the earlier reports (Eid *et al.*, 2008 and Sujatha *et al.*, 2010), they found significant increase in SOD activities of vitamin E supplemented groups. It is suggested that antioxidant vitamin E supplementation during heat stress enhances SOD activity to minimize oxidative stress in chicken by inhibiting the oxygen free radical production and scavenging the superoxide ions (Ozturk-urek *et al.*, 2001). As the stress period increased the enzyme activity in control, upward trend was observed in all supplemented groups. The result also clearly explains the synergistic effect of vitamin E (Sahin *et al.*, 2002 and 2003).

Combination with vitamin E and organic selenium increasing SOD activities, this result are confirms the findings of (Hill, 1992; Ogo *et al.* 1996; and Tras *et al.*, 2000).

In the cell, catalase reacts with generated hydrogen peroxide to form water and molecular oxygen thereby protecting the cells against hydrogen peroxide toxicity and lipid peroxidation (Yamaguchi, 1991). Erythrocyte CAT levels in the vitamin E supplemented groups under heat stress demonstrated an increasing trend, as compared to the control group, might be due to synergistic effect of vitamin E on scavenging free radicals and hydrogen peroxides, that is well supported by the findings of others (Panda *et al.*, 2007; Eid *et al.*, 2008 and Yardibi and Turkey *et al.*, 2008).

As environmental temperature increases respiration and evaporation increases to maintain optimal body temperature, which in turn increases their metabolism and energy consumption. If increased energy needs is not supplied with feed (less feed consumption during heat stress), mobilization of lipids from stored fat takes place. The MDA level, an indicator of lipid per-oxidation, increases with increases in lipid mobilization and oxidation of lipids. Level of erythrocyte MDA continued to rise in hens exposed to heat stress (control) and hens fed rations containing vitamin E separately or their combinations showed significantly lower MDA level. Feeding of the antioxidant vitamins for during summer stress decreased the MDA concentration from its level recorded at one month. This result is in accordance with the earlier findings (Puthongsiriporn *et al.*, 2001; Panda *et al.*, 2007; Sahin *et al.*, 2002 and Lin *et al.*, 2005). The observed decrease in erythrocyte MDA levels in the groups supplemented with vitamin E and their combinations might have been due to the inhibition of lipid peroxidation in erythrocyte membranes due to the antioxidant effect of vitamin E. This study suggests that lipid peroxides formed under heat stress conditions can be partially counteracted by dietary inclusion of antioxidants such as vitamin E.

Table (4). Effect of dietary supplements by organic chromium, organic selenium and Vitamin E on Antioxidant Status at 42 day of age.

Trait	Treatment				Sign.
	Control	Chromium	Selenium	Vit. E	
GPX, (U/ml)	3.86 ^c ±0.29	8.07 ^b ±0.06	9.38 ^a ±0.56	9.44 ^a ±0.66	*
SOD, (U/ml)	1.75 ^c ±0.07	2.63 ^b ±0.10	3.04 ^a ±0.21	3.03 ^a ±0.13	*
CAT, (U/ml)	24.64 ^d ±0.13	26.82 ^c ±0.09	34.04 ^a ±0.73	30.30 ^b ±1.48	*
MDA, (nmol/ml)	5.92 ^a ±0.35	4.93 ^b ±0.44	4.10 ^c ±0.02	4.13 ^c ±0.01	*

^{a,b and c} Means within a column with different superscripts are significantly different ($P \leq 0.05$).

NS = non-significant, * = $P \leq 0.05$; ** = $P \leq 0.01$

It is well known that Cr plays an important role as integral component of the Glucose Tolerance Factor (GTF), which potentate the action of insulin and regulate fat metabolism Mertz (1993). It has been well recognized that insulin metabolism influences lipid per-oxidation (Gallaher *et al.*, 1993). Cr is

insulin cofactor, therefore postulated to function as an antioxidant (Preuss *et al.*, 1997). According to antioxidant theory Klasing (1993), when the concentrations of antioxidant decrease, lipid per-oxidation increases in the plasma and tissues, leading to damage of cell membranes. (Sahin *et al.*, 2003) reported supplemental Cr resulted in decrease MDA in serum concentrations of heat-stressed broiler chicks, they added, when Japanese quails were fed by Cr, MDA concentration in serum decreased.

Carcass characteristics and some organs:

The overall means of carcass weight (%) and the proportional weights of some body organs are presented in Table 5, illustrated that at marketing age (42 DOA), Dressing carcass weight, breast, thigh, gizzard and heart as a percentage of live body weight was markedly significantly ($P \leq 0.05$) higher for supplemented diet with vitamin E, organic selenium and organic chromium compared with control, but this increase were insignificant in the relative weights of abdominal fats, thymus, bursa and spleen. However, the relative weights of liver were significantly ($P < 0.05$) decrease for all treatment compared with control.

These results may support the previous findings that broilers are able to compensate the retardation of growth by different magnitudes including the well documented relationships between the dietary supplements and some endocrine functions which include the stimulatory effects of vit.E, organic Sel. on thyroid gland activity under heat stress conditions. It is well known that the normal growth of all body organs needs a euthyroid status, In accordance with the results of the performance data of the present study It has been suggested that thyroid activity is affected by environmental temperature which made linear correlation between plasma T₃ concentration and feed intake and weight gain to a good improvement in carcass characteristics by increasing its weights (McNabb and King., 1993; Yahav *et al.*, 1997; Yahav 1999 and Sahin *et al.*, 2002).

In this study non significant effect of Cr supplementation on abdominal fat is in agreement with those of Lien *et al.* (1999), but is in incoherence with those of Ward *et al.* (1993), Hossain *et al.* (1998) and Sahin *et al.* (2003).

Table (5). Effect of dietary supplements by organic chromium, organic selenium and Vitamin E on Carcass characteristics and some organs of broiler chicks.

Trait (%)	Treatment				Sign.
	Control	Chromium	Selenium	Vit. E	
Caracas	66.17 ^c ±0.493	68.94 ^b ±0.519	70.87 ^a ±1.050	69.77 ^{ab} ±0.404	*
Breast	14.93 ^b ±0.904	15.60 ^{ab} ±0.163	15.97 ^a ±0.152	15.88 ^a ±0.224	*
Thigh	11.60 ^b ±0.904	12.40 ^{ab} ±0.190	12.91 ^a ±0.556	12.84 ^a ±0.550	*
Liver	2.08 ^a ±0.041	1.85 ^b ±0.060	1.83 ^b ±0.045	1.86 ^b ±0.092	*
Gizzard	1.10 ^b ±0.015	1.15 ^a ±0.037	1.16 ^a ±0.025	1.17 ^a ±0.015	*
Heart	0.34 ^b ±0.026	0.40 ^a ±0.015	0.42 ^a ±0.011	0.43 ^a ±0.030	*
A.F	1.13 ±0.011	1.16±0.010	1.18 ±0.026	1.20 ±0.106	NS
Thymus	0.23±0.015	0.25±0.017	0.24±0.015	0.24±0.005	NS
Bursa	0.14±0.035	0.13±0.051	0.13±0.020	0.12±0.011	NS
Spleen	0.23±0.020	0.22±0.015	0.21±0.005	0.22±0.005	NS

^{a, b, c} Means ± SD within a row with different superscripts are significantly different ($P \leq 0.05$).

NS = non-significant, * = $P \leq 0.05$.

A.F = abdominal fat (%)

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تأثير إضافة الكروم العضوي، السيلينيوم العضوي وفيتامين هـ الى العليقة على الاستجابة الفسيولوجية لدجاج التسمين تحت ظروف فصل الصيف.

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استهدفت الدراسة محاولة تخفيف التأثيرات الضارة لارتفاع درجات الحرارة على الأداء الفسيولوجي لدجاج التسمين خلال فترة الانتاج وذلك عن طريق اتباع اضافة الكروم العضوي ، والسيلينيوم العضوي وفيتامين هـ الى علائق دجاج التسمين و تم تربية كتاكيت التسمين فى بطاريات مناسبة مع تقديم كافة اساليب الرعاية المتكامله لهم.

وكانت اهم النتائج المتحصل عليها ان الاضافات العلفية (فيتامين هـ ، السيلينيوم العضوي والكروم العضوي) حسنت بالترتيب بصوره معنويه مقارنة بالكنترول

- معدلات الاداء (BW- BWG- FI- FC ratio).
- بعض قياسات الدم منها (Hb- HT- RBSc- MCH- MCV- MCHc- H/L ratio).
- الانزيمات المضادة للاكسده (GPX- CAT- SOD) وكذلك مادة المالوندايديد (MOD).
- قياسات الذبيحه ولكن كانت الفروق غير معنويه للاوزان النسبيه لبعض الغدد ودهن البطن