

The Eurasia Proceedings of Science, Technology, Engineering & Mathematics (EPSTEM), 2018

Volume 3, Pages 145-151

ICVALS 2018: International Conference on Veterinary, Agriculture and Life Science

# Effect of Vitamin E, C and Aspirin in the Performance, Lipid Peroxidation and Blood Biochemistry Traits of Broiler in Heat Stress

# Salim JEBUR

Ministry of Agriculture

# Thafer MOHAMMED

University of Anbar

#### Firas AL-KHALANI

State Board of Agricultural Research Ministry of Agriculture

**Abstract**: This Study was conducted Agricultural Research Department of the Ministry of Agriculture located in Abu Garib region from Oct 16th 2017 until Nov 17th 2017, where 294 chicks from the Ross-308 broiler, they were distributed in 7 treatments (42 chicks /treatment) with 3 replicate per treatment and 14 chicks per replicate, the first treatment was the control without added to ration, whereas, the vitamin E was added at rate of 250 and 500 mg / kg of feed in 2nd and 3rd treatment respectively , in 4th and 5th treatments the vitamin C was used at rate of 250 and 500 mg / kg of feed respectively, the aspirin (Asp) was added at rate of 250 and 500 mg / kg of feed in 2nd and 3rd treatment respectively, which amounted to 42 days and the temperature was 35 Celsius throughout the study .The results showed the following: There was an actual increase ( $P \le 0.01$ )in all additive treatments compared with control treatment in both live body weight, weight gain, and relative growth rate , while at feed consumption was significantly higher ( $P \le 0.05$ ) treatment T3 compared to T1, In the feed conversion factor ( $P \le 0.05$ ) was significantly improved in treatment T5 compared with T1. There was a significant decrease in ( $P \le 0.05$ ) treatment T6 in MDA compared with T1. At the glucose level, there was a significant difference ( $P \le 0.05$ ) in treatments T3 and T6 compared with T4. Add transactions and control treatment, indicating the role of additives used in the protection from the effect of exposure to high temperature and its ability to improve the performance and decrease Lipid peroxidation.

**Keywords:** Aspirin, Broiler performance, Lipid peroxidation

## Introduction

The high temperatures are so dangerous on birds and make them uncomfortable and lead to an increase in the rate of breathing immediately if exposed to them in addition to increase the consumption of water and consumption of feed decreases, and consequently the rise in temperature of the body of the chicken to more than 43 ° c and birds become not able to breathe normally and then lead to the loss of birds (hui zhang et al., 2016). sahin and others (2015) said that high temperature leads to a decrease in the defense system antioxidant in the body, leading to an increase in the level of free radicals that cause an increase in oxidative processes and the occurrence of oxidative stress and decrease in immunity. sohail et al. (2011) noted that high temperatures work more than twice as much as production performance through reduced feed consumption and feed conversion factors, followed by a decrease in body weight and high temperatures in the release of corticosteroid hormone, which causes endocrine disruptive disorders. low metabolic rate with oxidation of unsaturated fats in the body and weak immune response, making the body more susceptible to disease. therefore, many ways and means have to be used to reduce the high temperature of birds as an addition of aspirin (hassan, 2008). or vitamin e and c (rahawi, 2010). vitamin e is one of the most powerful natural antioxidants because it works to protect unsaturated fatty acids inside and outside the body's cells from free radicals and prevents free radicals reactions from respiratory processes (englmaierov et al., 2011). vitamin c clears all varieties oxygen is effective and has a

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protective effect against the inhibition of the antioxidant enzymes in the body which positively reflects on the productive performance (hajati et al., 2015). aspirin is a non-steroidal anti-inflammatory drug that is more effective in inhibiting the formation of prostaglandins by inhibiting the cyclo-oxygnase enzyme. this enzyme is composed of cox-2, which is responsible for stimulating inflammatory responses and cox-1, it is responsible for stimulating the formation of prostaglandins and thus free radicals are formed with the steps of prostaglandins synthesis. when this enzyme is inhibited, the function of aspirin here is as an antioxidant, protecting the body from free radical damage and lowering body temperature (Ghalib et al., 2011).

#### Method

This Study was conducted Agricultural Research Department of the Ministry of Agriculture located in Abu Garib, where 294 chicks from the Ross-308 broiler. And then randomly distributed to 21 pen  $(1.8 \times 2.50 \text{ m})$  were provided with nipples system drinker and hanging feeder during the experimental period (7 to 42 days of age). they were distributed in 7 treatments (42 chicks /treatment) with 3 replicate per treatment and 14 chicks per replicate, the first treatment was the control without added to ration, whereas, the vitamin E was added at rate of 250 and 500 mg / kg of feed in 2nd and 3rd treatment respectively , in 4th and 5th treatments the vitamin C was used at rate of 250 and 500 mg / kg of feed respectively, the aspirin was added at rate of 250 and 500 mg / kg of feed in 2nd and 3rd treatment respectively.

Where the continuous lighting system (24 hours / day) in the house to the end of the experiment, which amounted to 42 days and the temperature was 35 Celsius throughout the study. Feed and water was administered ad libitum for all treatments, (Starter diet :age 1-11 days; grower diet: age 11-22 days and Finisher diet : age 23-24 days) were the same for all the five treatments (Table 1), and formulated according to the recommendations of the National Research Council of the US (National Research Council, 1994)

Table 1. Composition and calculated nutrient content of the experimental diets (%)

Ingredients	Starter	Grower	Finisher
Corn	54.3	60	62.05
Soybean meal*	35	29.5	27
Concentrate protein**	5	5	5
Fat	3	3.55	4
Limestone	1.3	1.1	1.1
Di calcium phosphate	0.8	0.5	0.5
Salt	0.1	0.1	0.1
Methionine	0.3	0.15	0.15
Lysine	0.2	0.1	0.1
Calculated no	utrient content		
Protein Crude	23.4	21.3	20.2
Metabolism energy $(Kg)$	3048	3155	3203
Methionine+ Cysteine %	1. 2	1.3	1.23
Lysine	1.53	1.3	1.23
Calcium%	1	0.84	0.83
Phosphorus	0.46	0.4	0.39
Energy: protein ratio (C:P ratio)	130.38	158.06	157.94

<sup>\*</sup> Soybean meal used from Argentine origin Crude protein content 48% and 2230 kg / kg represented energy.

The vitamin E , C and aspirin were added in the form of powder (Juvedco Jordan), based on the active ingredient of vitamin E (alph-tochopherol acetate) and the active ingredient of vitamin C (L-ascorbic acid), and the active ingredient of aspirin (acetylsalicylic acid ) which were 50%, 98% and 50% respectively.

Mortality was recorded daily, and chick weight and feed intake per pen were measured on d 7,14,21,28,35 and 42 for determination of weight Gain, Feed Consumption, feed conversion rate (FCR), Relative growth rate

<sup>\*\*</sup> Proteins used in the production of Dutch Holland (imported) Wafi containing 40% crude protein, 2107 kgs / kg protein represented energy, 5% raw fat, 2.20% raw fiber, 4.20% calcium, 2.65% phosphorus , 3.85 Lysine, 3.70% methionine, 4.12% methionine + cysteine. It contains a mixture of rare vitamins and minerals that satisfy the bird's needs from these elements

(RGR) , production efficiency factor(PEF) , and economic indicator by Ross 308 Broiler Performance Objectives - Aviagen , 2016.

Liver blood samples from 3 chicks per replicate of each treatment were collected on Days 21 and 42 ,.the blood samples were collected from bronchial vein, for biochemical blood plasma parameters. The collected blood samples, during cervical dislocation, were distributed in tube have anticoagulant (EDTA). Blood samples in the tube were centrifuged (at  $2000 \times \text{rpm}$  for 10 min) and plasma was separated and then stored at -20 C until assayed for measuring blood parameters , 25 (g) of liver was taken from chicks and stored at -20 C for analysis.

The concentration of total protein ,albumin protein (gm /100ml) , globulin (g/dl) , glucose (dl/mg), cholesterol , Triglycerides, High density lipoproteins (HDL) , Low density lipoproteins (LDL) (mlg /100 ml) and the activities of ALT ,AST (IU/L) in plasma and The thiobarbituric acid reactive sbustances content, expressed as malondialdehyde (MDA) (Ml.m/Kg) equivalents, and the peroxid value(Mlg/Kg) and free fatty acid(%) in liver were determined using the commercial assay kits purchased from Nanjing Jiancheng Institute of Bioengineering (Jiangsu, China) following the standard procedures described by the manufacturer. The water used in the chemical analysis was ultrapurified.

#### Statistical

Data of percentage, lipid peroxidation and biochemical parameters , were first transfer to arcsine; then all transformed data were analyzed using One Way ANOVA analysis to assess the effect of Naturally and Synthetic antioxidant treatments on selected parameter of Layer Hens. The analysis were applied by using General Linear Model (GLM) procedure of statistical software package SAS version 9.1 (SAS Institute, 2012), P-values less than 0.05 and 0.01 were considered to be significant between treatments and the significant differences were examined using Duncan Multi Border Test, and the results were presented as mean/SEM (pooled).

#### **Results and Discussion**

#### Performance

The results of the study showed a significant superiority (P<0.01) for all additive treatments on the control treatment in both the mean body weight, the weight gain and the relative growth rate. In the feed consumption a significant improvement rate (P<0.05) in the T3 treatment on the T1 treatment. With treatment of T5 ,In the mortality The percentage of mortality was about 0.00% for all treatments, (P<0.01) in the T5 treatment on the T1 treatment, while not significantly different with the rest of the treatments except T7 , In the feed conversion the a significant improvement rate (P<0.05) in the T5 treatment on the T1.

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The reason for the ability of the addition treatment in improved the production performance compared with treatment of control is due to the effect of additives that contributed to reducing the heat stress on the birds elucidate Vitamin E is one of the most powerful antioxidants in the body. It prevents the formation of free radicals in the lipid cells. By blocking the formation of free radicals and thus provides an important cover in the protection of unsaturated fatty acids (Panda et al., 2008). Vitamin C acts as an antioxidant in the body through rapid oxidation to form a compound (Dehydroascorbic acid) this compound protects cell membranes and unsaturated fatty acids in membranes Oxidation due to oxidation For free radicals (Zahraa, 2008). Aspirin acts as an antioxidant because it has a role in reducing the production of free radicals by reducing the production of prostaglandins, as well as reducing the viscosity of blood resulting from a high rise in alkalinity of blood, where this increase in alkalinity of blood to rise in blood pressure, Aspirin plays a role in maintaining the structure of the heart and causes blood vessel expansion and improves blood flow to important organ such as the liver and kidneys, aspirin works to prevent inflammation and damage of free radicals by inhibiting an enzyme CoX-2, that enzymes work on oxygen consumption and conduct special oxidative reactions consisting of the group OH then the level of secretion of the stress hormone will decrease, resulting in a decrease in the disintegration of peroxides and thus decrease number of MDA (Madamanch et al., 2005 and huizhang, 2016). The reason for the significant improvement in dietary treatment Production efficiency factor and Economic Indicator in the treatment of vitamin T5 to the effect of vitamin A, elucidate Farooqi et al. (2005) that the addition of vitamin C to the diets of birds exposed to high temperature lead to a decrease in the proportion of hormone corticosterone

in the blood, An improvement in the secretion of thyroxine from the thyroid gland, which plays a role in maintaining the metabolism of the body and regulate the temperature in birds, which indicates the ability of the vitamin in regulating the temperature of the body of birds and thus an improvement in the coefficient of feed conversion of birds.

Table 2. Effect of dietary of vitamin E, C and aspirin on the performance of broilers (g).

		Diets treatments							
Traits T1		Vit E mg / kg		Vit C mg / kg		Asp. m	g / kg	SEM	P-
Traits	Control	T2 (250)	T3 (500)	T4 (250)	T5 (500)	T6 (250)	T7 (500)	SLM	value
Body Weight	1366 b	2028 a	2040 a	1950 a	1843 a	1835 a	1829 a	138.6	0.01
Weight Gain	1327 b	1990 a	2002 a	1913 a	1805 a	1796 a	1791 a	138.3	0.01
Feed Consumption	2480 с	3194 ab	3229 a	312 ab	2474 bc	3074 ab	ab 3078	378.9	0.05
Feed conversion	1.68 a	1.60 ab	1.61 ab	1.63 ab	1.37 b	1.71 a	1.72 a	0.170	0.05
Mortality %	2.38	0.00	0.00	0.00	0.00	0.00	2.38	2.06	N.S
Relative growth rate	188.9 b	192.6 a	192.6 a	192.4 a	191.7 a	191.7 a	191.7 a	0.6	0.01
Production efficiency factor	157.8* c	158.1 ab	259.1 ab	244.1 ab	286.8 a	219 abc	216.6 bc	35.28	0.01
Economic Indicator	161.6 c	158.1 ab	259.1 ab	244.1 ab	286.8 a	219 abc	220.7 bc	33.97	0.01

The different litters in the column referred to a significant differences between treatments at rate of 0.01 and 0.05.

#### **Lipid Peroxidation**

The results showed that the efficacy of aspirin to control lipid Peroxidation was significantly higher (P < 0.05) compared to the T1 treatment. Salicylic acid is the active substance in aspirin, which is the property of heat reduction in the body. Salicylic acid reduces the formation of prostaglandins by inhibiting the enzyme cycio-oxygnase, the enzyme responsible for the conversion of Arachidonic acid to prostaglandins which causing the pain , increase in heat body and Free roots , also Aspirin prevents thrombocytopenia from becoming sticky and aggregated (Valko et al., 2005; and Madamanch et al., 2005). Then the level of secretion of stress hormone corticosterone will decrease, leading to a decrease in the breakdown of peroxides and this leads to a decrease in the level of MDA, because the Malondialdehyde is a secondary by product of the process of oxidation and breakdown of peroxides. In the value of peroxide and in the level of free fatty acids, there were no significant differences between the additive and the control treatment table (3)

Table 3. Effect of dietary of vitamin E, C and aspirin on the lipid peroxidation in liver of broilers

Diets treatments									
Traits	T1 Control	Vit E r T2	ng / kg T3	Vit C: T4	mg / kg T5	Asp. n T6	ng / kg T7	SEM	P- value
MDA	0.7 a	(250) 0.395 ab	(500) 0.446 ab	(250) 0.482 ab	(500) 0.446 ab	(250) 0.290 b	(500) 0.376 ab	0.286	0.05
Peroxide Value	3.40	3.14	4.22	4.08	3.18	3.19	4.16	0.982	N.S
Free Fatty Acid	2.25	1.70	1.82	1.63	1.80	1.88	1.69	0.63	N.S

The different litters in the column referred to a significant differences between treatments at rate of 0.05.

#### **Blood Biochemistry**

Activities of ALT, AST in plasma

The concentrations of glucose and proteins in blood plasma are shown in table (4) , there were no significant difference between all addition treatments and control treatment for the activities ALT and AST.

Table 4. Effect of dietary of vitamin E, C and aspirin on activities of ALT, AST in plasma of broilers

Diets treatments									
Traits	T1	Vit E mg / kg		Vit C mg / kg		Asp. mg / kg		SEM	<i>P</i> -
	Control	T2 (250)	T3 (500)	T4 (250)	T5 (500)	T6 (250)	T7 (500)	2-2	value
ALT	25.20	26.50	23.43	24.42	24.80	25.03	25.36	1.7	N.S
AST	65.65	60.26	63.70	60.30	60.06	61.73	61.50	6.32	N.S

#### Concentrations of Glucose and Proteins in Blood Plasma

The concentrations of glucose and proteins in blood plasma are shown in table (5), there were no significant difference between the addition and the control treatment in the glucose level except T4, which was significantly lower (P <0.05) in the glucose level compared to the T1 treatment. while at the total protein , albumin and globulin, there was no significant difference between all addition treatments and control treatment The reason for the ability of vitamin C to lower the glucose level in serum may be caused by the decreasing of concentration of the corticosterone hormone, which is produced from the adrenal cortex, hence, increases its secretion during the exposure of birds to heat stress, working on the construction of glucose from non-carbohydrate sources (Gluconeogenesis), so the addition of this vitamin inhibits the operation of this process (Panda et al. , 2008).

Table 5. Effect of dietary of vitamin E, C and aspirin on Concentrations of glucose and proteins plasma of

				bı	oilers				
		Vit l	E mg / kg	Diets trea	tments C mg / kg	Δsn	mg / kg		P-
Traits	T1 Control	T2(2 50)	T3(500)	250) (T4	T5(500)	T6(250)	500) (T7	SEM	valu e
Glucose	169 ab	163 ab	174 a	127 b	170 ab	185 a	169 ab	23.94	0.0 5
Protein	3.60	3.61	4.05	3.79	4.38	4.13	4.37	0.560	N.S
Albumin	2.56	2.56	2.61	2.63	2.86	2.65	2.56	0.261	N.S
Globulin	1.04	1.05	1.43	1.16	1.52	1.48	1.81	0.473	N.S

The different litters in the column referred to a significant differences between treatments at rate of 0.05.

### Lipid Profile in Blood Plasma

Table 6 shows the effect of dietary of vitamin E, C and aspirin on Concentrations cholesterol, triglyceride, high density lipoproteins, and LDL plasma of broilers , there were no significant difference between all addition treatments and control treatment in the Concentrations cholesterol, triglyceride, high density lipoproteins, and LDL in plasma .

Table (6) Effect of dietary of vitamin E, C and aspirin on Concentrations of glucose and proteins plasma of broilers

			D	iets treatmer	nts				
Traits	T1 Vit E m		mg / kg Vit C mg / kg			Asp. mg	g / kg	SEM	P-
Traits	Contro 1	T2(25 0)	T3(500)	250) (T4	T5(500)	T6(250)	500) (T7	SEM	value
Cholesterol	112.7	97.1	124.1	105.5	116.8	108.8	127. 1	16.37	N.S
Triglyceride	125	116.8	129.7	134.8	145	141.1	154	23.16	N.S
HDL	29.45	17.23	33.63	24.13	23.64	35.43	35	11.44	N.S
LDL	58.27	56.6	64.57	64.43	66.9	45.2	61.4	15.51	N.S

# Conclusion

The role of Vit. E, C and Asp. in inhibiting the formation of free radicals and protection against damage caused by oxidative stress resulting from the effects of thermal stress (low concentration of MDA table 3) in tissues and liver cells. This leads to the maintenance of the metabolic functions of the hepatic cell in the representation of biological molecules (maintaining glucose, proteins and lipids within the normal concentration of table 5, 6)

which leads to the protection of fat and diet components from oxidation and thereby increasing the utilization of energy values of diet fat and other nutrients In the diet, thus providing nutritional requirements for improved production performance compared to control treatment (Table 2).

# Recommendations

Add Vitamin E , vitamin C and aspirin in level (500 mg / kg feed) for achieve the best performance of the production.

## Acknowledgements

I extend my thanks and appreciation to the Deanship of the Faculty of Agriculture / Anbar University, represented by Dr. Mohammed Oweid, Dean of the College for his unlimited support and noble positions. I also extend my thanks and appreciation to all members of the General Authority / Agricultural Research.

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Author Information					
Salim Jebur Ministry of Agriculture Abu Garib / Iraq	Thafer Mohammed University of Anbar Anbar / Iraq Contact E-mail: dafiralani@yahoo.com				
Firas Al-Khalani Ministry of Agriculture Abu Garib / Iraq					