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Artículo

Impact of increasing dietary oil concentrations with a constant energy level on the tolerance of broiler chickens to a high ambient temperature

Impacto de las concentraciones de aceite dietético con un nivel de energía constante en la tolerancia de pollos a una temperatura ambiente alta

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Abstract:

Broiler males (n= 140) were used in a straight-run experimental design and distributed randomly among four treatment groups with seven replicates per treatment and five broilers per replicate. During 21–42 d old, the chickens were fed iso-caloric and iso-nitrogenous diets containing four levels of dietary vegetable oils (DVO), of 2.7, 4, 6 and 8%. During d 25–27, 31–33, and 38–40 of age, broilers were exposed to heat stress for 4 h a day (1000–1400 h) at 34 ºC, 70–75 % relative humidity. Feeding an 8% DVO diet significantly increased body weight gain compared to the other DVO levels. The feed conversion ratio, protein conversion ratio, metabolizable energy conversion ratio and European production index were

significantly enhanced due to feeding an 8% DVO diet compared to a diet containing 6% DVO. Feeding 8% DVO significantly increased the meat protein and lipid percentages, compared to the control group (2.7 % DVO), but decreased the plasma low-density lipoprotein, very-low-density lipoprotein and lymphocytes. Feeding 8% DVO significantly increased the mean cell volume and mean cell hemoglobin, and bursa weight and percentage compared to the control. In addition, 6 and 8% DVO significantly increased the plasma total antioxidant capacity compared to the control group, but decreased the malondialdehyde. Thus, broilers fed a diet containing 8% DVO have an increased tolerance to heat stress, as evidenced by increasing the productive performance, meat quality, blood hematological and biochemical traits, antioxidants and immunity.

● **Key words**: Broilers, High ambient temperature, Growth performance, Physiological response, Immunity.

● **Resumen:**

Pollos de engorda (n= 140) se utilizaron en un diseño experimental de cuatro tratamientos distribuidos al azar con siete repeticiones por tratamiento y cinco pollos por repetición. Durante 21 a 42 días de edad, los pollos se alimentaron con dietas iso-calóricas e iso-nitrogenadas que contenían cuatro niveles de aceites vegetales alimenticios (AVA), 2.7, 4, 6 y 8%. Durante los días 25, 27, 31–33 y 38 a 40 de edad, los pollos se expusieron a estrés calórico durante 4 h al día (1000–1400 h) a 34 ºC, y 70-75 % de humedad relativa. La dieta AVA 8% significativamente incrementó la ganancia de peso corporal en comparación con los otros niveles. La conversión alimenticia, la tasa de conversión de proteína, tasa de conversión de energía metabolizable e Índice de la producción europea mejoraron significativamente debido a la alimentación con AVA 8% *versus* AVA 6%. Alimentación con AVA8%aumentó significativamente los porcentajes de proteína y lípidos de carne, en comparación con el grupo testigo (AVA2.7%), pero disminuyó la lipoproteína plasmática de baja densidad, lipoproteínas de muy baja densidad y los linfocitos. El nivel de AVA8% aumentó significativamente el volumen celular medio, hemoglobina celular media y peso y porciento de bursa comparado con el testigo. Además, 6% y 8% AVA aumentaron significativamente la capacidad antioxidante total plasmática en comparación con el grupo control, pero disminuyó el malondialdehído. Así, pollos de engorda alimentados con una dieta que contiene 8% de aceites vegetales, tienen una mayor tolerancia al estrés calórico.

● **Palabras clave:** Pollos, Estrés calórico, Crecimiento, Respuesta fisiológica, Inmunidad.

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Introduction

Poultry production in hot regions is challenged by a high environmental temperature causing heat stress that adversely impacts the productive and reproductive traits and welfare of chickens[1,2]. Adjusting the nutrient contents in chickens' diets is suggested as an effective way to negate the problems caused by a high ambient temperature[2,3,4] due to improving the appetite nutrient intake when experiencing a high temperature[5]. Feed, nutrients and metabolizable energy (ME) intake decreases during the period of high temperature as cited by National Research Council[1,3,6]. There is emerging evidence suggesting that including dietary fats/oils in broiler diets increases the ME intake and decreases the heat increment while increasing the animal's performance[7,8,9]. Energy is involved in all biochemical reactions, tissue growth and egg formation. Thus, chickens' growth during a period of high temperature may be restricted by the energy availability[1,10,11]. The availability of energy under a high temperature is more essential for growth than other dietary nutrients as energy is crucial for dissipating the metabolic heat production[3,12]. In the literature, the effect of energy concentration on broilers exposed to a high temperature is contradictory. Veldkamp *et al*[5] (with respect to turkeys), and Attia *et al*[3], Attia and Hassan[12] (both with respect to chickens) indicate that increasing dietary methionine, lysine, arginine, and threonine or protein did not improve broiler performance compared to their increasing energy concentration.

Carbohydrates and oils/fats are the principle energy sources; however, fats/oils show a greater boosting effect than carbohydrates due to their high energy value, low heat increment, extra calorific effect, fat soluble vitamins and improving digestibility, and thus better nutrient utilization[7,13]. Elevating the dietary energy level with fats or oils additionally boosted the economic traits of chickens' production in hot regions[14]. In addition, increasing the dietary ME concentration by using fats/oils in broiler diets during periods of high temperature increased their growth performance[15,16,17]. Thus, increasing oil levels may be a useful nutritional technique that may help overcome the negative effects of heat stress[1,3,10]. Nonetheless, fat supplements added to broiler diets under heat-stress conditions did not affect broiler performance[18]. Moreover, decreasing the metabolizable energy concentration during heat exposure is recommended[19,20]. There is a lack of study on the effect of increasing oil levels under a constant energy concentration on the performance of broilers exposed to a high ambient temperature. Thus, this research examines the effect of increasing the dietary oil

concentrations with a constant energy level on the tolerance of broiler chickens raised under a high ambient temperature.

Material and methods

Chickens, experimental design and diets

One hundred and forty (140), 21-d-old, Arbor Acres broiler chickens were randomly distributed, keeping their initial body weights equal, in a straight-run, completely randomized experimental design among four treatment groups. Each treatment consisted of seven replicates and five male chickens per replicate. Each replicate was kept in battery brooders (35×25×30 cm length-width-height). During the experiment period (chickens 21–42 d old), the chickens were fed iso-caloric and iso-nitrogenous diets containing four levels of vegetable oils, 2.7% (27 g/kg diet) (basal diet), 4%(40 g/kg diet), 6 % (60 g/kg diet) and 8% (80 g/kg diet) (Table 1). The oil is refining type oil certified for human consumption in Egypt that produced and purchased from Extracted Oils Company and products (Damanhour, Egypt). The estimated composition (NRC, 1994) of major fatty acids contained palmitic acid (8.9 %), stearic acid (4.1 %), oleic acid (24.95 %) linoleic acid (56.05 %), and linolenic (5.3 %).

Table 1: Ingredients and chemical composition of the experimental diets

Ingredients	Preliminary diets	Dietary vegetable oils, g/kg			
		27	40	60	80
Yellow corn	514	669.8	630	570	509
Soybean meal 48% CP	394	267	275	287	299
Di-calcium phosphate	20	15	15	15	15
Lime stone	12	10	10	10	10
NaCl	3	3	3	3	3
Vitamin+ mineral premix[1]	3	2	2	2	2
Dl-Methionine	2.5	3.7	3.6	3.4	3.3
L- Lysine	1.5	2.5	2.5	2.6	2.7
Soybean and sunflower oil[2]	50	27	40	60	80
Sand	0	0	18.9	47	76
Total	1000	1000	1000	1000	1000
Calculated[3] and analyzed[4] values (g/kg)					
ME MJ/kg[3]	12.80	12.97	12.97	12.97	12.97
Calcium[3]	10	8.0	8.0	8.0	8.0
Available phosphorus[3]	5.1	4.0	4.0	4.0	4.0
Methionine[3]	5.7	5.3	5.3	5.3	5.3
Sulphur amino acids[3]	9.2	8.2	8.2	8.2	8.2
Lysine[3]	12.8	11.7	11.7	11.7	11.7
Crude protein[4]	213	176	178	175	174
Ether extra[4]	55.9	55.7	84.4	115.9	133.3
Crude fiber[4]	4.23	5.21	4.91	5.02	5.03
Ash[4]	4.58	4.88	4.75	4.86	4.57
Dry matter[4]	906	897	899	901	910

[1] Vitamin A (retinyl acetate) 24 mg, vitamin E (dl-α-tocopheryl acetate) 20 mg, menadione 2.3 mg, Vitamin D3 (cholecalciferol) 0.05 mg, riboflavin 5.5 mg, calcium pantothenate 12 mg, nicotinic acid 50 mg, choline chloride 600 mg, vitamin B12,10 µg, vitamin B6 3 mg, thiamine 3 mg, folic acid 1 mg, d-biotin 0.50 mg. Trace mineral (mg per kg of diet): Mn 80 Zn 60, Fe 35, Cu 8, Se 0.60.

[2] A mixture of soybean oil and sunflower at 50 of each. According to NRC (1994), sunflower oil contained palmitic acid (6.7%), stearic acid (4.3%), oleic acid (27.4%) and linoleic acid (57.1), and linolenic (3.7%), and soybean meal oil contained palmitic acid (11.1%), stearic acid (3.9%), oleic acid (22.5%) linoleic acid (55%), and linolenic (6.9%). The estimated composition of major fatty acids of this mixture contained palmitic acid (8.9%), stearic acid (4.1%), oleic acid (24.95%) linoleic acid (56.05%), and linolenic (5.3%).

During the period when they were 25–27, 31–33, and 38–40 d old, the chickens were exposed to heat stress for 4 h a day (1000–1400 h) at 34 ºC, 70–75% relative humidity (RH) and, thereafter, during the unexposed period, the chickens were returned to a thermoneutral condition in which the average temperature and relative humidity were 24.9 °C and 66 % RH, respectively. The high temperature regimen was similar to those reported by Attia *et al*[(3,12,21)]. The chosen period experienced the highest environmental temperature. The average outdoor temperature was 29.5 °C with 32 % RH. The housing conditions and control for the high environmental temperature was as reported by Attia and Hassan[(12)]. During the period when they were 1–20 d old, the broilers were reared using common husbandry practices,

according to the broiler management guide, and fed a commercial diet containing 22 % crude protein (CP), 3,060 kcal, 1 % Ca and 0.5 % available phosphorus (Table 1). The scientific and ethics committee of the Animal and Poultry Production Department, Faculty of Agriculture, Damanhour University approved the experimental protocol.

Broiler husbandry

During the pre-experimental period, when the chickens were 1–20 d old, the birds were kept under similar managerial and hygienic conditions. The chickens were fed corn-soybean meal feeds in mash form during d 1–20, as shown in Table 1. The husbandry of the broilers was done according to the Arbor Acres broiler husbandry guide. Mash feed and water were provided *ad libitum*. The vaccination regimen was Hitchiner + IB on d 8, avian influenza (AI)(H5N2) on d 9, Gumboro on d 14 and d 24, and Newcastle disease virus (NDV) via Lasota on d 14, 20 and 30. The chickens were provided with a 23:1 light-dark cycle.

Measurements

At 21 and 42 d of age, the broilers were weighed (g) and feed intake was recorded for the same period. Subsequently, their feed, protein and ME were calculated using the feed intake data, and the CP and ME values of the experiment. In addition, the feed conversion ratio (FCR), protein conversion ratio (PCR) and metabolizable energy conversion ratio (MECR) were calculated used the intakes of feed, protein and ME divided by body weight gain.

At 42 d of age, five chickens from each treatment, representing all treatment replicates, were slaughtered according to the Islamic method to determine carcass criteria and inner organs, including lymphoid organs, which were expressed as a percentage of the pre-slaughter weight.

A meat sample (n= 5 per treatment), consisting of 50 % of the deboned thigh meat and 50 % of the deboned breast meat of the slaughtered chickens, was collected on d 42. About 200 g of each sample was wrapped and frozen at -18 °C until used for chemical analyses. A part of each of the fresh meat samples was used to determine the physical characteristics of the meat (n= 5 samples per treatment). The method of Volvoinskaia[22] was used to determine the water-holding capacity (WHC) and tenderness of the meat.

Colour intensity as the optical densities of the meat and drip were measured using the colorimetric method, and the pHs of the meat and drip were as reported by Husani *et al*[23] and Aitken *et al*[24], respectively. The chemical analyses of excreta samples (n= 5/treatment), and samples of breast and thigh meat (a mixture of 50 % breast and 50 % thigh meat) (n= 5/treatment), such as moisture, protein, ether extract and ash, were determined according to the Association Official Analytical Chemists (AOAC)[25] methods numbered 925.04, 990.3, 2003.06 and 942.05, respectively.

Five blood samples per treatment were collected on d 42, in unheparinised and heparinized tubes, to determine some of the hematological and biochemical constituents. Blood samples were centrifuged at 3,000 rpm for 20 min, and the plasma and serum were stored at -20 °C for further analyses. The blood's hematological characteristics, such as hemoglobin (Hgb) and PCV, were determined based on Eilers method[26]; red blood cells (RBCs) were determined as suggested by Hepler[27]; and the blood mean cell volume, mean cell haemoglobin and mean cell haemoglobin concentration (MCHC) were calculated. The white blood cells (WBCs) and WBCs' fractions were measured as described by Lucas and Jamroz[28]; the phagocyte index (PI) and activity (PA) were measured as suggested by Leijh *et al*[29]; and plasma glucose were determined according to the method of Trinder [30], serum total protein were determined as cited by Weichselbaum[31], serum albumin were measured according to the method of Doumas *et al*[32], and serum globulin were determined according to Coles[33]. In addition, the albumin-to-globulin ratio was calculated.

The serum aspartate amino transferase (AST) and alanine amino transferase (ALT) were gauged according to Reitman and Frankel's[34] method. Renal function, creatinine and urea were assessed in the serum based on the suggestions of Bartles *et al*[35] and Sampson *et al*[36], respectively, and the urea-to-creatinine ratio was calculated. Alkaline phosphatase (ALP) enzymes were measured according to the method of Kind and King[37]. The total plasma triglycerides, cholesterol, high-density lipoprotein, and low-density lipoprotein a were assessed according to the methods of Randrup *et al*[38], Watson[39], Friedwald *et al*[40] and Wieland and Seidel[41], respectively. Whereas, the very-low-density lipoprotein was determined as plasma triglycerides/5[42].

The methods of Koracevic *et al*[43] and Richard *et al*[44] were used to determine the total antioxidants capacity (TAC) and malondialdehyde (MDA), respectively. The serum antibody

body titres for NDV and AI were measured as suggested by Takats[45], and Kai *et al*[46] respectively, and the infectious bursal disease (IBD) was determined according to Cosgrove's method[47].

Statistical evaluation

An analysis of variance was done using a one-way analysis of variance, as described by SAS® [48], using the following model:

$$Yij = \mu + Fi + eij,$$ where

Y=the dependent variable,

μ=the overall mean;

Fi=the effect of dietary oil treatments and

***e*ij**=the random error.

The replicate was the experimental unit. All percentages were transformed to log10 to normalize the data distribution before analysis. The mean difference at $P \leq 0.05$ was tested using the Student-Newman-Keuls test. The survival rate was analyzed using the chi-square test.

Results

Growth performance

Results in Table 2 show a cubic-type pattern in bodyweight gain, FCR, PCR, MECR and EPI. In addition were significantly improved when feed was supplemented with 8% of oils during the high temperature period compared to the groups with 6% oil supplements. The feed, ME, and CP intakes were not significantly affected by the different oil concentrations. The survival rate was lower in 6% oils group than the other groups, but not significantly different (*P*=0.202).

Table 2: Effect of increasing dietary oils supplement on body weight gain, feed intake, feed, protein and energy conversion ratio, survival rate and European production index during 21-42 d of age[1]

Dietary vegetable oils, g/kg	Body weight gain, g	Feed intake, g	Protein intake, g	ME intake, MJ	FCR, g/g	PCR, g/g	MECR, j/g	Survival rate,%	European production Index
27	1380^{b}	3145	569.4	40.8	2.28^{ab}	0.414^{ab}	29.6^{ab}	91.4	270^{ab}
40	1343^{b}	2844	514.6	36.9	2.14^{ab}	0.387^{ab}	27.7^{ab}	97.1	297^{ab}
60	1255^{b}	2967	537.0	38.5	2.37^{a}	0.428^{a}	30.7^{a}	85.7	219^{b}
80	1569^{a}	3168	573.4	41.1	2.02^{b}	0.366^{b}	26.2^{b}	97.1	360^{a}
SEM	61.2	120.3	21.7	1.56	0.080	0.014	1.04	4.16	27.9
P-value	0.016	0.222	0.220	0.222	0.036	0.034	0.034	0.202	0.019

ME= Metabolzable energy; FCR= Feed conversion ratio; PCR= Protein conversion ratio; MECR= Metabolzable energy conversion ratio; SEM= standard error of mean.

[1]= Number of observation were 7 replicates/treatment.

[a,b] Means within a column with different letter superscripts are different (*P*≤ 0.05).

Carcass characteristics and inner body organs

Table 3 shows the effects of dietary oil supplementation on the dressing percentage, abdominal fat and inner body organs of broiler chickens at 42 d of age. There was a significant effect on most of the traits studied except for the gizzard and pancreas percentage. Broilers fed a 6% DVO diet show a significantly lower dressing percentage than the control and those on the 8% DVO diets, but these groups did not differ from those fed 4% DVO. The percentage abdominal fat was increased and proventriculus was reduced in a dose dependent manner.

Table 3: Effect of increasing dietary oils supplement within a constant metabolizable energy value on carcass characteristics and inner body organs[1]

Dietary vegetable oils, g/kg	Percentage							
	Dressing	Abdominal Fat	Proventriculus	Intestinal	Liver	Heart	Gizzard	Pancreas
27	73.1^{a}	1.23^{b}	0.409^{ab}	3.91^{c}	1.97^{b}	0.461^{b}	1.14	0.209
40	70.9^{ab}	1.20^{b}	0.481^{a}	5.39^{ab}	2.15^{b}	0.654^{a}	1.37	0.253
60	69.0^{b}	1.58^{ab}	0.378^{b}	6.07^{a}	2.64^{a}	0.422^{b}	1.25	0.246
80	72.0^{a}	1.90^{a}	0.366^{b}	4.56^{bc}	2.18^{b}	0.495^{b}	1.28	0.207
SEM	0.307	0.068	0.068	0.116	0.045	0.018	0.033	0.01
P value	0.0001	0.03	0.01	0.0003	0.02	0.006	0.583	0.651

SEM= standard error of mean.

[1]= Number of observation were 7 replicates/treatment.

[a,b,c] Means within a column with different letter superscripts are different ($P \leq 0.05$).

The intestine percentage was significantly increased feeding a 6% DVO diet as compared to the control and 8% DVO diets. In addition, the 4% DVO diets exhibited a significantly larger intestinal percentage than the control group (2.7% DVO). The liver percentage was significantly larger for the group fed 6% DVO than the other groups, but the heart percentage was significantly higher for groups fed 4% DVO than the other DVO groups.

Meat quality

There was no significant effect on most of the physical and chemical parameters of meat (Table 4), except for WHC, DM, protein and lipids in meat. The meat's WHC was significantly decreased progressively as a result of feeding 6 and 8% DVO diets compared to the control (2.7% DVO) and 4% DVO diets. On the other hand, meat's DM and lipid showed a stepwise significant increase with increasing DVO in broiler diets. Meat's protein exhibited a significant increase with feeding 8% DVO diet compared to the control group (2.7%) while the result for groups fed 4% DVO was intermediate.

Table 4: Effect of increasing dietary oils supplement within a constant metabolizable energy value on physical characteristics and chemical composition of meat[1]

Dietary vegetable oils, g/kg	Physical characteristics of fresh weight basis				Chemical composition on a dry matter basis			
	PH	Color (Optical density)	Tenderness (g/cm^2)	WHC (g/cm^2)	Dry matter (%)	Protein (%)	Lipid (%)	Ash (%)
27	6.09	0.184	9.89	16.3^a	25.7^d	19.2^b	5.53^d	0.958
40	6.07	0.189	10.3	16.2^a	25.9^c	19.3^a	5.68^c	0.96
60	6.07	0.191	10.2	15.6^b	26.2^b	19.3^a	5.98^b	0.97
80	6.05	0.217	10.2	15.2^c	26.4^a	19.3^a	6.07^a	0.96
SEM	0.103	0.002	0.116	0.048	0.012	0.011	0.013	0.004
P value	0.926	0.176	0.918	0.0001	0.0001	0.0001	0.0001	0.267

WHC= Water holding capacity; SEM= standard error of mean.

[1]= Number of observation were 7 replicates/treatment.

[a,b,c,d] Means within a column with different letter superscripts are different ($P \leq 0.05$).

Blood biochemistry of liver and renal function indices

There was no significant effect on most of the biochemical constituents except for the plasma total protein, excreta nitrogen, excreta lipids, plasma total cholesterol, LDL and VHDL (Table 5). The plasma total protein was significantly lower for the groups fed 6% DVO than for the control and 8% DVO groups, while the result for groups fed 4% DVO was intermediate. In addition, the percentage of excreta nitrogen was significantly higher for the control group compared to those in the groups fed 6 and 8% DVO. The groups fed 4% DVO had significantly decreased excreta lipids compared to the other groups. The plasma cholesterol was similarly and significantly increased due to increasing the DVO levels in broiler diets, but the plasma LDL decreased. In addition, the VLDL significantly decreased in groups fed 8% DVO compared with the other groups.

Table 5: Effect of increasing dietary oils supplement within a constant metabolizable energy value on plasma biochemical constituents and excreta nitrogen, and lipids, and indices of liver and renal functions[1]

Dietary vegetable oils, g/kg	Total protein (g/dl)	Blood biochemistry											Liver function			Renal function	
		Alb (g/dl)	Glo/ dl	Excreta (N,%)	Glu (mg /dl)	Excreta lipids (%)	Total Lip (mg/dl)	Trig (mg/ dl)	Cho (mg/ dl)	HDL (mg/ dl)	LDL (mg/ dl)	VLDL (mg/ dl)	ALT (U/L)	AST (U/L)	Alkaline phosphtase (U/L)	Urea (g/dl)	Creatinine, (g/dl)
27	6.38 [a]	3.32	3.06	6.42[a]	200.2	3.99[a]	466	185	196[b]	42.8	101[a]	37.0[a]	63.0	55.2[b]	11.2	23.8[b]	12.6
40	6.0 8[ab]	3.16	2.92	5.92[ab]	208.2	3.71[b]	470	185	209[a]	42.8	93.2[b]	37.0[a]	63.2	55.2[b]	11.0	25.0[ab]	12.6
60	5.8 8[b]	3.22	2.66	5.44[b]	210.8	3.97[a]	440	184	209[a]	42.2	94.6[b]	36.9[a]	62.0	57.6[a]	11.0	26.2[a]	12.6
80	6.36[a]	3.36	3.00	5.38[b]	210.8	4.06[a]	462	180	208[a]	42.0	95.8[b]	36.1[b]	61.6	54.6[b]	11.6	23.2[b]	13.0
SEM	0.059	0.024	0.064	0.229	2.76	0.076	4.89	1.21	0.603	0.375	0.426	0.242	0.194	0.293	0.212	0.246	0.232
P value	0.05	0.074	0.251	0.043	0.048	0.038	0.253	0.037	0.0001	0.866	0.002	0.041	0.082	0.02	0.785	0.007	0.929

Alb= Albumin; Glo= Globulin; Glu=Glucose; Lip=Lipid, Trig=triglycerides; Cho=Cholesterol; HDL= High Density Lipoprotein; LDL= Low-density lipoprotein and VLDL= very Low-density lipoprotein; ALT= Alanine aminotransferase; AST= Aspartate aminotransferase; AST/ALT= Aspartate aminotransferase to Alanine aminotransferase ratio. SEM= standard error of mean.

[1]= Number of observation were 7 replicates/treatment.

[a,b] Means within a column with different letter superscripts are different ($P \leq 0.05$).

The plasma AST was significantly decreased as a result of feeding 4 and 8% DVO compared to 6% DVO, but the other indices of liver function were not affected by the different DVO levels. In addition, the plasma urea was significantly lower for groups fed the control (2.7% DVO) and 8% DVO than that of group fed 6% DVO. There was no significant effect on plasma creatinine and the urea/creatinine ratio due to different levels of DVO.

Blood hematology

There was no significant effect on most of the hematological constituents, except for RBCs, PCV, MCV and MCH (Table 6). It was found that the value of RBCs was similarly and significantly decreased as a result of feeding 4 and 8% DVO compared to the control group, but the PCV was decreased as a result of feeding 4% DVO compared to the other DVO levels. In addition, the MCV and MCH were significantly increased for groups fed 8% DVO compared to the other groups and the control group, respectively.

Table 6: Effect of increasing dietary oils supplement within a constant metabolizable energy value on red blood cells parameters and white blood cells parameters[1]

Dietary vegetable oils, g/kg	Red blood parameters						White blood parameters						
	RBC,10^6 cell/mm^3	Hgb (g/dL)	PCV (%)	MCV (fl/cell)	MCH pg	MCHC (%)	WBC,10^3 cell/mm^3	Lymph (%)	Mono (%)	Baso (%)	Eosino (%)	Hetero (%)	H/L
27	1.96^{a}	11.2	34.4^{a}	179^{b}	58.3^{b}	32.6	21.8	46.6^{a}	14.2	0.6	10.8	27.8	0.597^{b}
40	1.58^{b}	10.2	31.0^{b}	197^{b}	65.1^{ab}	32.9	23.0	42.6^{b}	15.8	0.8	12.0	28.8	0.681^{ab}
60	1.78^{ab}	11.2	34.0^{a}	193^{b}	63.4^{ab}	32.9	22.8	42.2^{b}	14.8	0.4	11.2	31.4	0.746^{a}
80	1.46^{b}	11.2	33.8^{a}	232^{a}	77.0^{a}	33.1	21.0	42.4^{b}	15.2	1.0	12.2	29.2	0.689^{ab}
SEM	0.039	0.167	0.316	4.59	1.84	0.347	0.232	0.317	0.23	0.089	0.214	1.13	0.0348
P value	0.005	0.19	0.01	0.01	0.03	0.974	0.05	0.001	0.205	0.214	0.17	0.185	0.057

RBC= Red blood cells; PCV= Packed cell volume; MCV= Mean corpuscular volume; MCH= Mean corpuscular hemoglobin; MCHC= Mean corpuscular hemoglobin concentration. WBC= White blood cells; Lymph= Lymphocyte; Mono= Monocyte; Baso= Basophile; Eosino= Eosinophile; Hetero: Heterophile; H/L: Heterophylis to Lymphocytes ratio. SEM= Standard error of mean.

[1]= Number of observation were 7 replicates/treatment.

[a,b] Means within a column with different letter superscripts are different ($P \leq 0.05$).

In addition, there was no significant effect on most of the WBC parameters, except for lymphocytes and the stress index (Heterophile/ lymphocytes ratio; H/L ratio). Lymphocytes were significantly decreased for the 4, 6 and 8% levels of DVO groups compared to the control group. However, feeding 6% DVO significantly increased the H/L compared to the control group.

Immune status

The plasma TAC was significantly increased due to feeding diets containing 6 and 8% DVO compared to the control groups, but the MDA decreased compared to the control and 4% DVO groups (Table 7). Phagocyte activity was significantly increased from feeding diets containing 6 and 8% DVO compared to the control groups. However, PI was significantly higher of 4% DVO groups than the other groups.

Table 7: Effect of increasing dietary oils supplement within a constant metabolizable energy value on antibody titer to Newcastle disease, Avian influenza and Infection bursa disease[1]

Dietary vegetable oils, g/kg	Antioxidant status		Phagocytes		Lymphoid organs						Antibody titer, Log[1]		
	TAC (mg/dl)	MDA (µmol/l)	Activity	Index	Spleen		Bursa		Thymus		NDV	AI	IBDV
					g	%	g	%	g	%			
27	407^{c}	11.20^{a}	15.4^{b}	1.24^{b}	1.33	0.066	1.00^{b}	0.053^{b}	9.67^{a}	0.500^{a}	4.75	4.25	4.25
40	409^{bc}	10.80^{a}	17.8^{ab}	1.70^{a}	1.83	0.095	1.50^{b}	0.079^{b}	5.67^{b}	0.286^{b}	4.00	4.25	4.00
60	413^{a}	9.21^{b}	18.4^{a}	1.46^{b}	2.33	0.108	4.50^{ab}	0.215^{ab}	9.83^{a}	0.477^{a}	4.25	4.75	4.00
80	412^{ab}	9.60^{b}	19.0^{a}	1.32^{b}	2.17	0.094	6.83^{a}	0.332^{a}	6.83^{b}	0.332^{b}	4.75	4.00	4.25
SEM	0.554	0.17	0.311	0.029	0.112	0.004	1.0	0.077	0.263	0.014	0.332	0.167	0.135
P value	0.007	0.005	0.01	0.0003	0.114	0.134	0.02	0.03	0.0003	0.0006	0.077	0.38	0.906

TAC= Total antioxidants capacity; MDA= malondialdehyde; PA= Phagocytic activity; PI= Phagocytic Index; NDV= Newcastle disease; AI= Avian influenza; IBDV= Infection bursa disease. SEM= Standard error of mean.

[1]Geometric means.

[1]= Number of observation were 7 replicates/treatment.

[a,b,c] Means within a column with different letter superscripts are different ($P \leq 0.05$).

The bursa weight and percentage were significantly increased for groups fed 8% DVO compared with the control and 4% DVO groups. The thymus weight and percentages for groups fed the control diet (2.7% DVO) and 6% DVO were significantly higher than the 4% and 8% DVO groups. There were no significant effects from different DVO concentrations on the spleen weight and percentages and antibody titre for NDV, AI and IBD.

Discussion

Increasing DVO may be a useful nutritional technique to help negate the negative effects of a high ambient temperature due to the many beneficial effects of fat/oil supplementation[(1,3,10)]. The results indicate that feeding an 8% DVO diet to broilers exposed to a high ambient temperature significantly increased their growth rate; improved utilization of feed, protein and ME; and resulted in an increased EPI without affecting the intakes of feed, protein and ME, and survival rate. These show the increase in nutrient utilization for growth performance. Further evidence for the improvement in nutrient utilization for growth

and the extra caloric effect of oil are provided by the increase in the abdominal fat percentage of broilers fed 6 and 8% DVO diets. In addition, the low intestine percentage of the group fed 8% DVO suggests a lower energy expenditure for maintenance and higher expenditure for growth[12]. On the other hand, the decrease in growth performance of broilers fed 6% DVO coincided with decreasing dressed carcass percentage and plasma total protein, increasing liver and intestine percentage, plasma urea showing elevated energy expenditure for maintenance, and lower protein utilization. Moreover, there is an increase in the H/L, suggesting a decrease in animal welfare. The decreased performance of animals on 6% DVO may be due to the change in C:P ratio as a result of extra caloric effect of added oil, change in the saturated to unsaturated fatty acids ratio and formation of calcium-soaps complex that hindered nutrients unavailable. This suggests further research to determine the optimum level of oils/fats in broiler diets under heat stress condition.

The present results indicate that the effect of DVO on broiler performance seems to be a cubic type pattern as 8% DVO caused beneficial effects, 6% DVO showed negative effects and 4% DVO had no effects compared to the control containing 2.7% DVO. These results are in line with those reported elsewhere[3,4,10]. In addition, broilers housed at 29–36 °C and fed a diet supplemented with poultry fat at 5–8 % had enhanced growth performance[9,12]. Fats/oils have a greater boosting effect than carbohydrates due to their high energy value, low heat increment, extra calorific effect[49], fat soluble vitamins and improving digestibility, and thus nutrient utilization is increased due to the decreased feed-passage time and the increase in the organic matter digestibility[49,50,51]. In addition, under heat stress conditions, ME that is inadequate to sustain the processes of protein synthesis diverts energy and protein away from growth[5,8,52], and extra protein can contribute to the dietary heat increment[53]. However, in literature, the results are contradictory: in some studies, fat/oil additions boosted the economic traits of broiler chickens in hot regions[14]. In addition increasing the ME concentration by 10% in broiler diets by fat supplementation during heat stress increased growth performance of broilers exposed to heat stress[15,16,17]. However, in other studies, fat supplementation of broiler diets under heat-stress conditions did not affect broiler performance[18]. Moreover, decreasing the metabolizable energy concentration during heat exposure is recommended[19,20].

Meat quality has been shown to have an influence on customer preference, with poultry meat that has a greater WHC being more acceptable[54]. It was found that WHC was significantly decreased gradually due to feeding 6 and 8% DVO diets, and these negatively coincided with the increase in the meat's DM and lipids. Similarly, Attia and Hassan[12] found that there is a reduction in proventriculus, intestine and liver percentage, and in meat lipids, while abdominal fat, dry matter, protein, ash and physical characteristics were not affected by increasing energy concentrations. The decreases in the percentages of proventriculus, intestine and liver, for broilers on 8% DVO reveal the reassignment of nutrients towards growth, rather than maintenance[12]. Further evidence suggests that there is enhanced nutrient

utilization for growth as a result of increasing energy availability for net protein utilization, which is confirmed by increased plasma protein, decrease in excreta nitrogen and plasma urea[3,9,10].

Lipid metabolites are the primary index of lipid metabolism; however, DVO is a good source of polyunsaturated fatty acids since it composed mainly of sunflower and soybean oils. There was an unexpected increase in the plasma cholesterol as a result of increasing DVO levels in broiler diets, but plasma LDL was favorably decreased and VLDL decreased in 8% DVO group. However, there were increases in the weight and percentage of bursa, and the PA (cell-mediated immunity) of broilers fed 8% DVO. This coincided with increasing TAC and a decrease in MDA, indicating an improvement in the immune organs that may be due to the poly unsaturated fatty acids of DVO and fat soluble vitamins, such as vitamins A and E[12,55]. Further evidence for the improving health status of broilers fed 8% DVO was provided by the increased PCV, MCV and MCH compared to the control group. The increase in thymus percentage (T-cells) of groups fed 2.7 and 6% DVO are similar to those reported by Attia and Hassan[12].

Conclusions and implications

In conclusion broiler chickens fed a diet containing 8% DVO have an increased tolerance to a high ambient temperature (34 °C, 70–75 % RH.) during the period when they are 21–42 d old, as evidenced by increases in the productive performance, meat quality, blood hematological and biochemical traits, antioxidants, and immunity.

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