Acta Scientiarum
http://periodicos.uem.br/ojs
ISSN on-line: 1807-8672
https://doi.org/10.4025/actascianimsci.v48i1.74318

Serum biochemistry, lipid profile, and oxidative status of broiler chickens in the tropics fed diets supplemented olive-garlic extract oil

Gabriel Adedotun Williams

Department of Animal Science, School of Agriculture, Epe campus, Lagos State University, Lagos, Nigeria. E-mail: gabriel.williams@lasu.edu.ng

ABSTRACT. Broiler production in the tropics faces challenges from harmful residual effects of synthetic antibiotics and excessive fat accumulation. This study hypothesized that combining phytogenic products, like olive-garlic extract oil (OGEO), could enhance broiler health and lipid metabolism due to their bioactive compounds. The effects of OGEO supplementation on serum chemistry, lipid profile, and oxidative status were investigated using 240 Ross broilers assigned to four treatments: a control diet (no OGEO) and diets supplemented with 1% (10 g kg^{-1}), 3% (30 g kg^{-1}), and 5% (50 g kg^{-1}) OGEO. Each treatment consisted of six replicates with ten birds each. Blood samples were collected for Serum and lipid analysis on days 21 and 42 and oxidative status was assessed on day 42. Data were analysed using SAS Institute Inc. (2000). At day 21, 5% OGEO increased serum total protein (TP) (linear, $p = 0.003$) and globulin (linear, $p = 0.002$). By day 42, 5% OGEO improved TP, albumin, and reduced cholesterol. High-density lipoprotein cholesterol (HDL-C) increased with 5% OGEO, while meat cholesterol and triglycerides decreased. Malondialdehyde (MDA), an oxidative stress marker, was reduced in blood and meat at 1, 3, and 5% OGEO. The study concludes that 5% OGEO supplementation improves serum TP, reduces cholesterol, and enhances lipid profile and oxidative stability in broilers.

Keywords: tropical climate, broiler nutrition, antioxidants, blood parameters, fat metabolism.

Received on October 24, 2024.
Accepted on November 18, 2025.

Introduction

The demand for animal protein to meet the needs of a growing population has led to the development of broiler chickens that mature within 6-8 weeks, reaching an average weight of 1.8-2 kg (Havenstein et al., 2003). Commercial broiler production often employs intensive systems and controlled environments to enhance tissue growth. While this approach helps satisfy protein needs, it also presents challenges, notably poor immune responses and excessive fat accumulation. Rapid growth can hinder the immune system's ability to resist disease, as reported by Compassion in world farming (2019) and European Commission (2000), which noted that heavy broilers produce fewer antibodies. In tropical regions, high-energy diets promote fat deposition, leading to lower carcass quality and potential rejection by consumers, ultimately causing financial losses for farmers (Nikolova et al., 2007).

Historically, antibiotics have been used to address poor immune responses and combat disease in broilers. However, their use has contributed to antibiotic resistance in humans due to misuse and overuse (Khan et al., 2018). Agriculture, livestock production, and pharmaceutical waste are also recognized as significant sources of antibiotic resistance (Saima et al., 2020). The rise of multi-resistant bacteria has rendered antibiotic use in animal production increasingly unacceptable (World Health Organization, 2015). Although strategies like feed restriction have shown positive effects in reducing fat accumulation (Santoso et al., 1995; Mahmood et al., 2007), they can also lead to decreased weight gain, prolonging the time needed to reach marketable weight (Alkhair, 2019). Additionally, early feed restrictions may increase fat deposition later in growth (Velleman et al., 2014), highlighting the need for alternative dietary strategies to improve meat quality.

Recent studies have explored plant-based ingredients such as seeds, fruits, leaves, and roots for their active constituents, such as polyphenols, which lack harmful residues (Abudabos et al., 2016; Mech et al., 2021). These phytogenic compounds can serve as antibiotic alternatives due to their antimicrobial properties

and benefits for digestion and immunity (Hernandez et al., 2004). Garlic, known for its immune-boosting effects, contains bioactive compounds like alliin and allicin, which enhance immune response (Al-Shuwaili et al., 2015). Additionally, garlic can inhibit hepatic cholesterol synthesis by reducing the activity of lipogenic and cholesterogenic enzymes (Stanacev et al., 2011).

Olive oil, rich in phenolic compounds, also demonstrates immunomodulatory and antimicrobial properties (Sánchez-Fidalgo et al., 2013). Its polyphenols promote cytokine production and enhance white blood cell counts, supporting immune function against infections (Ding et al., 2018). Furthermore, the monounsaturated fatty acids (MUFA) in olive oil, particularly oleic acid, improve the fatty acid profile and inhibit cholesterol synthesis (Sarica & Toptas, 2014). Given these functional qualities, we hypothesize that the combination of olive and garlic extracts will synergistically mitigate pathogenic effects and reduce fat accumulation. Hence, this study aims to investigate the effects of dietary olive-garlic extract oil supplementation on the serum chemistry, lipid profile, and oxidative status of broilers.

Materials and methods

Experimental site

The experiment was conducted at the Teaching and Research Farm of the School of Agriculture, Lagos State University, Epe campus, Lagos State, Nigeria, in accordance with the approved animal research guidelines of the Nigeria Institute of Animal Science (NIAS). The farm is situated at Latitude 6°35'09.4"N and Longitude 3°59'54.7"E, within the lowland rainforest and savannah agro-ecological zone. The region experiences an average annual rainfall of 1694 mm and an average temperature of 27.1°C.

Preparation of Test ingredients

Extra-virgin olive oil was sourced from Shoprite, and garlic from Mile 12 Market, Lagos. The garlic bulbs were peeled, chopped, crushed, and macerated in 70% methanol. After filtering, the garlic extract was infused into olive oil (1 kg garlic 2 liters oil^{-1}), heated for 5 minutes at 35°C while stirring. The mixture was then sieved to remove garlic solids, leaving the olive-garlic extract oil (OGEO) to cool, which was stored in a plastic container for use.

Chemical composition and antioxidative capacity of test ingredients

Samples of the test ingredients were analyzed for their chemical composition, including phytochemicals, fatty acids, and antioxidant activity. Standard procedures were used for polyphenols (Wright et al., 2000), flavonoids (Arvouet-Grand et al., 1994), tannins (Hoff & Singleton, 1977), alkaloids (Siddiqui & Ali, 1997), and saponins (Edeoga et al., 2005), while fatty acids were determined using American Oil Chemists' Society methods (1998). Antioxidant capacity was assessed using the DPPH assay (Mimica-Dukic et al., 2004), and radical scavenging capacity (RSC) was measured spectrophotometrically and calculated.

Experimental birds and management

Two hundred and forty (240) day-old Ross 308 broilers were purchased from AGRITED hatchery in Ibadan, Nigeria. Poultry pens were cleaned, disinfected, and preheated using electric bulbs and charcoal. The brooding house temperature was maintained at 31°C for three days, then reduced by 2°C weekly to 27°C by the end of two weeks. The birds were reared on deep litter with wood shavings as bedding. Feeding trays and water troughs were adjusted as the birds grew. During the trial, the pen's temperature averaged 31.2°C, with 67.84% relative humidity.

Experimental design and dietary treatments

Broilers were assigned to four dietary treatments using a completely randomized design, with 16 pens (3 × 3 m) partitioned by wire mesh. Each treatment had 4 replicates of 15 birds. The diets were formulated for starter (0-21 days) and finisher (22-42 days) phases in line with Ross Broiler (2019). Diet 1 was the control, while diets 2, 3, and 4 were supplemented with olive-garlic extract oil (OGEO) at 1, 3, and 5%, respectively (Table 1). Birds were raised on a deep litter system, with feed and water provided *ad libitum*, and no antibiotics were used. The experiment lasted six weeks.

Table 1. Composition of experimental diet and test ingredients.

Gross composition of experimental diet			chemical and antioxidants constituents of ingredients			
Ingredients	(0-21 days)	(22-42 days)	Composition	Garlic	Olive oil	OGEO
Corn	55.00	63.00	Phenol (Total)	6012.40 mg kg^{-1}	452.00 mg L^{-1}	812.00 mg L^{-1}
Soya bean meal	18.00	13.50	Flavonoids	6.4.60 mg kg^{-1}	52.60 mg L^{-1}	240.10 mg L^{-1}
Wheat offal	5.00	4.00	Oleic acid	36.43 %	56.58 %	52.31 %
Groundnut cake	15.50	13.50	Linolenic acid	35.48 %	15.48 %	0.51 %
Fish meal (75%)	2.50	2.00	TA[1]	28.23 mg mL^{-1}	573.00 mg L^{-1}	657.20 mg L^{-1}
DL-Lysine	0.25	0.25	Saponin	17.80 mg kg^{-1}	95.00 g L^{-1}	64.70 g L^{-1}
Oyster	1.00	1.00	Alkaloids	53.20 mg kg^{-1}	110.40 mg L^{-1}	125.30 mg L^{-1}
DL-Methionine	0.25	0.25	Tannin	48000 mg kg^{-1}	52.30 g L^{-1}	73.50 g L^{-1}
Vitamin/Mineral premix*	0.25	0.25				
Salt	0.25	0.25				
Total	100	100				
Determined nutrients						
ME (kcal kg^{-1})**	2930.00	3097.00				
Crude protein (%)	22.70	19.84				
Fat (%)	3.98	4.36				
Crude fibre (%)	4.30	4.58				
Nitrogen free extract (%)	57.29	59.07				
Calcium	1.20	1.20				
Phosphorous (%)	0.50	0.40				
Lysine (%)	1.30	1.10				
Methionine (%)	0.60	0.60				
Ash (%)	2.20	1.90				

*Starter premix: vit. A10,000,000 IU, vit. D 32,500,000 IU, vit. E 23,000 mg, vit. K_3 2,000 (mg), vit. B1 1,800 (mg), vit. B_2 5,500 (mg), niacin 27,500 (mg), pantothenic acid 7,500 (mg), vit. D_6 3,000 (mg), vit. B_{12} (15 mg), folic acid (750 mg), biotin H_2 60 mg, chlorine chloride 300,000 mg, cobalt 200 mg, copper 3,000 mg, iodine 1,000 mg, iron 20,000 mg, manganese 40,000 mg, selenium 200 mg, zinc 30,000 mg; *Finisher phase: vit. 8,500,000 IU, vit. D_3 1,500,000 IU, vit. E 10,000 mg, vit K_3 1,500 mg, vit. B_1 1,600 mg, vit. B_2 4,000 mg, niacin 20,000 mg, pantothenic acid 5,000 mg, vit. D_6 1,500 mg, vit. B_{12} 10 mg, folic acid 500 mg, biotin H_2 750 mg, chlorine chloride 175,000 mg, cobalt 200 mg, copper 3,000 mg, iodine 1,000 mg, iron 20,000 mg, manganese 40,000 mg, selenium 200 mg, zinc 30,000 mg; **Estimated using the Nutrient Requirements of Poultry, National Research Council (1994) formulae, ME = 26.7 (%dry matter) + 77 (%ether extract) − 51.22 (%crude fibre); [1]TA Total Antioxidants ((% 2,2-diphenyl-1-picrylhydrazyl (DPPH) scavenged); OGEO= Olive garlic extract oil, ME= Metabolisable energy.

Blood collection for analysis

On the 21st and 42nd days, 5 mL blood samples were collected from two birds (through the brachial wing) per replicate (8 per treatment) into plain bottles for serum biochemical, lipid profile, oxidative status, and antioxidant enzyme analyses. Samples were centrifuged (1200 rpm for 15 min.), and aliquots were frozen at 20°C for later analysis.

Serum biochemical analysis

Serum total proteins and albumin were measured colorimetrically using Stanbio Company kits (Tietz, 1995). Globulin levels were calculated by subtracting albumin from total protein values. Serum enzymes (ALT and AST) were analyzed with Qualigens kits. Cholesterol was estimated using the Randox® diagnostic kit, glucose via the GOD PAD method (Dandekar & Rane, 2004), and serum uric acid was determined using the Quinica Clinica Spam kit (Wootton, 1964). Serum creatinine levels were measured according to Bonsnes and Taussky (1945).

Blood lipid profile

The serum lipid profiles which are triglyceride (TG), total cholesterol (TC), low density lipoprotein cholesterol (LDL-C), and high-density lipoprotein cholesterol (HDL-C) were analysed using an automatic analyser (Hitachi 902 Automatic analyser; Tokyo, Japan) and commercial lipid profile test kits (Crescent Diagnostic Lab). Very Low-density lipoprotein cholesterol (VLDL-C) was calculated using the formula (VLDL-C = triglycerides/5)

Meat and organ lipid profile

On day 42, 6 broilers per group were slaughtered and after the dissection of the carcass, 3 g of breast meat and liver tissue were collected. The meat and organ samples were homogenized using a blender and the samples were frozen and stored in a freezer at (-18°C) until further analysis. The TC, TG and HDL-C were determined spectrophotometrically by using commercial kits (Asan Pharm. Co., Ltd. Seoul, Korea) as

described by De Almeida et al. (2006). Very Low-density lipoprotein cholesterol (VLDL-C) levels were estimated using the formula (VLDL-C = triglycerides 5^{-1}). Low density lipoprotein (LDL) levels were estimated as the difference between total cholesterol and the sum of VLDL and HDL (Adler & Holub, 1997).

Blood and meat oxidative status

The blood oxidative status was carried out with the use of a spectrophotometer (GENESYS 180, Thermo Fisher Scientific, Waltham, MA, USA) according to the method detailed by Tsiplakou et al. (2017). The catalase (CAT) activity was carried out with the use of a commercial kit (Catalase Assay Kit; CAT100, Sigma-Aldrich, St. Louis, MO, USA). The malondialdehyde (MDA) and glutathione peroxidase (GPX) were determined according to the method described by Wyatt et al. (2001) and Mates et al. (2000) respectively. The Superoxide dismutase (SOD) activity was measured by the procedure of Kakkar et al. (1984) and recorded by monitoring the inhibition of cytochrome c oxidation at 550 nm.

For lipid peroxidation analysis, 5 g of breast meat was homogenized with 4 mL of a buffer made from 6.038 g and 21.95 g of sodium dihydrogen phosphate in 100 mL of distilled water. Oxidative status was assessed spectrophotometrically using commercial kits (Spectrum Diagnostics, Al Obour, Cairo, Egypt) and the Thio-barbituric acid (TBA)-Malondialdehyde (MDA) complex assay (Das et al., 1990). GPX activity was determined according to Paglia and Valentine (1967), SOD activity using Das et al. (2000) and CAT activity according to Clairborne (1986).

Statistical analysis

All data collected were analysed using one-way analysis of variance (ANOVA) in a completely randomised design using Statistical Analysis System (SAS Institute Inc., 2000). The significant means among treatments were separated using the Tukey test in the SAS statistical tool. The level of significance was considered at $p < 0.05$. Orthogonal polynomial contrast analysis was also done to determine the linear and quadratic response trend to varying levels of OGEO supplementation.

Results and discussion

The serum biochemistry parameters of broilers fed diet supplemented with OGEO are presented in Table 2. Broilers fed diet supplemented with 5% OGEO had increased (linear, $p = 0.003$) total protein (TP) and those fed diets supplemented with 0 and 1% had reduced TP and glucose while those fed 3% OGEO supplemented diet had intermediate TP. This is contrary to the report of Hlatshwayo et al. (2023) who observed no significant effect of olive pomace inclusion in the diet of jumbo quails on serum parameters. Elbaz et al. (2022) also observed no significant effect of garlic essential oil supplementation at 200 mg kg^{-1} in the diet of broilers compared to the control. However, Ismail et al. (2021) reported increased TP and globulin in the serum of broilers fed the diet supplemented with 0.75 g kg^{-1} garlic powder. The discrepancies observed could be due to the differences in the plant products used and differences in inclusion level. However, the increased TP and glucose observed in this study could be due to the positive synergistic effect of garlic and olive oil which increased nutrient utilisation through stimulation of digestive enzyme. The active constituents of garlic such as allicin are known to stimulate the activity of digestive enzyme promoting nutrient availability (Adibmoradi et al., 2006).

At day 42, the TP increased linearly ($p = 0.020$) with OGEO supplementation. Broilers fed the diet supplemented with 5% OGEO had higher TP than those fed diets supplemented with 0 and 1% OGEO while those fed 3% OGEO supplemented diet had intermediate TP. Serum albumin followed the same pattern with TP in which supplementation of OGEO at 5% in the diet of broilers resulted in increased (linear, $p = 0.002$; quadratic, $p = 0.022$) serum albumin. This is similar to the observation of Al-Harthi (2015) who reported increased serum albumin when olive cake was included in the diet of layers at 10 and 20%. Supplementation of 3 and 5% OGEO in the diet of broilers increased (linear, $p = 0.011$) serum glucose but 1% dietary OGEO supplementation resulted in reduced glucose which suggests that apart from nutrient adequacy of the diet the supplementation of OGEO promotes nutrient availability in circulating blood. It has been known that essential oil of phyto-additives enhances the activity of digestive enzymes for improved nutrient digestibility (Alagawany et al., 2021). It was also observed that the liver enzyme ALT and AST were not significantly affected on both days 21 and 42 indicating no negative effect on the hepatocytes as their increase in blood circulation is implicated in liver damage (Mehri et al., 2015). This is in agreement with the report of Pappas et al. (2019) who

observed no effect of olive pulp inclusion in the diet of broilers on serum AST and ALT. Pirozzi et al. (2016) also reported that oleic acid and polyphenols in olive oil exhibit hepatoprotective properties.

Table 2. Serum biochemistry parameters of broilers fed diet supplemented with olive-garlic extract oil.

Parameters	Olive-garlic extract oil supplementation level				Pooled	p-value	
	0%	1%	3%	5%	SEM	Linear	Quadratic
Day 21							
Total protein (g dL^{-1})	4.67^{b}	4.67^{b}	5.00^{ab}	6.00^{a}	0.19	0.003	0.067
Albumin (g dL^{-1})	3.37	3.50	3.24	4.00	0.14	0.620	0.282
Globulin (g dL^{-1})	1.30	1.17	1.76	2.00	0.13	0.058	0.500
Uric acid (mg dL^{-1})	13.33	14.00	12.00	12.00	0.55	0.269	0.776
Glucose (mg dL^{-1})	53.33^{b}	59.00^{b}	69.00^{ab}	78.33^{a}	3.41	0.002	0.679
Creatinine (mg dL^{-1})	2.67	2.33	2.00	2.67	0.42	0.940	0.616
AST (U L^{-1})	43.00	39.00	48.33	26.67	4.24	0.311	0.312
ALT (U L^{-1})	26.00	17.67	19.33	15.67	2.27	0.189	0.623
Day 42							
Total protein (g dL^{-1})	5.33^{b}	5.33^{b}	5.67^{ab}	6.67^{a}	0.22	0.020	0.172
Albumin (g dL^{-1})	3.33^{b}	3.46^{b}	4.04^{ab}	5.00^{a}	0.21	0.002	0.022
Globulin (g dL^{-1})	2.00	1.87	1.63	1.67	0.13	0.760	0.067
Uric acid (mg dL^{-1})	10.67	10.67	10.67	11.00	0.22	0.667	0.747
Glucose (mg dL^{-1})	94.33^{ab}	83.33^{b}	115.00^{a}	117.33^{a}	5.18	0.011	0.356
Creatinine (mg dL^{-1})	2.00	4.33	5.67	1.67	0.68	0.947	0.019
AST (U L^{-1})	53.33	54.33	46.33	55.33	0.22	0.925	0.410
ALT (U L^{-1})	24.00	25.33	18.67	24.00	0.21	0.020	0.172

$^{a-b}$Means on the same row with differing superscripts are significantly different ($p < 0.05$), SEM= Standard error of mean, AST= Aspartate aminotransferase, ALT= Alanine aminotransferase

Blood lipid profile of broilers

Table 3 presents the blood lipid profile of broilers fed the diet supplemented with OGEO. At day 21, supplementation of 1 and 5% OGEO in the diet of broilers resulted in reduced (quadratic, $p = 0.017$) blood cholesterol. Triglyceride reduced significantly (linear, $p = 0.008$) with supplementation of OGEO at 3 and 5% in the diet of broilers. Increased HDL-C was also observed for broilers fed 1 and 5% OGEO supplemented diet (quadratic, $p = 0.022$). Supplementation of 5% OGEO resulted in reduced (linear, $p = 0.012$) very low-density lipoprotein cholesterol (VLDL-C). The result of our study agrees with the report of Elbaz et al. (2022) who reported reduced serum cholesterol, triglyceride and LDL of broilers fed the diet supplemented with 200 mg kg^{-1} mixture of garlic and lemon essential oil. The authors observed that the supplementation of garlic and lemon essential oil mixture outperformed the individual supplementation of the essential oil which affirms the superior effect of combined use of phyto-additives.

Table 3. Blood lipid profile of broilers fed diet supplemented with olive-garlic extract oil.

Parameters (mg dL^{-1})	Olive-garlic extract oil supplementation level				Pooled	p-value	
	0%	1%	3%	5%	SEM	Linear	Quadratic
Day 21							
Cholesterol	120.00^{ab}	113.67^{b}	129.00^{a}	111.33^{b}	2.48	0.480	0.017
Triglyceride	121.57^{ab}	133.57^{a}	100.10^{b}	91.87^{b}	6.04	0.008	0.236
HDL-C	55.67^{b}	64.50^{a}	56.90^{b}	60.00^{ab}	1.25	0.487	0.022
LDL-C	32.80	37.73	36.87	40.13	1.08	0.125	0.640
VLDL-C	23.67^{ab}	26.73^{a}	19.67^{ab}	18.07^{b}	1.25	0.012	0.198
Day 42							
Cholesterol	133.33^{a}	123.00^{ab}	103.67^{b}	107.33^{b}	4.32	0.004	0.244
Triglyceride	119.80^{a}	110.87^{ab}	105.57^{ab}	95.00^{b}	3.33	0.004	0.862
HDL-C	58.43^{b}	60.50^{b}	60.23^{b}	72.27^{a}	1.92	0.004	0.060
LDL-C	34.73	37.10	38.27	43.73	1.94	0.147	0.703
VLDL-C	23.67^{a}	21.87^{ab}	21.97^{ab}	19.10^{b}	0.62	0.009	0.566

$^{a-b}$Means on the same row with differing superscripts are significantly different ($p < 0.05$), SEM= Standard error of mean, HDL-C= high density lipoprotein cholesterol, LDL-C= Low-density lipoprotein cholesterol, VLDL-C= Very low-density lipoprotein cholesterol

At day 42, a similar trend was observed in which broilers fed the diet supplemented with 3 and 5% OGEO had reduced serum cholesterol (linear, $p = 0.004$) and triglyceride (linear, $p = 0.004$) while groups of broilers fed the 5% OGEO supplemented diet had reduced VLDL-C (linear, $p = 0.009$) and increased serum HDL-C

(linear, p = 0.004). This agrees with the report of Zhang et al. (2013) who observed reduced triglyceride and increased HDL-C in the serum of broilers fed the diet containing 5% olive oil. Bölükbaşı and Erhan (2007) also reported decreased LDL-C and triglyceride without reduction in HDL-C for broilers fed diet with 3% olive oil. The reduction in these serum indices is also associated with the hypocholesterolemic effect of garlic through the activity of constituent bioactive compounds which inhibits the activity of enzymes necessary for cholesterol synthesis and lipogenesis (Issa & Omar, 2012). The observed reduction in serum cholesterol, triglyceride and VLDL-C suggests that the use of OGEO is capable of reducing these biochemical indices in circulating blood which is also a resultant function of increased HDL-C. The transportation of blood-circulating fatty acid and cholesterol from body tissues to the liver is facilitated by the lipoprotein HDL-C (Zhang et al., 2013). The observed result in this study further suggests that the dietary supplementation of OGEO will promote lipid stability thereby ameliorating the devastating effect of lipid oxidation which arises during heat stress. It is important to state that the increased HDL-C observed at days 21 and 42 in the serum of broilers confirms that the supplementation of OGEO in the diet of broilers will improve blood lipid profile which is supported by separate studies by Prasad et al. (2009) and Sarica and Toptas (2014) who earlier reported that olive oil and garlic can positively influence blood lipid profile.

Meat and organ lipid profile of broilers

The meat and organ lipid profile of broilers fed the diet supplemented with OGEO is shown in Table 4. The dietary supplementation of OGEO at 5% resulted in lower cholesterol (linear, p = 0.006) and triglyceride (linear, p = 0.008) in the meat of broilers compared to those fed diet without OGEO supplementation while those fed 1 and 3% OGEO supplemented diet had intermediate cholesterol and triglyceride. This is similar to what was observed in the serum of broilers which is not farfetched from the synergistic influence arising from OGEO supplementation. This observation is in agreement with the report of Crespo and Esteve-Garcia (2002) who observed increased digestibility of saturated fatty acid (SFA) and reduction in synthesis of endogenous SFA with inclusion of olive oil in the diet of broilers which has a consequential implication on serum fatty acid composition. This suggests that the profiles of fatty acids in tissues and fat deposits can be influenced by the inclusion of olive oil due to the possession of high quantity MUFAs as reported by Krejčí-Treu et al. (2010). This also confirms the report of Park et al. (1997) who indicated that lean tissue can be achieved with dietary inclusion of olive oil through the inhibition of lipid synthesis. The high constituent of MUFA in olive oil is less susceptible to lipid peroxidation which reduces the rate of meat deterioration and therefore promotes shelf life (Amaral et al., 2018). In addition, the phenolic constituent of olive oil exhibits antioxidant properties which aid in oxidation-reduction for the preservation of nutrients (Dermeche et al., 2013). The reduced triglyceride and LDL-C obtained in the meat of broilers is also due to the contributory effect of garlic extract which is effective in reducing fat accumulation through the activity of constituent sulfur compounds capable of reducing cholesterol amount in plasma and tissue (Stanaćev et al., 2012). The mechanism through which this is achieved is through the suppression of enzymes such as fatty acid synthase and malic enzyme associated with the liver necessary for the synthesis of fat and cholesterol (Yeh & Yeh, 1994). The HDL-C content of meat from broilers was higher (linear, p < 0.001) for broilers fed the diet supplemented with 5% OGEO than those fed diets supplemented with 0 and 1% OGEO while those fed 3% OGEO supplemented diet had intermediate HDL-C. Lower (linear, p = 0.008) LDL-C content was obtained from meat of broilers fed diets supplemented with 1 and 5% OGEO compared to those fed diet without OGEO supplementation and those fed 3% OGEO supplemented diet had intermediate LDL-C content. The increased HDL-C obtained in the meat of broilers with the dietary supplementation of OGEO in this study is a result of the positive influence of dietary treatment on lipid profile. Although, studies that investigated the effect of dietary inclusion of olive oil on meat lipid profile are scarce, however, research reports have indicated that dietary inclusion of olive oil for broilers increased serum HDL-C as well as reduced triglyceride and LDL-C (Omar, 2000; Rafei-Tari et al., 2021). Therefore, the observation in our study suggests that the increased HDL-C in the serum due to the dietary supplementation of OGEO consequently influenced the increase in HDL-C concentration of the meat

In the liver, contrary to what was observed in the meat of broilers in which dietary supplementation of OGEO reduced triglyceride and LDL-C, the liver lipid profile revealed that supplementation of 5% OGEO resulted in increased triglyceride and LDL-C in the liver. The supplementation of OGEO at 5% in the diet of broilers resulted in higher (linear, p = 0.012; quadratic, p = 0.015) triglyceride compared to other treatments. LDL-C also increased (linear, p=0.001) for broilers fed the diet supplemented with 5% OGEO. This disagrees with the report of Schuman et al. (2000) who reported a reduction in the liver lipid profile of laying hens in response to a flax oil-

supplemented diet. The discrepancy observed could be due to the different oils used as a result of differences in the fatty acid composition of the oil. The increase in the liver lipid fractions obtained in this study does not necessarily indicate a diseased state of the liver but could be because the liver is the site of lipid synthesis and in addition, due to the activity of OGEO which has limited the release of lipid into blood circulation. The increased HDL-C obtained in the serum of broilers fed the diet supplemented with 5% OGEO could also be a contributory factor to the rise in LDL-C and triglyceride in the liver as these lipoproteins are known to be responsible for the transport of cholesterol and fatty acids from the tissues to the hepatocytes. It is also known that fat and oil is a component of various cell membrane (cytoarchitecture) which is of high concentration in the liver cell (Zhang et al., 2013). Research studies on the effect of dietary inclusion of olive oil on the liver lipid profile of broilers are rare in literature however, earlier reports have indicated the hepatoprotective effect of olive oil which further suggests that the rise in triglyceride and LDL-C does not imply damage to the liver. It has also been documented that the consumption of olive limits the derangements of liver tissues and the reduction of fibrous tissue formation (Wang et al., 2014).

Table 4. Meat and organ lipid profile of broilers fed diet supplemented with olive-garlic extract oil.

Parameters (m dL^{-1})	Olive-garlic extract oil supplementation level				Pooled	p-value	
	0%	1%	3%	5%	SEM	Linear	Quadratic
Breast meat							
Cholesterol	98.57^{a}	90.33^{ab}	88.10^{ab}	70.57^{b}	3.78	0.006	0.392
Triglyceride	126.20^{a}	113.93^{ab}	111.03^{ab}	102.63^{b}	3.24	0.008	0.692
HDL-C	31.50^{b}	38.43^{b}	48.37^{ab}	50.60^{a}	2.55	<0.001	0.369
LDL-C	32.87^{a}	22.50^{b}	26.43^{ab}	20.03^{b}	1.74	0.008	0.399
VLDL-C	27.90	21.77	20.53	16.40	1.83	0.035	0.759
Liver							
Cholesterol	109.67	103.13	119.70	131.70	4.84	0.059	0.303
Triglyceride	146.43^{b}	146.10^{b}	149.07^{b}	185.23^{a}	6.36	0.012	0.015
HDL-C	47.87	46.07	58.37	55.83	3.07	0.227	0.954
LDL-C	23.50^{b}	24.50^{b}	33.53^{b}	41.73^{a}	2.53	0.001	0.229
VLDL-C	29.30	29.23	27.80	37.43	1.50	0.054	0.066

$^{a-b}$Means on the same row with differing superscripts are significantly different ($p < 0.05$), SEM= Standard error of mean, HDL-C= high density lipoprotein cholesterol, LDL-C= Low-density lipoprotein cholesterol, VLDL-C= Very low-density lipoprotein cholesterol

Blood and meat oxidative status of broilers

The blood and meat oxidative status of broilers fed the diet supplemented with OGEO is shown in Table 5. The blood oxidative status shows that broilers fed the diet supplemented with 0% OGEO had increased (linear, $p = 0.003$) MDA and those fed the diet supplemented with 5% OGEO had reduced MDA (linear, $p = 0.003$). This suggests that peroxidative damage which results in the generation of free radicals was inhibited with dietary supplementation of OGEO. The positive influence of OGEO supplementation on the oxidative status of broilers further implies that essential nutrients which include amino acids, vitamins and essential fatty acids are preserved due to the prevention of peroxidation as the oxidation of food protein causes nutrient loss due to reduced effectivity of digestive enzymes on oxidized proteins (Estévez, 2015). The dietary supplementation of OGEO at 1 and 5% resulted in higher (linear, $p < 0.001$) SOD than those fed the control diet. Broilers fed diet with OGEO supplementation at 5% had higher (linear, $p = 0.005$) CAT compared to other treatments. Increased SOD and catalase activity was observed in the serum of broilers fed the 5% OGEO supplemented diet. This is similar to the report of Oke et al. (2017) who observed elevated SOD activity in the plasma of broilers given water containing olive leaf extract. The increased SOD and catalase obtained in this study could be attributed to the high amount of MUFAs in olive oil which is known to be less susceptible to lipid peroxidation and it is also associated with the constituent bioactive hydroxytyrosol and phenolic compounds capable of scavenging free radicals (Laudadio et al., 2015).

In the meat, the supplementation of OGEO at 5% in the diet of broilers resulted in reduced (linear, $p = 0.042$) MDA but those fed the diet without OGEO supplementation had increased MDA. This is similar to the report of Branciari et al. (2017) who reported that the inclusion of olive cake at 165 g kg^{-1} in the diet of broilers increased antioxidative activity in the meat. Reduced MDA level in the meat of broilers was obtained when olive oil processing waste was supplemented in the diet of broilers (Gerasopoulos et al. 2015). The highest quadratic increase ($p = 0.049$) in CAT was observed for broilers fed OGEO supplemented diet at 3% and those fed diet without OGEO supplementation had the lowest CAT. The outcome of the current study is also in agreement with the findings of Shi et al. (2017) who indicated that the active constituent of olive oil

(oleuropein) is capable of elevating the expression of antioxidant enzymes and indicators (catalase, SOD and GSH-Px) in the liver. Hydroxytyrosol or oleuropein and their metabolites which are active phenolic compounds in olive oil are known to exhibit potent antioxidant properties (Omar, 2010). The report of Alagawany et al. (2016) also revealed increased activity of SOD, CAT, GSH-Px and reduced concentration of MDA in the liver of rabbits fed a diet supplemented with 6 g kg^{-1} garlic powder compared to those fed control diet. Based on the observed antioxidative effect of OGEO in the current study, it implies that its supplementation will promote shelf life of meat as well as promoting meat quality.

Table 5. Blood and meat oxidative status of broilers fed diet supplemented with olive-garlic extract oil.

Parameters	Olive-garlic extract oil supplementation level				Pooled	P-value	
	0%	1%	3%	5%	SEM	Linear	Quadratic
Blood							
MDA (U $L^{-1}\times10^{-9}$)	6.50^{a}	5.97^{ab}	4.75^{ab}	3.35^{b}	0.49	0.003	0.914
SOD (U L^{-1})	0.64^{bc}	0.43^{c}	1.36^{ab}	2.10^{a}	0.21	<0.001	0.038
GP_X (U L^{-1})	4.13	4.03	4.93	2.97	0.32	0.337	0.139
Catalase (U L^{-1})	1.23^{b}	1.40^{b}	1.43^{b}	3.17^{a}	0.28	0.005	0.050
Meat							
MDA (U $L^{-1}\times10^{-9}$)	6.40^{a}	4.74^{ab}	3.11^{ab}	2.71^{b}	0.55	0.042	0.449
SOD (U L^{-1})	0.05	0.59	0.22	0.64	0.12	0.205	0.805
GP_X (U L^{-1})	2.83	3.30	4.03	2.90	0.26	0.692	0.155
Catalase (U L^{-1})	1.47^{b}	2.00^{ab}	4.03^{a}	3.72^{a}	0.37	0.108	0.049

$^{a-b}$Means on the same row with differing superscripts are significantly different ($p < 0.05$), SEM= Standard error of mean, MDA= malondialdehyde, SOD= superoxide Dismutase, GP_X= Glutathione Peroxidase

Conclusion

The result from this study has revealed that supplementation of OGEO in the diet of broilers at 5% will sufficiently improve the health status of broilers through increased serum TP and glucose while at the same time improving the lipid profile by reducing cholesterol, triglyceride, VLDL-C and increasing HDL-C which will discourage fat accumulation. In addition, this study shows that dietary 5% OGEO supplementation enhances the oxidative status of the blood and meat of broilers by increasing the activity of SOD and catalase while reducing MDA.

Data availability

The data for the current study will be provided by the corresponding author upon reasonable request.

References

Abudabos, A. M., Alyemni, A. H., Dafalla, Y. M., & Khan, R. U. (2016). The effect of phytogenic feed additives to substitute infeed antibiotics on growth traits and blood biochemical parameters in broiler chicks challenged with Salmonella typhimurium. *Environmental Science and Pollution Research, 23*(23), 24151-24157. http://doi:10.1007/s11356-016-7665-2

Adibmoradi, M., forNavidshad, B., Saif, D. J., & Royan, M. (2006). Effect of dietary garlic meal on histological structure of small intestine in broiler chickens. *The Journal of Poultry Science, 43*(4), 378-83. https://doi.org/10.2141/jpsa.43.378

Adler, A. J., & Holub, B. J. (1997). Effect of garlic and fish oil supplementation on serum lipid and lipoprotein concentrations in hypercholesterolemic men. *The American Journal of Clinical Nutrition, 65*(2), 445-450. https://doi:10.1093/ajcn/65.2.445

Alagawany, M., Ashour, E., & Reda, F., 2016. Effect of dietary supplementation of garlic (allium sativum) and turmeric (curcuma longa) on growth performance, carcass traits, blood profile and oxidative status in growing rabbits. *Annals of Animal Science, 16*(2), 489-505. https://doi.org/10.1515/aoas-2015-0079

Alagawany, M., El-Saadony, M. T., Elnesr, S. S., Farahat, M., Attia, G., Madkour, M., & Reda, F. M. (2021). Use of lemongrass essential oil as a feed additive in quail's nutrition: its effect on growth, carcass, blood biochemistry, antioxidant and immunological indices, digestive enzymes and intestinal microbiota. *Poultry Science, 100*(6), 1-8. https://doi.org/10.1016/j.psj.2021.101172

Al-Harthi, M. A. (2015). The effect of different dietary contents of olive cake with or without Saccharomyces cerevisiae on egg production and quality, inner organs and blood constituents of commercial layers. *European Poultry Science, 79*(1), 1-14. https://doi.org/10.1399/eps.2015.83

Alkhair, S.M. (2019). The effect of physical feed restriction during the starter period on broilers performance. *International Journal of Livestock Production, 10*(1), 1-8. https://doi.org/10.5897/IJLP2018.0523

Al-Shuwaili, M. A., Ibrahim, E. I., & Al-Bayati, M. T. (2015). Effect of dietary herbal plants supplement in turkey diet on performance and some blood biochemical parameters. *Global Journal of Bioscience and Biotechnology, 4* (1), 85-89.

Amaral, A. B., Silva, M. V., & Lannes, S. C. S. (2018). Lipid oxidation in meat: mechanisms and protective factors – a eeview. *Food Science and Technology, 38*(1), 1-15. http://doi:10.1590/ fst.32518

American Oil Chemists' Society. (1998). *Official methods and recommended practices of the American oil chemists' society Method Ce 2-66. GLC ranges of Fatty acid composition*. AOCS Press

Arvouet-Grand, A., Vennat, B., Pourrat, A., & Legret, P. (1994). Standardization of propolis extract and identification of principal constituents. *Belgian Pharmacy Journal, 49*(6), 462-468.

Bölükbaşı, S. C., & Erhan, M. K. (2007). Effects of semi replacement of dietary olive oil and corn oil with conjugated linoleic acid (CLA) on broiler performance, serum lipoprotein levels, fatty acid composition in muscles and meat quality during refrigerated storage. *Journal of Animal and Veterinary Advances, 6*(2), 262-266.

Bonsnes, R. W., & Taussky, H. H. (1945). On the colorimetric determination of creatinine by the Jaffe reaction. *Journal of Biological Chemistry, 158*(1), 581-591. https://doi.org/10.1016/s0021-9258(19)51334-5

Branciari, R., Galarini, R., Giuseppon, D., Trabalza-Marinucci, M., Forte, C., Roila, R., Miraglia, D., Servili, M., Acuti, G., & Valiani, A. (2017). Oxidative status and presence of bioactive compounds in meat from chickens fed polyphenols extracted from olive oil industry waste. *Sustainability, 9*(9), 1566-1579. https://doi.org/10.3390/su9091566

Clairborne, A. (1986). Catalase activity. In R. A. Greenwald (Ed). *CRC Handbook of methods for oxygen radical research* (pp. 283-284) CRC Press Inc.

Compassion in world farming. (2019). *The life of: Broiler chickens*. https://www.ciwf.org.uk/media/5235306/the-life-of-broiler-chickens.pdf

Crespo, N., & Esteve-Garcia, E. (2002). Nutrient and fatty acid deposition in broilers fed different dietary fatty acid profiles. *Poultry Science*, *81* (10), 1533-1542. https://doi.org/10.1093/ps/81.10.1533

Dandekar, S. P., & Rane, S. A. (2004). *Practical and viva in medical biochemistry*. Elsevier.

Das, B. S., Thurnham, D. I., Patnaik, J. K., Das, D. B., Statpathy, R., & Bose, T. K. (1990). Increased plasma lipid peroxidation in riboflavin-deficient, malaria infected children. *American Journal of Clinical Nutrition, 51*(5), 859-863. http://doi.org/10.1093/ajcn/51.5.859

Das, K., Samanta, L., & Chainy, G. B. N. (2000). A modified spectrophotometric assay of superoxide dismutase using nitrite formation by superoxide radicals. *Indian Journal of Biochemistry and Biophysics, 37*(1), 201-204.

De Almeida, J. C., Perassolo, M. S., Camargo, J. L., Bragagnolo, N., & Gross, J. L. (2006). Fatty acid composition and cholesterol content of beef and chicken meat in Southern Brazil. *Brazilian Journal of Pharmaceutical Science, 42*(1), 109-117. https://doi.org/10.1590/S1516-93322006000100012

Dermeche, S., Nadour, M., Larroche, C., Moulti-Mati, F., & Michaud, P. (2013). Olive mill wastes: biochemical characterizations and valorization strategies. P*rocess Biochemistry, 48*(10), 1532-1552. http://doi.org/10.1016/j.procbio.2013.07.010

Ding, S., Jiang, H., & Fang, J. (2018). Regulation of immune function by polyphenols. *Journal of Immunology Research*, Article*, 1*(1), 1-8. http://doi.org/10.1155/2018/1264074

Edeoga, H. O., Okwu, D. E., & Mbaebie, B. O. (2005). Phytochemical constituents of some Nigerian medicinal plants, *African Journal of Biotechnology, 4*(7), 685-688.

Elbaz, A. M., Ashmawy, E. S., Salama, A. A., Abdel-Moneim, A. E., Badri, F. B., & Thabet, H. A. (2022). Effects of garlic and lemon essential oils on performance, digestibility, plasma metabolite, and intestinal health in broilers under environmental heat stress. *BMC Veterinary Research, 18*(1), 1-12. https://doi.org/10.1186/s12917-022-03530-y

Estévez, M. (2015). Oxidative damage to poultry: from farm to fork. *Poultry Science, 94*(6), 1368-1378. https://doi.org/10.3382/ps/pev094

European Commission. (2000). *The welfare of chickens kept for meat production (broilers)* (Report of the Scientific Committee on Animal Health and Animal Welfare. Adopted 21 March 2000). https://food.ec.europa.eu/system/files/2020-12/sci-com_scah_out39_en.pdf

Gerasopoulos, K., Stagos, D., Kokkas, S., Petrotos, K., Kantas, D., Goulas, P., & Kouretas, D. (2015). Feed supplemented with by products from olive oil mill wastewater processing increases antioxidant capacity in broiler chickens. *Food and Chemical Toxicology, 82*(1), 42-49. http://doi.org/10.1016/j.fct.2015.04.021

Havenstein, G. B., Ferket, P. R., & Qureshi, M. A. (2003). Carcass composition and yield of 1957 versus 2001 broilers when fed representative 1957 and 2001 broiler diet. *Poultry Science, 82*(10), 1509-1518. https://doi.org/10.1093/ps/82.10.1509

Hernandez, F., Madrid, J., Garcia, V., Orengo, J., & Megias, M. D. (2004). Influence of two plant extracts on broilers performance, digestibility, and digestive organ size. *Poultry Science, 83*(2), 169-174. http://doi.org/10.1093/ps/83.2.169

Hlatshwayo, I. S., Mnisi, C. M., & Egbu, C. F. (2023). Effect of dietary olive (Olea europea) pomace on productive performance, and physiological and meat quality parameters in Jumbo quail. *Scientific Reports, 13*(1), 1-9. http://doi.org/10.1038/s41598-023-33495-3

Hoff, J. E., & Singleton, K. E. (1977). A method for the determination of tannin in food. *Journal of Food Science, 42*(6), 1566-1569. https://doi.org/10.1111/j.1365-2621.1977.tb08427.x

Ismail, I. E., Alagawany, M., Taha, A. E., Puvača, N., Laudadio, V., & Tufarelli, V. (2021). Effect of dietary supplementation of garlic powder and phenyl acetic acid on productive performance, blood haematology, immunity and antioxidant status of broiler chickens. *Animal Bioscience, 34*(3), 363-370. https://doi.org/10.5713/ajas.20.0140

Issa, K. J., & Omar, J. M. A. (2012). Effect of garlic powder on performance and lipid profile of broilers. *Open Journal of Animal Science, 2*(2), 62-68. http://doi.org/10.4236/ojas.2012.22010

Kakkar, P., Das, B., & Viswanathan, P.N. (1984). A modified spectrophotometric assay of superoxide dismutase. *Indian Journal of Biochemistry and Biophysics, 21*(2), 130-132.

Khan, A., Miller, W.R., & Arias, C.A. (2018). Mechanisms of antimicrobial resistance among hospital-associated pathogens. *Expert Review of Anti-infective Therapy, 16*(4), 269-287. http://doi.org/10.1080/14787210.2018.1456919

Krejčí-Treu, T., Straková, E. Suchý, P., & Herzig, I. (2010). Effect of vegetable oil fortified feeds on the content of fatty acids in breast and thigh muscles in broiler chickens. Acta *Veterinaria Brno, 79*(suppl. 9), 21-28. https://doi.org/10.2754/avb201079S9S021

Laudadio, V., Ceci, E., Lastella, N. M., & Tufarelli, V. (2015). Dietary high polyphenols extra-virgin olive oil is effective in reducing cholesterol content in eggs. *Lipids Health Disease, 14*(1), 1-7. https://doi.org/10.1186/s12944-015-0001-x

Mahmood, S., Mehmood, S., Ahmad, F., Masood, A., & Kausar, R. (2007). Effects of feed restriction during starter phase on subsequent growth performance, dressing percentage, relative organ weights and immune response of broilers. *Pakistan Veterinary Journal, 27*(3), 137-141. http://www.pvj.com.pk/pdf-files/27_3/page%20137-141.pdf

Mates, J. M., Aledo, J. C., Perez-Gomez, C., Esteban del Valle, A., & Segura, J. M. (2000). Inter-relationship between oxidative damage and antioxidant enzyme activities: an easy and rapid experimental approach. *Biochemical Education, 28*(2), 93-95. https://doi.org/10.1111/j.1539-3429.2000.tb00031.x

Mech, A., Suganthi, U., Rao, S. B. N., Sejian, V., Soren, M., David, C., Awachat, V., & Kadakol, V. (2021). Effect of dietary supplementation of linseed oil and natural antioxidants on production performance, fatty acid profile and meat lipid per oxidation in broilers. *Asian Journal of Dairy Food Research, 40*(1), 62-68. https://doi.org/10.18805/ajdfr.DR-1592

Mehri, M., Sabaghi, V., & Bagherzadeh-Kasmani, F. (2015). Mentha piperita (peppermint) in growing Japanese quails' diet: Serum biochemistry, meat quality, humoral immunity. *Animal Feed Science and Technology, 206*(1), 57-66. https://doi.org/10.1016/j.anifeedsci.2015.05.022

Mimica-Dukic, N., Bozin, B., Sokovic, M., & Simin, N. (2004). Antimicrobial and antioxidant activities of Melissa officinalis L. (Lamiaceae) essential oil. *Journal of Agricultural and Food Chemistry, 52*(9), 2485-2489. http://doi.org/10.1021/jf030698a

Nikolova, N., Pavlovski, Z., Milosevic, N., & Peric, L. (2007). The quantity of abdominal fat in broiler chicken of different genotypes from fifth to seventh week of age. *Biotechnology in Animal Husbandry, 23*(5-6), 331-338. http://dx.doi.org/10.2298/BAH0702331N

National Research Council. (1994). *Nutrient Requirements of Poultry*. National Academic Press.

Oke, O. E., Emeshili, U. K., Iyasere, O. S., Abioja, M. O., Daramola, J. O., Ladokun, A. O., Abiona, J. A., Williams, T. J., Rahman, S. A., Rotimi, S. O, Balogun, S. I., & Adejuyigbe, A. E. (2017). Physiological responses and performance of broiler chickens offered olive leaf extract under a hot humid tropical climate. *Journal of Applied Poultry Research, 26*(3), 376-382. https://doi.org/10.3382/japr/pfx005

Omar, J. A. (2000). Effect of different levels of olive pulp on the digestibility of broiler chicks. *Bethlehem University Journal, 19*(1), 97-100.

Omar, S. H. (2010). Oleuropein in olive and its pharmacological effects. *Scientia Pharmaceutica, 78(*2), 133-154. https://doi.org/10.3797/scipharm.0912-18

Paglia, D. E., & Valentine, W. N. (1967). Studies on the quantitative and qualitative characterization of erythrocyte glutathione peroxidase. *Journal of Laboratory and Clinical Medicine, 70*(1), 158-169.

Pappas, A., Tsiplakou, E., Papadomichelakis, G., Mitsiopoulou, C., Sotirakoglou, K., Mpekelis, V., Haroutounian, S. A., Fegeros, K., & Zervas, G. (2019). Effects of olive pulp addition to broiler diets on performance, selected biochemical parameters and antioxidant enzymes. *Journal of the Hellenic Veterinary Medical Society, 70*(3), 1687-1696. https://doi.org/10.12681/jhvms.21793

Park, Y. K., Albright, J., Liu, W., Storkson, J. M., Cook. M. E., & Pariza, M. W. (1997). Effect of conjugated linoleic acid on body composition in mice. Lipids*, 32*(8), 853-858. https://doi.org/10.1007/s11745-997-0109-x

Pirozzi, C., Lama, A., Simeoli, R., Paciello, O., Pagano, T. B., Mollica, M. P., Canani, R. B., Raso, Calignano, A., & Meli, R. (2016). Hydroxytyrosol prevents metabolic impairment reducing hepatic inflammation and restoring duodenal integrity in a rat model of NAFLD. *Journal of Nutritional Biochemistry, 30*(1), 108-115. https://doi.org/10.1016/j.jnutbio.2015.12.004

Prasad, R., Rose, M. K., Virmani, M., Garg, S. L., & Puri, J. P. (2009). Lipid profile of chicken (*Gallus domesticus*) in response to dietary supplementation of garlic (*Allium sativum*). *International Journal of Poultry Science, 8*(3), 270-276. https://doi.org/10.3923/ijps.2009.270.276

Rafei-Tari, A., Sadeghi, A. A., & Mousavi, S. N. (2021). Inclusion of vegetable oils in diets of broiler chicken raised in hot weather and effects on antioxidant capacity, lipid components in the blood and immune responses. *Acta Scientiarum. Animal Sciences, 43*(1), e50587. https://doi.org/10.4025/actascianimsci.v43i1.50587

Ross Broiler. (2019). *Nutrition specifications*. Aviagen Brand. https://yestav.com/media/RossBroilerNutritionSpecs2019-EN.pdf

Saima, S., Fiaz, M., Zafar, R., Ahmed, I., & Arshad, M. (2020). Chapter 6-Dissemination of antibiotic resistance in the environment. In M. Z. Hashmi (Ed.). *Antibiotics and antimicrobial resistance genes in the environment.* (pp. 99-116). Elsevier

Sánchez-Fidalgo, S., Cárdeno, A., Sánchez-Hidalgo, M., Aparicio-Soto, M., & De la Lastra, C. A. (2013). Dietary extra virgin olive oil polyphenols supplementation modulates DSS-induced chronic colitis in mice. *Journal of Nutritional Biochemistry, 24*(7), 1401-1413. https://doi.org/10.1016/j.jnutbio.2012.11.008

Santoso, U., Tanaka, K., & Ohtani, S. (1995). Early skip-a-day feeding of female broiler chicks fed high-protein re-alimentation diets: performance and body composition. *Poultry Science, 74*(3), 494-501. https://doi.org/10.3382/ps.0740494

Sarica, S., & Toptas, S. (2014). Effects of dietary oleuropein supplementation on growth performance, serum lipid concentrations and lipid oxidation of japanese quails. *Journal of Animal Physiology and Animal Nutrition, 98*(6), 1176-1186. https://doi.org/10.1111/ jpn.12192þ

SAS Institute Inc. (2000). *SAS/ETS@ 9.1. User's Guide*. SAS Institute

Schuman, B. E., Squires, E. J., & Leeson, S. (2000). Effect of dietary flax seed, flax oil and n-3 fatty acid supplement on hepatic and plasma characteristics relevant to fatty liver haemorrhagic syndrome in laying hens. *British Poultry Science, 41*(4), 465-472. https://doi.org/10.1080/713654970

Shi, C., Chen, X., Liu, Z., Meng, R., Zhao, X., Liu, Z., & Guo, N. (2017). Oleuropein protects L-02 cells against H_2O_2-induced oxidative stress by increasing SOD1, GPx1 and CAT expression. *Biomedicine & Pharmacotherapy, 85*(1), 740-748 https://doi.org/10.1016/j.biopha.2016.11.092

Siddiqui, A. A., & Ali, M. (1997). *Practical Pharmaceutical Chemistry*. CBS Publishers and Distributors.

Stanaćev, V., Glamočić, D., Milošević, N., Perić, L., Puvača, N., Stanaćev, V., Milic, D., & Plavša, N. (2012). Influence of garlic (*Allium sativum* L.) and copper as phyto additives in the feed on the content of cholesterol in the tissues of the chickens. *Journal of Medicinal Plants Research, 6*(14), 2816-2819. http://dx.doi.org/10.5897/JMPR11.1255

Stanacev, V., Glamocic, D., Milosevic, N., Puvaca, N., Stanacev, V., & Plavsa, N. (2011). Effect of garlic (*Allium sativum L.*) in fattening chicks' nutrition. *African Journal of Agricultural Research, 6*(4), 943-948. http://dx.doi.org/10.5897/AJAR10.908

Tietz, N. W. (1995). *Clinical guide to laboratory tests*. W. B. Saunders Company.

Tsiplakou, E., Mitsiopoulou, C., Mavrommatis, A., Karaiskou, C., Chronopoulou, E. G., Mavridis, G., Sotirakoglou, K., Labor, N. E., & Zervas, G. (2017). Effect of under- and overfeeding on sheep and goat milk and plasma enzymes activities related to oxidation. *Journal of Animal Physiology and Animal Nutrition, 102*(1), 288–298. https://doi.org/10.1111/jpn.12741

Velleman, S. G., Coy, C. S., & Emmerson, D. A. (2014). Effect of the timing of post-hatch feed restrictions on the deposition of fat during broiler breast muscle development. *Poultry Science, 93*(10), 2622-2627. https://doi.org/10.3382/ps.2014-04206.

Wang, H., Sit, W. H., Tipoe, G. L., & Wan, J. M. F. (2014). Differential protective effects of extra virgin olive oil and corn oil in liver injury: a proteomic study. *Food and Chemical Toxicology, 74*(1), 131-138. https://doi.org/10.1016/j.fct.2014.09.002

World Health Organization. (2015). *Antimicrobial resistance: global report on surveillance*. WHO.

Wootton, T. D. (1964). *Micro analysis in medical bio chemistry*. Churchill Ltd.

Wright, L. P., Mphangwe, N. I. K., Nyirenda, H. E., & Apostolides, Z. (2000). Analysis of caffeine and flavan-3-ol composition in the fresh leaf of Camellia sinensis foe predicting the quality of black tea produced in Central and Southern Africa. *Journal of the Science of Food and Agriculture, 80*(13), 1823-1830. http://dx.doi.org/10.1002/1097-0010(200010)80:13<1823::AID-JSFA702>3.0.CO;2-E

Wyatt, T. A., Kharbanda, K. K., Tuma, D. J., & Sisson, J. H. (2001). Malondialdehyde–acetaldehyde-adducted bovine serum albumin activates protein kinase C and stimulates interleukin-8 release in bovine bronchial epithelial cells. *Alcohol, 25*(3), 159-166. https://doi.org/10.1016/s0741-8329(01)00177-x

Yeh, Y. Y., & Yeh, S. M. (1994). Garlic reduces plasma lipids by inhibiting hepatic cholesterol and triacylglycerol synthesis. *Lipids, 29*(3), 189-193. https://doi.org/10.1007/bf02536728

Zhang, Z. F., Zhou, T. X., & Kim, I. H. (2013). Effects of dietary olive oil on growth performance, carcass parameters, serum characteristics, and fatty acid composition of breast and drumstick meat in broilers. *Asian-Australasian Journal of Animal Science, 26*(3), 416-422. http://dx.doi.org/10.5713/ajas.2012.12486

Associate Editor in charge:
Leandro Dalcin Castilha
ORCID: https://orcid.org/0000-0003-4799-2839