

## Response of Broiler Chickens to the Dietary Fortification of Bile Acids

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### Abstract

The feeding trial was conducted for 31 days to investigate whether dietary energy modifications using bile acid feed additive (Runeon®) affected broiler performance, carcass characteristics, blood indices, intestinal lipase activity, and broiler's meat quality. A total of 1200 one-day-old Ross-308 broiler chicks (as hatch) were randomly distributed into three groups, each with five replicates (80 chicks/replicate). The first group was a control (T1) which fed a basal diet only. In the second group (T2), birds were fed the basal diet supplemented with bile acid (Runeon®) (on top application) at the rate of 200g/ton. In the third group (T3), birds were fed a basal diet reduced in energy requirements by 30kcal/kg and reformulated with 200g/ton of bile acid (Runeon®). Birds' diets fortified with bile acid in (T2) or (T3) significantly ( $P \leq 0.05$ ) improved body weight, body weight gain, feed conversion ratio (FCR), and European Production Efficiency Factor (EPEF) as compared to the control. The dressing%, breast, thigh, and drumstick yields were improved in T2 and T3 than in control. Supplementation of bile acid significantly ( $P \leq 0.05$ ) reduced abdominal fat%, as well, blood cholesterol, triacylglycerol, HDL, and LDL concentrations, but increased total protein concentration ( $P \leq 0.05$ ). Additionally, intestinal lipase levels significantly ( $P \leq 0.05$ ) increased in groups fortified with bile acid (T2 and T3). Besides, chicken meat moisture% and fat% were significantly ( $P \leq 0.05$ ) decreased in T3 compared to T1 and T2. Conclusively, dietary fortification of bile acid could improve growth performance, profitability, carcass traits, serum lipids profile, intestinal lipase secretion, and chicken meat quality in broiler chickens.

### KEYWORDS

Broiler performance, Bile acids, Abdominal fat, Intestinal lipase, Blood indices.

## INTRODUCTION

Birds, nutrition, management, and hygiene are the principal components of the poultry industry. Proper poultry nutrition represents about 75-80% of the total production costs (Chatterjee & Rajkumar, 2015). Energy is one of the main macronutrients that comprise about 55-60% of the feed cost (Pantaya *et al.*, 2020). According to (NRC, 1994), fats and vegetable oils provide almost 3-times more apparent metabolizable energy (AME) than other feedstuffs. Hence, they are the highest energy sources for poultry, with the highest caloric value of all nutrients. Additionally, they are the source of polyunsaturated fatty acids, which are essential for broiler chickens. As a result, fat is commonly added to poultry diets to meet the energy demands of modern broiler breeds, increase energy density, and improve nutritional outcomes (Prabakaran, 2003; Abudabos, 2014; Wu, 2017).

Globally, the price of vegetable oils is incredibly high, so the improvement of fat digestion and absorption is critically essential to reduce the cost and density of dietary energy through bile acid supplementation which aids in emulsifying the dietary fat. Also, it is important to consider the source of fat in the diet and the

level of bile secreted for fat digestion (Reshetnyak, 2013). Bile secretion is limited in young chicks, which causes digestion and absorption of dietary fat to be poorly developed (Krogdahl, 1985). This has led to the consideration of bile salts and synthesized bile acid in the diet of young broilers to increase fat utilization.

It has been reported that bile acids may enhance fat digestibility in broilers (Upadhaya *et al.*, 2019) and improve broiler performance (Parsaie *et al.*, 2007). A mixture of natural compounds produces bile acids, including deoxycholic acid, lithocholic acid, cholic acid, and chenodeoxycholic acid. Each species' bile acids differ in composition since the avian bile acids contain mostly cholic acid and chenodeoxycholic acid (Hofmann and Hagey, 2008). Birds synthesize natural bile acids from cholesterol, then conjugate them with taurine or glycine and store them in their gallbladders as primary bile acids. Upon ingestion of feed, they are carried into the duodenum to emulsify dietary lipids, along with secondary bile acids formed through bacterial alteration of primary bile acids via deconjugation (Lai *et al.*, 2018b). About 95% of bile acid is reabsorbed at the end of the ileum by the apical ileal sodium-dependent bile acid cotransporter (ASBT). The residual bile acid is then transported to the liver to be deconju-

gated to secondary bile acids (Chiang, 2017).

Therefore, this study aimed to investigate the influence of exogenous dietary fortification of bile acids (Runeon®) on growth performance, carcass traits, serum biochemical indices, intestinal lipase activity, and chicken meat quality in broiler chickens fed modified energy diets.

## MATERIALS AND METHODS

### *Birds, diets, and husbandry*

This study was conducted in the Animal and Poultry Research Center, Faculty of Veterinary Medicine, Cairo University, Egypt. The Faculty of Veterinary Medicine's Committee for the Institutional Animal Care and Use, Cairo University, Egypt, agreed on the experimental design (Vet CU 23052022452).

A total of 1200 one-day-old Ross 308 broiler chicks (as hatch) were randomly assigned into three different groups, each group was subdivided into five replicates, and each replicate contained 80 birds. The control group (T1) was fed the basal diet only without any supplementation. The basal diet was formulated according to Ross 308 broiler nutrient specification manual 2019 (AVIA-GEN, 2019) (Table 1). In the second group (T2), chickens were fed the basal diet supplemented with bile acid (on top application) added at the rate of 200g/ton of feed (Runeon®). In the third group (T3), chickens were fed the basal diet with 30kcal/kg reduced in energy requirements and reformulated with 200g/ton bile acid (Runeon®) (Table 1). Runeon® is a dietary bile acid supplementation used as a natural emulsifier for dietary fats (Shandong Longchang Animal Health Product Co., Ltd., Shandong, China). Feed and fresh water were provided ad libitum during the whole trial.

Birds were reared in deep litter floor pens bedded with wood shaving, with a stocking density of 10 birds/m<sup>2</sup>. Birds' diets were fortified with bile acid supplementation from day 1 to day 31 of age. Birds were housed in a semi-closed system. Birds were vaccinated against H5N1 Avian influenza at 7 days old and Newcastle disease at 7, 13, and 20 days old.

### *Growth performance*

The changes in body weights and feed consumption were recorded weekly to calculate the body weight, weight gain, feed intake, feed conversion ratio (FCR), and European Production Efficiency Factor (EPEF).

### *Carcass traits*

At the end of the experiment, three representative birds from each replicate (15 birds from each group) were weighted and euthanized after 4 h of fasting for complete evacuation of the gut. Each bird was scalded, de-feathered, and eviscerated after the exclusion of the head, neck, and legs. Weighted carcasses without giblets were expressed as a percentage of their live weight (carcass weight). Breast, thigh, and drumstick weights were measured after the carcass was dissected. In addition, the gizzard, heart, the weight of the liver (without gall bladder), spleen, and bursa of Fabricius were verified and their relations to the carcass weight of the birds were calculated in percentage (the relative organ weight) (Lai et al., 2018b). The abdominal fat which is located in tissues surrounding the proventriculus and gizzard, around the cloaca, and on the inside of the abdominal wall (Ricard et al., 1983) was also determined.

### *Blood biochemical indices*

At the end of the experiment, blood samples were collected from 15 birds/groups. Samples were permitted to clot at room temperature and then centrifuged for 15 min at 4000 rpm and clear serum was separated and preserved at -20 °C until used for biochemical analysis (Ali et al., 2022). Determinations of total protein (at wavelength 546nm), albumin (at wavelength 578nm), triacylglycerol (at wavelength 505nm), total cholesterol (at wavelength 500nm), LDL (at wavelength 500nm), and HDL (at wavelength 500nm) in sera were performed by using spectrum diagnostics kits (Spectrum Diagnostics Egyptian Company for Biotechnology) according to the manufacturer's instructions (UV-2100 Spectrophotometer, USA).

### *Colorimetric determination of intestinal lipase activity*

At the end of the experiment, broiler chickens were euthanized, and the small intestine was dissected, weighed, and homogenized in PBS for preparation of homogenate of 10 % concentration, which was then centrifuged at 12000 rpm for 15 min to obtain a clear supernatant (Hu et al., 2018). Intestinal lipase activity was determined in the separated supernatant by using a spectrum diagnostics kit (Spectrum Diagnostics Egyptian Company for Biotechnology) at a wavelength of 580nm according to the manufacturer's instructions (UV-2100 Spectrophotometer, USA).

### *Chicken meat quality*

At the end of the experiment during slaughter, five chicken meat samples from each bird group were collected to determine the moisture, crude protein (CP), ether extract (EE), and ash percentages (Thiex et al., 2012).

### *Statistical analysis*

Results were summarized in tables as means and standard errors of means (SEM). Statistical inference was tested using a one-way Analysis of Variance (ANOVA). Tukey's posthoc test was applied to compare the differences between means of the three groups. Significance was indicated at ( $P \leq 0.05$ ). Data analysis was performed using PASW Statistics, Version 18.0 software (SPSS Inc., Chicago, IL, USA). Boxplot was designed with ggplot2 and ggsignif packages (Wickham, 2016; Ahlmann-Eltze and Patil, 2021) using R for Statistical Computing (<https://www.r-project.org/>).

## RESULTS

### *Growth performance*

The changes in body weight, body weight gain, feed consumption and feed conversion ratio during different feeding phases, as well as the cumulative growth performance during the whole experimental period (1-31 days) of broiler chickens fed either basal or bile acid supplemented diets were illustrated in Tables 2 and 3. The data showed that supplementation of broiler chickens' diet with bile acid significantly ( $P < 0.05$ ) increased the final body weight and body weight gain by 8 and 9% and improved the feed conversion ratio by 5 and 7% in T2 and T3, respectively, compared to the control. Also, the European production efficiency factor was significantly ( $P < 0.05$ ) increased by 6 and 15 % in T2 and T3, respectively, compared to the control.

Carcass traits

The carcass traits of broiler chickens fed either basal or bile acid supplemented diets were shown in Table 4. The data showed that the dressing percent, breast, thigh, and drumstick yields were improved in T2 and T3 compared to T1; however, the differences were not significant. The abdominal fat percentage showed significant ( $P= 0.003$ ) reductions by 22 and 29% in T2 and T3, respectively, compared to T1. The giblets and immune organs percentages were similar among all groups with no significant variation.

Blood biochemical indices

Blood biochemical indices of broiler chickens fed either basal or bile acid supplemented diets were shown in Table 5. The data revealed that dietary bile acid in T2 and T3 significantly ( $P < 0.0001$ ) reduced the concentrations of total cholesterol by 20 and 28%, triacylglycerol by 27 and 39%, HDL by 17 and 26%, and LDL by 16 and 24%, respectively, compared to T1. Albumin levels were increased in T2 and T3 compared to T1, however, the differences were not significant. Also, total protein concentrations were significantly ( $P = 0.002$ ) increased by 2 and 16% in T2 and

Table 1. Ingredients composition and chemical analysis of the basal and experimental diets.

Ingredients	Basal diet			Experimental diet		
	Starter	Grower	Finisher	Starter	Grower	Finisher
Yellow corn	58.1	62.69	67.14	58.58	63.17	67.62
Soybean meal 46% CP	34.19	29.6	24.5	34.19	29.6	24.5
Corn gluten meal 60% CP	3.5	3	3	3.5	3	3
Soya oil	0.5	1.3	2	0	0.8	1.5
NaCl	0.35	0.35	0.35	0.35	0.35	0.35
Sodium bicarbonate	0.1	0.1	0.1	0.1	0.1	0.1
DCP	1.3	1.1	1	1.3	1.1	1
Limestone	1.3	1.2	1.2	1.3	1.2	1.2
DL-Methionine	0.2	0.18	0.2	0.2	0.18	0.2
L-Lysine	0.2	0.25	0.3	0.2	0.25	0.3
L-Threonine	0.1	0.07	0.05	0.1	0.07	0.05
Runeon®	0	0	0	0.02	0.02	0.02
Ronozyme proact	0.01	0.01	0.01	0.01	0.01	0.01
Ronozyme WX 2000	0.01	0.01	0.01	0.01	0.01	0.01
Hi Phos	0.01	0.01	0.01	0.01	0.01	0.01
Broiler premix <sup>1</sup>	0.13	0.13	0.13	0.13	0.13	0.13
Total	100	100	100	100	100	100
Chemical analysis						
ME (kcal/kg)	3009	3103	3202	3012	3100	3204
CP%	23.06	21.06	19.07	23.1	21.02	19.11
EE%	3.08	3.49	4.77	2.6	3.97	4.29
CF%	2.33	2.27	2.19	2.34	2.26	2.2
Calcium%	1.02	0.93	0.89	1.02	0.93	0.89
Available Phosphorus %	0.49	0.45	0.42	0.49	0.45	0.43

<sup>1</sup>Broiler premix contained per kg: Vit A 12,500,000 IU, Vit D3 5,000,000 IU, Vit E 70,000mg, Vit K3 3,500mg, Vit B1 3,000mg, Vit B2 7,000mg, Vit B6 4,000mg, Vit B12 20mg, Nicotinic acid 50,000mg, Pantothenic acid 15,000mg, Biotin 180mg, Folic acid 2,000mg, hy D 70mg, Iron 44,000mg, Copper 6,000mg, manganese 70,000mg, Zinc 75,000mg, Iodine 1,300mg, Selenium 230mg.

Table 2. Impact of dietary bile acid (Runeon®) fortification on broilers' performance.

Groups	Body weight (g)			Weight gain (g)			Feed intake (g)			FCR (g/g)		
	D 12	D 25	D 31	D 12	D 25	D 31	D 12	D 25	D 31	D 12	D 25	D 31
T1	442	1569	2058 <sup>b</sup>	400	1127	490 <sup>b</sup>	461 <sup>b</sup>	1434 <sup>b</sup>	966	1.15	1.27	1.97 <sup>a</sup>
T2	443	1617	2236 <sup>a</sup>	401	1174	620 <sup>a</sup>	475 <sup>ab</sup>	1514 <sup>a</sup>	1043	1.19	1.29	1.71 <sup>ab</sup>
T3	451	1612	2265 <sup>a</sup>	410	1161	652 <sup>a</sup>	485 <sup>a</sup>	1468 <sup>ab</sup>	967	1.18	1.27	1.49 <sup>b</sup>
SEM <sup>1</sup>	3.61	16.04	25.08	3.61	14.44	25.69	3.85	13.98	17.88	0.02	0.02	0.07
<i>p</i> -value	NS <sup>2</sup>	NS	0.000	NS	NS	0.01	0.018	0.05	NS	NS	NS	0.002

<sup>ab</sup> Mean values with different superscripts in the same column indicate a significant difference (Tukey's test;  $P \leq 0.05$ ).

T1, Control (basal diet); T2, basal diet + Runeon® (200 g/ton feed); T3, basal diet (-30 kcal/kg metabolizable energy) + Runeon® (200 g/ton feed).

<sup>1</sup>SEM: Standard error of the mean.

<sup>2</sup>NS: Not significant.

T3, respectively, compared to the control.

**Intestinal lipase enzyme activity**

The intestinal lipase levels of broiler chickens fed either basal or bile acid supplemented diets are demonstrated in Figure 1. The data showed that intestinal lipase enzyme concentration was significantly ( $P = 0.010$ ) increased by 14 and 17% in T2 ( $53.62 \pm 2.67$  U/L) and T3 ( $51.38 \pm 1.82$  U/L), respectively, compared to the control ( $44.16 \pm 1.26$  U/L).

**Chicken meat quality parameters**

The moisture, crude protein (CP), ether extract (EE), and Ash percentages in chickens' meat of broilers fed either basal or bile acid supplemented diet are illustrated in Table 6. The data showed that the moisture and EE percentages significantly ( $P \leq 0.05$ ) decreased in T3 compared to T1 and T2. Moreover, both CP and Ash recorded higher percentages in T2 and T3 compared to the

control, although the differences were not significant.

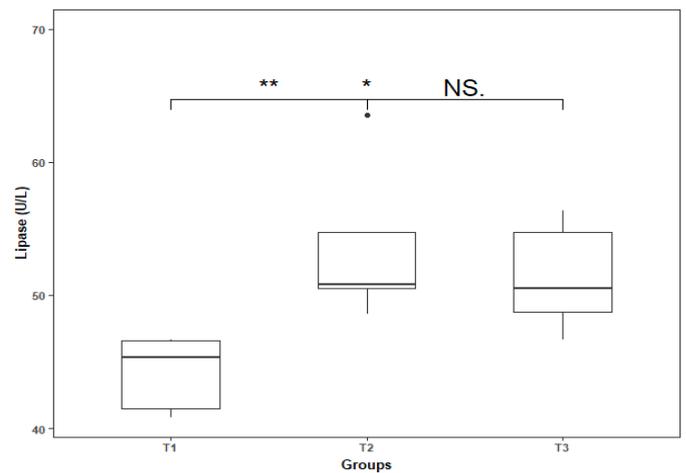


Fig. 1. Impact of dietary bile acid (Runeon®) fortification on broilers' intestinal lipase enzyme level.

Table 3. Impact of dietary bile acid (Runeon®) fortification on broilers' cumulative performance (days 1-31).

Groups	Body weight (g)	Weight gain (g)	Feed intake (g)	FCR (g/g)	EPEF	Mortality (%)
T1	2058 <sup>b</sup>	2017 <sup>b</sup>	2861	1.42 <sup>a</sup>	438 <sup>c</sup>	3.5
T2	2236 <sup>a</sup>	2195 <sup>a</sup>	3032	1.38 <sup>ab</sup>	482 <sup>b</sup>	4.75
T3	2265 <sup>a</sup>	2223 <sup>a</sup>	2919	1.31 <sup>b</sup>	515 <sup>a</sup>	4.5
SEM <sup>1</sup>	25.08	25.08	31.28	0.02	9.38	0.27
<i>p</i> -value	0.000	0.000	NS <sup>2</sup>	0.005	0.000	NS

<sup>a,b,c</sup> Mean values with different superscripts in the same column show a significant difference (Tukey's test;  $P \leq 0.05$ ).

T1, Control (basal diet); T2, basal diet + Runeon® (200 g/ton feed); T3, basal diet (-30 kcal/kg metabolizable energy) + Runeon® (200 g/ton feed).

FCR, Feed Conversion Ratio (g of feed / g of weight gain); EPEF, European Production Efficiency Factor= (body weight (kg) × % viability × 100 / feed conversion ratio (g feed/g gain) × Age (d)) (Marcu et al., 2013)

<sup>1</sup>SEM: Standard error of the mean.

<sup>2</sup>NS: Not significant.

Table 4. Impact of dietary bile acid (Runeon®) fortification on broilers' carcass traits.

Groups	Dressing (%)	Breast (%)	Thigh (%)	Drum (%)	Abdominal fat (%)	Liver (%)	Gizzard (%)	Heart (%)	Spleen (%)	Bursa (%)
T1	73.28	36.51	25.51	12.91	2.34 <sup>a</sup>	0.33	0.16	0.67	0.19	0.22
T2	74.51	37.01	25.88	13.12	1.66 <sup>b</sup>	0.3	0.16	0.65	0.16	0.19
T3	74.31	36.81	26.06	13.5	1.83 <sup>b</sup>	0.31	0.16	0.67	0.16	0.21
SEM <sup>1</sup>	0.3	0.24	0.15	0.11	0.07	0.01	0.02	0.01	0	0.01
<i>p</i> -value	NS <sup>2</sup>	NS	NS	NS	0.003	NS	NS	NS	NS	NS

<sup>a,b</sup> Mean values with different superscripts in the same column indicate a significant difference (Tukey's test;  $P \leq 0.05$ ).

T1, Control (basal diet); T2, basal diet + Runeon® (200 g/ton feed); T3, basal diet (-30 kcal/kg metabolizable energy) + Runeon® (200 g/ton feed).

<sup>1</sup>SEM: Standard error of the mean.

<sup>2</sup>NS: Not significant.

Table 5. Impact of dietary bile acid (Runeon®) fortification on broilers' blood biochemical indices.

Groups	Cholesterol (mg/dl)	TAG (mg/dl)	HDL (mg/dl)	LDL (mg/dl)	Albumin (g/dL)	Total protein (g/dL)
T1	114.76 <sup>a</sup>	74.33 <sup>a</sup>	75.26 <sup>a</sup>	24.31 <sup>a</sup>	1.24	1.99 <sup>b</sup>
T2	91.97 <sup>b</sup>	54.13 <sup>b</sup>	62.60 <sup>b</sup>	20.54 <sup>b</sup>	1.32	2.32 <sup>a</sup>
T3	82.10 <sup>c</sup>	45.32 <sup>c</sup>	55.97 <sup>c</sup>	18.41 <sup>b</sup>	1.42	2.37 <sup>a</sup>
SEM <sup>1</sup>	3.7	3.33	2.3	0.75	0.06	0.05
<i>p</i> -value	<0.0001	<0.0001	<0.0001	<0.0001	NS <sup>2</sup>	0.002

<sup>a,b,c</sup> Mean values with different superscripts in the same column show a significant difference (Tukey's test;  $P \leq 0.05$ ).

T1, Control (basal diet); T2, basal diet + Runeon® (200 g/ton feed); T3, basal diet (-30 kcal/kg metabolizable energy) + Runeon® (200 g/ton feed).

TAG: Triacylglycerol, HDL: High-density lipoprotein, LDL: Low-density lipoprotein.

<sup>1</sup>SEM: Standard error of the mean.

<sup>2</sup>NS: Not significant.

Table 6. Impact of dietary bile acid (Runeon®) fortification on chickens' meat quality parameters.

Groups	Moisture (%)	Crude Protein (%)	Ether Extract (%)	Ash (%)
T1	76.14 <sup>a</sup>	17.58	3.10 <sup>a</sup>	1.22
T2	76.08 <sup>a</sup>	18.15	2.47 <sup>a</sup>	1.45
T3	74.95 <sup>b</sup>	17.91	1.51 <sup>b</sup>	1.51
SEM <sup>1</sup>	0.21	0.29	0.2	0.06
<i>p</i> -value	0.01	NS <sup>2</sup>	0.001	NS

<sup>a,b</sup> Mean values with different superscripts in the same column indicate a significant difference (Tukey's test;  $P \leq 0.05$ ).

T1, Control (basal diet); T2, basal diet + Runeon® (200 g/ton feed); T3, basal diet (-30 kcal/kg metabolizable energy) + Runeon® (200 g/ton feed).

<sup>1</sup> SEM: Standard error of the mean.

<sup>2</sup> NS: Not significant.

## DISCUSSION

In the poultry industry, bile acids have been studied for their potential as dietary emulsifiers to increase fat digestibility (Upadhaya *et al.*, 2019) as well as enhance broiler performance (Parsaie *et al.*, 2007).

In the current study, on top application of 200g/ton bile acid to broiler basal diet (T2) or energy-reduced diet (-30kcal/kg energy) (T3) significantly increased the birds' performance in terms of body weight and body weight gain and improved FCR, although no significant differences were recorded in the feed consumption. The results agreed with Alzawqari *et al.* (2016) and Lai *et al.* (2018b) findings, who reported improved BWG and FCR in broiler chicks fed bile acid fortified diets, with no effect on average feed consumption. Supplementation of bile acids to broiler diets improved live body weight, weight gain, and FCR by improving nutrient digestion and dietary lipid absorption, which enhances the bioavailability of fat-soluble vitamins (A, D, E, and K) (Stamp and Jenkins, 2008). Moreover, chickens in T3 fed a diet that was reduced in 30kcal/kg of energy requirements and reformulated with 200g/ton bile acid recorded the best values for FCR and the European production efficiency factor (EPEF). Various measures of boiler performance are included in the EPEF, such as BW, survival rate, FCR, and production management. Recently, EPEF had become increasingly recognized by practitioners as an essential performance measurement method, as well as a significant profitability index (Bhamare *et al.*, 2016; Śliżewska *et al.*, 2020).

Broiler chickens fed diets supplemented with bile acids (T2, T3) recorded a significant ( $P \leq 0.05$ ) decrease in abdominal fat percent compared with the control. These results agreed with those obtained by Youssef *et al.* (2017), Lai *et al.* (2018a), and Ge *et al.* (2019), who reported that diets with different energy and bile acid levels significantly decreased abdominal fat. Lai *et al.* (2018a) attributed the decreased fat mass to the improved absorption of dietary lipids caused by bile acid supplementation, so fat is not stored in the abdominal fat pad. Becker *et al.* (1979) and Thomas *et al.* (1983) stated that the overall body fat level in birds is consistent with the abdominal fat pad. On the other hand, carcass traits (dressing percent, breast, thigh, drumstick yields) recorded better indices in both T2 and T3 regardless of the difference in energy levels compared to control, however, significance was not indicated. Similarly, Etop *et al.* (2020) observed strong positive relation between dressing percent and 100-200 g/ton bile acid supplementation in broiler diet, though differences between the broiler chickens fed diets supplemented with bile acids and the control were not significant. Additionally, Lai *et al.* (2018b) stated that birds supplemented with dietary bile acids at rates of 60 or 80 mg/kg reported higher percentages for dressing and thigh muscle weights ( $P < 0.010$ ) than control. The obtained relative weights of giblets and immune organs did not show significant differences between bird groups. Similar findings were reported previously by Ge *et al.* (2019) and Arshad *et al.* (2020) who stated that low energy diet did not affect the liver, spleen, gizzard, abdominal fat, and bursa of Fabricius. Results of liver weight percent exhibited 6 and 9% lower values in T3 and T2, respectively than in

control. This could be attributed to the applied inclusion rate, as Etop *et al.* (2020) previously stated that liver weight is negatively correlated with the level of supplemental bile acids.

In lipid metabolism, serum TG, LDL, and HDL concentrations can be utilized as diagnostic indicators (Lai *et al.*, 2018b). Dietary supplementation of bile acid in T2 and T3 significantly decreased the concentrations of HDL, triacylglycerol, LDL, and total cholesterol. The study of El-Katcha *et al.* (2019) found that containing 400g of dried bile acids (DBA)/ton of feed reduced serum total cholesterol, TG, and LDL concentrations insignificantly. At 21 and 42 days of age, Alzawqari *et al.* (2010) showed that birds that were fed 0.5% DBA had significantly higher serum triglyceride, cholesterol, LDL, and HDL concentrations than control. This may be due to the higher inclusion rate. Total protein concentration was significantly increased by bile acid supplementation in T2 and T3. We found similar results to those reported by Natsir *et al.* (2017), which showed that bile acids affected serum total protein and albumin more effectively. Several metabolic processes are regulated by bile acids, including the absorption of nutrients and the maintenance of cholesterol levels (Staels *et al.*, 2010).

It has been shown that intestinal lipase activity can serve as an indicator of poultry lipid utilization (Lai *et al.*, 2018b). In the current study, the action of intestinal lipase was amplified significantly due to the bile acid fortification in T2 and T3. This result agreed with Lai *et al.* (2018b) who reported that the activity of lipoprotein lipase was significantly increased by 60 and 80 mg/kg dietary levels of bile acids. This enzyme is essential for the metabolism of lipids, as it catalyzes triglyceride hydrolysis. Additionally, bile acid improves the digestion and absorption of dietary fats, hence raising the activity of the lipase enzyme and facilitating its action in fat digestion.

Chicken meat quality in the bile acid supplemented group showed a significant decrease in both moisture and ether extract percentages, however, crude protein and Ash percentages had not been affected. These findings were established by Kumar *et al.* (2018) and Hamada *et al.* (2021). The cause of decreasing fat percent in broiler chickens' meat may be attributed to the enhanced nutrients and lipids absorption caused by bile acid, which reduces fat storage in the abdomen (Lai *et al.* 2018a). Also, Suzuki *et al.* (2014) observed that after birds were fed a 400-kcal meal, stored body fat was adversely correlated with postprandial levels of bile acid.

## CONCLUSION

Conclusively, the dietary supplementation of bile acids (Runeon®) in broiler chickens' diets either on top dressing or reformulation could improve growth performance, carcass traits, and health status of broiler chickens.

## ACKNOWLEDGMENTS

The authors thank Dakahlia Poultry Company, which provided Runeon® (Shandong Longchang Animal Health Product Co., Ltd., Shandong, China).

**CONFLICT OF INTEREST**

The authors declare that they have no conflict of interest.

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