



## Growth Performance, Nutrients Digestibility, Immune System, and Blood Parameters in Broiler Chickens Fed on Diets Supplemented with Cumin (*Cuminum cyminum*) or Black Cumin (*Bunium persicum*) Seed Powders

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### Abstract

Effects of dietary inclusion of cumin (CUM) and black cumin (BCUM) powders were investigated on the performance, nutrient digestibility, lipid deposition, and immunocompetence of broiler chickens in a rearing period of 42 days. A total number of 240 male ROSS 308 day-old chicks were randomly allocated to six dietary treatments with four replicates. Dietary treatments consisted of a basal diet as control, control + 10 ppm avilamycin, control + 0.25% CUM, control + 0.75% CUM, control + 0.25% BCUM, and control + 0.75% BCUM. Feed intake, body weight gain (BWG), and feed conversion ratio (FCR) were recorded weekly. Total tract apparent digestibility (TTAD) of crude protein (CP) and ether extract (EE) were measured on day 21. Sheep red blood cells (SRBC) and cutaneous basophil hypersensitivity (CBH) tests were used to evaluate immune responses. On day 42, two chickens from each replicate were selected, bled, euthanized, and carcass, abdominal fat pad, and internal organs were weighted. CUM and also avilamycin improved BWG during the grower and whole period of the experiment. Also, FCR was improved by CUM (0.75%) as well as avilamycin compared to control. Also, CUM (0.75%) decreased serum total cholesterol and LDL, and increased anti-SRBC response compared to control. Supplementing the diet with 0.75% CUM also decreased abdominal fat pad percentage compared to other groups. There was an improvement in TTAD of CP and EE with dietary inclusion of CUM (0.75%) as well as avilamycin, compared to control. However, BCUM did not change the all measured parameters but increased FCR and decreased (0.75% BCUM) BWG and TTAD of CP compared to control. This study indicated growth-promoting, immunostimulatory, and hypolipidemic effects for cumin as a phytogetic feed additive. Then, it may act as an alternative for in-feed antibiotics in broiler nutrition.

### Introduction

From the first ban on the use of antibiotic growth promoters in animal nutrition (Hertrampf, 2001) up to now, different alternatives such as exogenous enzymes, organic acids, probiotics, prebiotics, synbiotics, and phytobiotics have been investigated (Lister, 2006). Phytobiotics are natural bioactive compounds with plant origin, mostly in the form of powders or extracts, which their application in feed exert beneficial effects concerning the performance

and well-being of animals (Windisch *et al.*, 2008). Many beneficial effects such as antioxidative, antimicrobial, antiviral, antiparasitic, and anti-inflammatory properties as well as palatability enhancement, digestion motivation, immunomodulation, and metabolism regulation have been reported for phytobiotics. These effects frequently have been ascribed to the phytogetic secondary metabolites (Windisch *et al.*, 2008).

Cumin (*Cuminum cyminum*) is an important herb belonging to the Apiaceae family, originated from the eastern Mediterranean and some Middle East parts of India (Lucchesi *et al.*, 2004). The seeds of cumin are applied as a well-known spice in human nutrition and feed industries (Hajlaoui *et al.*, 2010). Cumin seeds as a whole or ground form as well as its essential oil have long usage in the traditional medicine for the treatment of various diseases particularly digestive disorders (Muthamma *et al.*, 2008). Cuminaldehyde, cymene, and terpenoids are the major active components of cumin (Bettaieb *et al.*, 2011). Antioxidative, antibacterial, antifungal, and anti-inflammatory properties of cumin have been reported in the previous studies (Gachkar *et al.*, 2007; Hajlaoui *et al.*, 2010; Einafshar *et al.*, 2012). It is also indicated that cumin seed consumption could increase appetite, taste perception, and digestive activities of the intestine (Johri, 2011; Mnif and Aifa, 2015).

Black cumin (*Bunium persicum*) is another economically and medicinally important aromatic plant in the Apiaceae family. It is native to West Asia, particularly mountainous regions (Gachkar *et al.*, 2007). Black cumin is also called wild cumin (Hassanzadazar *et al.*, 2018) which refers mostly to non-cultivated varieties. Black cumin is usually used for culinary purposes as a spice and flavoring agent in foods and beverages. Black cumin seeds contain considerable amounts of flavonoids, phenolic acids, and aldehydes; and a high concentration of monoterpenes and sesquiterpenes have been detected in essential oil and extracts of this herb (Chizzola *et al.*, 2014). Anti-inflammatory activity as well as antioxidative, antimicrobial, anti-parasitic, and free radical scavenging effects have been detected for black cumin (Mandegary *et al.*, 2012; Agah *et al.*, 2013). In alternative medicine, black cumin is used as a carminative, diuretic, expectorant, anti-diarrhea, and antispasmodic agent (Miraj and Kiani, 2016).

This study aimed to evaluate the efficacy of cumin and black cumin as phytogetic feed additives in broiler nutrition. For this, characteristics such as growth performance, nutrients digestibility, immune system responses as well as blood parameters and carcass traits have been investigated.

## Materials and Methods

### *Experimental design, diets and bird management*

The animal experimental protocol was prepared according to guidelines for animal care and use of Shahrekord University, Shahrekord, Iran. A total of 240 day-old male broiler chicks (ROSS 308) were obtained from a commercial hatchery. At arrival, chicks were weighed and randomly assigned to 24 deep litter floor pens (1.00 × 1.50 m). A completely randomized design with six treatments and four

replicates was used. Dietary treatments were as follow: a corn-soybean meal-based diet (control), basal diet + 10 ppm avilamycin, basal diet + 0.25% cumin powder, basal diet + 0.75% cumin powder, basal diet + 0.25% black cumin powder, and basal diet + 0.75% black cumin powder. The basal diet for starter (days 1-21) and grower (days 22-42) periods were formulated according to the nutritional requirements of broiler chickens (NRC, 1994; Table 1). Grit was used as inert material in the basal diets and was replaced by the additives at an appropriate level to make dietary treatments. Cumin and black cumin seeds were purchased from a local retailer and were powdered to pass through a 1.0 mm mesh. Birds were reared in an environmentally controlled room. Diets and water were available *ad libitum*. The temperature was held at 30°C for the first week and then was gradually decreased to 22°C by the end of the third week. The light was continuously provided 23 h for the first week and then reduced to 20 h for the remaining period of the experiment. Feed intake (FI) and body weight gain (BWG) were determined weekly and feed conversion ratio (FCR) was calculated. Health status and mortalities were recorded daily during the experimental period.

### *Sample collection*

On day 42, two birds per pen were randomly selected and bled via a brachial vein for biochemical measurements. These birds were then weighted and euthanized by cervical dislocation. The abdominal cavity was opened and organs including the pancreas, liver, spleen, bursa of Fabricius, and also abdominal fat pad were dissected, weighed, and then expressed as a percentage of live body weight. Finally, the whole carcass as well as carcass parts including breast, legs (drumsticks + thighs), and wings were weighed and expressed as a percentage of live body weight.

### *Serum biochemical measurements*

To measure packed cell volume (PCV), hematocrit capillary samples were prepared. Microhematocrit tubes were then centrifuged at 8000 × *g* for 5 min. Packed cell volume was measured using a PCV auto-reader. Blood samples taken on day 42 were centrifuged (3000 × *g*, 10 min) and serum samples were collected and used for biochemical analysis. The concentration of triglycerides (TG), total cholesterol (Chol), high-density lipoprotein cholesterol (HDL-C), and low-density lipoprotein cholesterol (LDL-C) in serum samples were analyzed using a semi-automated biochemical analyzer (Stat Fax® 3300, Awareness Technology, Inc., USA) with appropriate kits (Pars Azmun Inc., Tehran, Iran) according to the kit instructions.

**Table 1.** Composition of the basal diets (as-fed basis)

Item	Starter (days 1-21)	Grower (days 22-42)
Ingredients (g/kg)		
Corn	526.7	593.1
Soybean meal (44% CP)	384.4	322.3
soybean oil	40.0	42.3
Limestone	14.4	14.5
Dicalcium phosphate	15.7	11.4
Salt	4.3	3.1
DL-methionine	2.0	0.8
Vitamin premix <sup>1</sup>	2.5	2.5
Mineral premix <sup>2</sup>	2.5	2.5
Grit <sup>3</sup>	7.5	7.5
Chemical composition		
Metabolizable energy (kcal/kg)	3000	3100
Crude protein (%)	21.55	19.4
Lysine (%)	1.25	1.09
Methionine + Cystine (%)	0.9	0.72
Calcium (%)	1.0	0.9
Available phosphorus (%)	0.45	0.36

<sup>1</sup>Vitamin premix provided per kg of diet: vitamin A (all-trans-retinyl acetate), 2.72 mg; vitamin D<sub>3</sub> (cholecalciferol), 0.05 mg; vitamin E (all-rac- $\alpha$ -tocopherol acetate), 4 mg; vitamin K<sub>3</sub> (menadione), 2 mg; thiamine, 1.8 mg; riboflavin, 6.6 mg; nicotinic acid, 9.8 mg; calcium pantothenate, 29.7 mg; pyridoxine, 1.18 mg; folic acid, 1 mg; cobalamin, 0.015 mg; D-biotin, 0.1 mg; choline chloride, 500 mg.

<sup>2</sup>Mineral premix provided per kg of diet: 76 mg Mn (as MnO<sub>2</sub>); 66 mg Zn (as ZnSO<sub>4</sub>); 40 mg Fe (as FeSO<sub>4</sub>·7H<sub>2</sub>O); 4 mg Cu (as CuSO<sub>4</sub>·5H<sub>2</sub>O); 0.64 mg I (as NaI); 0.2 mg Se (as Na<sub>2</sub>SeO<sub>3</sub>·5H<sub>2</sub>O).

<sup>3</sup>To make dietary treatments, grit was substituted for the herbal additive at the desired level.

### Digestibility of nutrients

For measurement of total tract apparent digestibility (TTAD) of nutrients, chromium oxide (Cr<sub>2</sub>O<sub>3</sub>) was used as an external marker. Briefly, Cr<sub>2</sub>O<sub>3</sub> was added and mixed with experimental diets (0.3%) and fed from 17 to 21 days of age. On day 21, samples of excreta were collected every 6 h (four samples during 24 h) and kept refrigerated (4°C). At the end of day 21, all four samples from each replicate were pooled and a sample was taken and kept at -20°C together with the corresponding feed sample until further analysis. Feed and excreta samples were then analyzed in duplicate for crude protein (CP, method 2001.11) and ether extract (EE, method 920.39) according to the AOAC (2002). Chromic oxide in feed and excreta samples was measured according to the method described by Fenton and Fenton (1979). TTAD of CP and EE were then calculated using the following equation:

$$\text{TTAD (\%)} = 100 - 100 \times \left( \frac{\% \text{ chromium in feed}}{\% \text{ chromium in excreta}} \times \frac{\% \text{ nutrient in excreta}}{\% \text{ nutrient in the feed}} \right)$$

### Antibody-mediated immunity

To evaluate the humoral immune response to dietary treatments, sheep red blood cells (SRBC) was used as an antigen. For this, 2% (v/v) SRBC suspension in

sterile normal saline was injected intramuscularly (2 mL/kg BW) into breast muscle (pectoralis) of two birds from each replicate (eight birds per treatment) on day 28 of age. Seven and 14 days after injection (35 and 42 days of age), blood samples were collected via a brachial vein. Serum samples were then used to measure antibody titer against SRBC using a direct hemagglutination test (Haghighi et al., 2005). The highest serum dilution which was able to agglutinate an equal volume of SRBC suspension was recorded as an anti-SRBC titer and expressed as log<sub>2</sub> of the reciprocal dilution factor.

### Cell-mediated immunity

Cutaneous basophil hypersensitivity (CBH) response was applied to investigate cellular immunity as described by Dibaiee-nia et al. (2017). In a brief, phytohemagglutinin-P (PHA-P) in phosphate-buffered saline (PBS) solution (100 µg/0.1 mL) was injected subcutaneously into the toe web of the right leg of two birds from each replicate. To correct the reaction to BPS alone, PBS was injected into the toe web of the left leg, simultaneously. The thickness of the skin at injection sites was measured 12 and 24 h after injection. Finally, CBH response was calculated by subtracting the thickness of the injection site in the left leg from the thickness of the injection site in the right leg at the corresponding measurement time.

### Statistical analysis

Data were statistically analyzed using the general linear model procedure of the SAS version 9.1 (SAS, 2002). Means comparison was done by the new Duncan multiple range test at  $P < 0.05$  (Duncan, 1955).

### Results

#### Growth performance

The results on FI, BWG, and FCR are shown in Table 2. No significant effect for dietary additives was seen on FI during periods of the experiment ( $P > 0.05$ ). The inclusion of avilamycin increased BWG during starter, grower, and entire (days 1-42) period of the experiment compared to control ( $P < 0.05$ ). Cumin also increased BWG during grower (0.75%) and entire (both levels) periods compared to control ( $P < 0.05$ ), while 0.75% black cumin decreased it ( $P$

$< 0.05$ ). In the starter period, FCR was not changed in the different experimental groups ( $P > 0.05$ ). The addition of avilamycin as well as 0.75% cumin improved FCR during the grower and whole period of the experiment ( $P < 0.05$ ) while 0.75% black cumin deteriorated it during the grower period compared to control ( $P < 0.05$ ).

#### Total tract apparent digestibility of nutrients

As is shown in Table 3, the addition of 0.75% cumin as well as avilamycin increased TTAD of EE, compared to control ( $P < 0.05$ ). The birds fed 0.75% cumin had the highest EE digestibility value (84.5%,  $P=0.012$ ). The addition of avilamycin or 0.75% cumin also improved TTAD of CP compared to the control diet ( $P < 0.05$ ). Supplementation with 0.75% black cumin decreased TTAD of CP in comparison with control diet ( $P < 0.05$ ).

**Table 2.** Broiler growth performance responses during starter (days 1 to 21), grower (days 22 to 42), and overall (days 1 to 42) periods of the experiment.<sup>1</sup>

Item	Control	Avilamycin (10 ppm)	Cumin powder		Black cumin powder		SEM <sup>2</sup>	P-value
			0.25%	0.75%	0.25%	0.75%		
Feed intake (g)								
1 to 21	855	881	872	857	829	887	21.1	0.45
22 to 42	3013	2883	2912	2922	2899	2888	84.4	0.89
1 to 42	3868	3764	3785	3779	3729	3775	79.6	0.88
Weight gain (g)								
1 to 21	555 <sup>bc</sup>	619 <sup>a</sup>	579 <sup>ab</sup>	605 <sup>ab</sup>	521 <sup>c</sup>	562 <sup>bc</sup>	17.7	0.01
22 to 42	1359 <sup>b</sup>	1521 <sup>a</sup>	1475 <sup>ab</sup>	1550 <sup>a</sup>	1372 <sup>b</sup>	1167 <sup>c</sup>	40.2	<0.001
1 to 42	1914 <sup>b</sup>	2140 <sup>a</sup>	2054 <sup>a</sup>	2155 <sup>a</sup>	1893 <sup>b</sup>	1729 <sup>c</sup>	38.8	<0.001
Feed Conversion Ratio								
1 to 21	1.55	1.42	1.51	1.42	1.61	1.58	0.065	0.22
22 to 42	2.21 <sup>b</sup>	1.90 <sup>c</sup>	1.97 <sup>bc</sup>	1.89 <sup>c</sup>	2.13 <sup>bc</sup>	2.49 <sup>a</sup>	0.086	0.001
1 to 42	2.02 <sup>ab</sup>	1.76 <sup>c</sup>	1.84 <sup>bc</sup>	1.75 <sup>c</sup>	1.98 <sup>b</sup>	2.19 <sup>a</sup>	0.062	0.001

<sup>a,b,c</sup> Means in a row not sharing common superscripts are different ( $P < 0.05$ ).

<sup>1</sup>Each value represents the mean of four replicates (ten birds per replicate).

<sup>2</sup>SEM, standard error of the means.

#### Antibody-mediated and cell-mediated immunity

As is depicted in Table 4, antibody titer against SRBC antigen was not affected by dietary treatments on 7 days post-immunization ( $P > 0.05$ ). Supplementation with 0.75% cumin increased anti-

SRBC titer on 14 days post-immunization, compared to control diet ( $P < 0.05$ ). Supplementation of the diet with dietary additives affected CBH response neither at 12 nor at 24 hrs after injection of PHA-P ( $P > 0.05$ , Table 4).

**Table 3.** Total tract apparent digestibility (%) of nutrients in broiler chickens measured on day 21.<sup>1</sup>

Item <sup>2</sup>	Control	Avilamycin (10 ppm)	Cumin powder		Black cumin powder		SEM <sup>3</sup>	P-value
			0.25%	0.75%	0.25%	0.75%		
EE	79.2 <sup>bc</sup>	83.3 <sup>a</sup>	80.3 <sup>b</sup>	84.5 <sup>a</sup>	78.6 <sup>bc</sup>	78.3 <sup>c</sup>	0.56	0.012
CP	62.9 <sup>bc</sup>	66.2 <sup>a</sup>	64.9 <sup>ab</sup>	65.8 <sup>a</sup>	60.9 <sup>dc</sup>	60.0 <sup>d</sup>	0.86	0.006

<sup>a,b,c,d</sup> Means in a row not sharing common superscripts are different ( $P < 0.05$ ).

<sup>1</sup>Each value represents the mean of four replicates (ten birds per replicate).

<sup>2</sup>EE, ether extract; CP, crude protein.

<sup>3</sup>SEM, standard error of the means.

#### Blood biochemistry profile

The effects of dietary additives on PCV and blood biochemistry biomarkers are shown in Table 5. PCV, serum triglycerides, and HDL-C were not affected by

dietary additives ( $P > 0.05$ ). However, cumin at 0.75% inclusion level decreased serum total cholesterol as well as LDL-C when compared to the control ( $P < 0.05$ ).

**Table 4.** SRBC and CBH responses in broiler chickens.<sup>1</sup>

Item <sup>2</sup>	Control	Avilamycin (10 ppm)	Cumin powder		Black cumin powder		SEM <sup>3</sup>	P-value
			0.25%	0.75%	0.25%	0.75%		
SRBC (log <sub>2</sub> RDF)								
7 dpi	4.37	5.25	4.62	5.50	4.87	4.50	0.42	0.34
14 dpi	4.25 <sup>b</sup>	4.87 <sup>ab</sup>	5.25 <sup>ab</sup>	6.25 <sup>a</sup>	5.00 <sup>ab</sup>	4.37 <sup>b</sup>	0.50	0.047
CBH (µm)								
12 hpi	519	672	726	987	848	710	161	0.46
24 hpi	585	546	926	773	518	839	169	0.21

<sup>a,b,c</sup> Means in a row not sharing common superscripts are different ( $P < 0.05$ ).

<sup>1</sup>Each value represents the mean of four replicates (two birds per replicate)

<sup>2</sup>SRBC, sheep red blood cells; RDF, reciprocal dilution factor; dpi, day post-immunization; CBH, cutaneous basophil hypersensitivity; hpi, hours post-injection.

<sup>3</sup>SEM, standard error of the means.

**Table 5.** Blood biochemistry profile (mg/dL) and PCV (%) of broiler chickens measured on day 42.<sup>1</sup>

Item <sup>2</sup>	Control	Avilamycin (10 ppm)	Cumin powder		Black cumin powder		SEM <sup>3</sup>	P-value
			0.25%	0.75%	0.25%	0.75%		
PCV	33.6	32.4	31.4	31.7	31.8	31.6	0.75	0.38
TG	105	103	97	96	105	101	7.98	0.96
Chol	139 <sup>a</sup>	132 <sup>a</sup>	127 <sup>a</sup>	110 <sup>b</sup>	130 <sup>a</sup>	128 <sup>a</sup>	3.68	0.001
HDL-C	49.0	56.0	48.2	41.5	47.5	51.2	4.65	0.42
LDL-C	71.7 <sup>a</sup>	65.0 <sup>ab</sup>	64.5 <sup>ab</sup>	55.2 <sup>b</sup>	68.2 <sup>a</sup>	64.5 <sup>ab</sup>	3.25	0.04

<sup>a,b</sup> Means in a row not sharing common superscripts are different ( $P < 0.05$ ).

<sup>1</sup>Each value represents the mean of four replicates (two birds per replicate).

<sup>2</sup>PCV, packed cell volume; TG, triglycerides; Chol, total cholesterol; HDL-C, high-density lipoprotein cholesterol; LDL-C, low-density lipoprotein cholesterol.

<sup>3</sup>SEM, standard error of the means.

### Carcass traits

As is indicated in Table 6, the effects of dietary additives on carcass traits including breast, legs (drumsticks + thighs), wings, and carcass yields were not significant when compared to control ( $P > 0.05$ ). There was a significant difference between 0.75% cumin and 0.75% black cumin on breast meat yield

(25.8% vs. 23.2%,  $P = 0.05$ ) and total carcass yield (76.1% vs. 73.9%,  $P = 0.03$ ). In other words, cumin powder at the level of 0.75% tended to increase breast meat yield and total carcass yield in comparison to the control diet whereas black cumin powder tended to show a negative effect in this case.

**Table 6.** Breast, legs (drumsticks + thighs), wings, and carcass yields (g/100 g live body weight) of broiler chickens at 42 days of age.<sup>1</sup>

Item	Control	Avilamycin (10 ppm)	Cumin powder		Black cumin powder		SEM <sup>2</sup>	P-value
			0.25%	0.75%	0.25%	0.75%		
Breast	24.6 <sup>abc</sup>	25.5 <sup>ab</sup>	25.1 <sup>abc</sup>	25.8 <sup>a</sup>	23.5 <sup>bc</sup>	23.2 <sup>c</sup>	0.65	0.045
Legs	29.8	28.8	29.5	29.2	28.6	27.8	1.16	0.83
Wings	11.0	11.2	12.9	11.5	10.9	11.4	1.45	0.41
Carcass	74.6 <sup>abc</sup>	76.0 <sup>ab</sup>	74.8 <sup>abc</sup>	76.1 <sup>a</sup>	74.5 <sup>bc</sup>	73.9 <sup>c</sup>	0.47	0.03

<sup>a,b,c</sup> Means in a row not sharing common superscripts are different ( $P < 0.05$ ).

<sup>1</sup>Each value represents the mean of four replicates (two birds per replicate).

<sup>2</sup>SEM, standard error of the means.

### Organs and abdominal fat weights

Although the relative weights of the spleen, bursa of Fabricius, and pancreas were not affected by dietary additives ( $P > 0.05$ ), effects of them on abdominal fat percentage was significant (Table 7). Addition of 0.75% cumin significantly decreased abdominal fat percentage compared to control ( $P < 0.05$ ). As for the liver relative weight, although no significant difference was seen between additives and control diet, the lowest weight belonged to the 0.75% cumin group, whereas the highest weight was seen in black cumin groups (1.24% vs. 1.53%,  $P = 0.05$ ).

### Discussion

This experiment was designed to investigate the effects of cumin and black cumin on growth performance, nutrient digestibility, immune system, and blood parameters in broiler chickens. Despite some evidence on appetite stimulatory effects of phytochemical additives (Johri, 2011), feed intake was not affected in this study by cumin or black cumin, that was consistent with other studies (Alimohamadi et al., 2014; Toriki et al., 2015; Habibi et al., 2016). On the other hand, some reports indicate a FI reduction due to the dietary application of cumin

seeds (Rafeeq *et al.*, 2016; Glamoclija *et al.*, 2017). In this study, supplementation of the diet with cumin powder partially improved BWG and FCR. This result could simply be attributed to the improvement in TTAD of EE and CP as were seen in this study. These data were also confirmed by Alimohamadi *et al.* (2014), Torki *et al.* (2015), Rafeeq *et al.* (2016), and Glamoclija *et al.* (2017). However, it has been indicated that cumin increases food digestibility via

the improvement of gut function and balance of intestinal microflora (Platel and Srinivasan, 2000a,b; Gachkar *et al.*, 2007; Hajlaoui *et al.*, 2010; Johri, 2011). On the other hand, it has been reported that black cumin has antihistaminic activity (Boskabady and Moghadas, 2004) that may negatively influence digestibility via the reduction of gastric secretion. This effect of black cumin may describe its lowering effect on TTAD of CP in this study.

**Table 7.** Organs and abdominal fat relative weights (g/100 g live body weight) of broiler chickens at 42 days of age.<sup>1</sup>

Item	Control	Avilamycin (10 ppm)	Cumin powder		Black cumin powder		SEM <sup>2</sup>	P-value
			0.25%	0.75%	0.25%	0.75%		
Spleen	0.120	0.117	0.100	0.112	0.115	0.117	0.014	0.93
Bursa	0.175	0.145	0.170	0.195	0.140	0.165	0.034	0.55
Pancreas	0.197	0.207	0.217	0.225	0.195	0.227	0.025	0.91
Liver	1.81 <sup>ab</sup>	1.89 <sup>ab</sup>	1.85 <sup>ab</sup>	1.62 <sup>b</sup>	2.18 <sup>a</sup>	2.15 <sup>a</sup>	0.146	0.049
Fat pad	1.42 <sup>ab</sup>	1.47 <sup>ab</sup>	1.35 <sup>bc</sup>	1.24 <sup>c</sup>	1.53 <sup>a</sup>	1.53 <sup>a</sup>	0.049	0.004

<sup>a,b,c</sup> Means in a row not sharing common superscripts are different ( $P < 0.05$ ).

<sup>1</sup>Each value represents the mean of four replicates (two birds per replicate).

<sup>2</sup>SEM, standard error of the means.

In the present study, supplementation of the diet with cumin powder (0.75%) increased antibody titer against SRBC. This immunity response of cumin has also been reported by other studies (Habibi *et al.*, 2016; Nandini *et al.*, 2016), although Aami-Azghadi *et al.* (2010) reported no significant effects of cumin on anti-SRBC titers. However, the Immunomodulatory effects of phytogetic feed additives could be attributed to their antioxidative properties which in turn is related to plants secondary metabolites (Al-Snafi, 2016).

A blood biochemical profile is one of the areas which is affected by medicinal plants. In this study, dietary cumin (0.75%) caused a reduction in serum cholesterol and LDL-C by 20.8% and 23%, respectively. In agreement with our results, Al-Kassi (2010), Torki *et al.* (2015), and Berrama *et al.* (2017) reported a decrease in serum cholesterol and triglycerides when broiler chickens were fed diets containing cumin. It is suggested that the cholesterol-lowering effects of phytogetic products might be associated with the inhibitory properties of these natural products on HMG-CoA reductase which is an allosteric enzyme with a central role in the cholesterol synthesis pathway (El-Dakhkhny *et al.*, 2000; Suganya *et al.*, 2017). Also, it has been demonstrated that competitive inhibitors of HMG-CoA reductase can up-regulate the expression of LDL receptors in the liver which increases the uptake and breakdown of plasma LDL by hepatocytes and eventually results in lower levels of LDL in plasma (Suganya *et al.*, 2017). Moreover, it has been reported that cumin seed acts as a choleric agent (Platel and Srinivasan, 2000b) which in turn could substantially increase the amount of excreted bile and the need for more hepatic bile production using cholesterol.

In this study, the additives comparing to control could not exert a significant effect on carcass traits. However, the highest breast and carcass yields were seen in the 0.75% cumin group while the lowest was seen in the 0.75% black cumin group. This discrepancy mostly could be attributed to the observed differences in CP and EE digestibility in these two groups. Berrama *et al.* (2017) reported that carcass yield was not affected when cumin was added to heat-stressed broiler diets. Similarly, Glamoclija *et al.* (2017) reported no significant effect on carcass, breast, and legs percentage when a phytogetic feed additive (a mixture of cumin, mint, clove, and anise) was added to broiler diets.

The bursa of Fabricius is a primary lymphoid organ with a major role in B cell development and the formation of antibody repertoire in birds (Boehm and Bleul, 2007). Spleen is also a vital organ that plays critical roles in both humoral and cell-mediated immunity responses (Swirski *et al.*, 2009). The relative weight of these organs is usually measured to judge the immune status of birds (pope, 1991). In this study, relative weights of the spleen, bursa of Fabricius, and pancreas were not affected by dietary treatments. Consistent with our results, Aami-Azghadi *et al.* (2010) and Berrama *et al.* (2017) reported no significant effects of cumin on the relative weights of these organs in broilers. On the other hand, there are some reports which indicated a significant increase in the relative weight of spleen, bursa of Fabricius, and thymus when cumin seed (Alimohamadi *et al.*, 2014; Berrama *et al.*, 2017) or cumin essential oil (Habibi *et al.*, 2016) was added to broiler diets.

In the current study, the addition of cumin powder into the diet (0.75%) caused a significant decrease in

the relative weight of liver and abdominal fat. Inconsistent with our result, the inefficacy of cumin seed (Alimohamadi *et al.*, 2014; Berrama *et al.*, 2017) or cumin essential oil (Aami-Azghadi *et al.*, 2010; Habibi *et al.*, 2016) on the reduction of liver and abdominal fat weight has been reported. It has been shown that many dietary spices possess potent inhibitory effects on fatty acid synthase, the enzyme which catalyzes *de novo* synthesis of long-chain fatty acids (Jiang *et al.*, 2015). The liver is the primary site of lipogenesis in avian species. Therefore, lower relative weights of liver and fat pad by dietary cumin may be due to the anti-lipogenic effects of this herb and so, less lipid synthesis and deposition in the liver and peripheral sites such as the abdominal cavity. Furthermore, stimulatory effects of this herb on protein digestion (as indicated in this study) could result in better provision of essential amino acids to body metabolism as well as improving energy to

protein ratio of the diet which both can result in more efficient growth and less lipid deposition in subjected birds.

### Conclusion

This study determined the beneficial effects of cumin in broilers and approved this herb as a potent alternative for in-feed antibiotics. Especially, the digestive stimulating effects and anti-lipogenic activity of cumin were more outstanding in this study. On the other hand, there were not considered positive effects on black cumin. However, these data suggest that cumin may be a useful dietary additive in broilers.

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