P-ISSN: 2304-3075; E-ISSN: 2305-4360
International Journal of Veterinary Science
www.ijvets.com; editor@ijvets.com

Research Article

Effects of Feeding Diets Supplemented with Different Levels of L-Carnitine on Growth Performance, Serum Metabolites, Histopathological Changes in Growing Japanese Quails

Rania E Mahmoud[1], Gehad E Elshopakey[2] and Walaa F Awadin[3]*

[1]Department of Nutrition and Nutritional Deficiency Diseases, Faculty of Veterinary Medicine, MansouraUniversity, Daqahlia, Egypt; [2]Department of Clinical Pathology, Faculty of Veterinary Medicine, Mansoura University, Mansoura, Egypt; [3]Department of Pathology, Faculty of Veterinary Medicine, Mansoura University, Daqahlia, Egypt
***Corresponding author:** ranianutrition@yahoo.com

Article History: 19-623 Received: July 08, 2019 Revised: July 30, 2019 Accepted: August 20, 2019

ABSTRACT

Under normal conditions, the endogenous L-carnitine (LC) synthesis together with its dietary intake is sufficient. However, in cases of increased metabolic rate, as in fasting-growth broilers, the energy demands are elevated and LC availability becomes a limiting factor for oxidative metabolism. In this study, our aim was to investigate the effect of dietary supplementation of LC at different levels on growth performance, carcass traits, blood biochemistry, lipid peroxidation, antioxidant profile and histopathology in growing Japanese quail. Two hundred day-old healthy Japanese quails were fed either basal diet containing no LC (control group) or the basal diet plus 200, 400 and 600mg/kg of LC. There is no significant impact of increasing levels of LC dietary supplementation from 200 to 600mg/kg on growth performance, carcass traits, levels of serum enzymes and albumin in growing quails. However, 600 mg LC/kg supplementation significantly increased levels of serum high density lipoprotein (HDL), total protein, globulin, immunoglobulin-G (IgG), lysozyme, bactericidal activity and tissue catalase (in liver and muscles) and significantly decreased levels of serum triglycerides, very low-density lipoprotein cholesterol (VLDL-C) and tissue malondialdehyde (MDA) (in liver and muscles). Histopathological examination revealed that dietary 600 mg/kg LC increased lymphocytes population in spleen, significantly increased duodenal villous height (VH) and decreased crypt depth (CD) and subsequently increased VH/CD ratio. In conclusion, supplementation of 600mg/kg of LC to diet of growing quails had no significant effect on growth performance, carcass traits however, this level decreased lipid peroxidation, had antioxidant effect and increased immunity.

Key words: L-carnitine, Growth performance, Serum metabolites, Growing quail

INTRODUCTION

There is an increasing need for protein of animal origin intended for human consumption, including meat and eggs. Therefore, farmers and researches search for new approaches to animal production in order to improve the productivity indices of livestock production (Faitarone *et al.,* 2005). The Japanese quail is a small avian species which support minimum space for rearing and rose primarily for meat and egg production. In poultry farms the introduction of Japanese quail has added a new and alternative means to expand the need for meat and eggs (Sreeranjini *et al.,* 2010).

Poultry producers are searching for ways to produce high growth and better feed conversion, as well as to decrease high amount of abdominal and subcutaneous fat deposition (Waller, 2007). In the recent years, L-carnitine (LC) has achieved attention as a potential food additive for raising chicken meat production and also as a substance for increasing physical performance (Zeyner and Harmeyer, 1999). LC is a vitamin-like material that is structurally like to amino acids. It can also be synthesized in vivo from two important amino acids, lysine and methionine in the company of ferrous ions and the vitamins, ascorbate, niacin and pyridoxine (Cave *et al.,* 2008). LC participates in the transport of long-chain fatty acids across the inner mitochondrial membrane from the cytosol to the mitochondrial matrix during lipid catabolism & facilitates the removal of short- and medium-chain fatty acids from the mitochondria that accumulate as a result of normal and abnormal metabolism so plays vital role in fat combustion and energy production (Jalali Haji-Abadi *et al.,* 2010).

Cite This Article as: Mahmoud RE, Elshopakey GE and Awadin WF, 2020. Effects of feeding diets supplemented with different levels of l-carnitine on growth performance, serum metabolites, histopathological changes in growing japanese quails. Int J Vet Sci, 9(1): 16-23. www.ijvets.com

Furthermore, LC increases the activity and levels of antioxidant enzymes such as, superoxide dismutase and glutathione peroxidase in the plasma of poultry through its antioxidant properties. Animal diets with high LC component may have more protein energy ready for growth (Dikel *et al.,* 2010). In addition, LC is used in poultry for several values including growth promoting, increasing the immune system, as an antioxidant and enhance semen quality (GolzarAdabi *et al.,* 2011). LC dietary supplementation enhanced β-oxidation of fatty acids to produce adenosine triphosphate energy and raise utilization of energy. As a consequence, diminish the amount of long-chain fatty acids availability for esterification to triacylglycerols and depot in the adipose tissue (Nouboukpo *et al.,* 2010).

Feedstuffs of plant origin such as cereal grains and their by-products generally contain very little LC compared to of animal origin. However, these plant-based feedstuffs usually constitute the major part of poultry diets, which resulted in deficiency of LC in the diet. Lysine & methionine are the precursors of LC biosynthesis and normally the most vital limiting amino acids in nutrition of poultry (Arslan, 2006).

Diets of quails contain a high amount of cereal grains such as yellow corn, corn glutens and soybean meal which supply low LC contents. Therefore, the present study was conducted to study the effect of supplementation of different levels of LC on growth performance, carcass trait and evaluation of serum biochemical parameters, lipid peroxidation, antioxidant enzymes activities and histopathology of duodenum and spleen in growing Japanese quails reared from 2 week to 5 weeks old age.

MATERIALS AND METHODS

This experiment was reviewed and approved by the nutrition and nutritional deficiency diseases department faculty of veterinary medicine, Mansoura University, Egypt from beginning of February until end of March 2018. It was conducted at the experimental unit related to department. This experimental unit was provided with all equipment necessary to meet the requirements for each experimental group such as light, water and good healthy condition of area.

Quails and dietary treatment

A total of 200-one day-old unsexed Japanese quails were reared till 14 d-old. Thereafter, the quails were randomly divided to one control group and 3 treatment groups each containing five replicate groups of 10 quails. Four isonitrogenous (24%CP) and isocaloric (2900 Kcal ME/kg) experimental diets were formulated by adding three levels of supplemental LC to "basal diets (200, 400, 600 mg/kg)" from 14 to 35 days of age. Water was provided via drinkers and feed was provided through feeders. The experimental diets were formulated on the basis of yellow corn, wheat bran, soybean meal, corn gluten and mixed oil to meet minimum nutrient requirements of growing Japanese quail, as provided by the National Research Council (NRC 1994). Each of the dietary treatments group contained 24% crude protein (CP) and 2900 kcal metabolic energy (ME)/kg (Table 1). The composition of the experimental diets and calculated nutrient content are presented in Table 1. All experimental diets were formulated (in mash form) and water was given ad libitum. All chicks were presented under the same environmental and hygienic conditions. Temperature of house was kept at 33°C and decreased 2°C weekly thereafter.

Performance traits

Quails were weighed at the beginning of the experimental period (day 14) then every week for calculating live weight gains. The average body weight gain (BWG) was calculated per chick in each subgroup every corresponding week. At the end of experiment, final body weight (FBW) was recorded. Feed consumption was recorded weekly and expressed as grams per bird per week and the feed conversion ratio (FCR) was calculated as g of feed per g of live weight gain.

Carcass trait parameters

At the end of experiment 20 quails from each group (4 from each replicate were randomly taken and slaughtered by cutting the neck with a sharp knife. The head, legs and skin were removed. Weights of internal organs were recorded. Carcass was weighed after removal of all internal organs to determine dressing carcass percent.

Samples collection

During slaughter five blood samples per treatment were taken in sterile tubes without anticoagulant, left 10 minutes to clot and centrifuged at 4000 rpm (4°C) for 15minutes for serum separation. The separated serum was stored at -80°C for later analysis. One gram of liver, breast muscle and thigh muscle were collected from each bird. The tissues were washed with ice-cold 0.9% NaCl solution and then homogenized in 9ml ice-cold sterilized PBS (PH 7.5). The homogenates were cold centrifuged for exactly 15 minutes at 3000 rpm and the supernatants were carefully collected to be used for measurement of lipid peroxidation and catalase activities (Ferdandez-Botran*et al.* 2002). Other tissue specimens were collected from duodenum and spleen and preserved in 10% neutral buffered formalin for histopathological examination.

Serum biochemical Parameters

The serum activities of alanine and aspartate aminotransferases (ALT & AST) (Randox, UK), serum total protein and albumin (Stanbio laboratory, USA) were estimated spectrophotometrically according to standard protocol in the enclosed pamphlets. Globulin was calculated by subtraction of serum albumin from total protein and the albumin/globulin (A/G) ratio was calculated.

Serum cholesterol, triglycerides and high-density lipoprotein cholesterol (HDL-C) were determined spectrophotometrically using available commercial kits (Spinreact, Spain). Very Low-density lipoprotein cholesterol (VLDL-C) was calculated from triglyceride by dividing the factor 5. Low-density lipoprotein cholesterol (LDL-C) was calculated using following formula:

LDL cholesterol = Total cholesterol - HDL cholesterol - VLDL cholesterol (Parizadian *et al.,* 2011).

Table 1: The composition of the basal experimental diets (g/ kg) & calculated nutrient content.

Ingredients kg	Diet
Yellow corn	554
Soybean meal	345
Corn gluten	65
Mixed oil	5
Limestone	15
Dicalcium phosphate	7
Min.Vit. premix*	3
Salt	3
Dl-methionine	1
Dl-lysin	2
Total	1000
Calculated nutrient content	
CP	24.16
ME	2905.19
Ca	0.82
P	0.54
Methionine	0.52
Lysine	1.29

*Minerals and vitamins premix each 2.5kg/ ton contained: choline, 300 gm; iron, 80 gm; manganese , 60gm; zinc, 50 gm; cupper, 8 gm; iodine, 400 mg; selenium, 170 mg; cobalt, 100 mg; vitamin A(retinol), 12,000,000 IU; vitamin D3 (cholecalciferol) 2,000,000 IU; vitamin E (tocopherol) 10 mg; vitamin K(phylloquinone) 1g; vitamin B1(thiamin) 2g; vitamin B2 (riboflavin) 5g; vitamin B6(pyridoxine) 1.5g; vitamin B12 (cyanocobalamin) 1g; biotin, 5g; niacin(nicotinic acid) 30gm; folic acid(folacin) 1gm; pantothenic acid, 10gm.

Antioxidant and oxidative stress Parameters

The level of malondialdehyde (MDA) and catalase activity was determined spectrophotometrically (5010, Photometer, BM Co. Germany) in liver, breast and thigh homogenates using commercial test kits (Bio-Chain, Inc., USA).

Immunoglobulins and innate immunity parameters
Immunoglobulins (IgG and IgM)

The level of IgG and IgM in serum was measured according to quantitative turbidimetric provided by Spinreactready-made kits using spectrophotometer (5010, Photometer, BM Co. Germany).

Lysozyme activity

Serum lysozyme activity was measured according to Akinbi *et al.* (2000); based on the lysis of *Micrococcus lysodeikticus* (Sigma Chemical Co), with some modifications. The serum (0.25 ml) was mixed with 0.75 ml *M. lysodeikticus* suspension (0.2 mg/mL in 0.05 M PBS, pH 6.2). The mixture reacted at 25°C for 5 min, and then the optical density (O.D.) was measured at 1 min intervals for 5 min at 540 nm (5010, Photometer, BM Co. Germany). The concentration of serum lysozyme was calculated using a calibration curve constructed using dilutions of lyophilized chicken egg- white lysozyme (Sigma) ranging from 2.0-20.0 µg/ml.

Bactericidal activity

One hundred ml of serum were added to 50 ml bacterial suspension of *E.coli* (1 X 10^8) suspension into duplicate wells of 96 round bottom well microtiter plate, and mixed before incubated for 2.5 h at 37°C. A blank control was also prepared by replacing serum with sterile Hank's Balanced Salt Solution. Fifty ml of diphenyltetrazolium bromide solution (MTT; 2 mg/ml) were added to all wells and incubated for 20 min at room temperature to allow the formation of formazan. Plates were again centrifuged for 10 min at 2000x g. The supernatant was discarded and the precipitate was dissolved in 200 ml dimethylsulfoxide (DMSO). The absorbance of the dissolved formazan was read at 560 nm with a microtiter plate reader (Optica, Mikura Ltd, UK). The bactericidal activity was calculated by subtracting the absorbance of samples from that of control and reported as absorbance units (Kampen *et al.*, 2005).

Histopathological Examination

All tissue specimens were routinely processed until being embedded in paraffin, sectioned at 5 µm thickness and stained with hematoxylin and eosin (H&E) according to Bancroft and Gamble (2007). Images were captured with an Olympus BX41 microscope supplied with a DVC 1300C color digital camera at 10X magnifications. The slides were coded and all measurements were made by one of the authors. Morphometric measurements were done using an image analysis (image J¼ http://imagejen.softonic.com). From each slide, one level of sectioning with the best orientation was chosen. The following parameters were measured: (i) villous height (VH), measured from the tip of the villus to the villus crypt junction, (ii) crypt depth (CD) defined as the depth of the invagination between adjacent villi. VH/CD ratio was calculated by dividing VH by CD (Kettunen *et al.* 2001). Values used for analysis were the means from 10 adjacent, vertically oriented villous-crypt units per section.

Statistical analysis

Statistical analysis was carried out using SPSS 20 to test the outcome of supplementation different levels of LC on growth performance, levels of some blood parameters and duodenal morphometric measurements in growing Japanese quails. One-way ANOVA and Duncan's multiple comparisons were applied to compare means and standard errors. Differences between treatments were considered significant when $P<0.05$.

RESULTS

Growth performance

The data concerned with growth performance of growing quails fed different levels of dietary LC was presented in Table 2. Live body weight (LBW) of growing Japanese quails were significantly improved for quails fed diet supplemented with 200, 400, 600 mg LC/kg (183.85, 204.75, 199, 60) respectively as compared with those fed the control diet had the lowest LBW (163.05) at 35day of age. Similarly, BWG was significantly improved by giving diets supplemented with 200 up to 600 mg LC/kg than the control group during the period of experiment. Generally, BWG was significantly improved by 144.45g, 163.40g, and 160.95g for quails fed diet supplemented with 200, 400 & 600 mg LC/kg; respectively. Meanwhile, quails fed the control diet had the lowest BWG (125.45). The impact of dietary supplementation of varying levels of LC on FCR &feed intake FI of growing Japanese quails was presented in

Table 2. There was significant improvement in FCR in group of growing quails which received diet supplemented with 6oo mg LC/kg from diet. Data concerned with FI revealed that group of growing quails which received diet supplemented with 4oo mg LC/kg from diet had the highest amount of FI compared with other experimental groups.

Carcass traits

The data concerned with dressing and organ percentages (expressed as percentages of LBW) of Japanese quail fed diets supplemented with varying levels of dietary LC throughout the test period are presented in Table 3. Data illustrated that; supplementation of varying levels of dietary LC did not significantly increase in the dressing carcass of quails. There were no significant differences in liver percent between control group and group fed diet supplemented with 600 mg LC/kg from diet. In addition group received 4oo mg LC/kg from diet had the lowest gizzard percent compared with other experimental groups. Finally, group fed diet supplemented with 600 mg LC/kg from diet had the highest heart percent.

Biochemical Parameters

As presented in Table 4, LC supplementation couldn't induce any significant effect on liver enzyme activities (ALT and AST) and serum albumin level (P<0.05). Total protein and globulin levels (P<0.05) were significantly higher in quails group fed 600 mg LC /kg than those of the control group. Albumin /globulin (A/G) ratio (P<0.05) was significantly decreased in quails group fed 600 mg LC /kg when compared the control group.

Table 2: Effect of dietary supplementation of different levels of LC on growth performance of growing Japanese quails.

	Dietary LC levels (mg/kg)			
Parameters	0	200	400	600
Initial BW	37.60± 1.03	39.40± 0.93	38.35± 0.74	38.65± 1.44
FBW(g/quail)	163.05± 6.75^{C}	183.85± 6.36^{b}	204.75 ± 4.83^{a}	199.60± 5.62^{ab}
BWG (g/quail)	125.45± 5.73^{C}	144.45± 5.49^{b}	163.40 ± 4.15^{a}	160.95± 4.37^{a}
FI	560± 5.7^{b}	535± 7.67^{c}	588± 6.8^{a}	540± 5.8^{c}
FCR	4.47± 0.22^{a}	3.71± 0.12^{b}	3.60± 0.09^{bc}	3.36± 0.09^{c}

^{a,b,c} Means in the same row with different superscripts are significantly different (P<0.05).

Table 3: Impact of different dietary LC levels supplementation on carcass trait of growing Japanese quails reared for 35d.

	Dietary LC levels mg/kg)			
Parameters	0	200	400	600
Dressed carcass	72.23± 0.89	71.81± 1.09	71.16± 1.81	71.50± 1.04
Liver %	2.17± 0.11^{a}	1.58 ± 0.06 ^{b}	1.57± 0.14 ^{b}	2.51± 0.28^{a}
Gizzard %	2.41± 0.16^{a}	2.38± 0.10^{a}	1.80± 0.03 ^{b}	2.54± 0.15^{a}
Heart %	1.09± 0.09^{b}	0.94± 0.06^{b}	0.92± 0.08 ^{b}	2.17± 0.11^{a}

^{a,b}Means in the same row with different superscripts are significantly different (P<0.05).

The effect of LC on serum lipids of quails was showed in Table 5. Serum cholesterol level *(*P<0.05) of quails fed diet supplemented with 200 and 400 mg/kg LC was significantly decreased in relation to the control group. Moreover, 600 mg/kg LC significantly elevated HDL and reduced levels of both triglycerides and VLDL (P<0.05) in comparison with control group. LDL level was significantly decreased (P<0.05) in all LC supplemented groups when compared with control group.

Antioxidant and oxidative stress parameters

As shown in **Table 6**, supplementation of quails with 600 mg/kg LC resulted in significant decrease in hepatic MDA level (P<0.05) when compared to control group. Non-significant differences were observed in hepatic catalase levels of all groups. The levels of MDA in breast muscle were significantly reduced (P<0.05) in all LC supplemented groups. Meanwhile, the MDA level in thigh muscle was significantly lower (P<0.05) in 400 and 600 mg/kg LC supplemented quails when compared with the control group. Significant increase in catalase levels (P<0.05) was noticed in both breast and thigh muscles of quail fed diet containing 400 and 600 mg/kg LC.

Immunoglobulins and innate immunity parameters

The serum IgG level was significantly higher (P<0.05) in quails fed with 600 mg/kg LC than in control group (Figure 1A). However, there was no considerable difference in levels of IgM in all groups (Figure 1B).

Table 4: Effect of dietary (LC) supplementation on serum biochemical parameters at 35 d of growing Japanese quails.

	Dietary LC levels (mg/kg)			
Parameters	0	200	400	600
ALT (U/L)	7.25 ± 0.61	6.20± 0.86	6.62± 1.03	6.68 ± 0.7
AST(U/L)	72.14± 3.73	69.20± 1.81	68.48 ± 3.63	68.03± 2.97
Total protein (g/dl)	2.82± 0.16^{b}	3.07± 0.15^{ab}	3.47 ± 0.39 ^{ab}	3.72± 0.26^{a}
Albumin (g/dl)	1.39± 0.10	1.54± 0.15	1.75 ± 0.19	1.56 ± 0.11
Globulin (g/dl)	1.43± 0.15^{b}	1.53± 0.27^{b}	1.72± 0.27^{ab}	2.16± 0.33^{a}
A/G ratio (%)	0.97± 0.14^{a}	1.01± 0.23^{a}	1.02± 0.18^{a}	0.72± 0.15^{b}

Data are expressed as Mean ± SEM (n=5). ^{a,b} Means in the same row with different superscripts are significantly different (P<0.05).

Table 5: Effect of dietary LC supplementation on serum lipid of Japanese quails.

	Dietary LC levels (mg/kg)			
Parameters	0	200	400	600
Cholesterol (mg/dL)	174.66± 6.23^{a}	154.43± 7.33^{b}	160.18± 6.19^{b}	167.08± 8.48^{ab}
Triglyceride (mg/dL)	191.30± 10.99^{a}	206.24± 14.09^{a}	197.46 ± 15.26^{a}	139.46± 16.41^{b}
HDL (mg/dL)	76.98± 4.44^{b}	79.27± 9.26^{ab}	83.78 ± 4.43 ^{ab}	95.98± 5.18^{a}
VLDL (mg/dL)	38.26± 2.19^{a}	41.25± 5.02^{a}	39.49 ± 5.05^{a}	27.89 ± 3.28^{b}
LDL (mg/dL)	59.42± 3.63^{a}	33.92± 4.83^{c}	36.90± 4.79^{c}	43.21± 2.82^{b}

Data are expressed as Mean ± SEM (n=5) ^{a,b,C} Means in the same row with different superscripts are significantly different (P<0.05).

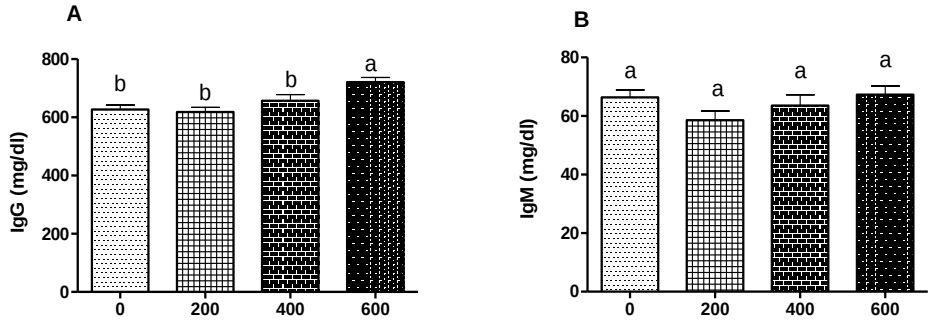

Fig. 1: (A) IgG and (B) IgM levels in serum of Japanese quails fed with diet containing LC (0, 200, 400, 600 mg/kg diet) for 35day. Data are expressed as Mean ± SEM (n=5). The different letters indicate significant difference (P<0.05) among groups.

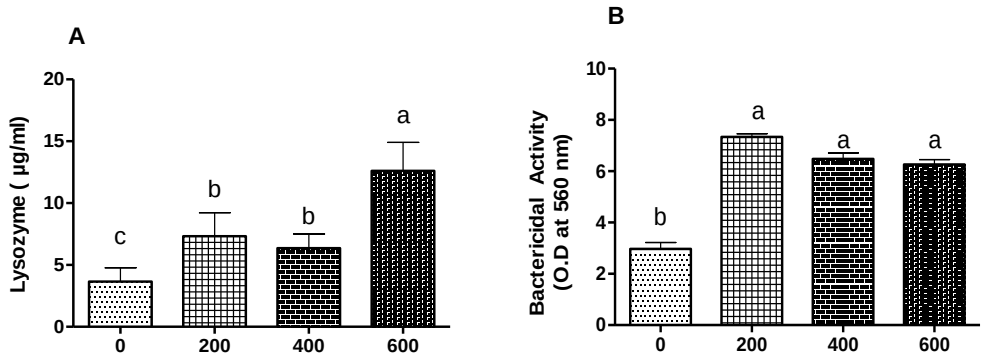

Fig. 2: (A) Serum lysozyme and (B) bactericidal activities of Japanese quails fed with diet containing LC (0, 200, 400, 600 mg/kg diet) for 35 days. Data are expressed as Mean ± SEM (n=5). The different letters indicate significant difference (P<0.05) among groups.

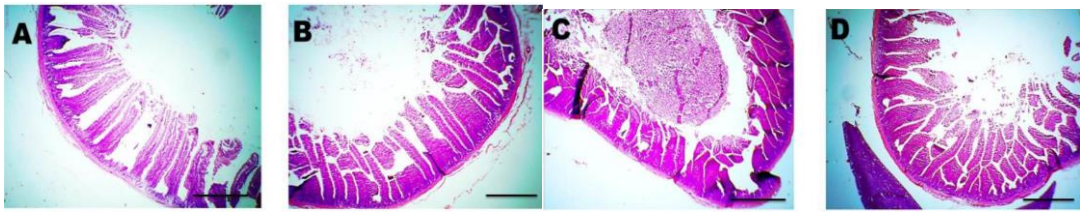

Fig. 3A-D: Microscopic pictures of duodenum in control group (A), group supplemented with 200mg/kg LC (B), group supplemented with 400mg/kg LC (C) and group supplemented with 600mg/kg LC (D). Density of villi increases in LC supplemented groups. H&E X: 40 bar 200.

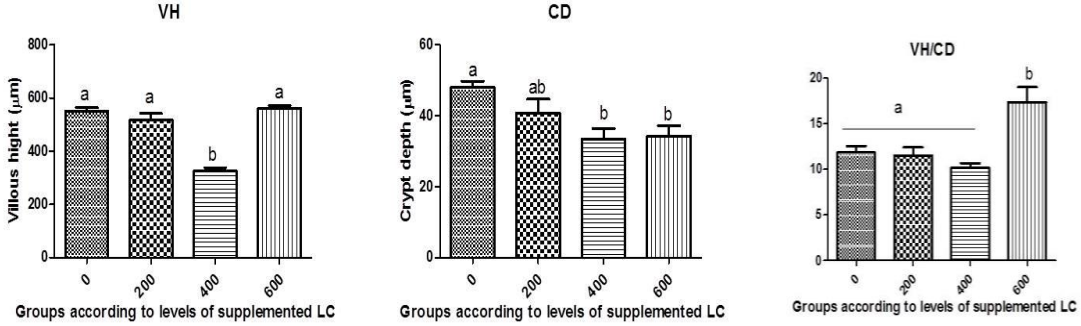

Fig. 4: Statistical analysis shows significantly increased VH and VH/CD with significantly decreased CD of duodenal villi in 600 mg/kg LC fed group. Different small alphabetical letters mean significant when level P<0.05.

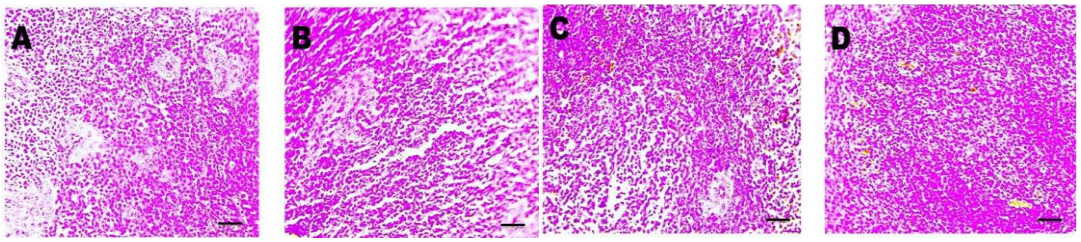

Fig. 5A-D: Microscopic pictures of spleen in control group (A), group supplemented with 200mg/kg LC (B), group supplemented with 400mg/kg LC (C) and group supplemented with 600mg/kg LC (D). Density of lymphocytes increases with 600mg/kg LC in (D) when compared with control group (A). H&E X: 400bar 50.

Table 6: Effect of dietary (LC) supplementation for 35 day on (MDA) and catalase levels in different tissues (liver, breast and thigh muscles) of Japanese quails.

	Dietary LC levels (mg/kg)			
Parameters	0	200	400	600
Liver tissue				
MDA (nmol/g. tissue)	36.24±2.73[a]	35.64±5.47[a]	34.94±4.09[a]	28.78±2.58[b]
Catalase (U/g. tissue)	1.76± 0.20	1.75±0.11	1.71 ± 0.14	1.90±0.06
Breast muscle				
MDA (nmol/g. tissue)	4.98±0.74[a]	2.97 ±0.48[b]	3.13 ± 0.77[b]	2.11± 0.78[c]
Catalase (U/g. tissue)	0.92±0.09[b]	1.06 ±0.06[b]	1.26 ± 0.03[a]	1.28 ± 0.13[a]
Thigh muscle				
MDA (nmol/g. tissue)	5.47±0.96[a]	5.77±0.72[a]	3.29± 0.53[b]	1.72± 0.60[c]
Catalase (U/g. tissue)	0.71±0.06[c]	0.65±0.13[c]	1.09± 0.21[b]	1.51± 0.08[a]

Data are expressed as Mean ± SEM (n=5). [a,b,C] Means in the same row with different superscripts are significantly different (P<0.05).

The serum lysozyme and bactericidal activities were significantly higher (P<0.05) in all LC supplemented quails than those of the control group (Figure 2A-B). The highest lysozyme activity was noticed in 600 mg/kg LC supplemented quail comparing with the other groups (Figure 2A).

Histopathology

Microscopic examination of duodenum and spleen revealed no structural damage in LC supplemented quails. However, densities of villi increased in LC supplemented groups. Significantly increased VH and VH/CD with significantly decreased CD of duodenal villi were statistically recorded in 600 mg/kg LC fed group (Figures 3&4). Lymphocytes population in spleen also increased in 600 mg/kg LC fed group when compared with other groups (Figure 5).

DISCUSSION

Our results suggested that the growing Japanese quails fed diet supplemented with 400 or 600 mg LC/kg diet recorded higher LBW and BWG values than the control in agreement with Taklimi *et al.* (2015); Awad *et al.* (2016). The improvements in LBW & BWG may be attributed to LC performs an important role in oxidation of mitochondrial long chain fatty acids and releasing of energy and improved the growth performance (Neuman *et al.* 2002). In addition, LC participates in increasing plasma insulin-like growth factor-I concentration, which serves as stimulating substances for chick's growth (Kita *et al.* 2005).

It was known that the improvement of FCR was associated with the decrease in FI and the increase in BWG of broiler chicks (El-Kelawy and Asmaa 2017).Our results showed that FCR was significantly improved by 3.71, 3.60 & 3.36 for growing quails fed diets supplemented with 200, 400 and 600 mg LC/kg than those fed the basal control diet in agreement with Abdel Fattah *et al.* (2014). This improvement may be related to enhancement of LC to burning of fatty acids which thus lowering calorie requirements, as well as, improve intestinal mucous membrane by active and passive mechanism (Fathi and Farahzadi 2014). Although, Xu et al. (2003) concluded that supplementation of dietary LC to commercial male broilers at different levels had no significant effect on FCR. Quails supplemented with 600 mg/kg LC showed decrease in FI. The reduction in FI returned to the ability of birds to compensate their FI according to their energy requirements since the experimental diets had similar metabolizable energy ME Awad *et al.* (2016). On the contrary, it was noticed that dietary LC (900 mg/kg diet) supplementation did not affect feed consumption in broilers during growing period (0-6 weeks) Murali *et al.* (2015).

Our results showed that the carcass traits were not significantly improved in Japanese quail fed on diet contained 200 ppm LC. Dressing carcass percentage significantly increased with increasing the inclusion levels of LC in broiler diets Oladele *et al.* (2011) noticed that. There was non-significant increase in liver and gizzard percentage between the control group and group received 600mg/kg LC. The enlargement in these organs may be attributed to hyperactivity where LC appeared to be extracted from the portal circulation into the systemic circulation by the liver after being traversed the mucosal intestinal membrane by active and passive transport mechanisms (Harris *et al.*, 1995). The highest heart percentage was achieved in group supplemented with 600mg/kg in agreement with Buyse *et al.* (2001). This may be attributed to appositive response to faster rate of metabolism for synthesis protein, glucose, cholesterol & LC (Harmeyer, 2002). However, Koksal *et al.* (2011) observed a reduction in heart and liver weights was shown in broiler diets supplemented with 100 ppm LC.

In the present study, dietary supplementation of LC for 35 days did not induce any significant change on liver enzymes activity (ALT and AST) and serum albumin level. Similar result has been reported by Kazemi-Fard *et al.* (2015) who found that LC did not affect ALT and AST activities of laying hens supplemented 50, 100 and 150 mg/kg LC in diets referring that to the protecting activity of it particularly in the liver parenchyma.

Meanwhile, dietary supplementation of LC to the Japanese quail significantly increased serum levels of total protein and globulin in accordance with Abedpour *et al.* (2017). The higher concentration of total protein and globulin levels in supplemented groups may be connected to protein sparing action of LC and reduced consuming amino acid precursor (lysine and methionine) for LC biosynthesis (Jalali Haji-Abadi *et al.* 2010).

In our study LC supplementation (200 and 400 mg/kg) decreased blood cholesterol level as been clarified by Parsaeimehr *et al.* (2012). The reduction of serum total cholesterol was mostly attained via a decrease of cholesteryl esters rather than by a decrease in free cholesterol. Moreover, it may be caused by modified repartition of whole body cholesterol, increased the

transformation of cholesterol to bile acids or rise excretion of biliary sterol (Arslan, 2006).

On other hand, dietary supplementation of Japanese quails with 600 mg/kg LC significantly reduced serum levels of triglycerides in agreement with Xu *et al.* (2003). The decrease of serum triglycerides may be due to the ability of LC to increase fatty acids oxidation by enhancing the transportation capacity of fatty acids to inner mitochondrial membrane and hence reduce serum non-esterified fatty acid and triacylglycerol contents (Shuenn *et al.,* 2012). Also, it may be due to the ability of LC to increase the action of lipase and reduce action of lipoprotein lipase, as a result of that leading to elevation concentration of fatty acid in serum by increasing hydrolysis of triglycerides to glycerol and fatty acid (Griffin and Whitehead 1982). Zhang *et al.* (2010) found that LC supplementation pick up lipid metabolism in broilers and decrease lipoprotein lipase activity which signified an increased hydrolysis of VLDL. This explained the decrease in VLDL and LDL levels and increase HDL level via LC supplementation as reported by Parsaeimehr *et al.* (2012).

Diets supplemented with LC decrease MDA level and increase catalase activity in hepatic tissue, breast and thigh muscles, especially with the higher dose (600 mg/ kg). This may be attributed to the free radical scavenging properties of LC (Packer *et al.* 1991). Similar results were demonstrated by Cao *et al.* (2011); Wang *et al.* (2013).

In our study, the serum level of IgG, lysozyme and bactericidal activities exhibited significant elevation in Japanese quail fed LC containing diets, especially at dose 600 mg/kg. Several previous studies proved that LC had a positive effect in enhancing the humoral immune response as mentioned by El-Kelawy and Asmaa. (2017).

Microscopic examination displayed an increased density; VH and VH/CD with decreased CD of duodenal villi in 600mg/kg LC supplemented quails. VH and CD are reflected on as a sign of good working intestine. Higher villus height produces a large surface area for absorption of nutrients and consequently performance enhancement (Awad *et al.,* 2008). In spleen, lymphocytes density increased in quails fed 600 mg/kg LC. Dietary LC supplementation significantly increased total WBC and lymphocyte counts in broilers as mentioned by Karadeniz *et al.* (2008). LC also inhibited apoptosis of lymphocytes and enhanced the proliferative response of human lymphocytes to mitogens (De Simone *et al.,* 1994). The positive effects of LC on inflammatory and immune cells were confirmed by Famularo and De Simone (1995).

Conclusions

Dietary supplementation of 600mg/kg LC to quails had no significant effect on growth performance, carcass traits but increased immunity, decreased lipid peroxidation level and exerted an antioxidant effect.

Abbreviations

LC: L-carnitine, FBW: final body weight, BWG: body weight gain, LBW: live body weight, DM: dry matter, OM: organic matter, CP: crude protein, ME: metabolizable energy, EE: ether extract, MDA: malondialdehyde, IgG immunoglobulin-G, A/G: albumin/globulin ratio, ALT: alanine aminotransferases, AST: aspartate aminotransferases, VH: villous height, CD: crypt depth, HDL-C: high-density lipoprotein cholesterol, VLDL-C: very low density lipoprotein cholesterol, LDL cholesterol: low density lipoprotein cholesterol.

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