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The effect of dietary L-carnitine and fat on performance, carcass traits and blood components in broiler chickens*

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The poultry industry has traditionally been selecting animals for improved performance without consideration for the effect on fat deposition. Dietary L-carnitine can alter lipid metabolism; nevertheless, when combined with fat, the effects are not clear. This study shows the effect of different dietary levels of L-carnitine (0, 200 and 400 mg/kg) and fat (0, 2.5 and 5%) on growth performance and slaughter traits of commercial broilers (Ross 308; n=270). The groups received the following dietary treatments: 1) 0 mg/kg L-carnitine + 0% fat, 2) 200 mg/kg L-carnitine + 0% fat, 3) 400 mg/kg L-carnitine + 0% fat, 4) 0 mg/kg L-carnitine + 2.5% fat, 5) 200 mg/kg L-carnitine + 2.5% fat, 6) 400 mg/kg L-carnitine + 2.5% fat, 7) 0 mg/kg L-carnitine + 5.0% fat, 8) 200 mg/kg L-carnitine + 5.0% fat, and 9) 400 mg/kg L-carnitine + 5.0% fat. Feed conversion ratio, growth performance, blood biochemical parameters, carcass traits and body composition were measured and analyzed. Levels

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of fat with L-carnitine had significant effects on the European Performance Efficiency Factor, wings weight, intestine length and weight, spleen and liver weight, full abdomen carcass and abdominal fat weight, as well as serum triglyceride levels. Dietary L-carnitine supplementation improved growth performance of broilers, thus it may be a promising solution to reduce fat storage in broilers and improve the quality of carcasses intended for human consumption.

KEY WORDS: blood lipids / carcass traits / fat / L-carnitine / performance / poultry

Over the last 50 years different studies have been conducted to determine the influence of diet supplementation on carcass composition and more recently, also on blood parameters [Pearson and Dutson 2013].The poultry industry has traditionally been selecting animals for improved performance (high growth rate, body weight, feed efficiency) without considering the effect on fat deposition. The intramuscular lipid accumulation affects mainly the abdominal adipose tissue, considered by the broiler industry a product of low economic value, which resulted in a re-evaluation of the improvement strategies incorporating new dietary technologies [Fouad *et al.* 2013]. L-carnitine (3-hydroxy-4-N-trimethylammoniobutanoate) is endogenously biosynthesized from lysine and methionine [Buyse *et al.* 2001]. Although its relative concentration in cells (in the form of free carnitine or acylcarnitines) is high, an exogenous dietary intake is needed. L-carnitine in the form of acylcarnitines facilitates the transport of fatty acids into the mitochondrial matrix [Furuno *et al.* 2001].The concept that dietary L-carnitine reduces cholesterol and triglyceride levels has been supported by the study of Wang *et al.* [2013], demonstrating the moderating influence of altering lipid metabolism on body fat. The dietary fat and L-carnitine combination effects on lipid metabolism and growth development of broiler chickens are not clear. Therefore, the objective of this study was to investigate the effects of dietary L-carnitine and fat on the performance, carcass characteristics and blood parameters in broiler chicks.

Material and methods

Animals

A total of 270 one-day old mixed Ross 308 chicks (weighing 44.0±1.5 g) were divided into 27 groups of 10 animals each. All birds were fed iso-caloric and iso-nitrogenous diets for 42 days (see the composition and nutritive values in Tab. 1). Furthermore, different L-carnitine (0, 200 and 400 mg/kg) and fat (corn oil 0, 2.5 and 5%) concentrations were added to the daily diet. The treatment groups (each treatment had three replicated pens) were as follows:

Treatment 1 (control) – 0% fat + 0 mg/kg L-carnitine, Treatment 2 – 0% fat + 200 mg/kg L-carnitine, Treatment 3 – 0% fat + 400 mg/kg L-carnitine, Treatment 4 – 2.5% fat + 0 mg/kg L-carnitine, Treatment 5 – 2.5% fat + 200 mg/kg L-carnitine, Treatment 6 – 2.5% fat + 400 mg/kg L-carnitine, Treatment 7 – 5% fat + 0 mg/kg L-carnitine, Treatment 8 – 5% fat + 200 mg/kg L-carnitine, Treatment 9 – 5% fat + 400 mg/kg L-carnitine.

Table 1. Feed ingredients and nutrient analysis of diets used during the starter (1^{st}-14^{th} days of age), grower (15^{th}-28^{th} days of age) and finisher periods (29^{th}-42^{nd} days of age)

Ingredient (g/kg)	Starter period	Finisher period	Finisher period
Feed ingredients			
corn	558.5	624	630
soybean meal	375.5	320.5	302.5
soybean oil	20	20	20
monocalcium phosphate ($CaH_4P_2O_8$)	17	11	13
$CaCO_3$	11.3	13	9
Mineral mixture[1]	3	2.5	3
Vitamin mixture[2]	3	2.5	3
Vitamin K3	1	1	1
Vitamin E	1	1	0.5
DL-Methionine	3.3	1.8	1.7
Lysine-hydro-chloride	2.2	1	1
NaCl	1.9	2	2.5
sodium bicarbonate ($NaHCO_3$)	2.5	1.5	1.5
multi-enzyme	0.35	0.35	0.35
phytase	0.1	0.1	0.1
total	100	100	100
Nutrient analysis			
metabolizable energy (kcal/kg)	2930	3050	3100
crude protein (%)	23	21.5	20
crude fiber (%)	3	3	2.90
calcium (%)	1	0.86	0.80
available phosphorus (%)	0.5	0.43	0.40
sodium (%)	0.16	0.16	0.16
lysine (%)	1.28	1.1	1.00
methionine (%)	0.58	0.45	0.45
Ca:P	2	2	2.00
linoleic acid (%)	1	1	1.00
potassium (%)	0.8	0.8	0.8
methionine + cysteine (%)	0.93	0.77	0.75
chlorine (%)	0.16	0.15	0.15

[1]Calcium pantothenate – 4 mg/g; niacin – 15 mg/g; Vitamin B6 – 13 mg/g; Cu – 3 mg/g; Zn – 15 mg/g; Mn – 20 mg/g; Fe – 10 mg/g; K – 0.3 mg/g.
[2]Vitamin A – 5000 IU/g; Vitamin D3 – 500 IU/g; Vitamin E – 3 mg/g; Vitamin K3 – 1.5 mg/g; Vitamin B2 – 1 mg/g.

Management of experimental birds

On the first day of age all birds were weighed individually and then randomly assigned to 27 floor pens. During the first three weeks of rearing the room temperature was set at 33°C in the first days, dropped to 30°C in the successive days of the first week, and subsequently lowered gradually by 2.8°C every week until 20°C was reached. Room temperature was monitored by three thermometers placed in the middle and two ends of the broiler house. The birds were kept under a 23-hour light regimen throughout the study period. Feed and water were provided *ad libitum*, during

the first week in feeder trays and conical drinkers, respectively. During the rest of the rearing period, cylindrical feeders and drinkers were used. The birds were vaccinated on the 1st day of age against infectious bronchitis and avian influenza, on the 1st, 8th and 18th day of age against Newcastle disease and on the 14th and 22nd days of age against Gumboro's disease. Body weight and feed intake were measured weekly. Feed conversion ratio (FCR) was calculated by dividing total feed consumption by total body weight gain (g/g bwg) and the European production efficiency factor (EPEF) was calculated weekly using the following equation: EPEF = Daily weight gain (g or kg) × Survival rate/10 × FCR.

On day 42 one representative chick per group was sacrificed. The carcass was then processed into edible parts and non-edible parts. Feet were separated from the carcass at the tibiotarsal joint. The neck, wingtips, gut and liver were removed and the empty or edible carcass was weighed.

Blood collection and analysis

On day 42 one bird with a weight close to the mean was selected, a 5ml blood sample was taken from the wing vein and held for 12 hours at room temperature, centrifuged at 5,000 rpm for 3 minutes (before serum separation). Serum was stored at -20˚C until biochemical analysis. Analysis was performed using Pars Azmoon commercial kits in an autoanalyzer (Hitachi 917, Japan). The levels of direct and total bilirubin, alkaline phosphatase, aspartate amino transferase (S.G.O.T) (EC 2.6.1.1), creatinine, blood urea nitrogen, low-density lipoprotein, high-density lipoprotein, triglycerides, total cholesterol and glucose were measured.

Statistical analysis

The data were analyzed using a general linear model (SAS Institute Inc. 2000., SAS Online Doc., SAS Institute Inc., Cary, NC.), which is robust enough to allow for the moderately imbalanced data from these experiments. The model included L-carnitine and fat as the main effects. A 3×3 factorial design was used to analyze the data. The interaction between the main effects was included in the model. Group differences were tested with Duncan's test. The significance level was set at $P<0.05$. The model used was:

$$y_{ij} = \mu + A_i + B_j + (AB)_{ij} + e_{ijk}$$

where:

μ – the overall mean;

A_i – the fixed effect of L-carnitine;

B_j – the fixed effect of fat;

$(AB)_{ij}$ – the fixed effect of interaction *A* by *B;*

e_{ijk} – the random error.

Before the statistical analysis of data, all data were tested by Shapiro-Wilk's and Levene's tests for normality and homogeneity of variance, respectively.

Results and discussion

Main effects

The effects of dietary fat on the growth performance of six-week old broilers are summarized in Table 2. The dietary supplementation of 5% fat decreased the feed conversion ratio (FCR, $P<0.05$) and increased the European Production Efficiency Factor (EPEF, $P<0.05$), while it had no effect on body weight (BW) when compared to the controls. Of all the analyzed carcass components (Tab. 4), the weight of abdominal fat was greater in broilers fed 5% supplemental fat compared to the controls, similarly as it was for full abdomen carcass weight ($P<0.05$) compared to birds fed the low-fat diet (2.5% fat). Weight of other carcass components as well as the spleen (Tab. 3) was not influenced markedly by the fat supplementation diets. The effect of fat rich diets on gastrointestinal components and organs is shown in Table 5. Fat rich diets (2.5% and 5%) increased the intestine weight ($P<0.05$) compared to the control birds. The low-fat diet increased the intestine weight, while it decreased liver weight (both $P<0.05$, Tab. 5). Other parameters as well as blood biochemistry parameters (Tab. 6) were not significantly affected by fat rich diets. High fat diets used in broiler nutrition to improve their BW lead to increased carcass fat deposition. In this study the high-fat diet (5% fat) had no effect on BW even though it improved FCR, which is in line with the study of Rezaei *et al.* [2007] in broiler chicks fed 5% supplemental fat. In turn, Burlikowska *et al.* [2010] found no effect of the diet fortified with lard on the BW gain and FCR.

The EPEF is used to evaluate growth performance of broilers [Kryeziu *et al.* 2018]. In the present study, only feeding the high-fat diet resulted in increased EPEF, which indicated a uniform BW gain as well as good health of birds [Bhamare *et al.* 2016]. The intestine, full abdomen carcass and abdominal fat weights were found to increase after feeding the 5% supplemental fat. The deposition of body fat depends on the net balance between fat absorption, synthesis and catabolism [Mohammad and Horniaková 2012]. In another study, dietary fat did not alter blood parameters in broilers fed with lard [Burlikowska *et al.* 2010].

Of the investigated carcass components (Tab. 4), only the neck and abdominal fat weights (both $P<0.05$) were decreased in broilers fed a diet supplemented with L-carnitine compared to the controls. Growth performance (Tab. 2), spleen weight (Tab. 3), gastrointestinal component and organ weights (Table 5) and blood serum biochemical indices (Tab. 6) were not altered markedly. Endogenous L-carnitine supports energy metabolism by mobilizing fat from body reserves [Rabie *et al.* 1997]. Supplemental L-carnitine (200 and 400 mg/kg) decreased the weight of the neck and abdominal fat, while no other carcass or blood parameters and growth performance of broilers were affected markedly. Our observations are in agreement with those of Wang *et al.* [2013], who found no effect of dietary L-carnitine on broiler performance, growth rate or feed efficiency, except for triglyceride mobilization properties. In contrast, Rabie and Szilágyi [1998] found a clear positive effect of L-carnitine on

Table 2. Growth performance means and standard errors (in parentheses) of Ross 308 male broilers fed different levels of dietary fat and L-carnitine in the first six weeks of age

Main effects	Fat (%)			L-carnitine (mg/kg)		
	0	2.5	5	0	200	400
FCR (g/g bwg)	1.83^{a}(0.04)	1.78^{ab}(0.10)	1.73^{b}(0.07)	1.77^{a}(0.08)	1.78^{a}(0.10)	1.79^{a}0.08)
Body weight (kg)	2.36^{a}(0.07)	2.41^{a}(0.13)	2.46^{a}(0.09)	2.44^{a}(0.08)	2.42^{a}(0.11)	2.37^{a}(0.12)
EPEF	308.00^{b}(13.47)	324.51^{ab}(42.35)	339.99^{a}(17.77)	330.32^{a}(22.87)	325.93^{a}(32.18)	315.64^{a}(30.15)

Interaction effects	T1	T2	T3	T4	T5	T6	T7	T8	T9
FCR (g/g bwg)	1.83^{a}(0.05)	1.80^{a}(0.03)	1.86^{a}(0.00)	1.73^{a}(0.05)	1.83^{a}(0.14)	1.76^{a}(0.12)	1.74^{a}(0.10)	1.70^{a}(0.07)	1.74^{a}(0.07)
body weight (kg)	2.42^{a}(0.07)	2.37^{a}(0.02)	2.28^{a}(0.04)	2.47^{a}(0.06)	2.41^{a}(0.19)	2.36^{a}(0.14)	2.43^{a}(0.12)	2.49^{a}(0.07)	2.46^{a}(0.09)
EPEF	318.40^{ab}(13.47)	313.16^{ab}(8.69)	292.42^{b}(6.54)	339.85^{ab}(15.73)	315.94^{ab}(46.65)	317.74^{ab}(42.35)	332.70^{ab}(36.14)	348.69^{a}(26.83	336.76^{ab}(17.77)

FCR – feed conversion ration; bwg – body weight gain; EPEF – European Production Efficiency Factor; T – treatment group;T1 (control) – 0% fat + 0 mg/kg L-carnitine; T2 – 0% fat + 200 mg/kg L-carnitine; T3 – 0% fat + 400 mg/kg L-carnitine; T4 – 2.5% fat + 0 mg/kg L-carnitine; T5 – 2.5% fat + 200 mg/kg L-carnitine; T6 – 2.5% fat + 400 mg/kg L-carnitine; T7 – 5% fat + 0 mg/kg L-carnitine; T8 – 5% fat + 200 mg/kg L-carnitine; T9 – 5% fat + 400 mg/kg L-carnitine.
^{ab}Means with different superscripts within each row of dietary treatments differ significantly at $P<0.05$.

Table 3. Immune organ means and standard errors (in parentheses) of Ross 308 male broilers fed different levels of dietary fat and L-carnitine in the first six weeks of age

Main effects	Fat (%)			L-carnitine (mg/kg)		
Organ weight (g)	0	2.5	5	0	200	400
Spleen	2.22^{a}(0.57)	2.55^{a}(1.00)	2.22^{a}(0.57)	2.55^{a}(0.52)	1.89^{a}(0.60)	2.55^{a}(0.72)

Interaction effects	T1	T2	T3	T4	T5	T6	T7	T8	T9
Spleen	2.33^{ab}(0.57)	2.00^{ab}(0.00)	2.33^{ab}(0.57)	2.66^{ab}(0.57)	2.00^{ab}(1.00)	3.00^{a}(1.00)	2.66^{ab}(0.57)	1.66^{b}(0.57)	2.33^{ab}(0.57)

T1 (control) – 0% fat + 0 mg/kg L-carnitine; T2 – 0% fat + 200 mg/kg L-carnitine; T3 – 0% fat + 400 mg/kg L-carnitine; T4 – 2.5% fat + 0 mg/kg L-carnitine; T5 – 2.5% fat + 200 mg/kg L-carnitine; T6 – 2.5% fat + 400 mg/kg L-carnitine; T7 – 5% fat + 0 mg/kg L-carnitine; T8 – 5% fat + 200 mg/kg L-carnitine; T9 – 5% fat + 400 mg/kg L-carnitine.
^{ab}Means with different superscripts within each row of dietary treatments differ significantly at $P<0.05$.

Table 4. Carcass component means and standard errors (in parentheses) of Ross 308 male broilers fed different levels of dietary fat and L-carnitine in the first six weeks of age

Main effects	Fat (%)			L-carnitine (mg/kg)		
Organ weight (g)	0	2.5	5	0	200	400
Drumsticks	633.11^{a}(109.57)	660.44^{a}(57.93)	659.44^{a}(62.95)	668.33^{a}(66.92)	622.11^{a}(74.39)	622.56^{a}(76.21)
Neck	53.00^{a}(7.00)	53.88^{a}(4.35)	55.88^{a}(3.60)	58.66^{a}(5.29)	52.22^{b}(6.61)	51.88^{b}(3.85)
Wings	122.77^{a}(26.53)	124.56^{a}(9.81)	128.00^{a}(17.38)	127.00^{a}(13.87)	118.00^{a}(14.60)	130.33^{a}(14.98)
Full abdomen carcass	1.96^{ab}(0.19)	1.78^{b}(0.53)	2.17^{a}(0.29)	1.90^{a}(0.35)	1.928^{a}(0.43)	2.01^{a}(0.46)
Breast	687.00^{a}(57.51)	698.33^{a}(38.00)	705.44^{a}(3.60)	704.44^{a}(62.13)	656.11^{a}(106.36)	730.22^{a}(46.81)
Abdominal fat	33.11^{b}(14.27)	45.55^{a}(14.16)	51.55^{a}(12.08)	51.55^{a}(9.24)	44.66^{ab}(14.16)	34.00^{b}(16.95)

Interaction Effects	T1	T2	T3	T4	T5	T6	T7	T8	T9
Drumsticks	642.00^{a}(109.57)	660.00^{a}(50.26)	597.33^{a}(75.79)	694.66^{a}(48.18)	577.33^{a}(61.37)	709.33^{a}(57.93)	668.33^{a}(38.83)	629.00^{a}(103.01)	681.00^{a}(62.95)
Neck	57.00^{a}(7.00)	49.33^{a}(4.16)	52.66^{a}(5.03)	59.66^{a}(6.65)	51.00^{a}(8.71)	51.00^{a}(4.35)	59.33^{a}(3.51)	56.33^{a}(6.42)	52.0^{a}(3.60)
Wings	131.66^{ab}(26.53)	120.66^{ab}(16.77)	116.00^{ab}(2.64)	124.00^{ab}(3.60)	109.00^{b}(11.26)	140.66^{a}(9.81)	125.33^{ab}(1.52)	124.33^{ab}(15.88)	134.33^{ab}(17.38)
Full abdomen carcass	1.96^{ab}(0.19)	2.09^{ab}(0.69)	1.82^{ab}(0.12)	1.63^{b}(0.41)	1.74^{ab}(0.22)	1.74^{ab}(0.53)	2.09^{ab}(0.36)	1.95^{ab}(0.37)	2.47^{a}(0.29)
Breast	704.33^{a}(57.51)	665.00^{a}(152.33)	691.66^{a}(62.64)	708.66^{a}(93.55)	640.66^{a}(129.24)	745.66^{a}(38.00)	700.33^{a}(57.72)	662.66^{a}(68.06)	753.33^{a}(3.05)
Abdominal fat	48.33^{ab}(4.72)	34.66^{bc}(4.16)	16.33^{c}(1.52)	49.33^{ab}(7.50)	51.66^{ab}(22.67)	35.66^{abc}(2.88)	57.00^{a}(14.00)	42.66^{ab}(5.85)	50.00^{ab}(16.82)

T1 (control) – 0% fat + 0 mg/kg L-carnitine; T2 – 0% fat + 200 mg/kg L-carnitine; T3 – 0% fat + 400 mg/kg L-carnitine; T4 – 2.5% fat + 0 mg/kg L-carnitine; T5 – 2.5% fat + 200 mg/kg L-carnitine; T6 – 2.5% fat + 400 mg/kg L-carnitine; T7 – 5% fat + 0 mg/kg L-carnitine; T8 – 5% fat + 200 mg/kg L-carnitine; T9 – 5% fat + 400 mg/kg L-carnitine.
^{abc}Means with different superscripts within each row of dietary treatments differ significantly at $P<0.05$.

Table 5. Gastrointestinal component and organ means and standard errors (in parentheses) of Ross 308 male broilers fed different levels of dietary fat and L-carnitine in the first six weeks of age

Main effects	Fat (%)			L-carnitine (mg/kg)		
	0	2.5	5	0	200	400
Intestine length (cm)	2.20^{a}(0.15)	2.29^{a}(0.20)	2.34^{a}(0.04)	2.21^{a}(0.14)	2.33^{a}(0.23)	2.28^{a}(0.12)
Intestine weight (g)	94.11^{b}(11.01)	112.44^{a}(12.12)	112.44^{a}(2.51)	104.88^{a}(12.49)	108.66^{a}(25.75)	115.44^{a}(16.21)
Heart weight (g)	10.88^{a}(2.64)	11.22^{a}(1.00)	11.33^{a}(1.52)	11.66^{a}(1.50)	10.33^{a}(2.54)	11.44^{a}(1.13)
Liver weight (g)	54.11^{a}(9.64)	53.22^{a}(2.64)	47.2^{a}(0.57)	51.22^{a}(10.33)	51.00^{a}(7.14)	52.33^{a}(5.78)
Gizzard weight (g)	35.88^{a}(2.64)	37.11^{a}(1.52)	37.77^{a}(6.42)	38.00^{a}(1.93)	35.33^{a}(3.57)	37.44^{a}(4.36)

Interaction effects	T1	T2	T3	T4	T5	T6	T7	T8	T9
Intestine length (cm)	2.14^{ab}(0.15)	2.13^{b}(0.11)	2.32^{ab}(0.08)	2.25^{ab}(0.19)	2.41^{ab}(0.29)	2.21^{ab}(0.20)	2.24^{ab}(0.10)	2.46^{a}(0.15)	2.32^{ab}(0.04)
Intestine weight (g)	93.33^{c}(11.01)	88.33^{c}(6.80)	100.66^{bc}(7.37)	113.33^{abc}(11.93)	112.00^{abc}(27.87)	112.00^{abc}(12.12)	108.00^{abc}(64.24)	125.66^{ab}(27.57)	133.66^{a}(2.51)
Heart weight (g)	12.00^{a}(2.64)	9.00^{a}(2.64)	11.66^{a}(1.15)	12.00^{a}(0.00)	10.66^{a}(2.88)	11.00^{a}(1.00)	11.00^{a}(1.00)	11.33^{a}(2.51)	11.66^{a}(1.52)
Liver weight (g)	52.00^{ab}(9.64)	58.66^{a}(3.51)	51.66^{ab}(10.96)	57.66^{ab}(10.69)	48.00^{ab}(3.60)	54.00^{ab}(2.64)	44.00^{b}(8.71)	46.33^{ab}(6.65)	51.33^{ab}(0.57)
Gizzard weight (g)	39.00^{a}(2.64)	33.66^{a}(2.30)	35.00^{a}(4.00)	38.00^{a}(2.00)	35.66^{a}(4.04)	37.66^{a}(1.52)	37.00^{a}(1.00)	36.66^{a}(4.72)	39.66^{a}(6.42)

T1 (control) – 0% fat + 0 mg/kg L-carnitine; T2 – 0% fat + 200 mg/kg L-carnitine; T3 – 0% fat + 400 mg/kg L-carnitine; T4 – 2.5% fat + 0 mg/kg L-carnitine; T5 – 2.5% fat + 200 mg/kg L-carnitine; T6 – 2.5% fat + 400 mg/kg L-carnitine; T7 – 5% fat + 0 mg/kg L-carnitine; T8 – 5% fat + 200 mg/kg L-carnitine; T9 – 5% fat + 400 mg/kg L-carnitine.
^{abc}Means with different superscripts within each row of dietary treatments differ significantly at $P<0.05$.

BW gain during the initial growing phase, while in a more recent study an increase of growth rate and greater BW, feed intake and FCR in broilers fed supplemental 60 ppm L-carnitine were observed by Oladele *et al.*[2011]. Our results are in agreement with other studies [Rabie *et al.* 1997, Rabie and Szilágyi 1998, Xu *et al.* 2003, Rezaei *et al.* 2007], which reported the effect reducing abdominal fat content for L-carnitine provided at doses of 50-250 mg/kg. Our results showed that L-carnitine decreased the weight of the neck, although breast and drumstick weights were not significantly affected in contrast to other studies [Rabie and Szilágyi 1998, Xu *et al.* 2003], in which supplemental 50 mg/kg L-carnitine increased breast meat and drumstick weights. Data showed that L-carnitine improved fatty acid oxidation, prevented esterification to triglycerides and fat storage in the abdomen.

Interaction effects

Data on the growth performance of broilers fed different diets are summarized in Table 2. No significant variations were observed between the different groups of feed supplementation and breeding parameters, i.e. FCR and BW. The EPEF was generally higher in the group receiving 5% fat + 200 mg/kg L-carnitine ($P<0.05$) compared to the control group and the group fed supplemental 400 mg/kg L-carnitine alone ($P<0.05$), in which EPEF was the lowest. The supplemented diet modified significantly the spleen weight (Tab. 3), which was greater in the group fed 2.5% fat + 400 mg/kg L-carnitine ($P<0.05$) than in the group fed the high fat (5%) diet with 200 mg/kg L-carnitine ($P<0.05$).

Of carcass components (Tab. 4), supplemental 5% fat + 400 mg/kg L-carnitine increased significantly the full abdomen carcass weight ($P<0.05$), while it was the lowest in broilers fed the low fat diet alone ($P<0.05$). The maximum abdominal fat weight was obtained in chickens fed 5% fat alone ($P<0.05$). When compared to the effect of the diet supplemented with L-carnitine, we observed abdominal fat weight reduced by approx. 40% at its lower dose (200 mg/kg, $P<0.05$) and by nearly 70% at its higher dose (400 mg/kg, $P<0.05$). The low fat diet combined with 400 mg/kg L-carnitine had a positive effect on the wings weight ($P<0.05$).The weights of gastrointestinal components and the heart are summarized in Table 5. The supplement of 200 mg/kg L-carnitine alone decreased the length and weight of the intestine (both $P<0.05$) when compared to the same dose supplementing the high fat diet. Moreover, the heaviest intestines were found in broilers fed the high fat diet with the higher L-carnitine dose compared to the controls and birds fed the lower dose of L-carnitine alone. We also found that the livers were heavier in the group fed with supplemental 200 mg/kg L-carnitine alone compared to the group fed the high fat diet alone.

Of the blood parameters (Tab. 6), the level of serum triglycerides ($P<0.05$) was altered depending on the dietary treatments. In general, formulations containing 400 mg/kg L-carnitine provided a greater reduction of plasma triglycerides. In contrast, the highest triglyceride level was observed in broilers receiving 200 mg/kg L-carnitine in the low fat diet ($P<0.05$). There were no significant variations in the other parameters

Table 6. Blood serum biochemical index means and standard errors (in parentheses) of Ross 308 male broilers fed different levels of dietary fat and L-carnitine in the first six weeks of age

Main effects	Fat (%)			L-carnitine (mg/kg)		
	0	2.5	5	0	200	400
DBIL (mg/dl)	0.66^{a}(0.00)	1.00^{a}(0.82)	0.48^{a}(0.17)	0.52^{a}(0.48)	0.44^{a}(0.26)	1.01^{a}(0.83)
TBIL (mg/dl)	1.75^{a}(0.37)	2.01^{a}(0.90)	1.72^{a}(0.51)	1.80^{a}(0.73)	1.74^{a}(0.74)	2.03^{a}(0.93)
ALP (mg/dl)	422.00^{a}(72.85)	405.00^{a}(35.67)	374.89^{a}(41.94)	379.44^{a}(48.47)	448.78^{a}(174.31)	393.78^{a}(32.53)
AST (U/l)	270.33^{a}(25.57)	293.67^{a}(32.33)	261.78^{a}(36.47)	279.33^{a}(37.37)	286.33^{a}(35.65)	268.11^{a}(31.02)
CREA (mg/dl)	0.32^{a}(0.00)	0.33^{a}(0.05)	0.30^{a}(0.00)	0.30^{a}(0.00)	0.33^{a}(0.07)	0.32^{a}(0.04)
BUN (mg/dl)	1.05^{a}(0.00)	1.67^{a}(1.51)	1.11^{a}(0.72)	0.11^{a}(0.05)	1.28^{a}(0.66)	1.50^{a}(1.03)
LDL (mg/dl)	60.55^{a}(1.52)	58.22^{a}(3.78)	57.66^{a}(6.11)	58.33^{a}(4.63)	59.00^{a}(5.29)	59.11^{a}(0.25)
HDL (mg/dl)	71.22^{a}(4.16)	66.55^{a}(4.58)	66.55^{a}(9.29)	67.22^{a}(5.65)	69.77^{a}(5.73)	67.33^{a}(7.14)
TG (mg/dl)	73.56^{a}(5.05)	97.67^{a}(9.45)	76.21^{a}(16.86)	78.22^{a}(15.24)	101.78^{a}(8.17)	67.44^{a}(10.64)
CHOL (mg/dl)	138.67^{a}(31.77)	138.00^{a}(9.50)	143.89^{a}(37.46)	139.00^{a}(28.19)	134.67^{a}(20.89)	146.89^{a}(28.45)
Glucose (mg/dl)	259.33^{a}(18.61)	250.88^{a}(6.35)	255.55^{a}(13.65)	250.88^{a}(19.12)	260.77^{a}(22.57)	254.11^{a}(23.67)

Interaction effects	T1	T2	T3	T4	T5	T6	T7	T8	T9
DBIL (mg/dl)	0.30^{a}(0.00)	0.30^{a}(0.00)	1.40^{a}(0.95)	0.83^{a}(0.83)	0.40^{a}(0.10)	1.23^{a}(1.00)	0.43^{a}(0.15)	0.63^{a}(0.41)	0.40^{a}(0.17)
TBIL (mg/dl)	1.36^{a}(0.38)	1.30^{a}(0.44)	2.60^{a}(1.15)	2.13^{a}(1.19)	2.06^{a}(0.64)	2.10^{a}(0.90)	1.90^{a}(0.36)	1.86^{a}(1.06)	1.40^{a}(0.51)
ALP (mg/dl)	401.66^{a}(72.85)	529.33^{a}(278.50)	395.00^{a}(15.00)	375.00^{a}(25.11)	429.00^{a}(41.39)	411.00^{a}(35.67)	361.66^{a}(47.05)	388.00^{a}(90.14)	375.00^{a}(41.94)
AST (U/l)	281.66^{a}(25.57)	283.33^{a}(50.33)	246.00^{a}(10.39)	299.66^{a}(53.87)	296.66^{a}(39.71)	284.66^{a}(32.33)	256.66^{a}(25.16)	279.00^{a}(26.85)	249.66^{a}(36.47)
CREA (mg/dl)	0.30^{a}(0.00)	0.33^{a}(0.06)	0.33^{a}(0.06)	0.30^{a}(0.00)	0.36^{a}(0.11)	0.33^{a}(0.05)	0.30^{a}(0.00)	0.30^{a}(0.00)	0.30^{a}(0.00)
BUN (mg/dl)	1.00^{a}(0.00)	1.16^{a}(0.29)	1.00^{a}(0.00)	1.06^{a}(0.11)	1.66^{a}(1.15)	2.30^{a}(1.51)	1.10^{a}(0.17)	1.03^{a}(0.05)	1.20^{a}(0.72)
LDL (mg/dl)	61.33^{a}(1.52)	60.00^{a}(5.52)	60.33^{a}(5.03)	57.66^{a}(5.85)	55.33^{a}(6.65)	61.66^{a}(3.78)	56.00^{a}(5.19)	61.66^{a}(2.08)	55.33^{a}(6.11)
HDL (mg/dl)	71.33^{a}(4.16)	71.00^{a}(8.19)	71.33^{a}(7.23)	66.33^{a}(7.02)	66.33^{a}(5.50)	67.00^{a}(4.58)	64.00^{a}(4.35)	72.00^{a}(2.64)	63.66^{a}(9.29)
TG (mg/dl)	132.33^{ab}(31.77)	81.00^{ab}(7.00)	64.33^{b}(7.23)	75.66^{ab}(10.06)	149.66^{a}(115.50)	67.66^{b}(9.45)	83.66^{ab}(27.13)	74.66^{ab}(3.51)	70.33^{b}(16.86)
CHOL (mg/dl)	132.33^{a}(31.77)	125.33^{a}(20.65)	158.33^{a}(20.41)	138.66^{a}(14.74)	150.00^{a}(26.45)	125.33^{a}(9.50)	146.00^{a}(42.57)	128.66^{a}(9.07)	157.00^{a}(37.46)
Glucose (mg/dl)	247.33^{a}(18.61)	258.33^{a}(13.20)	272.33^{a}(33.26)	239.33^{a}(10.06)	275.66^{a}(35.01)	237.66^{a}(6.35)	266.00^{a}(21.28)	248.33^{a}(8.02)	252.33^{a}(13.65)

DBIL – direct bilirubin; TBIL – total bilirubin; ALP – alkaline phosphatase; AST – aspartate amino transferase; CREA – creatinine; BUN – blood urea nitrogen; LDL – low density lipoproteins; HDL – high density lipoproteins; TG – triglycerides; CHOL – cholesterol; T1 (control) – 0% fat + 0 mg/kg L-carnitine; T2 – 0% fat + 200 mg/kg L-carnitine; T3 – 0% fat + 400 mg/kg L-carnitine; T4 – 2.5% fat + 0 mg/kg L-carnitine; T5 – 2.5% fat + 200 mg/kg L-carnitine; T6 – 2.5% fat + 400 mg/kg L-carnitine; T7 – 5% fat + 0 mg/kg L-carnitine; T8 – 5% fat + 200 mg/kg L-carnitine; T9 – 5% fat + 400 mg/kg L-carnitine.

^{ab}Means with different superscripts within each row of dietary treatments differ significantly at $P<0.05$.

of organs and blood. The current results indicated that supplemental L-carnitine has performance-improving and lipolytic effects in broiler chickens fed applying different fat levels, which are consistent with those reported in other studies [Rabie and Szilágyi 1998, Parsaeimehr *et al.* 2014].

The European Production Efficiency Factor (EPEF) is a simple and practical index to assess productive performance data, which was increased up to 349 in the group receiving 5% fat, suggesting that dietary supplementation may improve growth performance. Similar data were found in broilers fed diets containing probiotics [360, Biernasiak and Slizewska 2009]. We observed no response of FCR and BW during the experimental period, which is in line with findings of Murali *et al.* [2015], who used 900 mg/kg L-carnitine in a high fat diet of chickens. It contradicts findings of other studies, which reported a reducing [Rajabzadeh-Nesvan *et al.* 2013] or improving effect [Jalali *et al.* 2015] of L-carnitine supplementing different fat source diets on FCR, at increased BW and feed intake in chickens [Rajabzadeh-Nesvan *et al.* 2013, Jalali *et al.* 2015]. The weights of wings, spleen, intestine, liver, abdomen as well as intestine length were significantly increased in response to the experimental diets, which was attributable to the L-carnitine effect in a fat rich diet as a result of increasing energy efficiency of dietary fat. Our results are in line with the study of Rezaei *et al.* [2007] that revealed the same effect on the liver weight in broilers fed a combined diet. In contrast, Rajabzadeh-Nesvan *et al.* [2013] reported no significant effect of such a diet on the weight of the heart, liver, breast and thighs.

Energy requirements increase in the first period of broiler growth. Carnitine, as an essential compound in energy metabolism, facilitates the transport of fatty acids into the mitochondrial matrix mobilizing energy for cells [Furuno *et al.* 2001]. Elevated cellular energy utilization results in an increased metabolic rate, which makes the availability of L-carnitine particularly critical [Buyse *et al.* 2001]. High-energy intake generates higher serum levels of triglycerides and cholesterol as well as greater deposition of fat in the body [Parsaeimehr et al. 2014] as was reported in our study. Therefore, additional L-carnitine is favorable [Buyse *et al.* 2001].We found a decrease in the abdominal fat weight and serum triglycerides at the higher (400 mg/kg) L-carnitine dose, while the other blood parameters were not affected. Arslan *et al.* [2004] reported lowered serum triglycerides, but also cholesterol and total lipids in Japanese quails. By contrast, a low dose of dietary L-carnitine (60 mg/kg) increased cholesterol levels [Kheiri *et al.* 2011]. Moreover, there are reports on hypoglycaemic effects of L-carnitine in broilers [Buyse *et al.* 2001, Wang *et al.* 2013] and when given together with fat, resulting in markedly reduced cholesterol and LDL levels [Parasaeimehr *et al.* 2013]. However, Jalali *et al.* [2015] reported increased levels of cholesterol, HDL, LDL, and proteins in the serum of broiler chickens.

The results obtained in this study revealed that 400 mg/kg dietary L-carnitine markedly lower abdominal fat weight and serum triglycerides levels at no addition of fat to the diet. Feeding the high fat diet supplemented with 200 mg/kg L-carnitine showed the highest EPEF. Overall, the performance traits improved with the increase

in the fat inclusion level. It can be concluded that dietary L-carnitine improved growth performance of broilers, thus it may be a promising way to reduce fat storage in broilers and improve the quality of carcasses intended for human consumption.

REFERENCES

1. ARSLAN C., CITIL M., SAATCI M., 2004 – Effects of L-carnitine administration on growth performance, carcass traits and some serum components of Japanese quail (*Coturnix cot. japonica*). ***Archiv für Geflügelkunde*** 68, 111-114.
2. BHAMARE K.S., DILDEEP V., SENTHIL M.S., CHAVAN S.J., 2016 – Nutritive evaluation of cashew apple waste in broilers. ***International Journal of Science and Nature***7, 629-632.
3. BIERNASIAK J., SLIZEWSKA K., 2009– The effect of a new probiotic preparation on the performance and faecal microflora of broiler chickens. ***Veterinární Medicína*** 54, 525-531.
4. BURLIKOWSKA K., PIOTROWSKA A., SZYMECZKO R., 2010– Effect of dietary fat type on performance, biochemical indices and fatty acids profile in the blood serum of broiler chickens. ***Journal of Animal and Feed Sciences*** 19, 440-451.
5. BUYSE J., JANSSENS G.P.J., DECUYPERE E., 2001– The effects of dietary L-carnitine supplementation on the performance, organ weights and circulating hormone and metabolite concentrations of broiler chickens reared under a normal or low temperature schedule. ***British Poultry Science*** 42, 230-241.
6. FOUAD A.M., EL-SENOUSEY H.K., YANG X.J., YAO J.H., 2013– Dietary L-arginine supplementation reduces abdominal fat content by modulating lipid metabolism in broiler chickens. ***Animal*** 7, 1239-1245.
7. FURUNO T., KANNO T., ARITA K., ASAMI M., UTSUMI T., DOI Y., INOUE M., UTSUMI K., 2001– Roles of long chain fatty acids and carnitine in mitochondrial membrane permeability transition. ***Biochemical Pharmacology*** 62, 1037-1046.
8. JALALI S.M.A., RABIEI R., KHEIRI F., 2015– Effects of dietary soybean and sunflower oils with and without L-carnitine supplementation on growth performance and blood biochemical parameters of broiler chicks. ***Archives Animal Breeding*** 58, 387-394.
9. KHEIRI F., POURREZA J., EBRAHIMNEZHAD Y., NAZERADL K., HAJI-ABADI S.M.A.J., 2011– Effects of supplemental ractopamine and L-carnitine on growth performance, blood biochemical parameters and carcass traits of male broiler chicks. ***African Journal of Biotechnology*** 10, 15450-15455.
10. KRYEZIU A.J., MESTANI N., BERISHA S., KAMBERI M.A., 2018– The European performance indicators of broiler chickens as influenced by stocking density and sex. ***Agronomy Research*** 16, 83-491.
11. MOHAMMAD H.A., HORNIAKOVÁ E., 2012– Effect of using saturated and unsaturated fats in broiler diet on carcass performance. ***Slovak Journal of Animal Science*** 45, 21-29.
12. OLADELE O.A., ADEBOYE F., RICHARD S., ZAINOB H., 2011– Growth response, carcass yield and serum biochemistry of broiler chicken fed with supplemental L-carnitine in feed or drinking water. ***Journal of Poultry Science*** 48, 223-228.
13. PARSAEIMEHR K., AFROUZIYEH M., HOSEINZADEH S., 2014– The effects of L-carnitine and different levels of animal fat on performance, carcass characteristics, some blood parameters and immune response in broiler chicks. ***Iranian Journal of Applied Animal Science*** 4, 561-566.
14. PARSAEIMEHR K., FARHOOMAND P., AFROUZIYEH M., NAJAFI R., AHMADIE-NAGHDEHI A. A., 2013 – Effects of L-carnitine with different dietary fat sources on performance and some blood metabolites of broiler chickens. Animal Production Research 1(4), 27-34. (in Persian, with English summary).

15. PEARSON A.M., DUTSON T.R., 2013 – Production and processing of healthy meat, poultry and fish products (Vol. 11). Springer Science & Business Media.
16. RABIE M.H., SZILÁGYI M., 1998 – Effects of L-carnitine supplementation of diets differing in energy levels on performance, abdominal fat content, and yield and composition of edible meat of broilers. ***British Journal of Nutrition*** 80, 391-400.
17. RABIE M.H., SZILÁGYI M., GIPPERT T., VOTISKY E., GERENDAI D., 1997– Influence of dietary L-carnitine on performance and carcass quality of broiler chickens. ***Acta Biologica Hungarica*** 48, 241-252.
18. RAJABZADEH-NESVAN M., REZAEE M., ANSARI-PIRSARAEE Z., 2013 – Effect of L-carnitine supplementation to finisher diets with different sources of fat on the performance, carcass characteristics and body composition in broiler chickens. Journal of Management System 92(2), 51-68. (in Persian, with English summary).
19. REZAEI M., ATTAR A., GHODRATNAMA A., KERMANSHAHI H., 2007– Study the effect of different levels of fat and L-carnitine on performance, carcass characteristics and serum composition of broiler chicks. ***Pakistan Journal of Biological Sciences***10, 1970-1976.
20. WANG Y.W., NING D., PENG Y.Z., GUO Y.M., 2013 – Effects of dietary L-carnitine supplementation on growth performance, organ weight, biochemical parameters and ascites susceptibility in broilers reared under low-temperature environment. ***Asian-Australasian Journal of Animal Sciences*** 26, 233-240.
21. XU Z.R., WANG M.Q., MAO H.X., ZHAN X.A., HU C.H., 2003– Effects of L-carnitine on growth performance, carcass composition, and lipid metabolism in male broilers. ***Poultry Science*** 82, 408-413.