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Camelia (*Camelina sativa* L. Crantz Variety) Oil and Seeds as n-3 Fatty Acids Rich Products in Broiler Diets and Its Effects on Performance, Meat Fatty Acid Composition, Immune Tissue Weights, and Plasma Metabolic Profile

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ABSTRACT

The study was carried out to investigate the effects of Camelia (*Camelina sativa* L. Crantz) variety (CS) oil or seeds on performance, meat quality, immunity and plasma metabolic profile in broiler chickens. The broilers (n= 2,080, Cobb 500) were randomly allocated (sex ratio 1:1) for 32-day experimental periods (from 11d to 42 d) to 4 experimental groups: Control (corn-soybean meal-full fat soy based diet), Group I (with CS oil added at 2.5%), Group II (5% CS seeds), and Group III (10% CS seeds, respectively) of 520 birds each, and received the diets *ad libitum*. Gas chromatography method was used to determine the fatty acid profile of the ingredients and breast muscle. At slaughter (42 day), a simplified analysis of the carcasses was conducted. The thymus, spleen, and bursa of Fabricius were aseptically removed and weighed. Results indicated that the adding of 2.5% CS oil and 5% CS seed did not have a negative effect on performance and carcass characteristics (i.e. carcass yield, legs, and breast proportions). Chickens fed the diet containing 10% CS seed had significantly decreased (P< 0.001) BW gain. A significant decrease was also observed in the proportion of abdominal fat (P< 0.05) in carcasses with increasing levels of CS seeds in the diet. However, the diets with CS oil and seeds led to significant increases in omega *n*-3 fatty acids profile in the breast muscle, mainly *α*-linolenic acid (P< 0.0001), eicosapentaenoic acid (P< 0.0301), docosapentaenoic acid (P< 0.0123) and docosahexaenoic acid (P< 0.0026). The diets did not significantly affect the spleens and thymus weights, plasma enzymes activity, and total immunoglobulin (Ig) content. Plasma energy profile showed a tendency (P< 0.066) towards increased triglyceride content and significantly decreased total cholesterol (P< 0.019) and its fractions (P< 0.001) in the groups receiving CS oil and seeds in the diet.

Keywords: Blood parameters, Carcass characteristics, Chickens, Immunity, Meat quality.

INTRODUCTION

Camelia is the name of the first Romanian camelina (*Camelina sativa* L. Crantz) as a line of Calena, an Austrian camelina variety. Camelia (*Camelina sativa* L. Crantz) variety (CS) is an oilseed crop of the Brassicaceae (Cruciferae) family. Although CS or false flax (gold-of-pleasure) has been cultivated in Europe for over 2000 years for oil and livestock fodder, the crop has gained increased popularity recently as a biofuel source due to its oil content (Putnam *et al.*, 1993). Romania is one of the few countries where the production of CS variety is still carried on some farmland, with relatively low inputs and no irrigation. Its seeds contain 30.1-49.7% oil (Toncea *et al.*, 2013;

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Ayasan, 2014). Their composition varies with the agrotechnical measures used in their production but primarily α-linolenic acid (18:3n-3; 30.5-50.3%) and linoleic (18:2n-6; 16.60-19.82%) are found in the oil (Toncea *et al.*, 2013). Alpha-Linolenic acid is the precursor of long-chain *n*-3 PolyUnsaturated Fatty Acids (PUFA) such as EicosaPentaenoic Acid (EPA, 20:5n-3), DocosaPentaenoic acid (DPA, 22:5n-3), and DocosaHexaenoic Acid (DHA, 22:6n-3), which are commonly referred to as n-3 or omega-3 fatty acids. This makes CS oil a rich source of essential fatty acids and a very good source of omega-3 fatty acids, which are essential for humans since they cannot be synthesized in the organism and must be ingested in food. Protein and fibre content in CS seeds are also important nutritional parameters: the content of crude protein in seed ranges from 18.87-21.97% while the content of crude fibre ranges from 11.06-15.24%. Additionally, CS oil contains high amounts of vitamin E (25.83-28.21 mg 100 g^{-1}), a powerful antioxidant. Besides, Abramovic and Abram (2005) assert that *Camelina sativa* oil must contain an appreciable amount of antioxidant (400 mg of total phenolic per kg of fresh oil), which makes the oil quite stable in spite of its high PUFA content, without compromising the sensory quality of poultry products (Rokka *et al.*, 2002; Jaskiewicz and Matyka, 2003). Recent research shows that it is possible to use *Camelina sativa* oil and its by-products for animal nutrition (Flachowsky *et al.*, 1997; Moloney *et al.*, 1998; Jaskiewicz and Matyka, 2003; Peiretti *et al.*, 2007; Habeanu *et al.*, 2011; Ciuca *et al.*, 2013). Camelina utilization in poultry diets has been studied previously (Zubr, 1997) and it is well documented that the fatty acids composition of hen egg yolk can be modified through alteration of the diet. Recently, there has been some concern that diets enriched with *n*-3 PUFA may have detrimental effects on chicken immunity and impair their resistance to infection. However, it is not clear whether this concern is justified, given that some studies show an improvement (Korver and Klasing, 1997; Parmentier *et al.*, 1997; Sijben *et al.*, 2000; Yang and Guo, 2006), some show a detrimental effect (Fritsche *et al.*, 1991; Babu *et al.*, 2005), and some show no effect (Puthpongsiriporn and Scheideler, 2005). The main immune organs in poultry are the thymus, spleen and bursa of Fabricius. This immune tissue mass can in some cases indicate the immune status (Moller and Erritzoe, 2000; Smith and Hunt, 2004).

Therefore, the aim of this study was to investigate the effects of dietary *n*-3 PUFA-rich by addition of CS oil or CS seeds on the performances, breast muscle fatty acid composition, the plasma metabolic profile, and the immune tissue weight in broiler chickens.

MATERIALS AND METHODS

Broilers Diets and Performance Measurements

Birds were treated in accordance with the Romanian legislation for handling and protection of animals used for experimental purposes. This study protocol was approved by the Ethical Committee of the National Research Development Institute for Animal Biology and Nutrition Baloteşti, Romania (permission No. 257-30-3/2012).

A total of 2,080 one-day-old Cobb 500 broilers sexed at the local commercial hatchery (sex ratio 1:1) were randomly assigned for 32-day experimental diet periods (from 11d to 42 d) to four feeding groups. In total, 16 floor pens with wood shaving (130 chicks per pen, surface area 11 sq m) were used, to give 4 pens (replicates) and a total of 520 birds per group. Each replicate pen was considered the experimental unit. A lighting program of 23 L:1 D was used. Ambient temperature was gradually decreased from 32°C on day 1 to 22°C at the end of the experiment. Control parameters, such as temperature, humidity, light, ventilation, and vaccination were the same for all groups. The

broilers were given *ad libitum* access to feed and water.

Upon hatching, all chicks were given the same basal diet for 10 days. From 11 day, chicks (Average BW= 276±22.62 g), were fed (grower/finisher feeding phases) a corn-soybean meal-full fat soy based diet (Control), with CS oil added at 2.5% (Group I), or 2 different levels (5 or 10%) of CS seeds (Group II and Group III, respectively) (Table 1). All diets (Table1) were isocaloric, isonitrogenous and with similar content of digestible sulfur amino acids (met.+cys.), lysine, calcium and available P and in accordance with Cobb guidelines (Cobb-Vantress, 2008). The proximate composition of feed components and the diets (dry matter, crude protein, crude fiber, ether extract) was analyzed using the Kjeldahl (Foss Tecator) procedures (AOAC, 2000). The metabolizable energy content of

Table 1. Ingredients and chemical composition of the diets.

Ingredients (% as is)	Diets			
	Control	Group I	Group II	Group III
Corn	65.13	66.05	63.98	62.63
Soybean meal (45.5% CP)	8.78	23.50	12.30	15.45
Full fat soy (37.7% CP)	18.20	0.00	10.80	4.00
Corn gluten meal (62.8% CP)	3.00	3.00	3.00	3.00
Camelia seed	0.00	0.00	5.00	10.00
Camelia oil	0.00	2.50	0.00	0.00
Limestone	1.35	1.38	1.35	1.35
Mono calcium phosphate	1.54	1.56	1.54	1.54
Salt	0.30	0.30	0.30	0.30
Vitamin-mineral premix [a]	1.00	1.00	1.00	1.00
DL-methionine	0.26	0.26	0.25	0.25
L-lysine HCl	0.38	0.39	0.42	0.42
Choline chloride	0.06	0.06	0.06	0.06
Calculated nutrient composition (g kg^{-1})				
Crude protein	190.0	190.0	190.0	190.0
AME_n (MJ kg^{-1})	13.17	13.16	13.15	13.15
Crude fibre	28.1	25.6	31.2	34.5
Ether extract	61.4	56.0	66.6	72.7
Lysine (dig)	10.5	10.5	10.6	10.5
Methionine, (dig)	5.4	5.4	5.4	5.4
Met.+Cys. (dig)	8.0	8.0	8.0	8.1
Calcium	8.4	8.4	8.4	8.4
Available phosphorus	4.2	4.2	4.2	4.2
Analyzed (g kg^{-1})				
Dry matter	898.9	904.5	899.3	901.1
Crude fiber	28.5	26.7	29.9	33.3
Crude protein	192.6	191.8	189.3	189.7
Ether extract	59.7	54.6	65.8	71.8
Linoleic (C18:2n-6) as % of total FAME)	29.89	25.58	26.60	25.51
α-Linolenic (C18:3n-3) as % of total FAME)	1.32	14.77	13.53	23.76

[a] Supplied per kg diet: Retinyl acetate, 4.47 mg; Cholecalciferol, 0.12 mg; DL-α-tocopheryl acetate, 80 mg; Menadione sodium bisulphite, 4 mg; Thiamine mononitrate, 4 mg; Riboflavin, 9 mg; Pyridoxine-HCl, 4 mg; Cyanocobalamin, 0.020 mg; Ca-panthotenate, 15 mg; Niacin, 60 mg; Folic acid, 2 mg; Mn, 100 mg; Zn, 100 mg; Fe, 40 mg; Cu, 15 mg; I, 1.0 mg; Se, 0.30 mg, Co, 0.25 mg.

the diets was calculated from content of basic nutrients with regression equations proposed by Janssen (1989). In order to determine the performance of broilers, Feed Intake (FI) and the Body Weight (BW) gain were recorded weekly on a pen basis. FI, BW, and Feed Conversion Ratio (FCR) were calculated. In cases where mortalities were observed, the numbers and weights of such mortalities were recorded accurately to make necessary corrections in calculating FI and FCR.

Sample Collection

At the 42 day of age, 2 chick/ pen i.e. 8 chicks/ treatment, which represented an average pen weight were slaughtered by neck cut, severing the right carotid artery and jugular vein. Eviscerating and rinsing were subsequently done manually. Breast and legs (including, both, thigh and drumstick, with skin and bone) were separated from the carcass and were weighed individually. Thereafter, the remaining parts with breast and legs were added together to find the hot carcass weight for determining the carcass yield. To determine fatty acid profile the samples (n= 32) of left breast muscles were deboned, skin removed, packed into polyethylene bags, sealed, and immediately stored in the deep freezer at -20°C until the time of analysis. The pH at 24 hours post-mortem was directly measured in the breast tissue with a digital pH-meter (model HI 99163, Hanna Instruments, Romania), a portable HACCP (hazard analysis and critical control points) compliant pH meter for meat, equipped with internal temperature sensor (Model: FC 432D) and with a penetrating spear tip pH electrode (FC 099 stainless steel blade tip).

Lymphoid Organs Weight/Body Weight Ratio

The thymus, spleen, and bursa of Fabricius were aseptically removed and weighed separately to determine the lymphoid organs (Giamborne and Closser, 1990). All of these traits were calculated in relation to live BW.

Blood Analysis

These same 8 chicks from each treatment were also selected for blood sample collection for biochemical study. Blood samples were collected in heparinized tubes and, then, were centrifuged in the Multifuge 3L-R (Heraeus, Hanau, Germany) at 2,500 rpm for 10 minutes at room temperature (25°C) and the plasma was transferred to a fresh tube, frozen, and stored at −20°C until analyses. Plasma concentrations of glucose, cholesterol, High Density Lipoprotein Cholesterol (HDL-C), Low Density Lipoprotein Cholesterol (LDL-C), triglycerides, total protein, albumin, total globulin, creatinine, urea, and the Alkaline Phosphatase (AP) enzyme activity, γ-Glutamyl Transferase (GGT), Glutamate Pyruvate Transaminase (GPT), Glutamate Oxaloacetate Transaminase (GOT) were measured by specific commercially available Accent-200 MG kits (Cormay, Wiosenna, Poland) using an auto chemistry analyzer (model BS-130, Shenzhen Mindray Bio-Medical Electronics Co., Ltd., Shenzhen, PR China). The total concentration of Immunoglobulin (Ig) subsets was measured by ELISA kits (Bethyl Laboratories Inc., Montgomery, TX, USA) after plasma dilution: 1/4,000 (IgA), 1/60,000 (IgG) and 1/6,000 (IgM) as previously reported (Marin *et al.*, 2006), and according to the manufacturer's instructions. Absorbance was read at 450 nm using a microplate reader (Tecan Sunrise, Salzburg, Austria) and results were expressed as mg mL^{-1} of plasma.

Fatty Acid Analysis

The muscle tissue and ingredients fatty acid composition was analyzed using a gas chromatography system, according to Commission Regulation (EC) no.152/2009

(Official Journal of the European Union, 2009). Fatty Acid Methyl Esters (FAME) were prepared from total lipid extract using methanolic HCl as the derivatizing agent. Analyses of FAME were performed with a Clarus 500 gas chromatograph (PerkinElmer, Inc., SUA) equipped with an autos-ampler, Flame Ionization Detector (FID), and fused silica capillary column (cis/trans FAME), 60 m×0.25 mm×0.2 µm film thickness (PerkinElmer, Inc., SUA). The calibration and the peak determinations were based on authentic standards fatty acids from Sigma-Aldrich (St. Louis USA). The results were expressed for each fatty acid as % of total FAME.

Statistical Analysis

All data were analyzed by the General Linear Models (GLM) procedure using the SPSS software version 17 (SPSS, 2008). The pen was considered the experimental unit for performance criteria and the individual bird for carcass characteristics, chemical analysis and pH of breast muscle, immune tissue weight, and blood plasma parameters. The differences between means were considered significant when $P< 0.05$, and when significant main effects were observed, the differences between means were determined using Tukey's procedure.

RESULTS AND DISCUSSION

Nutrient Composition of Diets and Camelia Seeds

The experimental diets differed with regard to their fatty acid profiles (Table 1). Diets groups I, II, and III were characterized by the highest concentrations of *n*-3 PUFA, mainly *α*-linolenic acid. The *α*-linolenic acid in the diet reached 14.77% in group I, 13.53% in group II , and 23.76% in group III and was the result of the high levels of *C*18:3*n*-3 that was present in raw CS (Table 2).

Performance and Carcass Characteristics

Growth performance parameters such as BW, FI, and FCR, mortality and carcass characteristic are summarized in Table 3. The lowest BW was observed in group III, compared to the control, group I, and group II (2276.34 g *vs.* 2464.63 g; 2528.45 g; 2432.77 g, P< 0.001). However, during the total experimental period, the highest FCR noticed in groups II and III showed no significant difference (P> 0.05), compared with group I and the control (1.84 and 1.88 *vs.*

Table 2. Camelia (*Camelina sativa,* L. Crantz variety) seed composition.

Characteristics	Seeds
Dry matter (%)	93.66
Crude protein (%)	24.78
Crude fibre (%)	11.40
Ether extract (%)	36.84
Crude ash (%)	4.27
ME (MJ kg^{-1}) [a]	14.13[1]
Fatty acid content (as % of total FAME)	
Palmitic (C16:0)	6.07
Stearic (C18:0)	1.91
Oleic (C18:1n-9)	16.46
Linoleic (C18:2n-6)	18.84
α-Linolenic (C18:3n-3)	33.43
Eicosenoic (C20:1n-9)	12.99
Octadecatetraenoic (C18:4n-3)	0.36
Eicosadienoic (C20:2n-6)	1.47
Arachidonic (C20:4n-6)	1.02
Erucic (C22:1n-9)	5.02
Eicosapentaenoic (C20:5n-3)	0.12
Docosatetraenoic (C22:4n-6)	0.33
Docosapentaenoic(C22:5n-3)	0.04
Docosahexaenoic(C22:6n-3)	0.34
Total n-6 PUFA [b]	21.66
Total n-3 PUFA [c]	34.29

[a] Metabolizable Energy content was calculated based on the regression proposed by Janssen (1989). [b] Total *n*-6 PUFA= Sum percentage of C18:2*n*-6 trans; *C*18:2*n*-6 *cis*, *C*20:4*n*-6, *C*20:2*n*-6, *C*22:4*n*-6. [c] Total *n*-3 PUFA= Sum percentage of *C*18:3*n*-3; *C*18:4*n*-3; *C*20:5*n*-3; *C*22:5*n*-3, *C*22:6*n*-3.

Table 3. Effect of dietary treatment on the performance and carcass characteristics of broiler chickens from 42 days of age.[a]

Variable	Dietary treatment [b]				SEM	*P*-value
	Control	Group I	Group II	Group III		
BW gain (g)	2464.63[a]	2528.45[a]	2432.77[a]	2276.34[b]	7.89	<0.001
FI (g)	4261.98	4349.78	4396.83	4221.54	127.34	0.312
FCR (g g^{-1})	1.76	1.75	1.84	1.89	0.07	0.147
Mortality (%)	1.54	1.92	1.15	1.35	0.53	0.983
Carcass characteristic:						
Carcass yield (%)	73.8	74.2	73.6	73.1	2.17	0.083
Legs (g)	401	404	397	394	13.78	0.439
Breast (g)	334	336	332	330	12.60	0.127
Abdominal fat (%)	1.11[a]	1.02[a]	0.97[a]	0.59[b]	0.05	0.034

[a] Means in the same row with different superscripts differ significantly (P< 0.05). [b] Control= Represents the corn-soybean meal-full fat soy basal diet; Group I= Camelia (*Camelina sativa,* L. Crantz variety) oil at 2.5%; Group II= Camelia seeds at 5%, Group III= Camelia seeds at 10%.

1.75 and 1.76, respectively). One of the reasons for the lower broiler BW in group III may be due to the higher crude fiber content of diets that contained 10% CS seeds, which may have adversely affected nutrient digestibility. Another possible reason for this differential performance is the greater fat content and the reduced digestibility of fat and fatty acids reported for full-fat flaxseed by Ortiz *et al.* (2001) with broiler chickens. Results of feeding trials on *Camelina* meal in poultry are conflicting. Feeding camelina seed (10 and 20%) to broiler birds were associated with negative effects on performance (Ajuyah *et al.*, 1991; Gonzalez and Leeson, 2001). Impaired feed conversion, and decreased feed intake during the starter phase in birds fed 5% camelina cake or 10% camelina meal were reported by Ryhänen *et al.* (2007) and Pekel *et al.* (2009). However, Aziza *et al.* (2010) fed 2.5, 5, and 10% camelina meal to broiler birds and reported no difference in 42-day body weight gain, carcass weight, or feed efficiency when compared with maize-soybean-based control diet-fed birds. Also, Peiretti *et al.* (2007) and Prola *et al.* (2011) observed no significant differences in growth performance and carcass characteristics on rabbits when *Camelina* seed was fed at levels of up to 15% of the diet.

Throughout the present study, the mortality was negligible, with no significant difference (P> 0.05) among all groups (Table 3). Also, adding 2.5% CS oil (group I), or 5% and 10% CS seeds to the diet in groups II and III had no significant (P> 0.05) effect on the carcass characteristics i.e. carcass yield, legs and breast proportions. However, observations showed a significant decrease (P< 0.05) in the proportion of abdominal fat in carcasses with increasing levels of CS seeds in the diet (Table 3). These findings are similar to the results obtained by Crespo and Esteve-Garcia (2002), who found that PUFA reduce abdominal fat deposition. The mechanism by which the dietary PUFA modify body fat deposition is not completely understood. Some studies suggested that the lower fat deposition in broiler chickens fed on a diet containing oils rich in PUFA compared to those fed a diet poor in PUFA but with a higher level of SFA and MUFA was, in part, explained by a greater rate of lipid oxidation and by a decline in fatty acid synthesis (Sanz *et al.*, 2000).

Chemical Analysis, pH and Fatty Acid Profile of Breast Muscle

Chemical analysis of the breast muscle (Table 4) showed a significant increase (P< 0.0001) in crude protein content in groups fed CS seeds (II and III), compared to the control and group I (21.35 and 22.24% *vs.* 20.62 and 20.87%). The dry matter and crude fat content was at a similar level. No

Table 4. Effect of dietary treatment on the chemical analysis, pH, and fatty acid profile of breast muscle in broiler chickens.[a]

Analysis	Dietary treatment [b]				SEM	*P*-value
	Control	Group I	Group II	Group III		
Dry matter (%)	30.30	30.34	30.59	29.87	0.62	0.2031
Crude protein (%)	20.62^{c}	20.87^{c}	21.35^{b}	22.24^{a}	0.81	0.0001
Crude fat (%)	1.95	2.21	2.27	2.32	0.25	0.4236
pH	5.95	6.02	5.92	5.97	0.21	0.9872
Fatty acid content (As % of total FAME):						
Palmitic (C16:0)	22.39	22.15	21.97	22.26	0.58	0.3324
Stearic (C18:0)	11.55^{a}	7.43^{bc}	8.71^{b}	7.76^{bc}	0.77	0.0389
Oleic (C18:1n-9)	27.18^{b}	30.36^{a}	29.80^{a}	30.07^{a}	1.16	0.0441
Linoleic (C18:2n-6)	20.95	21.78	22.25	21.54	2.31	0.0617
α-Linolenic (C18:3n-3)	1.24^{c}	3.70^{ab}	2.97^{b}	3.62^{ab}	0.14	0.0001
Octadecatetraenoic (C18:4n-3)	0.19^{a}	0.14^{b}	0.12^{c}	0.10^{d}	0.04	0.0001
Arachidonic (C20:4n-6)	7.18^{a}	5.58^{b}	5.45^{b}	5.77^{b}	0.63	0.0232
Eicosadienoic (C20:2n-6)	0.87	0.55	0.61	0.52	0.26	0.0902
Eicosapentaenoic (C20:5n-3)	0.77^{c}	1.19^{a}	0.92^{b}	1.32^{a}	0.01	0.0301
Docosatetraenoic (C22:4n-6)	2.21^{a}	0.94^{b}	1.16^{b}	0.97^{b}	0.13	0.0194
Docosapentaenoic(C22:5n-3)	0.93^{b}	1.12^{a}	1.09^{a}	1.18^{a}	0.03	0.0223
Docosahexaenoic(C22:6n-3)	1.84^{c}	2.32^{a}	2.24^{ab}	2.30^{a}	0.01	0.0026
Total *n*-6 PUFA[c]	31.21^{a}	28.85^{b}	29.47^{b}	28.80^{b}	0.86	0.0288
Total *n*-3 PUFA[d]	4.97^{b}	8.47^{a}	7.34^{a}	8.52^{a}	3.45	0.0001

[a] Means in the same row with different superscripts differ significantly (P< 0.05). [b] Control= Represents the corn-soybean meal-full fat soy basal diet; Group I= Camelia (*Camelina sativa,* L. Crantz variety) oil at 2.5%; Group II= Camelia seeds at 5%, Group III= Camelia seeds at 10%. [c] Sum percentage of *C*18:2*n*-6 *trans*; *C*18:2*n*-6 *cis*; *C*20:4*n*-6; *C*20:2*n*-6, *C*22:4*n*-6. [d] Sum percentage of *C*18:3*n*-3; *C*18:4*n*-3; *C*20:5*n*-3; *C*22:5*n*-3, C22:6n-3.

significant differences were observed in pH value of the breast muscle in broilers fed a control diet than in the other three groups (Table 4). This is consistent with reports of other authors (Ajuyah *et al.*, 1991; Crespo and Esteve-Garcia, 2002).

The fatty acid profile of breast muscle indicated significant differences in fatty acid incorporation (Table 4). Dietary *n*-3 PUFA enrichment alters the fatty acid profile of breast muscle towards higher level of long chain PUFA. The significant increases in α-linolenic acid (P< 0.0001), EPA (P< 0.0301), DPA (P< 0.0223) and DHA (P< 0.0026) were observed in the breast muscle of broiler in the group I, II and III, compared with the control. Significant decrease (P< 0.0389) in stearic acid with a concomitant increase (P< 0.0441) in oleic acid was observed in breast muscle of broiler in the groups I, II, and III, than in the control group. No differences were observed in content of palmitic acid in broilers fed a control diet than in the other three groups. However, total n-3 PUFA (C18:3n-3+C18:4n-3+ C20:5n-3+C22:5n-3+C22:6n-3) were highest in the breast muscle from broiler chickens in the groups I, II, and III, mainly because of the increase in α-linolenic acid (C18:3n-3), a predominant n-3 PUFA in CS oil and seeds. These analysis demonstrated that the diets significantly increases (P< 0.0001) the proportion of PUFA, mainly total n-3 PUFA, in chicken breast muscle (Table 4). In agreement with the current results, the total n-3 PUFA, EPA, and DHA of muscle tissue significantly increased with the inclusion of full-fat flax seed (Ajuyah *et al.*, 1991) and camelina meal (Aziza *et al.*, 2010) in chickens and linseed oil (Jankowski *et. al.*, 2012) in turkeys diet.

Immunity

The effect of *n*-3 PUFA-enriched diets on immune tissue weight (as a percentage of BW) in broiler chickens is shown in Table 5. Immune tissue development, such as thymus, spleen and bursa of Fabricius, can in some cases reflect the immune system response and functionality. Our results show that CS oil or different levels of CS seeds did not affect the weights of the spleen and thymus of broiler chickens, which is in agreement with the study by Aziza *et al.* (2010). On the other hand, some studies suggested that feeding PUFA to chickens (Wang *et al.*, 2000) and mice (Ellis *et al.*, 1986) resulted in increased spleen weights. In the study of Wang *et al.* (2000), the authors used single-comb White Leghorn layers fed the 3 PUFA-rich diets (sunflower, linseed, or fish oils). Results in the current study showed that chickens fed diets containing 10% CS seeds had significantly lower (P< 0.001) bursa weights. This confirms the findings of Al-Khalifa *et al.* (2012), who found that *n*-3 PUFA diets significantly reduced bursa weights. Other studies have shown that consumption of n-3 PUFA incorporates these fatty acids into the lipid membrane of all tissues, including all cells and tissues of the immune system (Maroufyan *et al.*, 2012). Results of Korver and Klasing (1997) and Puthpongsiriporn and Scheideler (2005) were consistent with these findings, showing that *n*-3 PUFA influence macrophages to be less inflammatory, enhance antibody responses, and suppress cell-mediated response compared with *n*-6 PUFA. Thus, *n*-3 PUFA reduce immunity to diseases controlled by a strong inflammatory or cell-mediated response, but they also decrease morbidity in response to diseases caused by excessive inflammatory response (Klasing, 2007). These alterations are dependent on factors such as *n*-6 PUFA and *n*-6/n-3 PUFA ratio.

Plasma Metabolic Profile

Table 6 summarizes blood plasma metabolic profile of the experimental broiler chickens. Analysis of blood plasma energy profile showed a tendency (P< 0.066) towards increased triglyceride content and significantly decreased total cholesterol (P< 0.019) and its fractions (P< 0.001) in the groups receiving CS oil and seeds in the diet. There was no significant treatment effect on plasma protein (P> 0.05). Generally, the broilers fed diets containing CS had slightly higher plasma protein than the broilers fed on the control group. Serum urea concentration is a good indicator of protein status in chicks (Kaneko, 1997). Accordingly, the present results of plasma urea being within the normal values signified the good ability of diets to supply the protein requirements for chicks. Also, in our study, there was no significant treatment effect on plasma enzymes (AP, GGT, GPT or GOT) activity. Our results showed normal function of liver, kidney and pancreas and

Table 5. Effect of dietary treatment on the immune tissue weights in broiler chickens. [a]

Dietary treatment [b]	Tissue (% of BW)		
	Spleen	Thymus	Bursa
Control	0.13	0.18	0.28[a]
Group I	0.14	0.16	0.26[a]
Group II	0.11	0.15	0.30[a]
Group III	0.12	0.12	0.17[b]
SEM	0.01	0.02	0.04
P-value	0.387	0.343	<0.001

[a] Means in the same column with different superscripts differ significantly (P< 0.05).
[b] Control= Represents the corn-soybean meal-full fat soy basal diet; Group I= Camelia (*Camelina sativa,* L. Crantz variety) oil at 2.5%; Group II= Camelia seeds at 5%, Group III= Camelia seeds at 10%.

Table 6. Effect of dietary treatment on the blood plasma metabolic profile in broilers chickens at 42 days.[a]

Plasma profile	Parameters	Dietary treatment[b]					
		Control	Group I	Group II	Group III	SEM	P-value
Energy	Glucose (mg dl^{-1})	209.7	218.4	211.3	214.6	1.30	0.651
	Cholesterol (mg dl^{-1})	128.2[a]	110.1[b]	113.8[b]	102.5[c]	0.45	0.019
	HDL-C (mg dl^{-1})	92.1[a]	84.2[b]	86.6[b]	69.7[c]	0.85	<0.001
	LDL-C (mg dl^{-1})	23.6[a]	21.3[ab]	16.7[ab]	15.3[b]	0.37	<0.001
	Triglycerides (mg dl^{-1})	46.3	45.6	46.5	49.1	0.98	0.066
Protein	Total protein (g dl^{-1})	2.98	3.09	2.91	3.07	0.07	0.091
	Albumin (g dl^{-1})	1.52	1.34	1.26	1.28	0.02	0.077
	Total globulin (g dl^{-1})	2.13	2.21	2.18	2.27	0.03	0.067
	Creatinine (mg dl^{-1})	0.38	0.49	0.44	0.48	0.02	0.063
	Urea (mg dl^{-1})	1.43	1.58	1.64	1.67	0.12	0.561
Enzymatic	AP (U l^{-1})	943.29	869.39	884.39	877.56	39.75	0.099
	GGT (U l^{-1})	19.4	21.5	21.9	26.7	0.96	0.095
	GPT (U l^{-1})	10.67	11.64	12.11	11.78	0.23	0.135
	GOT (U l^{-1})	175.8	180.1	182.6	177.3	2.46	0.334
	IgA (mg dl^{-1})	0.27	0.31	0.29	0.33	0.01	0.073
	IgM (mg dl^{-1})	2.19	2.27	2.25	2.28	0.07	0.097
	IgG (mg dl^{-1})	4.34	4.48	4.39	4.51	0.09	0.131

[a] Means in the same row with different superscripts differ significantly (P< 0.05). [b] Control= Represents the corn-soybean meal-full fat soy basal diet; Group I= Camelia (*Camelina sativa,* L. Crantz variety) oil at 2.5%; Group II= Camelia seeds at 5%, Group III= Camelia seeds at 10%.

were in agreement with what was reported by Lebarcq-Verheyden *et al.* (1974). Also, these results confirmed the findings of Swennen *et al.* (2005) in an experiment with broiler chickens reared on similar isocaloric diets with a protein-fat substitution.

The diets did not significantly affect the total level of Ig (IgA, IgM, or IgG), indicating that the treatment did not affect the humoral immune status of the broilers (Table 6). Liver function tests for GPT, GOT and total Ig concentrations produced similar results in all treatments. This finding is congruent with the similar reference values reported for chicks by Kaneko (1997).

CONCLUSIONS

Feeding 2.5% CS oil and CS seeds to chickens at levels of up to 5% of the diet has no adverse effects on performance and on carcass characteristics i.e. carcass yield, legs and breast proportions and abdominal fat. In addition, these *n*-3 PUFA-enriched diets had no effect on pH and chemical analysis of the breast muscle (dry matter and crude fat content). Chickens fed diets containing 10% CS seeds had significantly lower BW, proportion of abdominal fat in carcasses and bursa weights. The weights of the spleens and thymus did not differ among groups. Addition of CS oil and CS seeds to chicken diets significantly enriched the breast meat in total *n*-3 PUFA, mainly α-linolenic acid, EPA, DPA and DHA. The diets did not significantly affect plasma enzymes activity, and IgA, IgM, or IgG values. It can be concluded that by using oil and seeds rich in *n*-3 PUFA, it is possible to produce chickens meat with an improved omega *n*-3 fatty acids profile.

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مصرف روغن و بذر کاملیا (رقم *Camelina sativa* L. Crantz)به عنوان مواد غنی از اسید چرب n-3 در جیره غذایی جوجه کبابی و کارآیی آن در عملکرد، ترکیب اسید چرب، وزن بافت ایمنی، و پروفیل متابولیک پلاسما

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چکیده

هدف این پژوهش بررسی اثر روغن یا بذر کاملیا (*Camelina sativa* L. Crantz)رقم (CS)روی عملکرد، کیفیت گوشت، ایمنی و پروفیل متابولیک پلاسما در جوجه های کبابی بود. تعداد ۲۰۸۰جوجه از نوع Cobb 500به طور تصادفی انتخاب شدند (نسبت جنسی ۱:۱) و برای یک دوره آزمایشی ۳۲ روزه به ۴ گروه دسته بندی شدند شامل: شاهد(جیره غذایی ذرت-سویا- وجیره کاملا چرب سویا full fat soy based diet) ، گروه I(با افزودن ۲/۵٪ روغن CS)، گروهII(با افزودن ۵٪ بذر CS)، و گروه III(با افزودن ۱۰٪بذر CS). هر گروه ۵۲۰ جوجه داشت و به طور آزاد تغذیه می شد. برای تعیین پروفیل اسید های چرب مواد متشکله و ماهیچه سینه ازروش گاز گروماتوگرافی استفاده شد. در روز کشتار (روز ۴۲) لاشه ها به گونه ای ساده شده تجزیه شدند. غده تیموس، طحال و کیسه فابریسیوس به صورت ضد عفونی شده برداشت شده و وزن شدند. نتایج حاکی از آن بود که افزایش۲/۵٪ روغن CSو ۵٪ بذر CSتاثیر منفی روی عملکرد و مشخصات لاشه (وزن لاشه، نسبت ران و سینه). درجوجه هایی که جیره حاوی ۱۰٪ بذرCS دریافت کرده بودند وزن بدن جوجه ها به طور معنی داری ($P<0.001$)کاهش یافت. همچنین با افزایش مقدار بذر CSدر جیره غذایی، کاهش معنی داری ($P<0.05$)در نسبت چربی شکمی لاشه مشاهده شد. با این همه، جیره های غذایی دارای بذر و روغن CSمنجر به افزایش معنی دار در اسید های چرب امگا 3-nبه ویژه آلفالینولنیک اسید ($P<0.001$)، eicosapentaenoic acid *($P<0.0301$)*، و docosahexaenoic acid ($P<0.0026$)و *docosapentaenoic acid* ($P<0.0123$) در ماهیچه سینه شد. جیره های غذایی مزبور تاثیر معنی داری روی وزن طحال و غده تیموس ، فعالیت آنزیم پلاسما و کل سیستم ایمنی (Ig) نداشتند. بررسی جزئیات انرژی پلاسما در گروه هایی که روغن یا بذر CS در جیره غذایی دریافت کرده بودند گرایشی ($P<0.066$)در جهت افزایش تری گلیسیرید و کاهش معنی دار ($P<0.019$)کلسترول کل و اجزای آن ($P<0.001$)نشان داد .