

## Effect of linseed co-extrudates addition into a broiler chickens diet on fatty acid composition of leg meat

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### SUMMARY

The experiment was conducted in order to investigate the influence of diet supplemented with linseed co-extrudates on fatty acid composition and sensory quality of chicken leg meat. Broiler chickens were fed a mash diet until slaughter at 35 days of age. Two diets, control (C) and linseed (Llin), were assessed with the aim of increasing the content of n-3 polyunsaturated fatty acids and evaluating their influence on proximate composition and sensory properties of leg meat. The L diet was formulated with two types of co-extrudates (5%), linseed-soybean meal (starter diet) and linseed-sunflower meal (finisher diet). 120 broiler chickens were assigned to each diet. The use of L diet enriched significantly ( $P<0.05$ ) the content of  $\alpha$ -linolenic (ALA) (3.37 % vs. 8.24%), EPA and DPA (EPA+DHA, 0.16% vs. 0.33%) fatty acids in meat and drastically reduced the n-6/n-3 ratio (10.5 to 3.55). Sensory attributes of roasted leg meat samples were negatively affected by supplementation with linseed co-extrudates.

**Keywords:** broiler chickens, leg meat, linseed, co-extrudates, fatty acids.

### INTRODUCTION

Omega-3 fatty acids have the beneficial influence on human health, including anti-atherogenic, anti-thrombotic and anti-inflammatory effects, as well as reduction of risk of coronary heart disease (CHD) (Ruxton et al., 2004; Givens and Gibbs, 2006).

Fat is one of the essential substances in human and animal nutrition, as it is an important source of energy, essential fatty acids and liposoluble vitamins (Krejčí-Treu et al., 2010). A number of experimental studies show that animal fat-rich diets are directly related to the risk of cardiovascular diseases and colon cancer (Jiménez-Colmenero et al., 2001; Roynette et al., 2004). However, meat is a primary source of dietary fat, especially saturated one. Hence, the

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increasing interest in meat fatty acid composition stems mainly from the need to find ways to produce healthier meat, i.e. with a higher ratio of polyunsaturated fatty acids (PUFA) to saturated fatty acids (SFA) and a more favorable balance between n-6 and n-3 PUFA (Wood et al., 2003).

It has been observed that the content of fatty acids in meat of food animals (including broiler chickens) depends largely on the content and composition of these acids in the diet (Schiavone et al. 2004; Krejčí-Treu et al., 2010). Due to its high  $\alpha$ -linolenic acid (ALA), linseed has gained attention as a dietary ingredient for animals (Nam et al., 1997; Wood et al., 2003; Juárez et al., 2011). However, its usage in animal nutrition is limited, due to the presence of antinutritive components - cyanogenic glycosides. One of the ways for detoxification of linseed is an extrusion process (Wu et al., 2008). Additionally, high oil content in linseed may cause extrusion difficulties. In order to overcome this problem, linseed is usually co-extruded with protein rich materials, which show great ability of oil adsorption (Thacker et al., 2004; Lević and Sredanović, 2012)

The objective of this study was to determine the influence of the consumption of diet formulated with linseed co-extrudates on fatty acid composition and sensory quality of chicken leg meat.

## MATERIAL AND METHODS

### *Experimental design and diets*

Broiler chickens were fed with a mash diet. A starter diet was given until 21 days, followed by a finisher diet, until slaughter at 35 days of age. Two different diets were assessed: control (C) and linseed diet (L). For L diet, two types of co-extrudates were prepared. Co-extrudate linseed-soybean meal (5%) was used for preparation of starter mash, since soybean meal has less content of cellulose than sunflower meal, which is limitation factor in breeding of young broiler chickens. Finisher mash was prepared with 5% of co-extrudate linseed-sunflower meal.

120 broiler chickens were assigned to each diet. Chickens were divided in four boxes for every diet (30 birds in one box), in order to eliminate the influence of breeding environmental conditions on broiler performances.

The animals were slaughtered in a commercial abattoir, according to the routine procedure. After chilling (24h *post mortem*), the carcasses of twelve birds per treatment (three per box) were transferred under refrigerated conditions (0-4°C) to the laboratory of Institute of Food Technology (FINS) in Novi Sad. The birds were chosen as a representatives of their experimental group and box, taking into account average mass of 30 birds from each box. After cutting and deboning, the leg meat samples were subjected to

physicochemical and sensorial analysis, while the rest of the samples were homogenized and stored at -20°C, pending fatty acid analysis.

#### *Physicochemical and sensory analysis*

Moisture, fat, protein (Kjeldahl N x 6.25) and ash contents in leg meat samples were quantified using the ISO recommended standards 1442:1997 (ISO, 1997), 1443:1973 (ISO, 1973), 937:1978 (ISO, 1978) and 936:1998 (ISO, 1998), respectively.

In order to analyze sensory characteristics of leg meat, the samples were roasted in a convection air oven at 175°C for 45 min. and cooled to room temperature for 1 h. Six trained panelists, experienced in the sensory evaluation of various meats were engaged. Sensory evaluation (appearance-color and smell-taste) was carried out according to the point system of analytical descriptive test using a scale from 1 to 5 (1-unacceptable, 5-excellent).

#### *Fatty acid analysis*

Supercritical fluid extraction with CO<sub>2</sub> was used for preparation of fat extracts. Extractions were performed on a LECO TFA2000 fat analyzer (LECO Corporation, MI, USA). Temperature, pressure and extraction flow rates were adopted from existing procedures for meat samples (LECO Corporation, 2003).

Fatty acid methyl esters were prepared from the extracted lipids by transmethylation method that uses 14% wt. boron trifluoride/methanol solution (Sigma Aldrich, MO, USA), as recommended method for this type of samples (Karlović and Andrić, 1996). Nitrogen gas (99.99%, Messer, Germany) was used for removing boron trifluoride/methanol solution and n-heptane (99.99%, J.T. Baker, NJ, USA) from fatty acid methyl esters. Obtained samples were analyzed by a gas chromatograph Agilent 7890A system (Agilent Technologies, Santa Clara, CA, USA) with flame ionization detector (GC-FID), auto-injection module for liquid, equipped with fused silica capillary column (DB-WAX 30 m, 0.25 mm, 0.50 µm). The oven temperature was programmed at 90°C for 4 min, increased from 70 to 150°C at rate of 15°C/ min and then held at 190 °C for 5 min. Carrier gas was helium (purity > 99.9997 vol %, flow rate = 1.26 ml/min, produced by Messer, Germany). The fatty acids peaks were identified by comparison of retention times with retention times of standards from Supelco 37 component fatty acid methyl ester mix and with data from internal data library, based on previous experiments and fatty acid methyl ester determination on GC-MS. Results were expressed as percentages of total fatty acids.

### Statistical analysis

One way (ANOVA), Post-hoc (Duncan test) was performed using the software package Statistica 12.0 for Windows (Stat Soft, Tulsa, Oklahoma, USA). Differences were considered significant at  $P < 0.05$ .

### RESULTS AND DISCUSSION

The obtained results of proximate chemical composition (Table 1) of leg meat showed a significant influence ( $P < 0.05$ ) of diet formulated with linseed co-extrudates (L) only on fat content in leg meat. Therefore, fat content in leg muscles of L group chickens was higher comparing to C group, being 1.75% and 1.63%, respectively.

Table 1. Proximate leg meat chemical composition of control (C) and linseed (L) diet fed broiler chickens

Group	Moisture (%)	Fat (%)	Protein (%)	Ash (%)
C	74.8 <sup>a</sup>	1.63 <sup>a</sup>	17.4 <sup>a</sup>	1.13 <sup>a</sup>
	±0.44	±0.01	±0.17	±0.05
L	75.1 <sup>a</sup>	1.75 <sup>b</sup>	17.5 <sup>a</sup>	1.10 <sup>a</sup>
	±0.42	±0.02	±0.27	±0.07

<sup>a, b</sup> Means within the same column with different superscript letters are different ( $P < 0.05$ ).

If compare fatty acid compositions of starter C and L diet (Table 2), it is obvious that linseed based diet contained almost 13% more essential ALA in comparison with C (18.6% vs. 5.98%). Statistically significant difference ( $P < 0.05$ ) also occurred in content of C18:2 n-6, which amounted to 51.1% in the control diet and to 38.0% in the experimental diet. n-6/n-3 ratio in starter control diet was approximately four times higher than in experimental diet (Table 2), which was consequence of C18:2 n-6 and ALA contents in the diets. Nevertheless, there were no significant ( $P > 0.05$ ) differences in contents of SFA, MUFA and PUFA.

Similarly, much higher content of ALA (almost 11%) was found in finisher L diet then in C (17.1 vs. 6.52). Also, the content of C18:2 n-6 was higher in finisher C diet (51.8% vs. 43.9), resulting in three times lower n-6/n-3 ratio in finisher L diet.

Fatty acids profile of leg muscles from broiler chickens fed a control and linseed diet is presented in Table 3. Dietary linseed increased ALA content of leg meat from 3.37% in control samples to 8.24% in experimental samples, which also affected n-6/n-3 ratio in leg meat of fed broilers. A more balanced dietary intake of n-6 relative to n-3 is desired for optimal health. Consuming

higher quantities of n-3 PUFAs is one approach to normalizing high n-6/n-3 ratios.

Table 2. Fatty acid composition of the diets used in the experiment

Fatty acids (%)	Starter C	Starter L	Finisher C	Finisher L
C 14:0	0.09	0.15	0.08	0.08
C 16:0	11.9	11.4	11.4	10.3
C 16:1	0.17	0.19	0.10	0.10
C 18:0	4.91	6.16	4.99	5.03
C 18:1 n-9	25.0	24.9	24.3	22.9
C18:2 n-6	51.1	38.0	51.8	43.9
C18:3 n-3	5.98	18.6	6.52	17.1
C 20:0	0.45	0.41	0.40	0.29
C 20:1	0.15	0.21	0.14	0.14
C 22:0	0.20	0.02	0.21	0.19
C 22:1 n-9	0.08	0.06	0.06	0.05
SFA	17.5	18.1	17.1	15.9
MUFA	25.4	25.4	24.6	23.2
PUFA	57.0	56.5	58.3	60.9
PUFA/ SFA	3.25	3.13	3.41	3.84
n-3	51.1	38.0	51.8	43.9
n-6	5.98	18.6	6.52	17.1
n-6/n-3	8.54	2.04	7.93	2.57

SFA – saturated fatty acids, MUFA – monounsaturated fatty acids, PUFA – polyunsaturated fatty acids, EPA - eicosapentaenoic acid, DHA – docosahexaenoic acid

There is mounting evidence that an increase in dietary n-3 PUFAs, is effective in treating and lowering the risk of developing inflammatory-related conditions such as arthritis, inflammatory bowel disease, asthma, sepsis, and cardiovascular heart disease (CVD) (McDaniel et al, 2013). According to our results, intake of linseed decreased n-6/n-3 ratio in leg meat from 10.5% to 3.55%. Related to cardiac health, several international organizations have recommended a dietary n-6/n-3 ratio of 4/1 to 7.5/1 to decrease the risk of CVD (McDaniel et al, 2013). Ratio of 10/1 was linked to negative consequences In the Lyon Heart Study, while the ratio of 4/1 led to a 70% decrease in total mortality from cardiovascular disease at the end of two years (Broughton et al., 1997).

The results of the sensory evaluation of roasted leg meat samples of C and L diet fed broiler chickens show a significant effect of linseed (Fig. 1). Both sensory attributes (appearance - color and smell - taste) gained lower scores for the samples obtained from L diet fed animals. The difference were particularly pronounced ( $P<0.05$ ) for smell - taste, as this sensory characteristic was graded 1.83 units lower for F group samples. This finding is in concordance

with results previously reported by several authors (Wood et al., 2003; Azcona et al., 2008; Woods et al., 2009; Juárez et al., 2011), who observed the negative effect of dietary linseed on sensory attributes of meat.

Table 3. Leg meat fatty acid composition of control (C) and linseed (L) diet fed chickens

Fatty acids (%)	C	L
C14:0	0.39	0.36
C16:0	19.4	18.9
C18:0	2.74	3.60
C16:1	7.77	6.50
C18:1 n-9	29.1	31.8
C18:2 n-6	23.6	29.1
C18:3 n-6	11.8	0.19
C18:3 n-3	3.37	8.24
C 20:0	0.13	0.14
C20:1	0.20	0.21
C20:2	0.29	0.23
C20:4 n6	1.10	0.50
EPA+DHA	0.16	0.33
SFA	22.6	23.0
MUFA	37.1	38.5
PUFA	40.1	38.2
PUFA/SFA	1.77	1.66
n-3	3.37	8.24
n-6	35.9	29.3
n-6/ n-3	10.5	3.55

SFA – saturated fatty acids, MUFA – monounsaturated fatty acids, PUFA – polyunsaturated fatty acids, EPA - eicosapentaenoic acid, DHA – docosahexaenoic acid

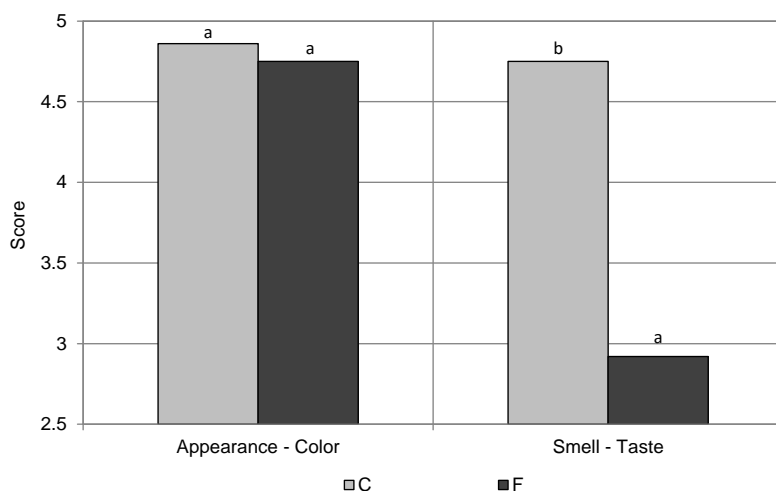


Figure 1. Leg meat sensory characteristics of control (C) and linseed (L) diet fed broiler chickens

## CONCLUSIONS

The use of linseed co-extrudates diet increased the content of essential  $\alpha$ -linolenic (ALA), EPA and DPA fatty acids in meat and drastically reduced the n-6/n-3 ratio (10.5 to 3.55). On the other hand, the supplementation with linseed negatively affected sensory attributes of roasted leg meat samples.

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