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Growth Performance, Carcass Traits and Breast Meat Fatty Acids Profile of Helmeted Guinea Fowls (*Numida meleagris*) Fed Increasing Level of Linseed (*Linum usitatissimum*) and Pumpkin Seed (*Cucurbita moschata*) Meals

ABSTRACT

This study was conducted to determine the effects of dietary inclusion with linseed and pumpkin seed meals on growth performance, carcass traits and breast meat fatty acids profile of helmeted guinea fowls. A total of 120 meat-type helmeted guinea fowl females of 12 weeks of age were fed with 0 (T0), 100 (T1) and 200 (T2) g/kg of linseed (LM) and pumpkin seed meals (PSM) for 56 days, in a completely randomized design with 10 replicates per treatment and four birds per replicate. The inclusion up to 200 g/kg of LM and PSM did not affect the livability, initial live weight and feed intake ($p>0.05$). However, final live weight and feed:gain ratio improved significantly ($p<0.05$). The T1 increased ($p<0.05$) the carcass yield and the T2 improved the breast weight and weight and yield of leg. Also, these experimental diets did not affect the carcass weight, breast yield and sensory quality of meat ($p>0.05$). The oleaginous seeds (LM and PSM) decreased ($p<0.05$) the myristic, palmitic and octadecanoic acids and the ω -6/ ω -3 ratio, as well as increased the linoleic, α -linolenic, eicosapentanoic and docosahexanoic acids ($p<0.05$), but did not modify the concentration of monounsaturated fatty acids ($p>0.05$) and the eicosatrienoic and arachidonic acids in breast meat ($p<0.05$). It is recommended the inclusion of 100 g/kg of LM and PSM to improve the live weight, weight and yield of the edible portions and the essential fatty acids in breast meat of guinea fowl, without affecting the sensory quality of the meat.

INTRODUCTION

Currently, lipid metabolism disorders are considered a serious health problem in the world population, mainly causing obesity, insulin resistance, inflammation, coronary artery disease and type 2 diabetes (Cao, 2014). The main risk factors for these types of diseases are high body mass index, consumption of alcohol and tobacco, lack of physical activity, low fruit and vegetable intake, high saturated fat intake and low in essential fatty acids, and high n-6/n-3 ratio (Yin *et al.*, 2017). In this sense, many international organizations have recommended the daily consumption of essential fatty acids omega 3 and omega 6, as well as a close ratio between them to stimulate brain development and decrease the induction of different types of cancer and metabolic diseases (British Nutrition Foundation, 1992; World Health Organization/FAO, 1995).

Poultry diets have been modified to incorporate the essential fatty acids in meat and egg, with the aim of incorporating these fatty acids and reducing harmful lipids (cholesterol and triacylglycerides) (Kanakri *et al.*, 2017), which favors the nutritional quality of the end product and offers greater benefits to the consumer (Simopoulos, 2002; Aguilar *et al.*, 2011).



Thus, the inclusion of essential fatty acids, such as α -linolenic and linoleic in poultry diets, reduces very-low-density lipoprotein (VLDL), low-density lipoprotein (LDL), triacylglyceride (TAG) and cholesterol, and decrease the atherogenic index and endothelial dysfunction in birds (Ayerza & Coates, 2000). Oilseeds such as linseed, rape (*Brassica napus* L.), chia (*Salvia hispanica* L.) and pumpkin which are rich in proteins are the main sources of these fatty acids of plant origin, and have been effectively used for their hypolipidemic action in poultry, as well as to increase the growth performance and the immune status of the animals (Aguilar *et al.*, 2011; Banaszkiwicz, 2013; Chen *et al.*, 2014; Apperson & Cherian, 2017). In addition, the use of these oilseeds can replace conventional energy and protein sources in diets and increase profitability. However, these feeds have biologically active compounds usually referred to as antinutritional factors when used in high concentration in poultry diets (Woyengo *et al.*, 2017).

The guinea fowl (*Numida meleagris*) is a bird native to the African continent (Central and Eastern Africa); it derives its name from the Coast of Guinea where it is believed to have originated (Teye & Abubakari, 2007). It has a high resistance to disease, it is not very demanding in terms of its diet and is highly prized for its meat and egg production (NCR, 1991; Nahashon & Tufarelli, 2012). In this sense, guinea fowl meat, as an alternative to chicken, has already proven to be a cost-effective activity in the United States, Canada, and Latin America (Tufarelli *et al.*, 2007; Laudadio *et al.*, 2012). However, to our knowledge few researches have been developed to enrich their meat with essential fatty acids and especially using linseed and pumpkin seed meals. The objective of this work was to determine the effects of dietary inclusion of linseed and pumpkin seed meals on growth performance, carcass traits and breast meat fatty acids profile of helmeted guinea fowls.

MATERIALS AND METHODS

Location

The Animal Care and Use Committee of the Faculty of Veterinary Medicine, National University of Pedro Ruiz Gallo, Lambayeque, Peru (Universidad Nacional Pedro Ruiz Gallo), approved the experiment. The experiments were carried out at the Poultry Experimental Unit, Lambayeque, Peru. An average relative humidity of 71%, and average minimum and maximum temperatures of 18.4 °C and 25.9 °C, respectively, was recorded using a hygro-thermometer placed inside the experimental house.

Linseed and Pumpkin seed meals

Five samples of linseed and whole pumpkin seed of different lots of the *Linum usitatissimum* species, reddish-brown Canadian variety and *Cucurbita moschata* species, Duchesne variety stored for 5 and 10 months, respectively, were collected, ground, and mixed into the diets. The chemical composition of linseed and pumpkin seed proposed by Ayerza & Coates (2000) and Martinez *et al.* (2010a), respectively, were taken into account for the preparation of the diets.

Birds and diets

A total of 120 meat-type helmeted guinea fowl females of 12 weeks of age were evaluated for 56 days. Birds were assigned to three dietary treatments according to a completely randomized design with 10 replicates per treatment and four birds per replicate.

Treatments consisted of diets containing 0 (T0), 100 (T1) or 200 (T2) g/kg of linseed meal (LM) and pumpkin seed meal (PSM). The experimental diets (Table 1) were

Table 1 – Ingredients and nutritional composition of the finisher (12-14 months) diets on as-fed basis.

Ingredients (g/kg)	Experimental diets		
	T0	T1	T2
Corn meal	642.0	601.7	555.7
Soybean meal	318.0	158.3	0.00
Pumpkin seed meal	0.00	100.0	200.0
Linseed meal	0.00	100.0	200.0
Dicalcium phosphate	18.40	17.00	14.70
Calcium carbonate	10.00	3.00	1.00
Salt	2.50	2.50	2.50
DL-Methionine	3.10	4.10	5.10
L-Lysine	0.00	4.20	8.60
L-Threonine	0.00	3.20	6.40
Choline	1.00	1.00	1.00
Vitamin and trace element premix ¹	5.00	5.00	5.00
<i>Calculated nutritional composition</i>			
Dry matter (g/kg)	928.2	925.4	922.9
Metabolizable energy (MJ/kg)	12.10	12.10	12.10
Crude protein (g/kg)	185.0	185.0	185.0
Ash (g/kg)	62.70	55.00	51.50
Calcium (g/kg)	10.00	13.40	18.60
Available phosphorus(g/kg)	4.00	4.00	4.00
Methionine (g/kg)	6.00	6.00	6.00
Lysine (g/kg)	9.90	9.70	9.70
Threonine (g/kg)	7.80	7.80	7.80
Crude fiber (g/kg)	30.90	46.10	61.30
Crude fat (g/kg)	29.10	61.00	92.70
Linoleic acid (g/kg)	12.90	30.40	47.90
Linolenic acid (g/kg)	0.50	17.8	35.10

¹Provided per kilogram of diet: vitamin A, 13500 IU; vitamin D₃, 3375 IU; vitamin E, 34 mg; vitamin K₃, 2.10 mg; vitamin B₁₂, 0.009 mg; vitamin B₂, 6.00 mg; riboflavin, 4.00 mg; pyridoxine, 3.00 mg; pantothenic acid, 16.00 mg; niacin, 56.00 mg; biotin, 0.10 mg; folic acid, 1.13 mg; choline, 2000 mg; manganese, 72.00 mg; zinc, 48.00 mg; iron, 60.0 mg; iodine, 0.80 mg; copper, 2000 mg; selenium, 0.20 mg; cobalt, 0.50 mg.



formulated to contain equal protein and energy levels and to supply the requirements recommended by Larbier and Leclercq (1994). The inclusion of LM and PSM was made at the expense of corn and soybean meals. A one-phase feeding program was used: finisher 12-14 weeks.

Experimental conditions

Each replicate consisted of a pen with deep corn stover litter. The pen was of 2 m x 6 m x 5 m including four helmeted guinea fowls. Feed and water were offered *ad libitum*, in tube feeders and nipple drinker, respectively. The light was at 12 hours per day. The adaptation period was two weeks and the birds were dewormed at the beginning of the research. No medication was added to the feed, nor were therapeutic drugs used during the experimental period.

Performance parameters

All birds were weighed at the start (12 weeks of age) and end of the experiment (14 weeks of age). Feed intake (g/bird/day) was measured daily during all the experimental weeks, and calculated as the difference between feed offer and feed residues. Feed conversion ratio was calculated as the amount of feed intake to gain one kg body weight. Livability was determined as the difference between the initial number of birds and recorded mortality.

Carcass traits

A total of 12 birds per treatment were sacrificed by bleeding of the jugular vein after four hours of feed fasting (water was offered *ad libitum*). Carcass weight was determined by weighing the birds before slaughter, after which breasts and legs plus thighs were weighed. Leg and breast samples were kept frozen at -20 °C.

Breast and leg sensory quality was determined by a panel of 30 tasters selected by the National University Pedro Ruiz Gallo (Universidad Nacional Pedro Ruiz Gallo). Meat samples were thawed, and cooked in water with no salt for 30 minutes at 80 °C (Ruiz *et al.*, 2001). Panel members in individual cabins were selected according to the following criteria: healthy,

non-smokers, did not have the habit of consuming coffee and/or alcoholic beverages, men and women between 25 and 55 years. The pieces of breasts and legs were served on plates with labels of different colors: blue for T0, green for T1 and light blue for T2. The sensory quality parameters surveyed were: aroma, flavor, tenderness, color (1=I really dislike it; 2=I am slightly displeased; 3=Neither like nor do I dislike me; 4=I like it lightly; 5=I like it a lot).

Fatty acids profile of breast

The extraction of the lipids from the breast of helmeted guinea fowls was carried out using a Soxhlet extractor. Ethyl ether anhydride was used as the solvent and the temperature was adjusted to 47 °C. To remove the residual anhydride ethyl ether in the samples, a Rotavapor Heidolph was used, with a temperature set at 38 °C, a pressure of 5 mm Hg and a speed of 90 to 100 rpm. Finally, the samples were placed in a solvent extraction hood in burners at 38 °C.

The quantification of the fatty acids was performed in Agilent gas chromatographer, Technologies 6890 (Palo Alto, California), equipped with flame ionization detector (FID) and fitted with autosampler HP 6890 Series. The equipment was controlled by a data operator GC Chemstation, version A.09.03. The fatty acids in the form of methyl-esters were separated in a capillary column HP-23 cis/trans (60 m x 250 mm ID x 0,25 mm layer thickness). The fatty acids profile was performed in the chemical laboratories at the Technological Institute of Production, Lima, Peru.

Statistical analysis

Data were submitted to one-way analysis of variance (Anova) according to a completely randomized experimental design. The test of Duncan (1995) was used to compare means, using the SPSS version 22.0 statistical package.

RESULTS AND DISCUSSION

Table 2 shows that livability (100%), feed intake and initial live weight was not influenced ($p>0.05$)

Table 2 – Growth performance in guinea fowls fed with increasing levels of linseed and pumpkin seeds.

Items	Inclusion levels of linseed and pumpkin seeds/g/kg			SEM±	p-value
	0	100	200		
Livability (%)	100.00	100.00	100.00		
Feed intake (g/bird/day)	131.21	122.97	127.95	1.891	0.061
Initial live weight (g)	1370.00	1375.36	1440.00	34.58	0.290
Final live weight (g)	1511.00 ^b	1618.00 ^a	1655.00 ^a	29.60	0.012
Feed:gain ratio (kg/kg)	4.95 ^a	4.40 ^b	4.45 ^b	0.130	0.008

^{a,b}Means within the same row with different superscript letters differ significantly ($p<0.05$).



by the dietary inclusion up to 200 g/kg of LM and PSM. However, final live weight and feed: gain ratio improved significantly ($p<0.05$) with the use of these oilseeds (LM and PSM).

Livability results (Table 2) show the lack of detrimental effects in linseed and pumpkin seed meals used in the present experiment in birds. These results coincide with Aguilar *et al.* (2011) and Ahmad *et al.* (2013) who did not find morbidity and mortality in the birds with levels of up to 100 g/kg of PSM and LM in the feed, respectively. Although, both oilseeds have secondary metabolites, high concentrations cause symptoms related to antinutritional factors, such as tannins, saponins and trypsin inhibitors and phytic acid, phytostereogenic and linamarin, respectively, and has been recommended by nutritionists up to 100 g/kg in the diet (Novak & Scheideler, 2001; Martínez *et al.*, 2010a). The use of up to 200 g/kg of LM and PSM had no negative influence on this productive indicator (Table 2), maybe due to their feeding habit and the experimental age of helmeted guinea fowls (Fraga *et al.*, 1997).

Although the feed intake was not different between treatments, the inclusion of LM and PSM rich in proteins, lipids and fiber increased the live weight and reduced the feed: gain ratio (Table 2). The contributions of the diet for the ethereal extract increased in 63.7 g/kg, due to the inclusion of up to 200 g/kg of these meals. One of the objectives of this research was to know the effect of diets rich in polyunsaturated fatty acids on growth performance of the guinea fowl with a digestive system that lacks a gallbladder (Kasperska *et al.*, 2012). This organ is important for the digestion of lipids and especially in birds, because it is clearly hepatic (Ravindran *et al.*, 2016). Unsaturated diets need less formation of micelles for the absorption of lipids, which could help in the digestion and absorption of this biomolecule and of other components of the ration, as well as helping the contribution of energy (Aguilar *et al.*, 2011).

Other results of Rosebrough *et al.* (1999) and Crespo & Esteve-García (2002) showed that an increase unsaturated fat level in bird diets, final live weight was higher than that obtained with the control treatment. Also, Mateos & Sell (1980), Latshaw (2008), Martínez *et al.* (2010a) and Aguilar *et al.* (2011) demonstrated a positive correlation between unsaturated fat levels and weight gain. As well as Murugesan *et al.* (2017) reported that lipids can improve feed palatability by masking undesirable flavors and reducing dust formation, thereby increasing feed efficiency. This

fact is important in guinea fowl, because the abrupt movement of the head to consume feed causes a significant loss of feed (in the form of dust) outside the feeders.

Also, a higher inclusion of LM and PSM in helmeted guinea fowl diets caused a higher fiber intake (from 30.90 to 61.30 g/kg). According to Savón *et al.* (2007), there is a high correlation ($R^2=0.75$) between dietary neutral detergent fiber (NDF) content and the volume they occupy in the gastrointestinal tract, which may trigger symptoms in poultry, reducing their voluntary feed intake. In addition, Walugembe *et al.* (2014) emphasize that the efficiency of fiber utilization in birds is related to intestinal transit time, which changes by poultry species. However, Martínez *et al.* (2015) shows that when unsaturated lipids are added to bird diets rich in fiber, they counteract the negative effect of these structural carbohydrates in the digestive system, because they may reduce the rate of intestinal transit. Aguilar *et al.* (2011), Martínez *et al.* (2015) and Kanakri *et al.* (2017) when including high concentrations of polyunsaturated fat in the bird diets reported similar results.

The inclusion of 100 g/kg of linseed and pumpkin seed meals increased ($p<0.05$) the carcass yield and the inclusion up to 200 g/kg improved the breast weight and leg yield. However, these experimental diets had no significant effect on carcass weight and breast yield ($p<0.05$).

It is known that the nutritive value of the guinea fowl meat is more favorable than broiler, especially as it has higher percentage of protein, essential amino acids and lean meat (Tufarelli *et al.*, 2015). Despite a greater contribution of lipids in the diet with up to 200 g/kg of linseed and pumpkin seeds this did not affect the carcass yield, on the contrary the inclusion with 100 g/kg improved this yield, perhaps due to the incorporation of polyunsaturated lipids and crude fiber, which increases lean meat by a reduction of VLDL and LDL, total cholesterol and serum triacylglycerols (Aguilar *et al.*, 2011).

The production of lean meat in poultry is one of the strategies of the market, the use of hypolipidemic ingredients and their effects on the key enzymes associated with lipid metabolism have shown a positive effect on carcass traits (Fouad & El-Senousey, 2014). In this sense, Lopez-Ferrer *et al.* (2001a), Lopez-Ferrer *et al.* (2001b) and Aguilar *et al.* (2011) found that diets rich in polyunsaturated fat improved the carcass and breast yields, respectively. In addition, according to Zubair & Leeson (1996) diets deficient in



protein and amino acids reduce lean meat in birds. In this sense, essential fatty acids can improve nutrient absorption and results of Tesseraud *et al.* (1999) showed that a higher absorption of lysine gradually increases breast weight compared to other muscles. However, we did not find an increase in breast yield (Table 3) with the new proposed feeds (LM and PSM) coinciding with other studies that using the inclusion levels of seeds and oils of LM and PSM in the diets

of the birds did not find beneficial effects on breast yield and other edible portions (Shen *et al.*, 2005; Pekel *et al.*, 2009; Hajati *et al.*, 2011; Mridula *et al.*, 2015; Wafar *et al.*, 2017), apparently related to the secondary metabolites and fiber levels in these seeds and their response on growth performance. Likewise, it was found that few researches have been done on the effect of these oilseeds (LM and PSM) in guinea fowl nutrition.

Table 3 – Carcass traits in guinea fowl fed with increasing levels of linseed and pumpkin seeds.

Items	Inclusion levels of linseed and pumpkin seeds (g/kg)			SEM±	p-value
	0	100	200		
Carcass weight (g)	1029.96	1127.78	1141.82	34.75	0.611
Carcass yield (%)	68.16 ^b	69.68 ^a	68.98 ^b	0.061	<0.001
Breast weight (g)	240.44 ^b	264.99 ^a	266.48 ^a	8.170	0.050
Breast yield (%)	23.34	23.49	23.34	0.037	0.009
Leg weight (g)	277.27 ^b	350.46 ^a	354.55 ^a	5.633	<0.001
Leg yield (%)	26.97 ^b	31.60 ^a	31.39 ^a	1.136	0.010

^{a,b}Means within the same row with different superscript letters differ significantly ($p < 0.05$).

The indicators for the evaluation of the sensory quality of the breast and leg of the helmeted guinea fowl fed with increasing levels of linseed and pumpkin seed are shown in Table 4. We did not find significant differences ($p > 0.05$) among the experimental treatments.

One of the main concerns of the use of foods rich in polyunsaturated lipids is the deterioration of the sensory quality of the poultry products (egg and meat), especially related to the induction of oxidative rancidity and the low concentration of natural antioxidants (like vitamin E) in the diets, which counteract this negative effect (Nkukwana *et al.*, 2014). It is known that fish oil easily becomes rancid due to its large polyunsaturated chains, such as eicosapentaenoic and docosahexaenoic fatty acids, and can cause unpleasant tastes in eggs and meat (Miyashita *et al.*, 2018). In this sense, Taga *et al.* (1984) and Lopez-Ferrer *et al.* (1999) found unpleasant odors in meats by including up to 82 g/kg fish oil in the feed. However, studies with oilseeds sources of linolenic and linoleic fatty acids in bird diets have not shown such effects, perhaps because the plants do not have

the enzymes delta 6 and 5 desaturase that elongates the polyunsaturated chains prone to the induction of the oxidative rancidity (Aguilar *et al.*, 2011). According to Shukla & Perkins (1998), linoleic and α -linolenic fatty acids are the most stable to oxidation within polyunsaturates, although they are more susceptible than saturated and monounsaturated fatty acids (Baudet *et al.*, 1984).

In this sense, other results using linseed and pumpkin seed meals in broiler found similar results (Gonzalez-Esquerria & Leeson, 2000; Lopez-Ferrer *et al.*, 2001a; Martínez *et al.*, 2010a; Aguilar *et al.*, 2011; Panda *et al.*, 2015). On the other hand, the inclusion of pumpkin seed meal in the diets with a high content of oleic acid (Martínez *et al.*, 2010a) maintained the sensory quality of breast, because it has been shown that this monounsaturated fatty acid has a high stability in cell membranes (Levental *et al.*, 2016). In addition, these oilseeds have a high concentration of unsaponifiable material and vitamin E, being the main antioxidant component of lipids (Herting & Drury, 1963; Stevenson *et al.*, 2007).

Table 4 – Sensory quality of breast and leg of guinea fowls fed with increasing levels of linseed and pumpkin seed.

Parameters	Inclusion levels of linseed and pumpkin seeds									
	Breast					Leg				
	T0	T1	T2	SEM±	p-value	T0	T1	T2	SEM±	p-value
Color	3.77	4.03	4.07	0.18	0.44	3.67	3.73	3.63	0.16	0.91
Aroma	4.06	4.10	3.70	0.17	0.25	4.03	4.07	3.77	0.14	0.26
Flavor	4.33	4.20	4.00	0.16	0.36	3.90	3.87	3.53	0.12	0.06
Tenderness	4.03	4.37	4.27	0.14	0.24	4.16	4.20	3.87	0.12	0.11

The values represent the opinion of the different panelists (30) that participated in the sensory evaluation.



Table 5 shows the profile of fatty acids in the breast of helmeted guinea fowl fed with different levels of linseed and pumpkin seed in the feed. Within the group of saturated fatty acids, myristic (C14:0), palmitic (C16:0) and octadecanoic acids (C18:0) are the most quantified and decrease ($p < 0.05$) with the inclusion of these oilseeds (LM and PSM) in the diet in relation to the control treatment. However, no significant differences were found for monounsaturated fatty acids, with oleic acid (C18: 1 ω -9) being the highest concentration for all treatments.

Likewise, the inclusion of LM and PSM in helmeted guinea fowl diets increased ($p < 0.05$) the concentration of linoleic (C18: 2 ω -6) and α -linolenic (C18: 3 ω -3) acids and the fatty acids derived from the elongation and desaturation process, such as eicosapentanoic (C20: 5 ω -3) and docosahexanoic (C22: 6 ω -3). However, the eicosatrienoic (C20:3 ω -3) and arachidonic (C20:4 ω -6) fatty acids did not show significant differences ($p > 0.05$) among treatments (Table 5). Also, the total concentration of the SFA decreased ($p < 0.05$) due to the effect of the oilseeds (LM and PSM) included up to 20% in the diet, with a progressive increase ($p < 0.05$) in the content of MUFA, PUFA, $\Sigma \omega$ -6 and $\Sigma \omega$ -3 and a significant reduction ($p < 0.05$) of the ω -6/ ω -3 ratio in the breast (Table 5).

The food intake with high content of saturated fats and low in polyunsaturated fatty acids is considered a risk factor for human health (Siri-Tarino *et al.*, 2015).

Our research corroborates the findings of Lopez-Ferrer *et al.* (2001a) and Martínez *et al.* (2010a), who found that these oleaginous seeds (LM and PSM) decrease the myristic, palmitic and stearic acids and in turn the SFA in birds, considered hypercholesterolemic fatty acids (Mir *et al.*, 2003). Therefore, this guinea fowl meat could be recommended for people with cardiovascular risk factors. In addition, other studies using seeds rich in linolenic and α -linolenic fatty acids such as chia (Ayerza *et al.*, 2002) and rapeseed (*Brassica napus* L.) (Kanakri *et al.*, 2018) showed similar results in poultry meat.

Monounsaturated fatty acids (MUFAs) are found in animal fats, olives, seeds, nuts and some vegetable oils (Schwingshackl & Hoffmann, 2014). The MUFAs remain without significant changes in the breast meat (Table 5), despite that the pumpkin seed has a high content of oleic fatty acid (Martínez *et al.*, 2010a). A decrease of stearic fatty acid (Table 5) precursor of oleic acid by the enzyme delta 9 desaturase (Lakhssassi *et al.*, 2017), could decrease this fatty acid in breast. Likewise, Cherian & Sim (1991) using 80 and 160 g/kg of linseed in the diets of laying hens observed a lower concentration of oleic acid in blood serum. Although, the mechanisms are not well known, Caston & Leeson (1990) had pointed out that the enzyme delta 9 desaturase, reduces its activity by including a high concentration of omega 3 fatty acids in the diets, thus the use of up to 200 g/kg (Table 1) of linseed (Lopez-

Table 5 – Fatty acids profile of breast in guinea fowls fed with increasing levels of linseed and pumpkin seed.

Fatty acids (mg/100 g)	Inclusion levels of linseed and pumpkin seed (g/kg)			SEM \pm	p-value
	T0	T1	T2		
C14:0	27.50 ^a	22.00 ^b	22.50 ^b	1.640	0.032
C16:0	965.50 ^a	811.00 ^b	831.50 ^b	46.24	0.028
C18:0	373.50 ^a	323.50 ^b	312.50 ^b	6.010	0.001
C18:1 ω -9	1213.00	1108.50	1268.50	91.98	0.500
C16:1 ω -7	123.50	88.50	112.50	11.18	0.157
C18:1 ω -7	43.00	35.50	39.50	2.220	0.135
C18:2 ω -6	732.50 ^c	809.50 ^b	927.00 ^a	57.68	0.035
C18:3 ω -3	22.50 ^b	225.00 ^a	208.50 ^a	37.29	0.015
C20:3 ω -3	90.50	77.00	99.50	5.200	0.068
C20:4 ω -6	80.52	81.25	79.82	0.952	0.079
C20:5 ω -3	ND ^c	5.12 ^b	8.64 ^a	1.561	0.047
C22:6 ω -3	ND ^c	15.00 ^b	17.58 ^a	2.179	0.013
Σ SFA	1366.50 ^a	1156.78 ^b	1165.50 ^b	53.22	0.015
Σ MUFA	1380.50 ^a	1232.20 ^b	1418.50 ^a	52.19	0.046
Σ PUFA	927.07 ^c	1213.82 ^b	1340.60 ^a	96.25	0.049
$\Sigma \omega$ -6	812.89 ^b	889.72 ^b	1005.98 ^a	47.68	0.035
$\Sigma \omega$ -3	112.58 ^b	321.89 ^a	334.10 ^a	33.43	0.005
ω -6/ ω -3	7.22 ^a	2.76 ^b	3.01 ^b	0.380	0.001

^{a,b,c}Means within the same row with different superscript letters differ significantly ($p < 0.05$)

ND: not detected; SFA: saturate fatty acids; MUFA: monounsaturated fatty acids; PUFA: polyunsaturated fatty acids; ω -6: ω -3.



Ferrer *et al.*, 2001a), could influence our results. Monounsaturated fatty acids have the highest oxidizing stability (Belingeri *et al.*, 2015), which research has shown that the replacement of saturated fatty acids by monounsaturates (Martínez *et al.*, 2010b, Praagman *et al.*, 2016) decreases the formation of LDL and the atherogenic index and as a result an increase of high density lipoproteins HDL.

Linseed and pumpkin seeds are found in the small and privileged group of oilseeds that possess essential fatty acids (Gómez-Cortés *et al.*, 2016; Potočnik *et al.*, 2016). The incorporation of essential fatty acids in helmeted guinea fowl meat increased according to the inclusion level of LM and PSM (Table 5). Authors such as Lopez-Ferrer *et al.* (2001a), Ayerza *et al.* (2002), Zelenka *et al.* (2008) and Apperson & Cherian (2017) reported an increase in α -linolenic acid by using the seed and linseed oil on bird diets, respectively. However, results of Martínez *et al.* (2010a) with the inclusion levels of 100 g/kg of pumpkin seed showed a slight increase of this essential fatty acid in the blood serum.

Likewise, in vegetable diets, the eicosapentanoic (C20:5n-3) and docosahexaenoic acids (C22: 6n-3) are detected when oilseeds are included as LM and PSM rich in α -linolenic acid precursor of these fatty acids. This could express that these feeds do not affect the activity of the enzyme delta 6 and 5 desaturase (with greater affinity for α -linolenic acid) that converts this fatty acid to EPA and DHA (Yary *et al.*, 2017). Although Grobas & Mateos (1996) have reported a low conversion of α -linolenic in DHA and EPA in laying hens. Apparently, guinea fowl have a greater enzymatic activity for the elongation and desaturation of omega 3, although other studies are necessary to justify this hypothesis. The EPA and DHA are important for the formation of cell membranes, essential for the growth and functioning of the organism (Kinsella *et al.*, 1990; Simopoulos, 2000).

Likewise, for the ω -6 family only the linoleic fatty acid (C18:2n-6) was found with notable differences, with 809.50 and 927.00 mg/100g in helmeted guinea fowl meat in the groups that received 100 and 200 g/kg of LM and PSM, respectively. Similar results were obtained by Martínez *et al.* (2010b), Chen *et al.* (2014) and Apperson & Cherian (2017), when diets were included with pumpkin, linseed and rapeseed seed, respectively. Likewise, Zelenka *et al.* (2008) found a progressive increase in the concentration of linoleic acid when using up to 70 g/kg of linseed oil in chicken diets. This shows a direct relationship

between the concentration of linoleic in the diet and its incorporation in the meat.

The content of arachidonic acid (C20:4n-6) did not decrease (Table 5) despite a higher incorporation of α -linolenic acid. According to Witte & Hardman (2015), the increase of the serum concentration of the omegas 3 can inhibit by competition the elongation and desaturation of the linoleic acid in its precursors, decreasing the arachidonic concentration. Studies in laying hens of Lopez-Ferrer *et al.* (2001a) and Lopez-Ferrer *et al.* (2001b) reported an indirectly proportional relationship of omega 3 fatty acids and the concentration of arachidonic acid. Apparently, the use of high percentages (up to 200 g/kg) of LM and PSM in the diets of helmeted guinea fowl, ingredients rich in lipids and omega 6 fatty acids, could cause these results (Table 5).

The European Commission Community Research (2000), specifies for humans a requirement of omega 3 fatty acids in the order of 1 g/day, however, Food and Nutrition Board in the United States (2005) reports an intake of 110-160 mg/day. In this sense, Simopoulos (2002) recommends an intake of 2.2 g/day of α -linolenic acid. Also, according to Kris-Etherton *et al.* (2002) the American Heart Association recommended for people with coronary heart disease a daily intake of DHA plus EPA of 900 mg/day. The helmeted guinea fowl fed up to 200 g/kg of LM and PSM has approximately 334.10 mg/100 g of omega 3, which turns this meat as a functional food, able to contribute to the daily need of these essential fatty acids in humans.

The ratio of omega 6/omega 3 (n-6/n-3) showed a tendency to decrease due to a higher inclusion of LM and PSM in the feed. This ratio decreased from 7.22 for the control to 2.76 for the diet added with 100 g/kg of LM and PSM (Table 5). Other authors found similar relationships (n-6/n-3), by the inclusion of linseed, rapeseed and chia seed in the diets of birds, justified to the content of α -linolenic acid in these oilseeds (Ayerza *et al.*, 2002; Apperson & Cherian, 2017; Kanakri *et al.*, 2018).

The World Health Organization (1995), in its report on fats and oils in human nutrition, recommended a n-6/n-3 ratio in the diet of 5 to 10/1 as advisable to prevent atherosclerosis and cardiovascular risk. However, the British Nutrition Foundation (1992) indicates a ratio of omega 6/omega 3 of 6/1, while Simopoulos (2000) recommended ratios of 2.1/1. The information reported in table 5 states that the values (2.76/1) are within the range proposed by these international references.



CONCLUSIONS

It is recommended the inclusion of 100 g/kg of linseed and pumpkin seed meals in partial replacement of corn and soymeal in the diet of helmeted guinea fowls (*Numida meleagris*) during the finisher phase (12-14 weeks) to improve the growth performance, weight and yield of the carcass and the omega 3 and 6 concentration in breast meat compared to the control treatment, without affecting the meat quality.

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