

EFFECT OF PARTIALLY REPLACING SOYBEAN MEAL AND SOYBEAN OIL WITH DEHULLED SUNFLOWER SEEDS ON MEAT QUALITY AND OXIDATIVE STABILITY OF BROILERS

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ABSTRACT

Broiler chicken (*Gallus gallus domesticus* L.) production in Mexico is fundamental for contributing to food security since it is inexpensive and has a high nutritional value. However, given the insufficient national production of ingredients such as soybeans to obtain oil and make balanced foods, it has been necessary to import them, thus increasing the cost of raising broilers. In this context, sunflower seed (*Helianthus annuus* L.), due to its nutritional makeup, can be used to replace soybean meal and soybean oil in broiler diets. The purpose of this study was to evaluate the effect of partially replacing soybean meal and soybean oil in broilers' diets with dehulled sunflower seeds on the physicochemical properties, fatty acid profile, and antioxidant capacity of meat. The experimental design was completely randomized: basal diet (sorghum-soybean meal); diet with 5 % sunflower seed and soybean oil; diet with 5 % sunflower seeds without soybean oil; and diet with 10 % sunflower seeds without soybean oil. There was no effect of the diet on the physicochemical characteristics ($p > 0.05$), but there was an effect on the fatty acid profile, which was better ($p \leq 0.001$) in the treatments with sunflower seed, with or without soybean oil. The antioxidant activity in raw and cooked meat decreased after 9 and 6 days of refrigerated storage ($p \leq 0.05$), respectively, without diet effect. Therefore, soybean meal can be substituted at 10 %, and soybean oil can be partially or completely replaced by dehulled sunflower seed in broiler diets as long as production parameters are not negatively affected.

Keywords: Carcass quality, chicken, fatty acid profile, nutrition, rancidity, shelf life.

INTRODUCTION

Broiler chicken (*Gallus gallus domesticus* L.) production in Mexico is fundamental for contributing to food security since it is the cheapest meat and has a high nutritional value. However, the insufficient national production of ingredients has made it necessary to import soybeans for oil and feed production, increasing the cost of raising broilers. In this context, sunflower seeds (*Helianthus annuus* L.), given their nutritional composition, represent an alternative to substitute soybean meal and soybean oil in

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broiler diets (Karkelanov *et al.*, 2020). Sunflower is grown in Mexico, and the amount imported is smaller than soybean (Bye *et al.*, 2009).

Research on the use of sunflower seeds and sunflower meal on broiler performance has shown positive results (Ciurescu *et al.*, 2019; Sosa-Montes *et al.*, 2021). Moreover, given that sunflower seeds have a high amount of polyunsaturated fatty acids (PUFA), their effect on the metabolism of broilers has been examined, concluding that it can decrease the ratio of saturated fatty acids in abdominal fat as well as breast and thigh meat (Kalakuntla *et al.*, 2017) while increasing PUFA contents (Mahfoudh *et al.*, 2016). The high fiber content of sunflower seeds is a disadvantage; therefore, it is recommended not to use in a proportion higher than 25 % (Sosa-Montes *et al.*, 2021). Alternatively, dehulling the seed can improve its digestibility (Karkelanov *et al.*, 2020). A 15 % inclusion level of sunflower meal in broiler diets has been proposed with no any negative effects on performance, or even higher levels with the addition of enzymes, which can improve body weight gain of broilers raised up to 35 days of age (Mbukwane *et al.*, 2022).

This work hypothesized that using dehulled sunflower seeds can enhance the fatty acid profile in broiler meat. Nevertheless, it could potentially reduce chicken meat production, especially when considering the added value of the meat. Its shelf life could be negatively affected since polyunsaturated fatty acids are highly susceptible to oxidation, requiring the addition of antioxidants. Based on these facts, the present study evaluated the effect of partially replacing soybean meal and soybean oil by dehulled sunflower seeds on the physicochemical characteristics, fatty acid profile, and antioxidant activity of broiler breast meat.

MATERIALS AND METHODS

Study site

The study was conducted at the Poultry Research Unit of the Experimental Farm of the Colegio de Postgraduados, Montecillo Campus, located in Texcoco, State of Mexico, Mexico (19.3° N, 98.53° W, at an altitude of 2250 m), under the international, national, and institutional guidelines for the care and use of animals according to NOM-033-ZOO-1995 (DOF, 1995) (human slaughter of domestic and wild animals) and approved by the Animal Welfare Committee of the Colegio de Postgraduados (02.11.16).

Animals, treatments, and feeding

A total of 200 eighty-one-day-old broilers (Ross 308[®]) were used, which were distributed into 28 experimental units of 10 broilers each and fattened for 49 days. Four treatments were randomly assigned to the 28 experimental units, with seven replicates per treatment. The treatments were: DBSS, basal diet based on sorghum-soybean meal; D5SO, diet with 5 % dehulled sunflower seeds + soybean oil + soybean meal; D10WO, diet with 10 % dehulled sunflower seeds without soybean oil + soybean meal; and

D5WO, diet with 5 % dehulled sunflower seeds without soybean oil + soybean meal. The sunflower seeds were dehulled to improve their digestibility and were added to the diet to replace soybean meal and soybean oil, according to the experimental treatments. All diets had isoproteic and isoenergetic contents. The ingredient and chemical composition of the diets and dehulled sunflower seeds are shown (Tables 1 and 2). The broilers had *ad libitum* access to feed and water.

Sampling

Fourteen broilers were randomly selected from each treatment (n = 56) and slaughtered at 49 days of age. Breast meat samples (*Pectoralis major*) were collected and stored in hermetically sealed polyethylene bags at -10 °C for laboratory analysis 48 hours after slaughtering.

Table 1. Ingredient composition of the experimental diets.

| Item | DBSS | D5SO | D10WO | D5WO |
|-----------------------------|-------|-------|-------|-------|
| Starter diet (0–21 days) | | | | |
| Sorghum | 53.09 | 53.88 | 54.53 | 56.72 |
| Soybean meal | 37.06 | 33.61 | 30.26 | 33.12 |
| DSS | 0.00 | 5.00 | 10.00 | 5.00 |
| Dicalcium phosphate | 2.08 | 2.05 | 2.03 | 2.04 |
| Calcium carbonate | 1.18 | 1.17 | 1.19 | 1.21 |
| Biolys® | 0.59 | 0.67 | 0.76 | 0.69 |
| Methionine | 0.45 | 0.45 | 0.44 | 0.44 |
| Threonine | 0.18 | 0.19 | 0.20 | 0.19 |
| Salt (NaCl) | 0.30 | 0.30 | 0.30 | 0.30 |
| Vitamin/mineral premix | 0.30 | 0.30 | 0.30 | 0.30 |
| Oil | 4.79 | 2.38 | 0.00 | 0.00 |
| Finishing diet (22–49 days) | | | | |
| Sorghum | 60.62 | 61.64 | 62.36 | 64.5 |
| Soybean meal | 30.13 | 26.64 | 23.20 | 26.14 |
| DSS | 0.00 | 5.00 | 10.00 | 5.00 |
| Dicalcium phosphate | 0.94 | 0.93 | 0.98 | 0.97 |
| Calcium carbonate | 1.58 | 1.56 | 1.54 | 1.55 |
| Biolys® | 0.37 | 0.46 | 0.55 | 0.47 |
| Methionine | 0.34 | 0.33 | 0.32 | 0.33 |
| Threonine | 0.08 | 0.09 | 0.11 | 0.09 |
| Salt (NaCl) | 0.30 | 0.30 | 0.30 | 0.30 |
| Vitamin/mineral premix | 0.30 | 0.30 | 0.30 | 0.30 |
| Oil | 5.00 | 2.41 | 0.00 | 0.00 |
| Coccidiostat | 0.05 | 0.05 | 0.05 | 0.05 |
| Pigment | 0.30 | 0.30 | 0.30 | 0.30 |

DBSS: basal diet based on sorghum-soybean meal; D5SO: diet with 5 % dehulled sunflower seeds + soybean oil; D10WO: diet with 10 % dehulled sunflower seeds without soybean oil; D5WO: diet with 5 % dehulled sunflower seeds without soybean oil; DSS: dehulled sunflower seeds.

Table 2. Chemical composition of the experimental diets and dehulled sunflower seeds.

| Item | DBSS | D5SO | D10WO | D5WO | DSS |
|---|-------|-------|-------|-------|--------------------|
| Dry matter (%) | 93.44 | 93.06 | 93.45 | 93.51 | 97.69 |
| Metabolizable energy (kcal kg ⁻¹) [†] | 3149 | 3148 | 3158 | 3019 | 4250 |
| Crude protein (%) | 21.68 | 21.69 | 21.70 | 21.69 | 28.34 [†] |
| Ether extract (%) | 5.96 | 4.46 | 6.30 | 3.67 | 48.72 |
| Ash (%) | 5.71 | 5.33 | 6.01 | 5.82 | 4.69 |
| NDF (%) | 14.72 | 15.99 | 16.64 | 15.79 | 34.03 |
| ADF (%) | 14.39 | 15.28 | 16.32 | 15.60 | 29.09 |
| Fatty acid profile (g 100 g ⁻¹ fatty acids) | | | | | |
| Caproic acid (C6:0) | 8.72 | 0.59 | 0.37 | NI | NI |
| Tridecanoic acid (C13:0) | 5.32 | NI | NI | NI | NI |
| Palmitic acid (C16:0) | 11.40 | 11.27 | 13.86 | 7.61 | 5.83 |
| Stearic acid (C18:0) | 2.60 | 3.55 | 2.52 | 3.71 | 6.88 |
| Oleic acid (C18:1n9) | 22.13 | 26.72 | 27.58 | 21.04 | 21.52 |
| Linoleic acid (C18:2n6) | 45.62 | 56.86 | 52.03 | 51.14 | 63.90 |
| Arachidonic acid (C20:4n6) | NI | NI | NI | NI | 0.42 |
| Linolenic acid (C18:3n3) | NI | 0.81 | 3.62 | NI | NI |
| Eicosenoic acid (C20:1n9) | NI | NI | NI | 16.48 | NI |
| DGLA (20:3n6) | 4.17 | NI | NI | NI | NI |
| Behenic acid (C22:0) | NI | NI | NI | NI | 0.98 |
| Lignoceric acid (C24:0) | NI | NI | NI | NI | 0.26 |
| ∑SFA | 28.07 | 15.44 | 16.75 | 11.32 | 13.97 |
| ∑MUFA | 22.13 | 26.77 | 27.58 | 27.52 | 21.56 |
| ∑PUFA | 49.79 | 57.78 | 55.65 | 51.14 | 64.45 |
| ∑n-3 | NI | 0.81 | 3.62 | NI | NI |
| ∑n-6 | 49.79 | 56.97 | 52.03 | 51.14 | 64.45 |
| Antioxidant activity (mmol TroloxEq kg ⁻¹ sample) | 15.24 | 13.93 | 23.07 | 21.20 | 9.45 [‡] |

DBSS: basal diet based on sorghum-soybean meal; D5SO: diet with 5 % dehulled sunflower seeds + soybean oil; D10WO: diet with 10 % dehulled sunflower seeds without soybean oil; D5WO: diet with 5 % dehulled sunflower seeds without soybean oil; DSS: dehulled sunflower seeds; NDF: neutral detergent fiber; ADF: acid detergent fiber; NI: not identified; DGLA: dihomogamma linolenic acid; SFA: saturated fatty acids; MUFA: monounsaturated fatty acids; PUFA: polyunsaturated fatty acids; n-3: omega 3 acids; n-6: omega 6 acids. [†]Datum provided by EVONIK (Evonik Industries de México, S.A. de C.V.). [‡]Seeds treated with BHA (butylated hydroxyanisole) and BHT (butylated hydroxytoluene) at a ratio of 1 g kg⁻¹ sunflower seeds.

pH and color

The pH of the breast meat was measured 30 min after bleeding and 24 h post-mortem by inserting the electrode of a HI 99163 portable potentiometer (HANNA Instruments, Mexico) to a depth of 1 cm in the muscle. Immediately after, the color was measured in the internal part of the breasts using a Konica-Minolta colorimeter (CR-410 Sensing Inc.,

Tokyo, Japan). The results were reported using the color system of the International Commission on Illumination (CIE) for the trichromatic coordinates: lightness (L^*), red index (a^*), and yellow index (b^*).

Texture

The texture of raw and cooked breast meat was measured through shear force using a Warner-Bratzler blade and the TA-XT2i stable texture analyzer (Microsystems, Godalming, England) (Honikel, 1998). To do this, 10 rectangular prisms were cut ($4 \times 2 \times 0.7$ cm) parallel to the muscle fibers in each breast sample at an average temperature of 5°C . Five of these prisms were kept raw, and the others were cooked in water at 75°C for 30 min. The meat samples were refrigerated at 4°C until analysis. The evaluation conditions were: pre-essay speed of 4 mm s^{-1} , essay speed of 1 mm s^{-1} , post-essay speed of 5 mm s^{-1} , and 30 mm of distance. The shear force was expressed in newtons (N).

Water holding capacity

Five grams of finely chopped meat with 8 mL of sodium chloride solution 0.6 M were placed in ice for 30 min and centrifuged (Beckman Coulter J2-HS, CA, USA) (Honikel, 1998) for 15 min at 10 000 rpm. The released water was poured out and its volume was measured, and the difference was reported in millimeters of NaCl (0.6 M) solution retained per 100 g of meat.

Proximal analysis

The proximal and cellulosic fractions of the dehulled sunflower seeds and diets (Table 2) were determined according to the Association of Official Analytical Chemists (AOAC, 2003) and van Soest *et al.* (1991) methodologies. The contents of protein, fat, moisture, and collagen of each breast sample were determined using the FoodScan™ (FOSS, MN, USA) near infrared (NIR) spectrophotometer (Analytical AB, Sweden), according to the Official Method 2007.04 (AOAC, 2003). To do so, 200 g of meat were ground and homogenized in a food processor for 30 s, so the fat, connective tissue, and muscle fibers were distributed uniformly. The ground meat sample was placed in the glass cup inside the support of the instrument, and three readings were taken per sample.

Fatty acid profile

To measure the fatty acid profile of the lyophilized samples (Labconco Free Zone 6, Kansas City, KS, USA), the methylation technique proposed by Palmquist and Jenkins (2003) was used, where the fatty acids are shown as methyl esters. The fatty acids were determined with a gas chromatograph (Hewlett-Packard 6890, USA) with automatic injector and capillary column ($100\text{ m} \times 0.25\text{ mm} \times 0.20\text{ }\mu\text{m}$ width, Sp-2560, Supelco). To identify the fatty acids, the standard retention times were compared (Supelco 37 FAME Components) against those of the samples. The results were expressed in g of fatty acids contained in 100 g of fatty acids.

Antioxidant activity

The antioxidant activity of the raw and cooked broiler breast meat was determined using the conventional 2,2-diphenyl-1-picrylhydrazyl (DPPH) method described by Brand-Williams *et al.* (1995). This was evaluated at 0, 3, 6, and 9 days of refrigeration storage. All the samples were kept at 4 °C in hermetically sealed polyethylene bags. The amount of DPPH (Sigma Aldrich, USA) was determined using a visible UV light spectrophotometer (Cary 1-E Varian, USA) at 517 nm. To determine the antioxidant activity or free radical scavenging, a standard curve was used (absorbance reduction vs. Trolox concentration). Trolox ((S)-(-)-6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic, Sigma Aldrich, USA) reacts with DPPH, a stable free radical. The results were expressed as mmol of TroloxEq kg⁻¹ meat sample. The higher the TroloxEq, the higher the antioxidant activity of the sample (Normah and Hanapi, 2019).

Statistical analysis

The data were analyzed through analysis of variance (ANOVA) under a completely randomized experimental design. The Tukey test for mean comparison was performed at a $p \leq 0.05$ significance level. The data of the antioxidant activity were analyzed through the PROC MIXED procedure in SAS 9.4 (SAS Institute). The Schwarz and Akaike Bayesian information criteria were obtained to determine the most appropriate covariance structure for each variable. The variance components procedure was used for the repeated measures design.

RESULTS AND DISCUSSION

Physicochemical characteristics of the meat

The physicochemical characteristics of the breast meat samples were not significantly altered ($p > 0.05$) by the treatments (Table 3 and 4).

Although there was no effect of the treatments on the physicochemical characteristics of the meat, according to Cavitt *et al.* (2005), the texture of both raw and cooked meat in this study (19.6 and 26.6 N, respectively) is classified as extremely soft. In their study, these authors found an increase in broiler meat tenderness as post-mortem hours increased, from 0.25 to 24 h, with values from 78.9 to 35.9 N, classifying them as slightly tough to moderately tough. Those values are highly related to the ease of chewing and palatability (Jiang *et al.*, 2018).

Color, whose mean L* and a* values were 45.5 and 5.94, respectively, correspond to dark meat according to the classification by Soares *et al.* (2009) and Oda *et al.* (2009), which might be influenced by the composition of the diet (Wideman *et al.*, 2016), particularly by the contents of flavonoids, tocopherols, carotenoids, and chlorogenic acid that sunflower seeds contain (Velasco and Ruiz-Mendez, 2015; Pajak *et al.*, 2014). Wildermuth *et al.* (2016) point out that chlorogenic acid reacts oxidatively with proteins, forming greenish colorations that affect the color of meat, although the present study did not support this idea.

Table 3. Physical characteristics of breast meat of broilers (*Gallus gallus domesticus* L.) fed with or without dehulled sunflower seeds in their diet.

| Attribute | Treatments | | | | SEM | p |
|-----------------------------|------------|-------|-------|-------|-------|-------|
| | DBSS | D5SO | D10WO | D5WO | | |
| Shear force cooked meat (N) | 27.07 | 27.66 | 28.15 | 23.54 | 0.163 | 0.079 |
| Shear force raw meat (N) | 19.81 | 21.48 | 19.12 | 18.05 | 0.384 | 0.210 |
| Lightness L* value | 45.17 | 44.57 | 45.88 | 46.38 | 7.296 | 0.317 |
| Color a* value | 6.09 | 5.97 | 5.67 | 6.04 | 1.526 | 0.817 |
| Color b* value | 19.70 | 18.41 | 18.33 | 18.10 | 3.740 | 0.169 |
| pH (30 min post-mortem) | 6.26 | 6.28 | 6.17 | 6.23 | 0.030 | 0.404 |
| pH (24 h post-mortem) | 5.26 | 5.17 | 5.25 | 5.46 | 0.080 | 0.140 |
| WHC (%) | 25.10 | 31.65 | 30.19 | 32.46 | 1.161 | 0.323 |

DBSS: basal diet based on sorghum-soybean meal; D5SO: diet with 5 % dehulled sunflower seeds + soybean oil; D10WO: diet with 10 % dehulled sunflower seeds without soybean oil; D5WO: diet with 5 % dehulled sunflower seeds without soybean oil; SEM: standard error of the mean; WHC: water holding capacity; L*: lightness; a*: red index; b*: yellow index ($p \leq 0.01$). All attributes, except shear force, were measured in raw meat.

Table 4. Chemical composition of raw breast meat from broilers (*Gallus gallus domesticus* L.) fed partially with or without dehulled sunflower seeds in their diet.

| Fraction (%) | Treatments | | | | SEM | p |
|------------------|------------|-------|-------|-------|-------|-------|
| | DBSS | D5SO | D10WO | D5WO | | |
| Collagen | 0.75 | 0.70 | 0.71 | 0.68 | 0.007 | 0.229 |
| Protein | 23.20 | 23.35 | 23.31 | 22.92 | 0.772 | 0.534 |
| Fat | 2.00 | 2.10 | 2.09 | 2.00 | 0.008 | 0.727 |
| Moisture | 74.80 | 74.88 | 74.81 | 74.99 | 0.625 | 0.919 |
| Ash [†] | 1.19 | 1.02 | 1.24 | 0.96 | 0.021 | 0.315 |

DBSS: basal diet based on sorghum-soybean meal; D5SO: diet with 5 % dehulled sunflower seeds + soybean oil; D10WO: diet with 10 % dehulled sunflower seeds without soybean oil; D5WO: diet with 5 % dehulled sunflower seeds without soybean oil; SEM: standard error of the mean ($p \leq 0.01$). [†]Determined directly according to AOAC (2003). The other fractions were indirectly determined by near infrared (NIR) spectrophotometry, according to AOAC (2003).

The water holding capacity (WHC) values found in this study were lower than those obtained by other authors (Barbin *et al.*, 2015; Carvalho *et al.*, 2017). This is presumably due to the pH value close to the isoelectric point of meat at 24 h post-mortem, when the WHC was measured. Zhao *et al.* (2017b) reported an isoelectric point of 5.5 for pale soft exudative meat of broilers. This value was very close to the average pH

at 24 h post-mortem (5.29) (Table 3). At that point, the net charge of the protein is zero, meaning that the numbers of positive and negative charges on the proteins are essentially equal. Consequently, there is a reduction in the amount of water that can be attracted and held by those proteins (Ghimire and Parajuli, 2020). All treatments produced very similar pH of the breast meat ($p > 0.05$); however, from 30 min to 24 h post-mortem, these pH values decreased on average from 6.24 to 5.29 ($p \leq 0.05$). To this regard, Veeramuthu and Sams (1999) found that pH values decreased from 6.67 to 5.65 after 24 h postmortem in broiler carcasses.

The fact that the chemical composition of the breast meat samples was not different between treatments could indicate that the experimental broilers have the capacity to regulate their feed intake and therefore the nutrient intake, especially energy, whose levels are equaled when access is *ad libitum* and, consequently, protein and fat deposition in the muscle tend to be similar, independently of the diet. Kalakuntla *et al.* (2017) evaluated diets containing different polyunsaturated fatty acid sources and found no differences in the chemical composition of thighs and breasts. This is concordant with the fact that including dehulled sunflower seeds did not modify the chemical composition of the meat (Table 4).

Fatty acid profile

The fatty acid profile of the breast meat samples was influenced by the experimental diets (Table 5). Treatments D5SO, D10WO, and D5WO showed similar contents of polyunsaturated fatty acids (PUFA) ($p > 0.05$), which were higher than the content of meat samples under the DBSS treatment ($p \leq 0.05$). Contrarily, treatments D5SO, D10WO, and D5WO showed similar contents ($p > 0.05$) of saturated fatty acids (SFA), which were lower than the content of meat samples under the DBSS treatment ($p \leq 0.05$). The first trend with PUFA was also observed with the omega-6 fatty acids and the PUFA:SFA ratio. However, the MUFA content of meat under the D5WO diet was higher than the corresponding content under the other diets.

The addition of 5 and 10 % dehulled sunflower seeds with or without soybean oil caused higher concentrations of polyunsaturated fatty acids, as well as higher omega-3 and omega-6 content and lower levels of saturated fatty acids in the meat than the corresponding values of the DBSS diet. Consequently, these meat samples showed a higher PUFA:SFA ratio. It is important to point out that C18:3n3 is the fatty acid that is most used as an energy source throughout the muscle β -oxidation pathway (Smink *et al.*, 2010). The treatments had no effect on the concentrations of C18:0, C20:4n6, C20:2n6, and 20:3n6 fatty acids of the breast meat samples ($p > 0.05$). The C18:2n6 acid was the most abundant under treatments D5SO, D10WO, and D5WO ($p \leq 0.05$), since soybean oil and sunflower oil (Kalakuntla *et al.*, 2017) provide high amounts of polyunsaturated fatty acids, mainly C18:2n6 (Table 2).

Very long chain omega-3 fatty acids are a result of the lengthening and desaturation of 18:3n-3 acid, like the correspondent omega-6 fatty acids are formed from 18:2n6 acid. In this study, there was a higher concentration ($p \leq 0.05$) of omega-6 acids than

Table 5. Fatty acid profile of raw breast meat of broilers (*Gallus gallus domesticus* L.) fed partially with or without dehulled sunflower seeds in their diet.

| Fatty acid (g 100 g ⁻¹ fatty acids) | Treatments | | | | SEM | p |
|---|------------|----------|---------|----------|-------|-------|
| | DBSS | D5SO | D10WO | D5WO | | |
| Myristic acid (C14:0) | 0.53 a | 0.45 b | 0.43 b | 0.44 b | 0.003 | 0.001 |
| Palmitic acid (C16:0) | 22.73 a | 19.22 b | 19.10 b | 18.59 b | 0.001 | 0.001 |
| Palmitoleic acid (C16:1n7) | 1.90 b | 2.62 b | 2.53 b | 4.32 a | 0.689 | 0.001 |
| Stearic acid (C18:0) | 7.17 | 7.04 | 7.85 | 7.54 | 1.606 | 0.406 |
| Oleic acid (C18:1n9) | 25.35 c | 27.68 b | 27.71 b | 32.25 a | 5.84 | 0.001 |
| Linoleic acid (C18:2n6) | 26.00 b | 34.56 a | 34.77 a | 34.83 a | 8.30 | 0.001 |
| α -linolenic acid (C18:3n3) | 0.30 c | 1.78 a | 1.56 a | 0.53 b | 0.158 | 0.001 |
| Arachidonic acid (C20:4n6) | 2.80 | 3.27 | 3.55 | 2.77 | 1.791 | 0.403 |
| Eicosadienoic acid (C20:2n6) | 0.60 | 0.68 | 0.68 | 0.50 | 0.042 | 0.108 |
| DGLA (20:3n6) | 0.52 | 0.53 | 0.52 | 0.60 | 0.043 | 0.740 |
| Σ SFA | 31.56 a | 27.38 b | 27.53 b | 28.22 b | 4.540 | 0.001 |
| Σ MUFA | 28.44 b | 30.92 b | 30.85 b | 37.33 a | 7.510 | 0.001 |
| Σ PUFA | 34.01 c | 41.68 ab | 40.91 b | 41.10 ab | 6.50 | 0.001 |
| Σ n-3 | 0.45 b | 1.82 a | 1.58 a | 0.54 b | 0.152 | 0.001 |
| Σ n-6 | 30.55 b | 39.86 a | 40.33 a | 40.55 a | 6.30 | 0.001 |
| PUFA:SFA | 0.98 b | 1.52 a | 1.60 a | 1.46 a | 0.024 | 0.001 |

Means in a row with different letters indicate differences ($p \leq 0.01$). DBSS: basal diet based on sorghum-soybean meal; D5SO: diet with 5 % dehulled sunflower seeds + soybean oil; D10WO: diet with 10 % dehulled sunflower seeds without soybean oil; D5WO: diet with 5 % dehulled sunflower seeds without soybean oil. DGLA: dihomo-gamma linolenic acid; SFA: saturated fatty acids; MUFA: monounsaturated fatty acids; PUFA: polyunsaturated fatty acids; n-3, omega-3 acids; n-6, omega-6 acids; SEM: standard error of the mean.

omega-3 acids in the breast samples under treatments with dehulled sunflower seeds. The deposition and distribution of fatty acids in the meat depend on absorption, *de novo* synthesis, and β -oxidation of fatty acids, processes that in turn depend on the concentrations and types of fatty acids contained in the diet (Villaverde *et al.*, 2006). To this regard, Smink *et al.* (2010) found that diets with high concentrations of polyunsaturated fatty acids inhibit lipogenic enzymes ($\Delta 9$ desaturase) and decrease the *de novo* synthesis of saturated and monounsaturated fatty acids in chickens. Additionally, during absorption, there is a greater affinity of fatty acid-linking proteins in intestinal cells for polyunsaturated fatty acids (Ravindran *et al.*, 2016). The low percentage of polyunsaturated fatty acids in the breast meat samples of the DBSS treatment could be caused by the reduced concentration of unsaturated fatty acids in the diet. The Δ -5 and Δ -6 desaturase and elongase enzymes break the bonds of saturated fatty acids, insert double bonds, and elongate the chains of C18:2n6 and C18:3n3 acids (Wood and Enser, 2017). The DBSS treatment did not have enough substrate to carry out these functions. The acids C18:3n3 and C18:2n6 compete for the same enzymes and interfere in the elongation and desaturation processes (Betti *et al.*,

2009); at the same time, the β -oxidation pathway decreases their availability (Smink *et al.*, 2010).

It is important to point out that the diet of the D5WO treatment had a lower concentration of metabolizable energy compared with the other treatments. According to Villaverde *et al.* (2006), this promotes the synthesis of saturated and monounsaturated fatty acids to maintain an adequate ratio of saturated to unsaturated fatty acids in the cell membranes. As noted above, sunflower seed oil has a high concentration of polyunsaturated fatty acids. This fact was confirmed by Crespo and Esteve-García (2002), who found that the main acids resulting from hepatic lipogenesis are C16:0, C18:0, C18:1n9, and C16:1n7. For this reason, the breast meat samples under the D5WO treatment had a higher amount of monounsaturated fatty acids. Comparing the results of the breast samples under treatment D5SO against DBSO, it is deduced that soybean oil mixed with sunflower seed oil can improve (increase the polyunsaturated acids) of the meat.

Antioxidant activity

The antioxidant activity of the raw or cooked breast meat samples (Figures 1 and 2) was not different between treatments ($p > 0.05$), although it did vary throughout the time ($p \leq 0.01$). The antioxidant activity of meat, measured as the activity of free radical

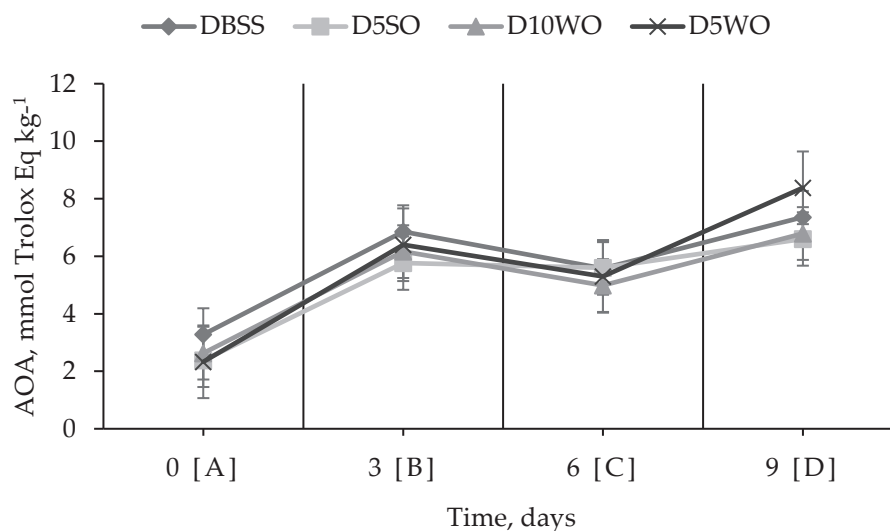


Figure 1. Antioxidant activity of raw breast meat of broilers (*Gallus gallus domesticus* L.) partially fed with dehulled sunflower seeds in the diet. DBSS: basal diet based on sorghum-soybean meal; D5SO: diet with 5 % dehulled sunflower seeds + soybean oil; D10WO: diet with 10 % dehulled sunflower seeds without soybean oil; D5WO: diet with 5 % dehulled sunflower seeds without soybean oil; AOA: antioxidant activity or free radical scavenging as Trolox equivalents. Means with different letters indicate differences ($p \leq 0.01$).

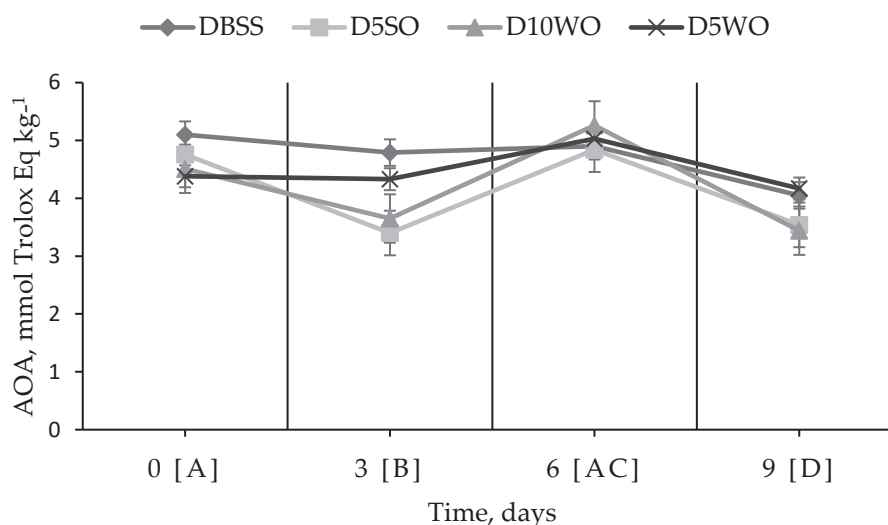


Figure 2. Antioxidant activity of cooked breast meat of broilers (*Gallus gallus domesticus* L.) partially fed with dehulled sunflower seeds in the diet. DBSS: basal diet based on sorghum-soybean meal; D5SO: diet with 5 % dehulled sunflower seeds + soybean oil; D10WO: diet with 10 % dehulled sunflower seeds without soybean oil; D5WO: diet with 5 % dehulled sunflower seeds without soybean oil; AOA: antioxidant activity or free radical scavenging as Trolox equivalents. Means with different letters indicate differences ($p \leq 0.01$).

scavenging, was only different over time. This could be attributed to the sunflower seed used in the experimental diets, which was treated with BHA (butylated hydroxyanisole) and BHT (butylated hydroxytoluene) at a ratio of 1 g kg^{-1} sunflower seeds, as was the commercial soybean oil, which also contained synthetic antioxidants. This suggests that the diets contributed significantly to the antioxidant activity of the meat due to their content of exogenous antioxidants (Sampaio *et al.*, 2012). To this regard, it has been reported that sunflower seeds contain phenolic acids, flavonoids, tocopherols, and carotenoids that function as potent natural antioxidants (Velasco and Ruiz-Mendez, 2015), although their effect on chicken meat is still unknown.

Oxidative rancidity is one of the main causes of food deterioration, measured by the low free radical scavenging activity in $\text{mmol TroloxEq kg}^{-1}$; this technique is used to predict the optimum storage time or shelf life of foods for humans and animals. According to the results, raw meat loses its oxidative stability on day 9 of storage in refrigeration ($4 \text{ }^\circ\text{C}$), while cooked meat does so on day 6. This result is probably because the heat treatment causes structural and functional changes in the meat. For example, heat denatures proteins, breaks cell membranes, and begins the degradation of endogenous antioxidants like vitamin C and vitamin E (Serpen *et al.*, 2011; Sampaio *et al.*, 2012).

When the meat contains high levels of polyunsaturated fatty acids, heat can produce lipid oxidation due to the susceptibility and instability of their double bonds. Lipid oxidation increases free radicals, which in turn decreases antioxidants (Zhao *et al.*, 2017a). Therefore, antioxidant activity or free radical scavenging activity decreases due to the excess of free radicals. These results agree with Serpen *et al.* (2011), who found a decrease in the antioxidant capacity of chicken meat as an effect of heat treatment. As time passes, the concentration of fatty acids in the meat decreases (Sampaio *et al.*, 2012), and secondary and tertiary compounds increase as a consequence of the oxidation processes. Cooked meat, unlike raw meat (Figures 1 and 2), shows lower free radical scavenging activity as time progresses because heat decreases the initial concentration of fatty acids (Serpen *et al.*, 2011), increasing the production of free radicals.

CONCLUSION

Including dehulled sunflower seeds in the diets of broilers did not modify the physicochemical characteristics of the meat but improved the fatty acid profile. Soybean meal can be substituted by 10 %, and soybean oil can be substituted partially or completely by dehulled sunflower seeds in the diet of broilers as long as the production parameters are not negatively affected.

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