The effects of replacing corn with low-tannin sorghum in broiler’s diet on growth performance, nutrient digestibilities, lipid peroxidation and gene expressions related to growth and antioxidative properties

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ABSTRACT
The purpose of this study was to examine the effects of substituting yellow corn with low-tannin sorghum on growth performance, nutrient digestibilities, and some gene expressions. Chickens were divided into the four groups: 1) the basal diet containing 100% yellow corn, 2) 25% yellow corn was replaced with low-tannin sorghum, 3) 50% yellow corn was replaced with low-tannin sorghum, and 4) 100% yellow corn was replaced with low-tannin sorghum. Body weight significantly improved (P < 0.05) by low-tannin sorghum substitutions, however, feed conversion ratio (FCR) was improved numerically (P = 0.672). Moreover, a significant decrease in the thiobarbituric acid reactive substance in muscle was found in all sorghum groups (P < 0.05). The mRNAs of genes related to growth and antioxidative properties insulin growth factor (IGF), β-Actin, fatty acids synthesis (FAS), glutathione peroxidase (GPX) and superoxide dismutase (SOD) were significantly increased due to low-tannin sorghum substitutions. Interestingly, feeding the 50% yellow corn + 50% low-tannin sorghum diet led to a significant increase in mRNA expressions of IGF, β-Actin, FAS, GPX and SOD in broilers. It could be concluded that substitution of 50% yellow corn with low-tannin sorghum might improve performance, modify plasma lipids and enhance the antioxidative status in broilers.

Abbreviations: IGF: Insulin growth factor; ACC: Acetyl-CoA carboxylase; FAS: fatty acids synthesis; GPX: glutathione pre-oxidation; HDL: high density lipoprotein; GOT: Glutamic-Oxaloacetic Transaminase; T3: triiodothyronine concentration; TBARS: thiobarbituric acid reactive substance; HPLC: High-performance liquid chromatography; TG: triglyceride; TC: total cholesterol; SOD: superoxide dismutase; LDL: low-density lipoprotein cholesterol; HMGCoA: 3-hydroxy-3-methylglutaryl CoA; TP: Total protein; T-AOC: total antioxidant capacity; MDA: malondialdehyde

1. Introduction
Broilers require a major proportion of grains in their diets to supply crude protein and metabolizable energy. Sorghum is the fifth most important crop after yellow corn, rice, wheat and barley (Bryden et al. 2009). Yellow corn remained the chief energy source in compounded diets and constituted about 50–60% of broiler ration (Ajaja et al. 2002) and it has similar nutritive value to that of sorghum grain (Hancock 2000) and wheat (Mikkelsen et al. 2008).

Sorghum is cultivated in low water areas and it can be produced in arid areas worldwide, as it is adapted to low-quality soils (Guaitieri and Rapaccinin 1990). In comparison with yellow corn, sorghum seed had higher rates of protein, while the energy or fat content of sorghum is relatively lower (Etuk et al. 2012). The amino acids profile of sorghum matches well with yellow corn. Also, the moderate digestible lysine content of sorghum is also similar to yellow corn. Sorghum seed is a prosperous exporter of diverse phytochemicals including phytosterols, anthocyanins, tannins and phenolic acids. These phytochemicals play a vital role in leverage animal authenticity (Awika and Rooney 2004). In vitro studies showed that sorghum has a superiority antioxidative properties which could be involved in offering health benefits to animals (Hahn et al. 1983). It was reported that sorghum consumption reduced the hazard of certain types of cancer in humans (Rhodes and Kresovich 2016). Moreover, the high concentricity of phytochemicals content in sorghum seed may be slightly responsible for these healthy benefits (Awika and Rooney 2004). These phytochemicals also promote cardiovascular health in human and animals (Anderson 2003).

Low-tannin sorghum may replace yellow corn as an alternative ingredient in broiler’s diet. Several studies showed that growth performance was not negatively affected by substitutions of yellow corn by low-tannin sorghum in pigs and poultry (Garcia et al. 2005; Campos 2006). On contrarily, Pour-
Reza and Edriss (1997) found a negative influence on growth performance of broilers when fed on high-tannin sorghum. Moreover, low-tannin sorghum has good beneficial effects on poultry health due to its phytochemical constituents (Awika et al. 2005). To our knowledge, very few previously conducted research has investigated the effect of substitutions of yellow corn diets with low-tannin sorghum on growth performance, plasma lipids profile and gene expression related growth and antioxidative properties in broiler chickens. Therefore, the objective of this study was to examine the effect of substitutions of yellow corn with low-tannin sorghum on growth performance, nutrient digestibilities, lipid peroxidation, and gene expression related growth and antioxidative properties in broiler chickens.

2. Materials and methods

This experiment and procedures were conducted in accordance with the guidelines of Animal Ethics Committee, Kaferelsheikh University, Egypt. All efforts were made to minimize suffering. A total of 100, 1-day-old, male broiler chickens (ROSS 308) were offered by a mercantile hatchery (Kagoshima Chickens Foods Co., Ltd., Kagoshima, Japan). The chickens were settled in an electrically heated battery brooder and offered free access to water and a commercial starter diet (22% crude protein and 3000 kcal/kg ME) until 12 days of age. On day 12, 28 male chicks with comparable body weight were chosen and housed individually in wire-bottomed aluminum cages. The birds were preconditioned for 3 days by feeding a basal diet. This system for choosing and grouping the chicks’ optioned to have very low standard error between treatments. The compositions of control and low-tannin sorghum diets used in this experiment are shown in Table 1. The chemical composition and mineral content of the low-tannin sorghum used in this study were as follows: (dry matter 92.5%, crude protein 8.7%, Ether extract 3.01%, calcium 0.051% and phosphorus 0.23%). The sorghum used in this experiment had low tannin content (0.56%). The 28 birds at 15 days of age were divided into 4 experimental groups: (i) the basal diet containing 100% yellow corn and served as control; (ii) 25% yellow corn was replaced with low-tannin sorghum; (iii) 50% yellow corn was replaced with low-tannin sorghum and (iv) 100% yellow corn was replaced with low-tannin sorghum. The broilers were fed on the experimental diets from 15 to 27 days of age. The trial was managed in a temperature-controlled room with a 14-h light/10-h dark cycles. The animal room temperature is preserved at 25 ± 1°C with proportional humidity between 50% and 70% through the experiment time.

2.1. Sampling

Chickens body weight (BW) and body weight gain (BWG) were registered every 3 days, and feed intake (FI) was recorded every day during the experimental period. At the end of the experimental period, the chickens were slaughtered and then dissected to gauge the organs weights (breast muscle ‘pectoral superficial muscle’, thigh muscle, abdominal fat and liver). Blood specimens were collected at heparinized test tubes and centrifuged at 5900×g for 10 min at 4°C to separate plasma and stocked at −30°C until tests.

Blood plasma concentrations of total cholesterol, triglycerides, high density lipoprotein (HDL-C), glucose, total protein and glutamic-oxaloacetic transaminase (GOT) in were analysed by automated Fuji DRY-CHEM 3500 (Fuji Medical Systems, Tokyo, Japan) according to the industrialist’s prescripts. Plasma triiodothyronine (T3) concentration was analysed by a commercial enzyme-immunoassay kits (ELISA-T, International Reagents Corp., Kobe, Japan) as reported previously by Hayashi et al. (1994). The analysis of muscle biochemistry and genes expression was performed in six samples per treatment. Breast muscle content of thiobarbituric acid reactive substance (TBARS) was analysed by the method of Ohkawa et al. (1979).

2.2. Nutrients digestibility

The diets and faeces were dried on oven at 105°C. After drying, all of the specimens were grinded and pushed through a 0.5-mm screen for smooth analysis of the dry matter content. Chromic oxide was measured according to the procedure qualified by Dansky and Hill (1952). The nitrogen content of the samples was fixed by using the Kjeldahl method. Ash digestibility was analysed according to the procedure described by AOAC (2005).

Calculation of digestibility

The following equation was used for the calculation of nutrient digestibility:

$$\text{Digestibility} = \frac{\text{Intake} - \text{Output}}{\text{Intake}} \times 100$$

Table 1. Composition of the experimental diet*.

| Ingredient | Basal diet | Sorghum diets |
|------------|------------|---------------|
|            | 0%         | 25%           | 50% | 100% |
|            | g/kg diet  | g/kg diet     | g/kg diet | g/kg diet |
| Yellow Corn| 566        | 425           | 287 | –    |
| Sorghum    | 143        | 283           | 566 | –    |
| Soybean meal, 46% | 346 | 344 | 342 | 346 |
| Corn oil   | 50.2       | 50.2          | 50.2 | 50.2 |
| CaHPO₄     | 20         | 20            | 20   | 20   |
| DL-Methionine | 1.2 | 1.2           | 1.2  | 1.2  |
| CaCo₃      | 6.6        | 6.6           | 6.6  | 6.6  |
| Vitamins and minerals mixture | 5 | 5 | 5 | 5 |
| Nac        | 5          | 5             | 5    | 5    |
| Nutrient composition (calculated values) | | | | |
| Metabolizable energy, Mcal/kg | 3.2 | 3.2 | 3.2 | 3.2 |
| Crude protein, % | 19.60 | 19.62 | 19.62 | 19.64 |
| Calcium, % | 1.10 | 1.11 | 1.12 | 1.12 |
| Total phosphorus, % | 0.80 | 0.81 | 0.81 | 0.82 |
| Sodium, % | 0.261 | 0.262 | 0.262 | 0.263 |
| Chloride, % | 0.25 | 0.25 | 0.25 | 0.25 |

*The mixture minerals supplied (mg/kg feed) of iron sulfate, 400; copper sulfate, 31.5; zinc sulfate, 176; manganese sulfate, 152; sodium iodate, 0.55; sodium selenium, 0.27. The mixture vitamins supplied (mg/kg feed): retinol, 1.4; DL-α-tocopherol acetate, 6.5; thiamine hydrochloride, 2.6; riboflavin, 6.5; pyridoxine hydrochloride, 1.30; calcium D-pantothenate, 10.4; nicotinic acid, 26; (mg/kg feed): menadione sodium bisulphite, 650; D-biotin, 70; choline chloride, 780; pteroylglutamic acid, 520; cyanocobalamin, 26; cholecalciferol, 13. The basal diet fed to the chicks was formulated to meet the NRC (1984) recommendations for broiler chickens.
Digestibility (%) = 100 – [100 × (diet Cr2O3/faces Cr2O3) × (faces/diet nutrient)].

2.3. RNA extraction and real-time PCR

Total RNA was extracted from the pectoralis superficial muscle using an RNasefree Fibrous Tissue Mini Kit (Qiagen, Tokyo, Japan) according to the manufacturer’s protocol. The RNA concentration and purity were determined spectrophotometrically using a photometer (BioPhotometer, Eppendorf, Hamburg, Germany) to obtain the A260 and A280 values. The A260/A280 ratios for all samples were between 1.8 and 2.0. cDNA was synthesized with 800 ng RNA per 20 ml of reaction solution using the PrimeScript® RT reagent Kit (Perfect Real Time, Takara, Shiga, Japan) and the Program Temp Control System PC320 (Astec, Fukuoka, Japan) with the following conditions: reverse transcription at 42°C for 15 min, inactivation of reverse transcriptase at 85°C for 5 s, and refrigeration at 4°C for 5 min. Real-time PCR primers were prepared as previously described. Gene expression was measured via real-time PCR using the 7300 Real Time PCR system (Applied Biosystems, Foster, USA) with SYBR® Premix Ex Taq™ (Perfect Real Time, Takara, Shiga, Japan) and specific primers for candidate genes. The thermal cycler conditions were as follows: 1 cycle at 95°C for 10 s and then 30 cycles at 95°C for 5 s and 60°C for 30 s. The expression of glyceraldehyde-3-phosphate dehydrogenase (GAPDH) mRNA was used as an internal standard, and it was not significantly different among the experimental groups. The gene expression results were shown as the % of the control value (Ohtsuka et al. 2011). The values are means of six samples and each sample was repeated three times described formerly by Saleh et al. (2013).

2.4. Statistical analysis

The collected data were evaluated with ANOVA for a complete randomized block design, using the general linear models procedure of SAS software (version 9.2, SAS Institute Inc., 2008). Tukey’s multiple range test was applied for mean to compare the treatment means when the treatment effect was significant at P < 0.05.

3. Results

Results presented in Table 2 show the influence of replacing yellow corn with low-tannin sorghum on growth performance, nutrient digestibilities and internal organ weights in broilers. Replacement of yellow corn with low-tannin sorghum increased BWG and FI significantly (P < 0.05), however, FCR was improved numerically (P = 0.672). Broilers fed 50% yellow corn + 50% low-tannin sorghum diet had higher significant BWG than those fed control diet. In addition, metabolizability, nitrogen and ash digestibilities were not significantly affected by low-tannin sorghum substitutions.

Replacement of yellow corn by low-tannin sorghum increased the breast and thigh muscles weights insignificantly compared with the control group. Substitutions of 25% or 50% low-tannin sorghum resulted in a significant increase in the abdominal fat weight compared with 100% yellow corn or 100% low-tannin sorghum, while sorghum substitutions did not influence both liver and intestine weights.

Table 3 presented blood plasma concentrations of total cholesterol, triglycerides, HDL-C, glucose, GOT, total protein and T3. Plasma concentrations of triglycerides and total cholesterol were significantly decreased by low-tannin sorghum substitutions. While plasma glucose, GOT, total protein and T3 were not significantly influenced by treatments.

Muscle TBARS was significantly decreased, while muscle total fat content was significantly increased by low-tannin sorghum substitutions. However, muscle a-tocopherol content was not significantly affected by dietary treatments (Table 4).

As shown in Figure 1, the mRNAs of genes related to growth (IGF and β-Actin), fatty acids metabolism (ACC and FAS) and antioxidative properties (GPX and SOD) were significantly increased by low-tannin sorghum substitutions. Interestingly, feeding the 50% yellow corn + 50% low-tannin sorghum diet led to a significant increase in mRNA expression of IGF, β-Actin, ACC, FAS, GPX and SOD in broilers.

4. Discussion

In this study, BWG was significantly improved by feeding low-tannin sorghum but FCR was not significantly influenced in broiler chickens (Table 2). Several investigators studied the utilization of low-tannin sorghum in broiler diets and found that body weight gains of chicks fed diets containing corn or low-tannin sorghum did not differ significantly (Reddy 1993; Makled and Affi 2001). The FI was significantly increased in this study and this is in agreement with Nyachoti and Atkinson (1995) who found high FI for birds eating high-tannin sorghum (1.8%). It was reported that high tannins content in sorghum resulted in a significant reduction in FI (Ibrahim et al. 1988). However, the low-tannin sorghum which used in this study has low tannin content (0.56%) and had no negative effect in FI. These results are in disagreement with Attia (1998) who found that low-tannin sorghum variety (0.24–0.50%) had insignificant effect on FI.

In Table 3, FCR was not significantly affected by low-tannin sorghum substitutions and these results are in accordance with Lucbert and Castaing (1986) who concluded that low-tannin sorghum (less than 1%) could be used in broiler diets without any adverse effects on FCR and its nutritional value was similar to that of corn. Also, Attia (1998) found that broiler chicks fed 100% low-tannin sorghum instead of yellow corn had similar FCR to the corn control diet. Moreover, Torres et al. (2013) reported that FCR were not significantly affected by yellow corn replacement with low-tannin sorghum at ratio 50% in broiler diets.

Replacing yellow corn with low-tannin sorghum had no significant effect on muscles and organs weights except abdominal fat weight which was significantly increased by replacing 50% yellow corn with sorghum (Table 2) and this may be due to the highest metabolizability in comparison with the control group. Furthermore, by taking into account our results in mRNA expression of fatty acids synthases (FAS and ACC), it could be mentioned that dietary low-tannin sorghum may increase the abdominal fat in broilers. Results of muscles and
organisms weights are in agreement with the report of Torres et al. (2013) who found that feeding broilers with sorghum at 50% instead of corn had no significant effect on muscle and organ weights. In a similar way, Kumar et al. (2005) reported that carcass and breast muscle weights were not influenced by different low tannin red sorghum levels in broiler diets.

Digestibilities of crude protein, ash and ME were not significantly improved by low-tannin sorghum substitutions (Table 2). Epithelial integrity and intestinal membrane enzymes are essential to secure digestion and absorption of nutrients such as amino acids from the intestinal lumen. Thus any improvements in the intestinal mucosa could enhance nutrient digestion and absorption (Saleh et al. 2013). Torres et al. (2013) found that feeding sorghum diets did not affect the villus height and crypt depth. These results indicate the similarity of the nutritive value of diets based on corn or low-tannin sorghum to broilers.

Feeding low-tannin sorghum decreased plasma triglycerides and total cholesterol concentrations significantly but HDL-C, glucose, GOT, total protein and T3 were not significantly affected (Table 3). Sorghum contains phytosterols which are cholesterol-like composition and involved in the structural components of all plant cell membranes. There is a large interest in these phytosterols due to their advancement of human and animal health like cardiovascular health, especially out of their cholesterol-lowering properties (Fang et al. 2003). Also, sorghum contains flavones and polyflavans (Krueger et al. 2003) which could play an important role in lowering cholesterol level. Ouyang et al. (2016) found that feeding broiler with flavones resulted in decreased total cholesterol and low-density lipoprotein cholesterol (LDL-C) levels and increased HDL-C level in the serum. Moreover, Nyamambi et al. (2007) reported that there was a significant unfavourable association between plasma lipid contents (total cholesterol and triglycerides) and sorghum level in broiler diets. This demonstrates the ability of sorghum grain ingredient to change plasma lipids and cholesterol concentrations when fed to birds. Moreover, sorghum is a good source of other phytochemicals such as policosanols and some plant sterols. These chemicals are more probably to be connected with lipid content changes and cholesterol metabolism in rats (Carr et al. 2005). Phenolic compounds and proanthocyanidins in sorghum were explained as modest cholesterol lowering ability in animal and human studies (Santos-Buelga and Scalbert 2000). In vivo studies on the effects of sorghum on blood lipids content in poultry are scarce. Klopfenstein et al. (1981) reported that total cholesterol was decreased by feeding 58% of low-tannin sorghum in pig’s diets and they found that the effect of sorghum was better than feeding wheat or pearl millet. Also, Cho et al. (2000) found that when sorghum was fed to rats at 30% of feed, the fecal bile acid excretion and 3-hydroxy-3-methylglutaryl CoA (HMGCoA) reductase, as well as levels of HDL-C were increased, without a change in plasma total cholesterol. They also reported that when whole sorghum was fed to rats at 30% of feed the faecal bile acid excretion, as well as levels of HDL-C were increased, without a change in plasma total cholesterol. Moreover, sorghum might enhance broiler health due to its content of anthocyanins. Anthocyanins from fruits and sorghum had been shown to possess various curative

| Item                        | 100% corn + 0% sorghum | 75% corn + 25% sorghum | 50% corn + 50% sorghum | 0% corn + 100% sorghum | SEM    | P-values |
|-----------------------------|------------------------|------------------------|------------------------|------------------------|--------|----------|
| Initial body weight, g      | 412b                   | 412                    | 412                    | 412                    | 4      | 0.942    |
| Final body weight, g        | 1064.9b                | 1230a                  | 1264.7a                | 1233.7a                | 34.5   | 0.032    |
| Body weight gain, g/12 day  | 652.9b                 | 818.0b                 | 852.7b                 | 821.7b                 | 32.8   | 0.021    |
| Feed intake, g/12 day       | 1103b                  | 1342*                  | 1332*                  | 1334*                  | 19.3   | 0.032    |
| Feed conversion ratio, g gain/g feed (12d) | 1.69 | 1.64 | 1.55 | 1.62 | 0.075 | 0.672 |
| Nitrogen digestibility, %   | 61.2                   | 66.5                   | 67.9                   | 62.9                   | 1.7    | 0.878    |
| Metabolizability, %         | 65.9                   | 67.6                   | 68.6                   | 68.1                   | 1.3    | 0.889    |
| Ash digestibility, %        | 34.4                   | 37.3                   | 39.0                   | 32.5                   | 12.6   | 0.657    |
| Organ weight, % body weight |                        |                        |                        |                        |        |          |
| Breast muscle               | 16.0                   | 17.1                   | 17.2                   | 17.1                   | 0.525  | 0.645    |
| Thigh muscle                | 16.7                   | 17.1                   | 17.6                   | 17.4                   | 0.525  | 0.621    |
| Abdominal fat               | 0.67b                  | 0.83ab                 | 0.94a                  | 0.65b                  | 0.0683 | 0.050    |
| Liver                       | 1.8                    | 1.91                   | 1.73                   | 1.82                   | 0.048  | 0.643    |
| Intestine                   | 5.2                    | 4.9                    | 5.3                    | 5.3                    | 0.25   | 0.723    |

**Table 2.** The effects of replacing corn with low-tannin sorghum in broiler diets on growth performance, nutrients digestibilities and internal organ weights in broilers.

| Item                        | 100% corn + 0% sorghum | 75% corn + 25% sorghum | 50% corn + 50% sorghum | 0% corn + 100% sorghum | SEM    | P-values |
|-----------------------------|------------------------|------------------------|------------------------|------------------------|--------|----------|
| TG, mg/dL                   | 29.1a                  | 25.6ab                 | 21.1b                  | 23.4ab                 | 1.78   | 0.05     |
| TC, mg/dL                   | 140.3a                 | 131.6b                 | 129.6b                 | 120.3c                 | 1.77   | 0.013    |
| HDL-C, mg/dL                | 81.9                   | 87.6                   | 91.1                   | 91.9                   | 2.35   | 0.674    |
| Glucose, mg/dL              | 201.9                  | 204.3                  | 216.4                  | 175.6                  | 10.8   | 0.782    |
| GOT, I/U                    | 246                    | 241                    | 250                    | 243                    | 8      | 0.884    |
| Total protein, mg/dL        | 2.46                   | 2.60                   | 2.49                   | 2.34                   | 0.138  | 0.739    |
| T3, mg/dL                   | 5.7                    | 5.2                    | 5.1                    | 5.9                    | 0.25   | 0.884    |

**Table 3.** The effects of replacing corn with low-tannin sorghum in broiler diets on plasma concentrations of TG, TC and HDL-C, glucose, GOT, total protein and T3.

**Notes:** Means within the same row with different superscripts differ (P < 0.05). Results are presented as means ± SEM (n = 7).

Triglyceride (TG); Total Cholesterol (TC); High Density Lipoprotein (HDL); Glutamic-Oxaloacetic Transaminase (GOT); Trilodothrynone concentration (T3).
advantages, inclusively vasoprotective and anti-inflammatory properties (Lietti et al. 1976), anti-cancer and chemoprotective properties (Karaivanova et al. 1990), as well as anti-neoplastic properties (Kamei et al. 1995).

In Table 3, plasma GOT, total protein and T3 concentrations were not significantly affected by dietary treatments. These results are in correspondence with Makled and Affi (2001) who reported that feeding sorghum to broiler chicks did not significantly affect blood GOT and total protein. Plasma glucose level tends to be decreased by sorghum substitutions; and this may be due to the polyphenolic content in sorghum (Krueger et al. 2003).

Muscle fat content was significantly increased, while muscle TBARS was significantly decreased by low-tannin sorghum substitution treatments (Table 4). Jimenez-Ramsey et al. (1994) reported that fat and ash ratios were higher in muscles of quails fed 50% or 100% sorghum instead of yellow corn. The lower molecular weight of polyphenols associated with tannin in the sorghum grain which can be absorbed and distributed in various tissues may be partially responsible for the observed altering in tissue lipid contents. Sorghum contains flavones and polyflavans which play an important role as antioxidants (Krueger et al. 2003). Ouyang et al. (2016) found that feeding broilers with flavones resulted an enhanced the total antioxidant capacity, and GPX, and reduced the malondialdehyde concentrations in the blood. However, the phenolic acids of low-tannin sorghum broadly exist as benzoic, cinnamic acid protocatechuic, caffeic, p-coumaric acid, and sinapic-

### Table 4. The effects of replacing corn with low-tannin sorghum in broiler diets on TBATS, α-tocopherol and total fat content in breast muscle.

| Treatments                        | TBARS, nmol MDA/g | α-Tocopherol, mg/100 g | Total fat, g/100g muscle |
|-----------------------------------|-------------------|------------------------|--------------------------|
| 100% corn + 0% sorghum            | 14.5<sup>a</sup>  | 0.304                  | 0.793<sup>b</sup>       |
| 75% corn + 25% sorghum            | 6.8<sup>b</sup>   | 0.360                  | 1.151<sup>ab</sup>      |
| 50% corn + 50% sorghum            | 5.3<sup>b</sup>   | 0.378                  | 1.427<sup>a</sup>       |
| 0% corn + 100% sorghum            | 7.5<sup>b</sup>   | 0.360                  | 1.081<sup>ab</sup>      |

<sup>a,b</sup> Means within the same row with different superscripts differ (P < 0.05). Results are presented as means ± SEM (n = 6).

TBARS, ThioBarbituric Acid Reactive Substance.

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![Figure 1](image.png)

**Figure 1.** The effects of replacing corn with low-tannin sorghum in broiler diets on relative expression of IGF (A), β-Actin (B), ACC (C), FAS (D), SOD (E) and GPX (F) contents in pectoral superficial muscle. Values are means with standard errors represented by vertical bars. The values are means for six samples and each sample was repeated three times. (a–c) Mean values with unlike letters were significantly different (P < 0.05).
derivatives show better antioxidant activity in vitro and thus may give a share significantly to the health benefits related with whole grain intake (Adom and Liu 2002). Also, sorghum contains anthocyanins and the most common anthocyanins in sorghum are the 3-deoxyanthocyanidins (Gous 1989). Anthocyanins are flavonoids in fruits and seeds offering them the antioxidative activities and health benefits (Clifford 2000). Tannins from sorghum show powerful antioxidant activity in vitro (Prior et al. 1998; Riedl and Hagerman 2001). However; Awika and Rooney (2004) reported that sorghum contains higher levels of oxygen radical absorbance capacity compared to blueberries. Moreover, by taking into account our results in mRNA expression of GPX and SOD, it could be concluded that dietary low-tannin sorghum may reduce the meat lipid peroxidation in broilers.

As graphically presented in Figure 1, mRNA expression related to growth, antioxidative status and fatty acids syntheses were significantly improved. Rama-Rao et al. (1995) found that amino acid profile in sorghum was almost similar to that in corn except for tryptophan content, which was higher in sorghum compared to corn (0.09–0.12% vs. 0.07%). Moreover, different researchers (Pan et al. 2013; Saleh 2014; Saleh et al. 2018) reported that dietary tryptophan affected proteolysis by decreasing the mRNA expression of atrogin-1, MAFbx and MuRF1, and increased protein synthesis by increasing mRNA expression of IGF-1 and intracellular kinases such as protein kinase B. Also, sorghum has higher content of phytosterols which are important to membrane permeability and for nutrients transportation from the blood to cells. Moreover, phytosterols enhanced protein synthesis and this may be the reason of increasing of the mRNAs expression related growth including IGF and β-actin (Naji et al. 2014; Saleh et al. 2019). The FAS and ACC play an important role in the lipogenic passage and are involved in enhancing the maximal ability of a muscle tissue to synthesize fatty acids (Saleh et al. 2013). In common physiological case, nutritional factors such as high-fat feed and hormone could organize the enzyme activity and gene expression of the FAS and ACC (Rosebrough et al. 2011). The levels of ACC and FAS in this study were higher in sorghum experimental groups than in control group which resulted in an increase in muscle fat content and abdominal fat weight.

Sorghum contains phytochemicals including phytosterols, anthocyanins, flavonoids, tannins and phenolic acids which possesses superior antioxidative activities (Awika and Rooney 2004). Thus it could be mentioned that dietary low-tannin sorghum could increase mRNA expression of SOD and GPX (Figure 1E and F). Flavonoids are polyphenols that are amply and closely found in sorghum and give orange and red colours to fruits and vegetables (Pikulski and Brodbelt 2003). These complexes are not produced by the bird’s body and need to be ingested (Kamboh et al. 2015) to get their antioxidative properties and health benefits (Urso and Clarkson 2003; Saleh et al. 2017). Ouyang et al. (2016) found that feeding broiler with flavones resulted in enhancing the SOD and GPX and this finding might explain the enhancement of mRNA expression of SOD and GPX by feeding sorghum in this study.

5. Conclusion
It could be concluded that the replacement of 50% yellow corn by an equal amount of low-tannin sorghum might improve growth performance, modify plasma lipids and enhance gene expression related growth and antioxidative status in broilers.

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