

PERFORMANCE AND TIBIA CHARACTERISTICS OF BROILERS FED DIETS CONTAINING RAW OR TREATED OAK ACORN (*QUERCUS BRANTII* LINDL)

L. Ghaedi, M. Houshmand and S. Parsaei

Department of Animal Science, Faculty of Agriculture, Yasouj University, Yasouj, Iran

Corresponding Author Email: hooshmand@yu.ac.ir

ABSTRACT

Using a completely randomized design, a total of 360 one-day-old male and female broiler chicks were allocated to 6 experimental diets with four replicates (15 birds each), to evaluate the effects of raw or treated oak acorn (OA) on the performance and tibia characteristics of broilers. First diet was a corn-based diet (without OA) as the control, while other 5 diets contained 150 g/kg OA (raw or treated with distilled water, acetic acid, sodium hydrogen carbonate (Alkali) and traditional method). Birds were given starter and finisher diets from 1 to 21 and 22 to 42 d of age, respectively. The results indicated that all treatment methods significantly reduced the content of phenolic compounds. Dietary inclusion of raw OA had no deleterious effect on performance and tibia characteristics when compared to the control. Treatment with Alkali caused the highest overall body weight gain while traditional method resulted in worse performance compared to the raw OA. In conclusion, 150 g/kg raw OA could be included in broiler diets, without adverse effects on their performance or tibia characteristics. Chemical treatment, particularly using Alkali, has the potential to reduce the content of phenols and maybe improve the nutritional value of OA.

Key words: Broiler, Oak acorn, Performance, Tibia characteristic, Treatment.

INTRODUCTION

Corn is one of the most expensive feed ingredients in poultry nutrition. Thus, finding and inclusion of unconventional local feedstuffs which have potential to be used as alternative to corn will reduce the production cost. One of such feedstuffs is oak acorn, seed of acorn, an abundant tree in some regions of the world including Iran. Therefore, huge amounts of oak acorn are produced and available to include in animal and poultry diets.

Due to high levels of carbohydrates, particularly starch (Kekos and Kaukios, 1985), oak acorn has the potential to serve as an energy source in poultry diets (Midilli *et al.*, 2008; Saffarzadeh *et al.*, 1999; Bouderona *et al.*, 2009). Beside nutritional advantages, oak acorn is a tannin-rich feedstuff (72.8- 117.2 g/kg DM) (Shimada, 2001). Tannins are water-soluble polyphenolic compounds with a molecular weight between 500 and 3000 Da. They are divided into hydrolyzable and condensed tannins (Jansman, 1993). Different anti nutritional effects of tannins including reduction in diet palatability and feed intake (Butler *et al.* 1984), pancreatic hypertrophy, reduced protein and starch digestibility (Mahmood *et al.*, 2006; 2007; 2008), reduction in minerals and vitamins availability (Jansman, 1993) as well as histological lesions in the intestines and liver (Ortiz *et al.*, 1994) have been documented. Thus, it is expected that feeding with oak acorn, as a tannin-rich feedstuff, negatively influences broilers. For example, recent results have indicated that broilers fed diet

containing 150 g/kg raw oak acorn had poorer performance than the control group (Houshmand *et al.*, 2015). Detrimental effects of oak acorn on broilers performance have also been shown in other studies (Hamou *et al.* 2012; Bouderoua and Selselet-Attou, 2003).

On the other hand, due to the potential of high-tannin feedstuffs to use in animal nutrition, different strategies such as chemical treatment have been used as detanninification approaches to alleviate the anti nutritive consequences of tannins, thereby improve the nutritive value of those feedstuffs (Medugu *et al.*, 2012). Sharif *et al.* (2012) reported that feeding with diets containing high-tannin sorghum reduced broilers performance. However, treatment of sorghum with water reduced its tannin content and resulted in an improved weight gain. In a study conducted by Mahmood *et al.* (2008), treatment of high tannin salseed (*Shorea robusta*) meal with water or alkali improved the digestive enzymatic activity and performance of broilers. In another study, chemical treatment of salseed meal with distilled water, acetic acid and sodium hydrogen carbonate reduced the content of tannins and therefore prevented their negative consequences (Mahmood *et al.*, 2006).

A traditional method has been used by Iranian local people, particularly nomad, to reduce the tannin level in oak acorn and thereby improve its taste and nutritional value (Ghaderi *et al.*, 2011). In this method, oak fruit is dehulled and then ground. Ground fruits are put inside bags and soaked in water for 2-3 days. Treated materials are used to cook bread for human nutrition.

Minerals availability and absorption can be negatively influenced by tannins (Jansman, 1993). Hassan *et al.* (2003) reported that feeding with sorghum grain containing high levels of tannins, significantly reduced minerals (Ca, P, Mg, Na, K, Fe and Co) absorption. Considering the critical role of dietary minerals, particularly calcium, in bone structure and health (Rath *et al.*, 2000), it is expected that tannins have negative effects on bones, as it has been indicated by Houshmand *et al.* (2015). Few studies have been conducted on the influences of oak acorn on broilers performance and tibia characteristics. Also, there is no report on the effects of chemically treated oak acorn on broiler chickens. Thus, the current experiment was conducted to evaluate above-mentioned effects.

MATERIALS AND METHODS

Birds and experimental groups: All procedures used in the current experiment were approved by the Institution Animal Care Committee of the Yasouj University. The experiment was arranged as a completely randomized design. A total of 360 one-day-old male and female Cobb 500 broiler chicks with same initial body weight (around 43 g) were obtained from a local commercial hatchery.

Upon arrival, birds were weighed in groups and randomly distributed among six experimental diets, with 4 replicates of 15 birds each. First diet was a corn-based diet (without oak acorn) and served as the control (Ctrl), while other 5 diets contained 150 g/kg oak acorn (raw oak acorn or oak acorn treated with distilled water (Water), acetic acid (Acid), sodium hydrogen carbonate (Alkali) and traditional method (Trad). Starter and finisher diets were formulated to meet the NRC (1994) requirements and were fed from 1 to 3 and 4 to 6 weeks of age, respectively. The composition of the experimental diets is shown in Table 1. Birds had free access to feed and water throughout the experiment. The birds were reared in floor pens (150 cm length × 150 cm width) with rice straw as litter. Feed intake was calculated weekly on a pen basis. Birds in each pen were weighed weekly as a group and FCR was calculated. Mortality was recorded daily and FCR was adjusted for mortality.

Oak acorn preparation: Oak acorns were collected from the forest of Yasouj, Kohgeluyeh and Bovir Ahmad Province, Iran. In this forest, *Quercus brantii* Lindl is the most common species. The seed coat of fruits was removed and fruits were dried then fine ground.

Table 1. Composition of the experimental diets.

| Ingredients (g/kg) | *Starter | | *Finisher | |
|------------------------------|----------|-----------|-----------|-----------|
| | Control | Oak acorn | Control | Oak acorn |
| Corn | 562.4 | 380.6 | 656.0 | 476.3 |
| Oak acorn | ... | 150 | ... | 150 |
| Soybean meal (440 g/kg CP) | 342.1 | 359.9 | 273.7 | 291.0 |
| Meat meal | 30 | 30 | 20 | 20 |
| Vegetable oil | 29.6 | 44.5 | 18.6 | 31.8 |
| Limestone | 11.5 | 10.7 | 13 | 12.3 |
| Dicalcium phosphate | 14 | 13.5 | 10.2 | 9.7 |
| Common salt | 4.2 | 4.3 | 3 | 3.1 |
| ¹ Vitamins premix | 2.5 | 2.5 | 2.5 | 2.5 |
| ² Minerals premix | 2.5 | 2.5 | 2.5 | 2.5 |
| DL-Methionine | 1.3 | 1.5 | 0.6 | 0.8 |
| Calculated composition | | | | |
| ME (MJ/kg) | 12.56 | 12.56 | 12.56 | 12.56 |
| Crude protein (g/kg) | 215.6 | 215.6 | 187.5 | 187.5 |
| Calcium (g/kg) | 9.4 | 9.4 | 8.5 | 8.5 |
| Available phosphorus (g/kg) | 4.2 | 4.2 | 3.3 | 3.3 |
| Lysine (g/kg) | 10.3 | 10.3 | 9.4 | 9.4 |
| Methionine (g/kg) | 4.7 | 4.7 | 3.6 | 3.6 |

*Control: control diet, Oak acorn: diet containing 150 g/kg oak acorn

¹ The vitamin premix supplied the following per kilogram of diet: vitamin A (retinyl acetate), 8,000 IU; vitamin D₃, 1,000 IU; vitamin E (dl- α -tocopherol), 30 IU; vitamin K₃, 2.5 mg; vitamin B₁, 2 mg; vitamin B₂, 5 mg; vitamin B₆, 2 mg; vitamin B₁₂, 0.01 mg; niacin, 30 mg; d-biotin, 0.045 mg; vitamin C, 50 mg; d-pantothenate, 8 mg; folic acid, 0.5 mg.

² The mineral premix supplied the following per kilogram of diet: Mn, 70 mg; Fe, 35 mg; Zn, 70 mg; Cu, 8 mg; I, 1 mg; Se, 0.25 mg; Co, 0.2 mg.

Treatment was performed following the method of Mahmood *et al.* (2006). In this method, Oak powders were mixed with distilled water (pH 5.3), 0.67 M acetic acid (pH 2.4), or 0.67 M sodium hydrogen carbonate (pH 8.2) solutions at 820 mL/kg of oak DM and incubated for 12 h at 37° C. The treated material was dried after incubation at the same temperature in oven. Oak acorn was analyzed for contents of DM, EE, CF, CP, Ash and NFE according to the procedure of AOAC (1995). To measure the phenolic components of oak acorn, in the first stage, extraction process was done. About 1 g ground oak acorn was weighed using a digital electronic balance (1mg accuracy) and put in tubes and then, 3 ml of acetone (75%) and 3 ml methanol (50 %) were added to the samples. The tubes were vortexed and then centrifuged (3000 g at 4° C) for 20 min. After that, upper parts of the samples were taken by sampler and transferred to 2 ml tubes. The extracts were analyzed following the procedure of Makkar (2003).

Sample collection: On d 21 and 42 (end of the starter and finisher phases of the study, respectively), one bird from each pen was sacrificed by cervical dislocation and immediately digestive system was carefully removed and weight of liver, pancreas and abdominal fat pad were measured. The relative organ weight was calculated as the weight of the organ as a percentage of live body weight. In addition, at the same times (d 21 and 42), right and left tibia bones were removed as drumsticks. They were kept frozen in plastic bags at -20° until further measurements. After thawing, drumsticks were put inside boiling water for 10 min. After cooling, bones were defleshed by hand and dried for 24 h in room temperature. Right and left tibia bones were used to determine the tibia breaking strength and ash, respectively. Tibia length, weight and volume were accurately detected. Tibia breaking strength was measured using a Machine. In order to determine bone ash, tibia bones were dried at 105° C for 24 h and then put in a furnace at 550° C for 24 h. Bone ash was calculated on the basis of dry weight of tibia bone. Tibia bone ash weight/tibia bone length index was calculated by dividing the tibia ash weight by its length (Seedor *et al.*, 1991). Tibia robusticity index was calculated using following formula: tibia length/cube root of tibia weight (Reisenfeld, 1972).

Statistical analysis: Data were analyzed by analysis of variance using the General Linear Models (GLM) procedures of SAS software (SAS Institute, 2005). The means were compared by Duncan's multiple range test. The level of statistical significance was set at P< 0.05.

RESULTS AND DISCUSSION

Proximate analysis of oak acorn: Proximate analysis of oak acorn is presented in Table 2. As expected, nitrogen

free extract (NFE) was the main component (735.2 g/kg DM) of oak acorn. Previously, high levels of NFE (874 - 903 g/kg DM) were detected in 3 species of acorn (Shimada, 2001).

Table 2. Proximate analysis of oak acorn (g/kg DM).

| Component | Ash | CP | EE | CF | NFE |
|-----------|------|------|-------|------|-------|
| Level | 10.6 | 50.3 | 116.7 | 52.0 | 735.2 |

CP: crude protein, EE: ether extract, CF: crude fiber, NFE: nitrogen free extract

Content of the phenolics components: The current results (Table 3) suggest that high levels of phenolics components including tannins are present in this feedstuff. In agreement with our results, Shimada (2001) indicated that acorn contained considerable amounts of tannin (72.8 – 117.2 g/kg DM). Also, our findings (Table 3) indicated that all treatment methods significantly reduced the amount of total phenols, condensed tannins and total tannins. The most reductions were observed in Alkali treatment. Similar to these findings, Mahmood *et al.* (2006) reported that chemical treatment with acid acetic, sodium carbonate hydrogen and water reduced the contents of tannins in the salseed meal. Also, in their study, alkali treatment was more effective in reducing tannins compared with acid and water treatments.

Performance: The effects of experimental diets on broilers performance [body weight gain (BWG), feed intake (FI) and feed conversion ratio (FCR)] are shown in Table 4. It is interesting to note that dietary inclusion of raw oak acorn resulted in a higher BWG than the Ctrl group in the starter (d 1-21) period. Birds in the raw group significantly consumed more feed than the Ctrl group (1075 vs. 924 g). Thus, the reason for the higher BWG of raw group was probably due to higher FI. Significant differences in FI and BWG were not observed between these 2 groups during the finisher period. However, feeding birds with raw oak acorn significantly increased overall (d 1-42) BWG compared to the Ctrl group. Although, overall FI was significantly not different between 2 groups, but raw group numerically had more FI than the Ctrl group (3379 vs. 3137 g). Hence, a possible reason for the difference in BWG between raw and Ctrl groups may be related to their FI. Our results showed that dietary inclusion of raw oak acorn had no adverse effects on FCR, throughout the experiment. Previously, Kaushal and Singh (1982) reported that dietary inclusion of 100 and 150 g/kg oak acorn had no detrimental effects on BWG, while 200 and 250 g/kg acorn significantly reduced this parameter. The results of Hamou *et al.* (2012) indicated that dietary inclusion of green oak acorn (at the rate of 324 g/kg), as a replacement to corn, negatively influenced broilers final body weight and feed efficiency. In another study, feeding with different levels (50, 100, 150 and 200 g/kg)

of acorn seed (*Quercus cerris*) had no negative effects on performance traits of Japanese quail (Midilli *et al.*, 2008).

Bouderoua *et al.* (2009) reported that at 35 day of age, body weight of birds fed diets containing 335 g/kg green

Table 3. Effects of treatment method on phenolics components of oak acorn (g/kg DM).

| Parameter | Treatment method | | | | | SEM |
|------------------|------------------|-----------------|-----------------|-----------------|-----------------|-----|
| | Raw | Acid | Alkali | Water | Trad | |
| Total phenols | 106 ^a | 59 ^b | 7 ^d | 23 ^c | 63 ^b | 2 |
| Condensed tannin | 54 ^a | 45 ^c | 12 ^c | 32 ^d | 48 ^b | 0.8 |
| Total tannin | 61 ^a | 43 ^b | 18 ^d | 36 ^c | 42 ^b | 2 |

Means within a row with different superscripts are significantly different at P<0.05.

Raw: raw oak acorn; Acid, Alkali, Water, Trad: treated-oak acorn with acetic acid, sodium hydrogen carbonate, distilled water and traditional method, respectively.

Table 4. Effects of experimental diets on body weight gain, feed intake and feed conversion ratio of broilers at different phases of the experiment.

| Parameter | ¹ Experimental diets | | | | | | SEM |
|-----------------------|---------------------------------|--------------------|--------------------|---------------------|--------------------|--------------------|------|
| | Ctrl | Raw | Acid | Alkali | Water | Trad | |
| Body weight gain (g) | | | | | | | |
| d 1-21 | 491 ^c | 597 ^b | 615 ^b | 688 ^a | 591 ^b | 471 ^c | 15 |
| d 22-42 | 1172 ^b | 1204 ^b | 1138 ^b | 1422 ^a | 1197 ^b | 1184 ^b | 37 |
| d 1-42 | 1663 ^c | 1802 ^b | 1753 ^{bc} | 2110 ^a | 1788 ^{bc} | 1655 ^c | 42 |
| Feed intake (g) | | | | | | | |
| d 1-21 | 924 ^b | 1075 ^a | 1102 ^a | 1103 ^a | 1121 ^a | 974 ^b | 21 |
| d 22-42 | 2214 ^c | 2304 ^{bc} | 2605 ^{ab} | 2931 ^a | 2645 ^a | 2737 ^a | 105 |
| d 1-42 | 3137 ^c | 3379 ^{bc} | 3706 ^{ab} | 4036 ^a | 3765 ^a | 3711 ^{ab} | 110 |
| Feed conversion ratio | | | | | | | |
| d 1-21 | 1.882 ^b | 1.801 ^b | 1.792 ^b | 1.603 ^c | 1.897 ^b | 2.068 ^a | 0.04 |
| d 22-42 | 1.889 ^b | 1.914 ^b | 2.289 ^a | 2.061 ^{ab} | 2.210 ^a | 2.312 ^a | 0.08 |
| d 1-42 | 1.886 ^b | 1.875 ^b | 2.114 ^a | 1.913 ^b | 2.106 ^a | 2.242 ^a | 0.06 |

Means within a row with different superscripts are significantly different at P<0.05.

¹ Ctrl: corn based diet (without oak acorn); Raw: diet containing raw oak acorn; Acid, Alkali, Water and Trad: diets containing treated-oak acorn with acetic acid, sodium hydrogen carbonate, distilled water and traditional method, respectively.

oak acorns was 12% lower than the control. However, final weight at day 56 was similar between two groups. Treatment with Alkali significantly increased BWG during the starter phase (d 1-21) of the study compared with the raw group. Birds of Alkali diet had the highest BWG in all phases of the study. Interestingly, treatment with traditional method significantly reduced BWG from 1 to 21 and 1 to 42 d of the study compared with the raw group which means that traditional method had negative effects on BWG.

The current results indicated that dietary inclusion of 150 g/kg raw oak acorn did not reduce feed intake. In addition, all treated groups (except for Trad group during d 1-21) consumed more feed than the Ctrl during all phases of the study. Tannins can make stringent or bitter taste which influences diet palatability and thereby causing decrease in feed intake (Nyachoti *et al.*, 1996). Thus, less feed intake was expected for birds fed diets containing oak acorn, but such inhibitory effect was not observed in the current study. Significant

differences in FCR were not observed between the Ctrl and raw group throughout the experiment. It suggests that 150 g/kg oak acorn could be included in broiler diets without negative effects on feed efficiency. In contrast with this finding, Houshmand *et al.* (2015) reported that dietary inclusion of same level of raw oak acorn resulted in a worse performance compared to the control.

Treatment method had significant effects on FCR in all phases of the experiment. In starter phase, the best and the worse FCR were observed in the Alkali and Trad groups, respectively. Acid, Water and Trad groups had worse FCR than the Ctrl or raw group during the finisher or whole period of the experiment. However, at the same times, significant differences in FCR were not observed among Alkali, Ctrl and raw groups. It could be concluded that treatment with Alkali and traditional method had the best and the worst effects on FCR, respectively. The results show that consumption of 150 g/kg raw oak acorn had no deleterious effect on broilers. It means that tannin level was not high enough to affect

performance and they were able to tolerate the tannin content of this level of oak acorn. Most likely, if higher levels of oak acorn are included in broiler diets, negative effects of tannins will appear. Probably, under this condition, treatment of oak acorn has more beneficial effects on broilers.

Beneficial consequences of chemicals treatment on the improvement of nutritional value of tannin-rich feedstuffs have previously been shown in several studies. In a study conducted by Sharif *et al.* (2012) water treatment reduced the tannin content of sorghum and improved feed efficiency of broilers. Mahmood *et al.* (2007) reported that addition of water, acetic acid, sodium bicarbonate and polyvinyl-pyrrolidone solutions to high-tannin salseed meal and subsequent anaerobic incubation at 37°C significantly reduced its tannin content. Also, treatment with alkali or water improved the digestive enzymatic activity. This improvement may be related to reduction in the formation of tannin-dietary protein and tannin-enzyme complexes in the broiler gut (Mahmood *et al.*, 2008). In another study, treatment of salseed meal with alkali and water significantly improved the protein digestibility and nitrogen retention in colostomized hens and broiler cockerels. Probably, in the presence of moisture, the tannins are polymerized to larger molecules that are insoluble and lose their ability to precipitate proteins (Mahmood *et al.*, 2006).

It is possible that under treatment conditions, hydrolysable tannin be hydrolysed and converted to smaller units which do not have tannin like effect. The mechanism of tannin deactivation by water or acid treatment may be similar to the reaction which proceeds in grain as it approaches maturity. As the seed ripens, polymers of procyanidins are formed from monomers present in the seed resulting in an increase in the concentrations of soluble and insoluble polymers (Mahmood *et al.*, 2007). Treatment of high tannin sorghum with an alkaline substance (wood ash) was effective in reducing the tannin content and thus improved its nutritive value and resulted in a better broiler performance. It was suggested that due to polymerization of tannins in an alkaline media, they can form none-toxic compounds (Kyarisiima *et al.*, 2004). The results of Banda-Nyirenda and Vohra (1990) indicated that broilers fed diets based on tannin-containing sorghum had poorer performance compared with the control. Dietary supplementation with sodium bicarbonate decreased tannin contents and thus inhibited those deleterious effects. They suggested that sodium bicarbonate can improve dietary electrolyte balance, creating conditions for an improvement in feed efficiency without influencing AME and apparent nitrogen

retention. In the study of Muindi *et al.* (1981), anti nutritive effects of tannin sorghum were inhibited by treatment with sodium bicarbonate (soaking for 3 days in Magadi soda solutions).

A possible explanation for the poor performance in birds of Trad group may be related to the nutrient leaching. As stated earlier, in Trad method, oak acorn was soaked and washed in water for a long period (2-3 days). Probably, under this condition, in addition to tannins, dissolved valuable nutrients have lost. This condition can cause lower nutritional quality and therefore poor performance.

Tibia characteristics: The effects of experimental diets on tibia characteristics at 21 and 42 d of age are shown in Table 5. Different parameters could be used to determine the bone mineralization and hence bone structure and health in poultry, including bone ash, bone breaking strength, bone weight and bone volume (Rao *et al.*, 1993). Also, two indexes are used to evaluate bone density: 1- the bone ash weight/bone length index. The higher index indicates more density (Monteagudo *et al.*, 1997). 2- robusticity index. Lower index indicates more strong bone (Reisenfeld, 1972).

On d 21, significant differences in tibia characteristics (except for bone volume) were not observed between Ctrl and raw group ($P>0.05$). Also, on d 42, there were no significant differences in tibia characteristic among experimental diets. It means that oak acorn, had no negative effect on tibia bone ($P>0.05$). This result is not in agreement with previous report (Houshmand *et al.*, 2015). However, birds in raw group had lower bone ash and strength than those in Alkali group, at 21 d of age. These reductions may be attributed to the presence of the high levels of tannins in raw oak acorn. This could be because, minerals availability and absorption and thus bone structure and health are negatively influenced by tannins (Jansman, 1993; Hassan *et al.*, 2003).

Pancreas, liver and abdominal fat weight: The results (Table 6) indicate that abdominal fat pad and liver weight were significantly not influenced by experimental diets at 42 d of age ($P>0.05$). There was no significant difference in pancreas weight between the Ctrl and other groups at 21 d of age. However, treatment with Alkali and Acid significantly reduced the pancreas weight compared with the raw group at 21 d of age. It is reported that, high tannin diets have inhibitory effects on pancreatic enzymes activities. Thus, pancreas needs to increase enzymes production. As a result, pancreatic hypertrophy will occur (Ahmed *et al.*, 1991).

Table 5. Effects of experimental diets on tibia characteristics of broilers at 21 and 42 d of age.

| Parameter | ¹ Experimental diets | | | | | | SEM |
|-------------------------------|---------------------------------|--------------------|--------------------|-------------------|--------------------|--------------------|------|
| | Ctrl | Raw | Acid | Alkali | Water | Trad | |
| Weight (g) | | | | | | | |
| d 21 | 1.5 ^{bc} | 1.7 ^{abc} | 1.8 ^{ab} | 2.0 ^a | 1.9 ^{ab} | 1.4 ^c | 0.09 |
| d 42 | 5.7 | 5.6 | 5.7 | 5.7 | 6.8 | 5.2 | 0.58 |
| Length (cm) | | | | | | | |
| d 21 | 6.3 ^a | 6.4 ^a | 6.4 ^a | 6.5 ^a | 6.4 ^a | 6.0 ^b | 0.07 |
| d 42 | 9.8 | 9.4 | 9.4 | 9.3 | 9.6 | 9.2 | 0.20 |
| Volume (ml) | | | | | | | |
| d 21 | 3.5 ^a | 2.5 ^b | 3.5 ^a | 3.5 ^a | 4.0 ^a | 2.5 ^b | 0.26 |
| d 42 | 8.5 | 8.3 | 9.0 | 8.0 | 9.2 | 9.4 | 1.1 |
| Ash (%) | | | | | | | |
| d 21 | 39.5 ^b | 38.1 ^b | 41.5 ^b | 51.8 ^a | 40.7 ^b | 41.2 ^b | 2.1 |
| d 42 | 41.3 | 43.0 | 45.7 | 52.0 | 46.6 | 41.1 | 3.2 |
| Strength (kg/m ²) | | | | | | | |
| d 21 | 27.5 ^{ab} | 21.0 ^b | 24.5 ^{ab} | 32.0 ^a | 26.0 ^{ab} | 24.5 ^{ab} | 3.3 |
| d 42 | 39.0 | 35 | 36.5 | 37.5 | 40.5 | 33.3 | 3.5 |
| ² W/L (mg/mm) | | | | | | | |
| d 21 | 25 ^{bc} | 26 ^{abc} | 28 ^{ab} | 30 ^a | 29 ^a | 24 ^c | 2 |
| d 42 | 62 | 60 | 61 | 61 | 63 | 57 | 6 |
| Robusticity index | | | | | | | |
| d 21 | 1.9 ^a | 1.9 ^a | 1.9 ^a | 1.7 ^b | 1.9 ^a | 1.8 ^b | 0.13 |
| d 42 | 2.7 | 2.7 | 2.7 | 2.5 | 2.7 | 2.7 | 0.02 |

Means within a row with different superscripts are significantly different at P<0.05.

¹ Ctrl: corn based diet (without oak acorn); Raw: diet containing raw oak acorn; Acid, Alkali, Water and Trad: diets containing treated-oak acorn with acetic acid, sodium hydrogen carbonate, distilled water and traditional method, respectively.

² W/L: tibia ash weight / tibia length index

Table 6. Effects of experimental diets on relative weight of pancreas (at 21 and 42), liver and abdominal fat at 42 d of age (% body weight).

| Parameter | ¹ Experimental treatments | | | | | | SEM |
|---------------|--------------------------------------|-------------------|-------------------|-------------------|--------------------|--------------------|------|
| | Ctrl | Raw | Acid | Alkali | Water | Trad | |
| Pancreas | | | | | | | |
| d 21 | 0.31 ^{ab} | 0.38 ^a | 0.30 ^b | 0.28 ^b | 0.31 ^{ab} | 0.31 ^{ab} | 0.02 |
| d 42 | 0.24 | 0.25 | 0.22 | 0.21 | 0.21 | 0.22 | 0.01 |
| Liver | | | | | | | |
| d 42 | 2.8 | 2.4 | 2.6 | 2.7 | 2.4 | 2.8 | 0.19 |
| Abdominal fat | | | | | | | |
| d 42 | 1.7 | 1.9 | 2.7 | 1.9 | 2.1 | 2.2 | 0.32 |

Means within a row with different superscripts are significantly different at P<0.05.

¹ Ctrl: corn based diet (without oak acorn); Raw: diet containing raw oak acorn; Acid, Alkali, Water and Trad: diets containing treated-oak acorn with acetic acid, sodium hydrogen carbonate, distilled water and traditional method, respectively.

Finally, it should be mentioned that effects of tannins are influenced by different factors including response parameters chosen (weight gain, feed intake and feed efficiency), source and concentration of tannins, animal factors (species, age and production level) and diet composition (Jansman, 1993). Thus, differences in our results with others may be attributed to these factors.

Conclusion: Treatment with substances such as distilled water, acetic acid, sodium hydrogen carbonate as well as traditional method are effective in reducing the phenolic

components of oak acorn. Raw oak acorn at level of 150 g/kg could be included in broiler diets, without adverse effects on their performance or tibia characteristics. Chemical treatment of oak acorn, particularly using Alkali, has the potential to improve the nutritional value of oak acorn. However, traditional treatment had negative influences on broilers performance.

Acknowledgments: The authors would like to thank the Yasouj University, Iran for the providing supports to this study.

REFERENCES

- Ahmed, A.E., R. Smithard, and M. Ellis (1991). Activities of enzymes of the pancreas, and the lumen and mucosa of the small intestine in growing broiler cockerels fed on tannin-containing diets. *Br. J. Nut.* 65, 189-197.
- AOAC (1995). Official methods of analysis (16th ed). Association of Official Analytical Chemists, Washington, USA.
- Banda-Nyirenda D.B.C., and P.Vohra (1990). Nutritional improvement of tannin-containing sorghums (*Sorghum bicolor*) by sodium bicarbonate. *Cereal Chem.* 67, 533-537.
- Bouderoua, K., J. Mourot, and G. Selselet-Attou (2009). The effect of green oak acorn (*quercus ilex*) based diet on growth performance and meat fatty acid composition of broilers. *Asian-Aust. J. Anim. Sci.* 6, 843 – 848.
- Bouderoua, K., and G. Selselet-Attou (2003). Fatty acid composition of abdominal adipose tissue in broilers fed green-oak (*Quercus ilex*), cork oak acorn (*Quercus Suber L.*) based diets. *Anim. Res.* 52, 377-382.
- Butler, L.G., D.J. Riedl, D.G. Lebryk, and H.J. Blytt (1984). Interactions of proteins with sorghum tannin: Mechanism, specificity and significance. *J. Am. Oil Chem. Soc.* 61, 916-920.
- Ghaderi, M., A. Sadeghi Mahoonak, M. Aalami, M. Ghorbani, and M.H. Azizi (2011). Effect of soaking in water, alkali and acetic acid to remove polyphenol from kernel of two Iranian acorn varieties. *Iranian Food Sci. Technol. Res. J.* 7, 50-59.
- Hamou, H., K. Bouderoua, I. Sisbane, and J. Mourot (2012). Effect of green oak acorn based diet on performance and fatty acid composition of cooked breast meat. *Int. J. Appl. Anim. Sci.* 1, 94-101.
- Hassan, I.A.G., E.A. Elzubeir, and A.H. El Tinay (2003). Growth and apparent absorption of minerals in broiler chickens fed diets with low and high tannin contents. *Trop. Anim. Health Prod.* 35, 189-196.
- Houshmand M., F. Hojati, and S. Parsaie (2015). Dietary nutrient manipulation to improve the performance and tibia characteristics of broilers fed oak acorn (*Quercus brantii Lindl.*). *Braz. J. Poult. Sci.* 17, 17-24.
- Jansman, A.J.M. (1993). Tannins in feedstuffs for simple-stomached animals. *Nut. Res. Rev.* 6, 209–236.
- Kaushal, N.A. and B. Singh (1982). Potentials of (*Quercus leucotrichophora*) A. Camus ex Bahadur acorns in poultry feed. *J. Tree Sci.* 1, 120-122.
- Kekos, D. and B. Kaukios (1985). Acid hydrolysate of acorn polysaccharid as substrates of *Candida utilis* growth. *Biotechnology* 7, 345-348.
- Kyarisiima, C.C., M.W. Okot, and B. Svihus (2004). Use of wood ash in the treatment of high tannin sorghum for poultry feeding. *S. Afr. J. Anim. Sci.* 34, 110-115.
- Mahmood, S., M. Ajmal Khan, M. Sarwar, and M. Nisa (2006). Chemical treatments to reduce antinutritional factors in Salseed (*Shorea robusta*) Meal: Effect on nutrient digestibility in colostomized pullets and intact broilers. *Poult. Sci.* 85, 2207- 2215.
- Mahmood, S., M. Ajmal Khan, M. Sarwar, M. Nisa, W.S. Lee, S.B. Kim, T.Y. Hur, H. J. Lee, and H.S. Kim (2007). Use of chemical treatments to reduce tannins and trypsin inhibitor contents in Salseed (*Shorea robusta*) meal. *Asian-Aust. J. Anim. Sci.* 20, 1462 – 1467.
- Mahmood, S., M. Ajmal Khan, M. Sarwar, and M. Nisa (2008). Use of chemical treatments to reduce antinutritional effects of tannins in salseed meal: Effect on performance and digestive enzymes of broilers. *Livestock Sci.* 116, 162-170.
- Makkar, H.P.S. (2003). Quantification of tannins in tree and shrub foliage. A laboratory manual. FAO/IAEA.
- Medugu, C.I., B. Saleh, J.U. Igwebuike, and R.L. Ndirmbita (2012). Strategies to improve the utilization of tannin-rich feed materials by poultry. *Int. J. Poult. Sci.* 11, 417-423.
- Midilli, M., Ö.H. Muglali, L. Altıntas, H. Erol, and S. Cakır (2008). Shelled acorn seed (*Quercus cerris*) as a diet ingredient on the performance of growing Japanese Quail. *S. Afr. J. Anim. Sci.* 38, 38-42.
- Monteagudo M.D., E.R. Hernandez, C. Seco, J. Gonzales-Riola, M. Revilla, L.F. Villa, and H. Rico (1997). Comparison of the bone robusticity index and bone weight/bone length index with the results of bone densitometry and bone histomorphometry in experimental studies. *Acta Anatomica.* 160, 195–199.
- Muindi, P., J. S. Thomke, and R. Ekman (1981). Effect of Magadi soda treatment on the tannin content and in vitro nutritive value of grain sorghums. *J. Sci. Food Agric.* 32:25-34.
- NRC (1994). Nutrient Requirements of Poultry. National Academy Press, Washington, DC.
- Nyachoti, C.M., J.L. Atkinson, and S. Lesson (1996). Response of broiler chicks fed a high-tannin sorghum diets. *J Appl. Poult. Res.* 5, 239-245.
- Ortiz, L.T., C. Alzueta, J. Trevino, and M. Castano (1994). Effects of faba bean tannins on the growth and histological structure of the

- intestinal tract and liver of chicks and rats. *Br. Poult. Sci.* 35, 743-754.
- Rao S.K., M.S. West, T.J. Frost, J.I. Orban, M.M. Bryant and S.R.D.A. Roland (1993). Sample size required for various methods of assessing bone status in commercial leghorn hens. *Poult. Sci.* 72, 229–235.
- Rath, N.C., G.R. Huff, W.E. Huff, and J.M. Balog (2000). Factors regulating bone maturity and strength in poultry. *Poult. Sci.* 79, 1024–1032.
- Reisenfeld, A. (1972). Metatarsal robusticity in bipedal rats. *Am. J. Phys. Anthropol.* 40, 229–234.
- Saffarzadeh, A., L. Vincze, and J. Csap (1999). The effects of different levels of acorn seeds on laying hens performance in first phase of egg production. *Acta Agraria Kaposváriensis.* 3, 369-377.
- SAS (2005). *Statistical Analysis Systems users guide* (9th ed.). SAS Institute Inc., Cary, N.C., USA.
- Seedor, J.G., H.A. Quarruccio, and D.D. Thompson (1991). The bisphosphonate alendronate (MK-217) inhibits bone loss due to ovariectomy in rats. *J. Bone and Min. Res.* 6, 339–346.
- Sharif, M., M. Idrees, N.A. Tauqir, M.A., Shahzad, M.F. Khalid, M. Nisa, M. Sarwar, and M.L. Khan (2012). Effect of water treatment of sorghum on the performance of broiler chicks. *S. Afr. J. Anim. Sci.* 42, 189-194.
- Shimada, T. (2001). Nutrient compositions of acorns and horse chestnuts in relation to seed-hoarding. *Ecol. Res.* 16, 803-808.