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The Metabolizable Energy Content and Effect of Grape Pomace with or without Tannase Enzyme Treatment in Broiler Chickens

Research Article

S.K. Ebrahimzadeh[1*], B. Navidshad[1], P. Farhoomand[2] and F. Mirzaei Aghjehgheshlagh[1]

[1] Department of Animal Science, Faculty of Agricultural Science, University of Mohaghegh Ardabili, Ardabil, Iran
[2] Department of Animal Science, Faculty of Agriculture, Urmia University, Urmia, Iran

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*Correspondence E-mail: sk.ebrahimzadeh@uma.ac.ir

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ABSTRACT

Two experiments were conducted to determine the chemical composition, metabolizable energy content and effect of grape pomace (GP) with or without tannase enzyme treatment on growth performance of broiler chickens. In experiment 1, the apparent metabolizable energy (AME) and apparent metabolizable energy corrected for nitrogen (AMEn) of GP were determined as 2642.19 and 2641.08 kcal/kg, respectively. Supplementation of tannase enzyme at 0.05 and 0.1 percent of experimental diets significantly ($P \leq 0.01$) affected the metabolizable energy content of the diets and the highest improvement was observed with diets containing 0.01 percent tannase enzyme ($P \leq 0.01$). True metabolizable energy (TME) and true metabolizable energy corrected for nitrogen (TMEn) were determined based on Sibbald's procedure. The TME and TMEn of grape pomace were 1844.14 and 1839.83 kcal/kg, respectively. In experiment 2, four dietary treatments (50 birds/treatment) were conducted to study the effect of inclusion of 10% GP with or without tannase enzyme supplementation on growth performance in broiler chicks (0 to 42 days of age). At 10 d of age, the body weight (BW) of the control groups was higher ($P<0.05$) compared with other experimental groups. Furthermore, broilers fed the control diet had higher ($P<0.05$) average daily gain (ADG) in the starter (days 0-10) experimental periods. Addition of GP in the chicken diets did not impair growth performance (BW, ADG, average daily feed intake (ADFI) and feed-to-gain ratio (F:G)) of birds fed the grower (days 11-24), finisher (25-42) and the overall (days 0-42) experimental periods compared with other treatments. Among diets supplemented with GP, feed intake and body weight of broilers feeding diet containing 0.1% tannase were higher and the addition of tannase enzyme, improve the metabolizable energy content of GP.

KEY WORDS grape pomace, growth performance, metabolizable energy, tannase enzyme.

INTRODUCTION

Population growth, economic and social development caused higher demand for livestock products in many developing countries. A large portion of agricultural by-products has no direct human consumption, but it can be used indirectly to produce human food. Effective usage of agricultural by-products as animal feed depends on some factors such as nutrient composition compared to animal needs (McDonald *et al.* 1995). In order to enhance energy efficiency and optimize poultry performance it is suggested that the proximate analysis and metabolizable energy value of feedstuffs must be measured before feed formulation (McDonald *et al.* 1995). Grapes (*Vitis vinifera*) are one of the world largest fruit crops with annual production of 77 million metric tons (FAO, 2013). Esteeming, crushing, and

pressing of grapes during processing for ethanol, fruit juice and wine production results in huge quantities of grape pomace (GP) including stems, skins, seeds, and peels and accounts for about 20% of the weight of the grape processed into wine (Llobera and Canellas, 2007). The GP are rich in a wide range of polyphenols like flavonoids, monomeric phenolic compounds, catechins, and epicatechins (Dorri *et al*. 2012). The available GP as a by-product of food processing industry in Iran exceeds 50000 ton/year, causing problems in both economical and ecological terms. Thus, any useful application for these by-products could represent an interesting advance in the maintenance of the environmental equilibrium and also an economic revaluation of the raw material (Abarghuei *et al*. 2010). The GP contains some antinutritional factors such as tannins that may decrease the GP feeding value (Pirmohammadi *et al*. 2007). A variety of protocols has been suggested by some scientists to improve the feeding value of tanniniferous feeds such as storage, drying, ensiling and adding exogenous enzymes (Makkar, 2003). Enzyme supplementation is a technique with increasing applicability for improving the nutritional characteristic of by-products and it is widely used in animal nutrition. However, results of GP utilization on growth performance in livestock were inconsistent.

For example, Hughes *et al*. (2005) found the addition of grape seed extract (90.2% total phenolics) at the level of 30 g/kg decreased the growth performance in chickens. However, Brenes *et al*. (2008) and Goni *et al*. (2007) found GP supplementation enhanced antioxidant capacity without any negative effect on growth performance in broilers (4.86% total phenolics; added at 30 g/kg). The aim of this study was to determine the chemical composition, metabolizable energy, and effect of GP with or without tannase enzyme addition on growth performance of broiler chickens.

MATERIALS AND METHODS

Experiment 1

Grape pomace preparation and analysis

The red GP was obtained from fruit juice factory located in the Urmia city (TATAO factory, Urmia city, Iran) and were completely dried under sunlight. Three samples were taken and ground to pass through a 1 mm screen for chemical analyses. Proximate composition (Table 1) of GP samples were analyzed according to procedures described by the Association of Official Analytical Chemists (AOAC, 2000). Gross energy (GE) content was determined by a Parr adiabatic calorimetric bomb. Extraction of the phenolic compounds from GP was performed by ultrasonication, using methanol/HCl 99/1 (v/v) as extraction solvent. Total phenolic content of extracts was determined by using Folin-Ciocalteu colorimetric method (Singleton and Rossi, 1965).

Gallic acid was employed as a calibration standard and results were expressed as gallic acid equivalents (mg gallic acid/g of dried samples).

Apparent metabolizable energy assay

In this experiment, 16 male broilers (Ross 308) from 37 to 45 d of age were used. Birds were distributed randomly to 4 treatments with 4 replicates in battery cages. Basal diets were formulated to meet or exceed NRC (1994) nutrient recommendations for broilers (Table 2). The amount of 10% GP without enzyme or supplemented with 0.05 or 0.1% tannase enzyme (supplied by Kikkoman Foods Products Company, Edogawa Plant, Japan, containing tannin acylhydrolase, 500 U/g, EC 3.1.20) was substituted by the corn and soybean in the experimental diets. Then the experimental diets were as follows: 1) GP free diet (100% basal corn, soybean diet), 2) 90% basal diet + 10% GP, 3) 90% basal diet + 10% GP + 0.05% tannase, 4) 90% basal diet + 10% GP + 0.1% tannase. Birds were fed *ad libitum*. The following procedures were common to all experiments. Body weight was determined when broilers were allocated to battery cages and also at the end of experimentation to ensure that dietary treatments did not limit growth. A 72-h total excreta collection period was conducted to evaluate AME and AMEn of GP. After a 3-d acclimation period, feed refusal and feed allocation were weighed daily throughout the 72-h collection period. The total amount of excreta voided at the end of the collection was weighed (wet basis). Multiple subsamples were collected from the total amount of excreta and homogenized, and then a 250-g representative sample was placed in a plastic bag for analysis. Representative samples of feed and excreta were frozen and subsequently dried at 70 ˚C. Dry matter and nitrogen content of feed and excreta were determined according to AOAC (2000). The GE was determined by a Parr adiabatic calorimetric bomb. Feed consumption and excreta weights during the 72-h collection period were used to calculate energy and nitrogen intake and excretion. AME and AMEn experimental diets were calculated using the following equations:

$$AME/g\ of\ feed = [(Fi \times GEf) - (E \times GEe)] / Fi$$

$$AMEn/g\ of\ feed = [(Fi \times GEf) - (E \times GEe) - (NR \times K)] / Fi$$

$$NR = (Fi \times Nf) - (E \times Ne)$$

Where:

AME: apparent metabolizable energy (kcal/g).

AMEn: apparent metabolizable energy corrected for nitrogen (kcal/g).

F_i: feed intake (g).

E: excreta (g).

GE_f: gross energy of feed sample (kcal/g).

GE_e: gross energy of excreta (kcal/g).
NR: nitrogen retention (g).
K: nitrogen retention correction coefficient (8.22 kcal/g for each g N).
NF: feed nitrogen (%).
N_e: fecal nitrogen (%).

The AME and AMEn content of GP calculated using the following modified equations (Anison *et al.* 1996):

AME_{GP}= $AME_{testdiet}$ - [$AME_{basaldiet}$ × (corn+$soybean_{inclusionrate}$)] / grape pomace $_{inclusion\ rate}$
$AMEn_{GP}$= $AMEn_{testdiet}$ - [$AMEn_{basaldiet}$ × (corn+$soybean_{inclusionrate}$)] / grape pomace $_{inclusion\ rate}$

Where:
AME_{GP}: apparent metabolizable energy of grape pomace (kcal/g).
$AMEn_{GP}$: apparent metabolizable energy corrected for nitrogen for grape pomace (kcal/g).
Corn + $soybean_{inclusion\ rate}$= 82. 74%.
Grape $pomace_{inclusion\ rate}$= 10%.

True metabolizable energy assay

The precision-fed cockerel assay of Sibbald *et al.* (1986) was used for determining the true metabolizable energy (TME and TMEn) of the GP. Seven adults Leghorn roosters were housed in individual metabolism cages that were 0.40-m wide, 0.40-m long, and 0.50-m high. Following a period of 24 h without feed, 25 g of the GP samples were fed by intubation to 6 adult Leghorn roosters. Another rooster was fasted to estimate endogenous losses. Total excreta voided over the following 48-h period was dried and ground for subsequent analyses. The GP and excreta samples were freeze-dried before analysis and the DM and Nitrogen content of feed and excreta were determined according to AOAC (2000). The GE was determined by a Parr adiabatic calorimetric bomb. The TME and TMEn content of GP samples were calculated using the following equations:

TME/feed= {[(Fi×GEf) - (E×GEe)] + (FEm+UEe)} / Fi
TMEn/feed= {[(Fi×GEf) - (E×GEe) - (NR×K)] + [(FEm+UEe) + (NR×K)]} / Fi
NR= (Fi×Nf) - (E×Ne)

Where:
TME: true metabolizable energy (kcal/g).
TMEn: true metabolizable energy corrected for nitrogen (kcal/g).
F_i: feed intake (g).
E: excreta (g).
GE_f: gross energy of feed sample (kcal/g).
GE_e: gross energy of excreta (kcal/g).
FE_m: metabolic fecal energy (kcal/g).
UE_e: indigenous urinary energy (kcal/g).
NR: nitrogen retention (g).
K: nitrogen retention correction coefficient (8.22 kcal/g for each g N).
NF: feed nitrogen (%).
N_e: fecal nitrogen (%).

Experiment 2

Dietary treatments and feeding schedules

A total of 200 newly hatched male broiler chicks (Ross 308) were purchased from a local hatchery. On arrival, all birds were weighed and randomly assigned to 1 of 4 treatments, with 5 replicate pens/treatment and 10 chickens/pen.Dietary treatments (Table 6) were formulated to meet or exceed the nutrient requirements of broilers provided by Ross Broiler Manual (Aviagen, 2014). The MEn value for GP was assumed 1839.83 kcal/kg based on experimen1. Experimental diets were as follows: 1) control corn, soybean diet (C); 2) C + 10% of GP; 3) C + 10% GP + T1 (500 mg/kg tannase enzyme) and 4) C + 10% GP + T2 (1000 mg/kg tannase enzyme).

The straw was substituted by GP in the experimental diets.The feeding regimen consisted of a starter (1 to 10 d), grower (11 to 24 d), and finisher diet (25 to 42 d). Chickens were raised in floor pens (100×120 cm) and had free access to feed and water for the entire experimental period (days 0-42). The room temperature gradually decreased from 33 to 22 ˚C on day 28 and then remained constant thereafter. The lighting program was a period of 20 h of light and 4 h of darkness. All experimental procedures were evaluated and approved by the Institutional Animal Care and Ethics Committee of the Mohaghegh Ardabili University. An enzyme with tannase activity (T) supplied by Kikkoman Foods Products Company (Edogawa Plant, Japan) containing tannin acylhydrolase (500 U/g, EC 3.1.20) was used.

Performance data

Birds and feed were weighed at 10, 24 and 42 d of age. The values of AFDI and ADG were recorded in different periods, and the feed-to-gain ratio (F:G) was calculated. Mortality was also recorded as it occurred. However, AFDI and F:G was corrected for the mortality of related groups.

Statistical analysis

Data were analyzed in a completely randomized design using the General Linear Models procedures of SAS (2001). When the differences were significant ($P<0.05$), mean values between treatments were compared using the Duncan test.

Table 1 Chemical composition (%) , gross energy (kcal/kg) and total polyphenol (mg gallic acid equivalent/g) of grape pomace samples

Item	Grape pomace samples			Mean	SD	CV
	1	2	3			
Dry matter	91.82	91.07	91.46	91.45	0.37	0.41
Crude protein	9.13	8.87	8.83	8.94	0.16	1.82
Ether extract	6.71	6.28	8.03	7	0.91	13.01
Ash	3.75	3.09	2.94	3.26	0.43	13.21
Crude fiber	31.08	27.94	31.58	30.20	1.97	6.53
Calcium	0.59	0.56	0.42	0.52	0.09	17.23
Phosphorus	0.39	0.26	0.24	0.29	0.08	27.45
Gross Energy	4346.72	4264.88	4581.28	4397.63	164.22	3.73
Total polyphenol	34.17	34.36	33.22	33.92	0.48	0.56

SD: standard deviation and CV: coefficient of variation.

Table 2 Ingredients of basal and experimental diets (experiment 1)

Ingredients (%)	Dietary treatment[1]			
	Diet 1	Diet 2	Diet 3	Diet 4
Corn	61.38	55.24	55.24	55.24
Soybean meal (44% CP)	30.55	27.50	27.50	27.50
Soybean oil	4.28	4.28	4.28	4.28
Dicalcium phosphate	1.4	1.4	1.4	1.4
Limestone	1.17	1.17	1.17	1.17
Sodium chloride	0.25	0.25	0.25	0.25
Vitamin and mineral premix[2]	0.5	0.5	0.5	0.5
DL-methionin	0.251	0.251	0.251	0.251
L-lysin, HCL	0.144	0.144	0.144	0.144
Grape pomace (GP)[3]	-	10	10	10
Enzyme	-	-	+	+

[1] The experimental diets were as follows: 1) GP free diet (100% basal corn, soybean diet); 2) 90% basal diet + 10% GP; 3) 90% basal diet + 10% GP + 0.05% tannase; 4) 90% basal diet + 10% GP + 0.1% tannase.
[2] Each kg of vitamin and trace mineral premix provided: vitamin A: 900 IU; vitamin D: 2000 IU; vitamin E: 18 IU; vitamin K: 2 mg; vitamin B_1: 1.8 mg; vitamin B_2: 6.6 mg; vitamin B_6: 3 mg; vitamin B_{12}: 15 µg; Niacin: 30 mg; Pantothenic acid: 10 mg; Biotin: 0.1 mg; Folic acid:1.25 mg; Choline chloride: 200 mg; Fe: 50 mg; Cu:10 mg; Mn: 100 mg; Zn: 85 mg; I: 0.8 mg and Se: 0.2 mg.
[3] 10% GP without enzyme or supplemented with 0.05 or 0.1% tannase enzyme was substituted by the corn and soybean in the experimental diets.

RESULTS AND DISCUSSION

Experiment 1

The chemical composition of GP samples is presented in Table 1. The mean value of dry matter (DM), crude protein (CP), crude fiber (CF), ash, ether extract (EE), calcium (Ca), phosphorus (P), gross energy and total polyphenols content were 91.45, 8.94, 30.2, 3.26, 7, 0.52, 0.29%, 4397.63 kcal/kg and 33.92 mg gallic acid/g ,respectively. The obtained data from this study was similar to values reported by Pirmohammadi *et al*. (2007). Unfortunately, there are no data for a GP in the NRC (1994) to compare with the results of this study.

Mean CP, CF, EE and total polyphenols content, obtained in the present study was less (8.94, 30.2, 7 and 33.92 *vs*. 13.79, 32.5, 10.26 and 48.70, respectively) than that reported by Goni *et al*. (2007). Brenes *et al*. (2016) reported the Ca and P content of GP samples to be 0.5 and 0.3%, respectively. In the current study, mean Ca and P content was 0.52 and 0.29%, respectively. Ash content averaged 3.26% and was higher (3.26% *vs*. 2.41%) than that reported by Goni *et al*. (2007).

However, any differences in chemical composition and mineral content between plant material were due to the agronomic and climatological conditions of the area that the plants are grown. Feed intake, nitrogen retention, AME and AMEn of experimental diets are presented in Table 3. The feed intake of the birds that fed the diet containing 10 % GP was less than those fed other diets (P<0.01). The enzyme supplementation increased feed intake (P<0.01) and birds fed on a diet containing 0.1% tannase had an increased feed intake compared to birds fed diet containing 0.05% tannase enzyme (P<0.01). Similarly, AME and AMEn were significantly reduced (P<0.01) on the diet containing 10% GP but, increased by diets with enzyme supplementation (P<0.01). Nitrogen retention was significantly affected by experimental diets (P<0.01). The nitrogen retention of birds fed the diet containing 10% GP, was significantly less than those fed other diets (P<0.01). The GP contains high levels of fiber and polymeric polyphenols as tannins with the capacity to bind and precipitate both dietary and endogenous proteins, and therefore the incorporation of GP at high doses in chicken diets might impair feed intake and nitrogen retention (Chamorro *et al*. 2015).

Table 3 Feed intake, nitrogen retention and apparent metabolizable energy of experimental diets

Experimental diets[1]	Feed intake(g)	AME (kcal/kg)	AMEn (kcal/kg)	Nitrogen retention (g)
Diet 1	541.162[a]	3215.95[a]	3203.77[a]	0.802[ab]
Diet 2	410.659[c]	2859.68[d]	2852.52[d]	0.359[c]
Diet 3	492.098[b]	2901.66[c]	2890.01[c]	0.697[b]
Diet 4	551.452[a]	3012.07[b]	2998.31[b]	0.924[a]
SEM	14.655	35.880	35.530	0.057
P-value	< 0.01	< 0.01	< 0.01	< 0.01

[1] The experimental diets were as follows: 1) GP free diet (100% basal corn, soybean diet); 2) 90% basal diet + 10% GP; 3) 90% basal diet + 10% GP + 0.05% tannase; 4) 90% basal diet + 10% GP + 0.1% tannase.
AME: apparent metabolizable energy and AMEn: apparent metabolizable energy corrected for nitrogen.
The means within the same row with at least one common letter, do not have significant difference (P>0.05).
SEM: standard error of the means.

Although the information on enzyme application to GP is scarce, the results of the present study showed that utilization of enzymes with the capacity to hydrolyse polymeric polyphenols present in GP, might improve the feed intake and AME content of the diets.

Apparent metabolizable energy content(AME) and metabolizability(AMEn/GE) of GP samples are presented in Table 4. The AME and AMEn content of GP samples were significantly affected by experimental diets (P<0.01). The AME and AMEn of GP in diet with 10% GP, was significantly less than GP samples in diets containing 90% basal diet + 10% GP + 0.05% tannase and 90% basal diet + 10% GP + 0.1% tannase (P<0.01). The mean value of AME and AMEn content was 2642.19 and 2641.08 kcal/kg, respectively.

The metabolizability in diet containing 90% basal diet + 10% GP + 0.1% tannase was higher (79.34%) than other diets (P<0.01). The addition of tannase enzyme, improved the AME content and metabolizability (AMEn/GE) of GP samples. Moreover, AME determination can be a highly variable measurement (Dozier *et al*. 2001; Batal and Dale, 2006). In addition, variability associated with feed intake and excreta measurements in balance experiments can make differences due to treatments with low inclusion levels of a test ingredient.

True metabolizable energy content and metabolizability of GP samples are presented in Table 5. The mean value of TME and TMEn content of GP was 1844.14 and 1839.83 kcal/kg, respectively. The metabolizability (TMEn/GE) of GP was 56.41%. Unfortunately, there are no data for AME and TME of GP in the NRC (1994) to be compared with the results of this study. It seems that high level of fiber and tannins are the main cause of low TMEn and metabolizability of GP. Fiber may interfere with protein and mineral digestion, whereas tannin binds and precipitate both dietary and endogenous proteins (Pirmohammadi *et al*. 2007).

Experiment 2

The effect of feeding diets containing GP and tannase enzyme on broiler BW, ADG, AFDI, and F:G is shown in Table 7.

At 10 d of age, the BW of the control groups was higher (P<0.05) compared with other experimental groups. Furthermore, broilers fed the control diet had higher (P<0.05) ADG in the starter (days 0-10) experimental periods compared with other treatments, but the addition of GP in the chicken diets did not impair growth performance (BW, ADG, ADFI and F:G) of birds fed the grower (days 11-24), finisher (25-42) and the overall (days 0-42) experimental periods compared with other treatments.

There are few references in the literature in relation to the use of grape byproducts in chicken feed. Hughes *et al*. (2005) and Lau and King (2003) reported growth depression in chickens fed diets containing grape seed extract. The effect of polyphenols has also been studied in chickens using ingredients like sorghum and faba bean. In general, relatively high dietary concentrations of polyphenols by the addition of these ingredients reduced performance in chickens as well as other livestock (Gualtieri and Rapaccini, 1990; Jansman *et al*. 1989; Nyachotti *et al*. 1997). Polyphenolic compounds are also known for their ability to interact with different molecules such as proteins. Binding of polyphenolic compounds to both dietary and endogenous proteins, such as digestive enzymes and proteins located at the luminal side of the intestinal tract, have been used to explain the reduced apparent digestibility of protein in the polyphenol-containing diets. GP contains a high level of fiber and polymeric polyphenols as procyanidins with the capacity to bind and precipitate both dietary and endogenous proteins, and therefore the incorporation of GP at high doses in chicken diets might impair nutrient digestion and growth. Thus, in the present study, we used high doses of GP and because all diets were formulated to contain the same fiber content, any difference should be attributed to the polyphenol content. The effect of feeding diets containing GP supplemented with the tannase enzyme on growth performance of chickens is shown in Table 7. No effect (P>0.05) of dietary treatments were observed on body weight and feed consumption. There are several methods which can be used to reduce or neutralize the negative effects of tannins in poultry feeds so as to increase the efficiency of feed utilization.

Table 4 Apparent metabolizable energy content and metabolizability of grape pomace samples*

Grape pomace samples	AME (kcal/kg)	AMEn (kcal/kg)	Metabolizability (%)[1]
Diet 2	2030.1[c]	2001[c]	75.47[c]
Diet 3	2420.8[b]	2405.1[b]	76.47[b]
Diet 4	3504.8[a]	3488[a]	79.34[a]
SEM	198.69	193.47	0.512
P-value	< 0.01	< 0.01	< 0.01
Mean	2642.19	2641.08	77.09

[1] AMEn/GE.
* The experimental diets were as follows: 1) GP free diet (100% basal corn, soybean diet); 2) 90% basal diet + 10% GP; 3) 90% basal diet + 10% GP + 0.05% tannase; 4) 90% basal diet + 10% GP + 0.1% tannase.
AME: apparent metabolizable energy and AMEn: apparent metabolizable energy corrected for nitrogen.
The means within the same row with at least one common letter, do not have significant difference (P>0.05).
SEM: standard error of the means.

Table 5 True metabolizable energy (TME) content and metabolizability of grape pomace samples*

Energy content	**Grape pomace samples**						**Mean**	**SD**	**CV**
(kcal/kg)	1	2	3	4	5	6			
TME	1831.71	1827.14	1799.16	1939.88	1891.63	1775.33	1844.14	61.029	3.30
TMEn	1827.60	1822.83	1794.90	1935.52	1887.23	1770.92	1839.83	61.006	3.31
Metabolizability (%)[1]	56.04	55.89	55.04	59.35	57.87	54.30	56.41	1.871	3.31

[1] TMEn/GE.
* The experimental diets were as follows: 1) GP free diet (100% basal corn, soybean diet); 2) 90% basal diet + 10% GP; 3) 90% basal diet + 10% GP + 0.05% tannase; 4) 90% basal diet + 10% GP + 0.1% tannase.
TME: True metabolizable energy and TMEn: True metabolizable energy corrected for nitrogen.
SD: standard deviation and CV: coefficient of variation.

Table 6 Ingredients and nutrient composition of basal diets (experiment 2)

Ingredients	**Starter (days 0-10)**	**Grower (days 11-24)**	**Finisher (days 25-42)**
Corn	42.53	44.52	48.75
Soybean meal (44% CP)	41.18	37.87	32.76
Soybean oil	5.99	7.27	7.77
Straw[1]	5.93	6.40	7.06
Grape pomace[2]	0	0	0
Dicalcium phosphate	1.75	1.57	1.40
Limestone	1.10	0.99	0.94
Sodium chloride	0.35	0.35	0.35
Vitamin and mineral premix[3]	0.5	0.5	0.5
DL-methionine	0.36	0.32	0.29
L-lysine, HCl	0.22	0.15	0.14
L-threonine	0.10	0.07	0.05
Calculated analysis			
Metabolizable energy	2900	3000	3100
Crude protein	22.22	20.81	18.89
Methionin + cystine	1.04	0.96	0.88
Lysine	1.39	1.25	1.12
Ether extract	7.87	9.19	9.80
Crude fiber	6.20	6.20	6.20
Ca	0.93	0.84	0.77
Available P	0.46	0.42	0.38

[1] The straw was substituted by grape pomace in the experimental diets.
[2] Experimental diets were as follows: 1) Control corn, soybean diet (C); 2) C + 10% of GP; 3) C + 10% GP + T1 (500 mg/kg tannase enzyme); 4) C + 10% GP + T2 (1000 mg/kg tannase enzyme).
[3] Each kg of vitamin and trace mineral premix provided: vitamin A: 900 IU; vitamin D: 2000 IU; vitamin E: 18 IU; vitamin K: 2 mg; vitamin B_1: 1.8 mg; vitamin B_2: 6.6 mg; vitamin B_6: 3 mg; vitamin B_{12}: 15 µg; Niacin: 30 mg; Pantothenic acid: 10 mg; Biotin: 0.1 mg; Folic acid:1.25 mg; Choline chloride: 200 mg; Fe: 50 mg; Cu:10 mg; Mn: 100 mg; Zn: 85 mg; I: 0.8 mg and Se: 0.2 mg.

The effectiveness of enzymes to enhance nutrient digestibility has been reported (Choct, 2006). However, Torki and Farahmand-Pour (2007) observed no significant effect of enzyme supplementation on performance of broiler chickens fed sorghum based diets.

The utilization of enzymes with the capacity to hydrolyse complex polyphenols present in GP, might allow the use of higher doses of GP in chicken diets.

Table 7 The effects of different dietary treatments on broiler body weight (BW), average daily gain (ADG), average daily feed intake (ADFI) and feed-to-gain ratio (F:G)

Item	Dietary treatment[1]				SEM	P-value
	Control	C + 10 GP	C + 10 GP+T1	C + 10 GP + T2		
BW, g						
10 d	248.32[a]	214.32[b]	202.64[b]	217.04[b]	5.836	0.022
24 d	979.68	902.61	910.87	912.96	15.754	0.291
42 d	2416.20	2272.46	2275.92	2358.74	40.280	0.560
ADG, g						
Starter (0-10 d)	20.52[a]	17.15[b]	15.96[b]	17.17[b]	0.580	0.019
Grower (11-24 d)	52.24	49.16	50.58	49.70	1.089	0.796
Finisher (25-42 d)	79.80	76.09	75.83	80.31	2.273	0.867
Overall (0-42 d)	56.50	53.08	53.16	55.13	0.959	0.560
ADFI, g						
Starter (0-10 d)	32.80	24.60	23.37	21.88	1.587	0.052
Grower (11-24 d)	101.68	93.96	98.17	97.34	1.544	0.392
Finisher (25-42 d)	161.95	150.30	151.45	165.28	5.435	0.732
Overall (0-42 d)	113.46	101.90	104.26	109.24	2.087	0.207
F:G, g:g						
Starter (0-10 d)	1.60	1.42	1.46	1.26	0.058	0.235
Grower (11-24 d)	1.96	1.91	1.94	1.96	0.026	0.933
Finisher (25-42 d)	2.03	1.98	1.98	2.05	0.038	0.909
Overall (0-42 d)	2.00	1.92	1.96	1.98	0.015	0.241

[1] Experimental diets were as follows: 1) Control corn, soybean diet (C); 2) C + 10% of GP; 3) C + 10% GP + T1 (500 mg/kg tannase enzyme); 4) C + 10% GP + T2 (1000 mg/kg tannase enzyme).
The means within the same row with at least one common letter, do not have significant difference (P>0.05).
SEM: standard error of the means.

Thus, in the present study, we used high doses (10%) of GP and hypothesized that the use of enzymes with tannase activities might hydrolyze the complex polyphenol structure of grape pomace into more simple phenols reduce the negative effects of complex polyphenols in feeds. In summary, the degradation and absorption of polyphenols within the gastrointestinal tract depend on the nature not only of the phenolic compound, but also of the intestinal microflora, which fermentative effect on other dietary components will be affected, conversely, by the type of polyphenolic compound (Bravo, 1998). Among diets supplemented with GP, feed intake and body weight of broilers feeding diet containing 0.1% tannase were higher. It seems that the tannase supplementation increased feed intake compared to birds fed diet containing 0.05% or unsupplemented tannase enzyme. Higher BW of broiler feed diet supplemented with 0.1% tannase can related to improving in ME content of the diet that we detected in experiment 1.

CONCLUSION

The results of this research showed that GP has 2030.1 and 1839.83 kcal/kg AMEn and TMEn, respectively. The addition of 10 percent GP in the chicken diets did not impair growth performance and the supplementation of tannase enzyme, improve the metabolizable energy content of GP and increase the nutritive value of grape pomace in poultry diets.

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