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Effect of enzyme addition on energy utilization and performance of broiler chickens fed wheat-based diet with different metabolizable energy levels

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ABSTRACT. An experiment was carried out to investigate the effect of multi-enzyme in high and low levels of metabolizable energy (13.81 and 11.51 MJ kg^{-1} diet) on performance and energy utilization of broilers fed wheat-soybean meal diets from 0 to 21 days of age. Result showed that birds fed diets containing 11.51 MJ kg^{-1} consumed significantly ($p < 0.05$) more feed than diets containing 13.81 MJ kg^{-1}, whereas daily gain and feed conversion ratio improved ($p < 0.05$) when enzyme was added to 11.51 MJ kg^{-1} diet. There was significant improvement in metabolizable energy, net energy for production, organic and dry matter digestibility in 0-21 when diets supplemented with enzyme ($p < 0.05$). Addition of enzyme to 11.51 MJ kg^{-1} containing diet significantly ($p < 0.05$) reduced heat production of birds in 0-10 d, whereas heat production was not changed in 21 days. Supplementation of 11.51 MJ kg^{-1} diet with enzyme improved the efficiency of ME use for carcass energy and protein retention of broilers ($p < 0.05$). Generally, the results of current study demonstrated that addition of enzyme to wheat-soybean diets improved NEp of broiler chickens while MEI was not changed and it seems NEp is a more sensitive energy utilization response criterion to use in evaluating broilers response to enzyme supplementation.

Keywords: enzyme; net energy; wheat; body composition; broilers.

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Introduction

Anti-nutritional compounds known as non-starch polysaccharides (NSPs) are the most important factor has confined feeding wheat to broiler chickens especially at early days of growing period. The presence of soluble NSP can cause highly viscous conditions in the gastrointestinal tract and decrease contact between digestive enzymes and substrates, resulting in poor nutrient digestion and performance broiler chickens (Choct, Sinlae, Al-Jassim, & Pettersson, 2006; Moftakharzadeh, Moravej, & Shivazad, 2017; Nian, Guo, Ru, Li, & Peron, 2011). Supplementation of wheat-based diet with exogenous glycanase appear to enhance growth performance and energy utilization alongside with (Hashemipour, Khaksar, Rubio, Veldkamp, & Van Krimpen, 2016; Kiarie, Romero, & Ravindran, 2014; Pirgozliev et al., 2015; Wu, Choct, Wu, Liu, & Swick, 2017) or without reduction of intestinal viscosity of broilers (Amerah, Ravindran, Lentle, & Thomas, 2008). Some reports indicate that enzyme supplementation is more effective in younger broiler chickens, which is may related to inefficient production of endogenous digestive enzymes at this period (Almirall, Francesch, Perez-Vendrell, Brufau, & Esteve-Garcia, 1995). Olukosi, Cowieson, and Adeola (2008) demonstrated that broiler chickens benefited more from enzyme supplementation at younger age and contribution of the enzymes to nutrient retention reduce with age in chickens. Another key factor for rapid growth of broiler chickens that promote enzyme effects is energy concentration of diet. Many researchers noted that improvement occurred in metabolizable energy (ME) availability with enzyme supplementation is greater at low-energy diet compared to high-energy food (Cho, Zhao, & Kim, 2012; Gitoee, Janmohammadi, Taghizadeh, & Rafat, 2015; Zhou, Jiang, Lv, & Wang, 2009).

ME has been commonly accepted and extensively used to energy evaluation of diets for poultry, which calculate energy loss of excreta. However, this system is not capable of accounting for losses of chemical energy as heat production (Pirgozliev & Rose, 1999). Another measure of the energy value of a feed is net

energy (NE) system in which efficiency of ME utilization is considered and it might be more sensitive compared to ME for determination of energy utilization efficiency in poultry (De Groote, 1974; Pirgozliev, Rose, Kettlewell, & Bedford, 2001). Pirgozliev, Bedford, Acamovic, Mares, and Allymehr (2011) confirmed a high correlation between BW and NE for production. According to definition, NE is the amount of energy that is available for animal after ME has been used to provide the heat increment of feeding. The NE of a sample feed represents the energy available for productive purposes (NEp), and takes account of the losses in the metabolism of absorbed nutrients (NEm). It seems that NEp may be more sensitive and accurate evaluation of energy truly used by the chickens receiving enzyme since it takes into account the efficiency of ME utilization for growth. There are two different methods identified for NE determination including carbon-nitrogen method and comparative slaughter technique and both of them can be used to determine NE when diet include exogenous enzyme as reported by many researchers (Barekatain, Antipatis, Choct, & Iji, 2013; Nian et al., 2011; Olukosi et al., 2008). However, in comparison with carbon-nitrogen, the comparative slaughter method is more economical and simulates the real rearing environment (Sakomura, Silva, Couto, Coon, & Pacheco, 2003).

The aim of this study was to evaluate the effect of enzyme addition on performance, ME, NEp, heat production, protein and fat retention in broilers fed wheat-based diets with low and high energy concentration.

Material and methods

Diets and enzyme

The experimental diets were formulated with two levels of ME (11.51 and13.81 MJ kg^{-1}) representing high and low concentration of diet energy content for broiler chickens and two enzyme inclusion rates (0 and 150 mg kg^{-1}). The basal diet was wheat–soybean and birds were fed the experimental diets for 21 days. The ingredients compositions of diets are shown in Table 1. Throughout the experiment, feed and water were available for *ad libitum* consumption. Titanium oxide was added to the diets as an indigestible marker to enable determination of digestibility. The enzyme preparation used in this study was a commercial multi-enzyme complex Grindenzyme, provided 1500 BGU endo-1,4-b-glucanase, 3600 FXU endo-1,4-b-xylanase. One unit (U) of xylanase was defined as the quantity of the enzyme that liberates 1 mmol xylose equivalent min^{-1}.

Table 1. Ingredient composition of experimental diets of the experimental diets (g kg^{-1} of diet).

Ingredients	T_1 (A_1B_1)	T_2 (A_1B_2)	T_3(A_2B_1)	T_4(A_2B_2)
Wheat	673.2	673.2	581.5	581.5
Soybean meal	245.1	245.1	285.9	285.9
Gluten meal	11.8	11.8	40.4	40.4
Soya oil	23.2	23.2	38.2	38.2
Dicalcium phosphate	11.7	11.7	12.3	12.3
Limestone	7.3	7.3	11.8	11.8
Sodium chloride	3.1	3.1	3.2	3.2
Titanium oxide marker	17	17	17	17
Vitamin premix[1]	2.5	2.5	2.5	2.5
Mineral premix[2]	2.5	2.5	2.5	2.5
L-lysine-HCL	0.5	0.5	1	1
L-threonine	1.1	1.1	1.2	1.2
DL-methionine	1	1	2.5	2.5
Enzyme, mg kg^{-1} of diet	-	150	-	150
Calculated nutrient contents				
ME, MJ kg^{-1}	11.51	11.51	13.81	13.81
Crude protein, g kg^{-1}	204.4	204.4	231.1	231.1
Calcium, g kg^{-1}	9.4	9.4	10.6	10.6
Available phosphorus, g kg^{-1}	4.3	4.3	4.9	4.9
Arginine, g kg^{-1}	14.1	14.1	14.9	14.9
Lysine, g kg^{-1}	13	13	14.2	14.2
Methionine+cyctine, %	9.6	9.6	10.9	10.9
Dietary Cation-Anion balance	250	250	250	250

[1]Provided the following (per kg of diet): vitamin A (transretinyl acetate), 9,000 IU; vitamin D3 (cholecalciferol), 2,000 IU; vitamin E (allrac- tocopherol acetate), 18 IU; vitamin K (bisulfate menadione complex), 2 mg; riboflavin, 6.6 mg; pantothenic acid (D-calcium pantothenate), 10 mg; pyridoxine (pyridoxine HCl), 3 mg; folic acid, 1 mg; thiamin (thiamin mononitrate), 1.8 mg; vitamin B12 (cyanocobalamin), 15 µg; D-biotin, 0.1 mg; niacin, 30 mg; choline (choline chloride), 500 mg and ethoxyquin, 0.1 mg. [2]Provided the following (per kg of diet): Se (Na2SeO3), 0.2 mg; I (KI), 1 mg; Cu (CuSO4 .5H2O),10 mg; Fe (FeSO4 .7H2O), 50 mg; Zn (ZnO), 85 mg and Mn (MnSO4.H2O),100 mg.

Birds and management

A total of 224-day-old male Ross broiler chickens were used for the current study. At the first day, the slaughter groups, comprising 24 chicks, were killed before starting the trial. The remaining 200 birds were allocated into four dietary treatments in a randomized complete design. Each treatment had five replicate cages of equal body weight, with ten birds per replicate cage. The other slaughter groups of 40 chicks each made up the final slaughter groups that were killed by cervical dislocation at days 10 and 21. On slaughter days, after weighing the birds, feed was withdrawn for 5 hours before cervical dislocation. The birds were subsequently frozen after slaughter and prior to processing. Parameters measured at the end of each period were feed intake (FI), feed conversion ratio (FCR) and average daily gain (ADG). In order to determine ME, excreta were collected from each cage in the last 5th day of each week. Before being dried, the excreta were immediately frozen in a forced air oven to a constant weight. Then, samples were pooled within each pen and ground prior to analyses.

Chemical analysis

To determine the ME, grab samples excreta and diets samples were analyzed for gross energy, dry matter (DM) and organic matter. Titanium concentration in the diets and excreta samples was determined using the method of Short, Gorton, Wiseman, and Boorman (1996). The whole intact chicken (feathers, head, feet and all organs) was frozen immediately after being killed and later processed. Frozen birds were partially thawed, coarse-ground, and thoroughly mixed in a blender to obtain a homogenous subsample. To calculate DM of carcasses, the wet subsamples were accurately weighed before and after freeze-drying and finely ground and kept in an air-tight container before further analyses. Gross energy, fat, and protein contents of samples were determined according to the methods of Association Official Analytical Chemist (AOAC, 2005).

Calculations

ME (MJ kg^{-1}) was calculated as follows:

(A) $$ME = GE_i - \left[GE_0 - C_i / C_0\right]$$

where GE_i is gross energy (MJ kg^{-1}) in feed; GE_0 is the gross energy (MJ kg^{-1}) in excreta, C_i is the concentration of chromium in the diets; and C_0 is the concentration of chromium in the excreta.

Net energy for production (NEp) was calculated as follows:

(B) Initial GE of carcass (kJ)= carcass GE $\left(\frac{kJ}{g}\right)$ × body weight of bird (g)

(C) Final GE content of carcass (kJ) = carcass GE (kJ g^{-1}) × body weight of bird (g)

(D) NEp (kJ) = (C) – (B).

Heat production (HP), which consists of the heat increment of feeding and fasting HP is calculated as the difference between NEp and ME intake:

(E) HP (kJ) = MEI – NEp

where ME intake (MEI) was calculated using the following formula:

(F) MEI (kJ) = ME (kJ g^{-1}) × feed intake (g).

Energy retained as fat (RE_f) and as protein (RE_p) were calculated as follows:

(G) RE_f (kJ) = Carcass fat (g) × 38.2 kJ g^{-1}

(H) RE_p (kJ) = Carcass crude protein content (g) × 23.6 kJ

The values 38.2 and 23.6 kJ g^{-1} are energy values per gram of fat and protein, respectively, and were according to Olukosi et al. (2008).

ME intake, DM and OM digestibility were determined for each period (10 and 21 days). Therefore, these parameters for chickens killed at day 10 were calculated as shown earlier using days 0–10 feed intake. The ME intake, DM and OM digestibility for chickens killed at day 21 was calculated by adding the ME intakes, DM and OM digestibility from 0 to 10 and 11 to 21 periods.

(I) Efficiency of ME use for energy retention ($K_{RE)}$ = NE_p / MEI

(J) Efficiency of ME use for lipid retention (K_{REf}) = RE_f / MEI

(K) Efficiency of ME use for protein retention (K_{REp}) = RE_p / MEI

Statistical analysis

The study was conducted in a completely randomized factorial with two levels of enzyme and ME. Data obtained were analyzed using GLM procedure of Statistical Analysis System (SAS, 2004). For all variables, when a significant difference was detected, means were separated using least squares means option of SAS (2004) at $p < 0.05$.

Results

Growth performance and ME

The growth performance data are presented in table 2. In the 0–10 d and 0-21 d periods, FI was significantly ($p < 0.05$) affected by energy level and as amount of energy of diet increased, feed consumption of birds were reduced ($p < 0.05$). On the other hand, there was significant ($p < 0.05$) main effect of enzyme on ADG and FCR of broilers at the 0–10 d and in 0-21 d periods and only addition of enzyme to 11.51 MJ kg^{-1} energy containing diet significantly ($p < 0.05$) improved ADG and FCR of broilers. Birds fed 11.51 MJ kg^{-1} diet without enzyme supplementation, however, had the poorest ($p < 0.05$) FCR and there was no significant difference among other treatments in 0-21 d ($p > 0.05$).

According to table 3, there were significant main effects ($p < 0.05$) of enzyme and energy level as well as enzyme × energy interaction ($p < 0.05$) on ME of the diets. ME improvement increased as energy concentration of diet for chicken reduced, and the highest improvement ($p < 0.05$) occurred when broilers consumed diet containing 11.51 MJ kg^{-1} with enzyme.

Digestibility of DM and OM

The effects of dietary treatments on digestibility of OM and DM are shown in Table 3. In the 0-10 d and in 0-21 d periods, there was only significant main effect of enzyme on digestibility of OM and DM and these criterions improved when enzyme was added to both 11.51 MJ kg^{-1} and 13.81 MJ kg^{-1} energy containing diets ($p < 0.05$). There was no significant difference between enzyme treated groups ($p > 0.05$).

Energy utilization

Based on Table 4, there was only main effect of enzyme ($p < 0.05$) on MEI in 0-10 and in 0-21 d periods. Feeding diets containing enzyme significantly improved NEp in 0-10 d period. Overall, in 0-21 d period, only feeding diet containing 11.51 MJ kg^{-1} with enzyme increased ($p < 0.05$) NEp of birds and there was no significant difference among other treatments ($p > 0.05$).

Heat production

As it can be seen in table 4, HP was significantly ($p < 0.05$) decreased when 11.51 MJ kg^{-1} diets supplemented with enzyme in 0-10 d. There were no effects of enzyme, energy as well as enzyme × energy interaction ($p > 0.05$) on this parameter in 0-21 d period.

Table 2. Effects of ME, Enzyme and their interactions on FI, ADG and FCR of broilers in 10 and 21 days of age.

Treatments	Performance					
	Feed intake (g $bird^{-1}$ day^{-1})		Average daily (g $bird^{-1}$ day^{-1})		Feed conversion ratio	
	Period		Period		Period	
	0-10d	0-21d	0-10d	0-21d	0-10d	0-21d
T_1 (A_1B_1)	27.43^a	39.12^a	16.53^b	20.13^b	1.66^a	1.94^a
T_1 (A_1B_2)	27.43^a	38.90^a	21.24^a	24.07^a	1.32^b	1.61^b
T_2 (A_2B_1)	23.97^b	34.48^b	16.67^b	20.11^b	1.41^b	1.72^{ab}
T_2 (A_2B_2)	24.52^b	34.61^b	19.40^{ab}	21.61^{ab}	1.28^{ab}	1.60^b
SEM	0.438	0.854	0.793	0.702	0.0651	0.0597
ME	<0.001	<0.001	0.135	0.096	0.090	0.066
Enzyme	0.323	0.966	<0.001	<0.001	0.002	<0.001
ME × Enzyme	0.986	0.837	0.098	0.100	0.104	0.086

Means with different superscripts on same column differ significantly ($p < 0.05$); A: ME concentration (A_1: 13.81; A_2: 11.51); B: Enzyme inclusion rate (B_1: 0; B_2: 150 mg kg^{-1} diet).

Table 3. Effects of ME, Enzyme and their interactions on metabolizable energy (MJ kg^{-1} DM) content and digestibility of dry matter and organic matter of treatments

Treatments	MEI (kJ d^{-1})				Dry matter digestibility		Organic matter digestibility	
	Period				Period		Period	
	0-10d	Improvement	0-21d	Improvement	0-10d	0-21d	0-10d	0-21d
T_1 (A_1B_1)	11.49^{d}		11.54^{d}		69.55^{b}	69.97^{b}	71.70^{b}	72.40^{b}
T_1 (A_1B_2)	12.01^{c}	0.51	12.01^{c}	0.47	76.83^{a}	76.83^{a}	78.91^{a}	78.82^{a}
T_2 (A_2B_1)	13.14^{b}		13.16^{b}		70.83^{b}	71.36^{b}	72.76^{b}	73.71^{b}
T_2 (A_2B_2)	13.42^{a}	0.28	13.38^{a}	0.22	76.52^{a}	75.23^{a}	79.14^{a}	78.83^{a}
SEM	0.040		0.048		0.962	0.998	0.948	0.971
ME	<0.001		<0.001		0.621	0.915	0.503	0.481
Enzyme	<0.001		<0.001		<0.001	<0.001	<0.001	<0.001
ME × Enzyme	<0.001		<0.001		0.420	0.158	0.664	0.488

Means with different superscripts on same column differ significantly ($p < 0.05$); A: ME concentration (A_1: 13.81; A_2: 11.51); B: Enzyme inclusion rate (B_1: 0; B_2: 150 mg kg^{-1} diet).

Table 4. Effects of ME, Enzyme and their interactions on metabolisable energy intake, net energy for production and heat production (HP) of broilers.

Treatments	MEI (kJ d^{-1})		NEp (kJ d^{-1})		HP (kJ d^{-1})	
	Period		Period		Period	
	0-10d	0-21d	0-10d	0-21d	0-10d	0-21d
T_1 (A_1B_1)	315.21^{a}	498.30^{a}	125.47^{b}	187.79^{c}	189.74^{a}	310.51^{a}
T_1 (A_1B_2)	335.49^{a}	516.52^{a}	178.84^{a}	237.52^{a}	156.64^{b}	279.01^{a}
T_2 (A_2B_1)	315.06^{a}	501.14^{a}	129.98^{b}	191.26^{bc}	185.08^{a}	309.88^{a}
T_2 (A_2B_2)	329.05^{a}	514.25^{a}	160.31^{a}	216.86^{ab}	168.74^{ab}	293.71^{a}
SEM	5.352	11.358	6.260	6.759	6.50	10.711
ME	0.533	0.980	0.243	0.290	0.686	0.497
Enzyme	0.014	0.194	<0.001	<0.001	0.003	0.032
ME × Enzyme	0.617	0.828	0.064	0.117	0.272	0.461

Means with different superscripts on same column differ significantly ($p < 0.05$); A: ME concentration (A_1: 13.81; A_2: 11.51); B: Enzyme inclusion rate (B_1: 0; B_2: 150 mg kg^{-1} diet).

Body Composition and efficiency of ME

Table 5 shows the result of the energy deposited as fat and protein in the carcass. Although energy retained as protein significantly improved ($p < 0.05$) with enzyme supplementation in 0-10 d, but energy retained as fat had not changed among treatments during the same period ($p > 0.05$). In general, carcass energy deposited as fat and protein significantly affected by main effect of enzyme ($pt < 0.05$) in 0-21 d and these criterions were significantly higher when diet containing 11.51 MJ kg^{-1} supplemented with enzyme ($p < 0.05$).

The data on efficiency of use of MEI for NEp, RE_p or RE_f are presented in Table 6. In the 0-10 d period, exogenous enzyme significantly ($p < 0.05$) increased RE_p, and efficiency of MEI use for NEp at both energy levels, whereas RE_f was not changed ($p > 0.05$). During the whole period, only efficiency of use of MEI for NEp significantly improved with enzyme at 11.51 MJ kg^{-1}.

Table 5. Effects of ME, Enzyme and their interactions on retained energy as fat (RE_f) and protein (RE_P) of broilers.

Treatments	RE_f (kJ d^{-1})		RE_p(kJ d^{-1})	
	Period		Period	
	0-10d	0-21d	0-10d	0-21d
T_1 (A_1B_1)	56.21^{b}	84.94^{b}	62.89^{b}	98.95^{b}
T_1 (A_1B_2)	64.71^{a}	101.18^{a}	82.44^{a}	117.59^{a}
T_2 (A_2B_1)	55.68^{b}	84.40^{b}	60.98^{b}	96.38^{b}
T_2 (A_2B_2)	60.86^{a}	90.86^{ab}	77.44^{a}	107.81^{ab}
SEM	4.869	3.182	2.677	3.226
ME	0.659	0.107	0.217	0.074
Enzyme	0.179	0.002	<0.001	<0.001
ME × Enzyme	0.738	0.143	0.569	0.283

Means with different superscripts on same column differ significantly ($p < 0.05$); A: ME concentration (A_1: 13.81; A_2: 11.51); B: Enzyme inclusion rate (B_1: 0; B_2: 150 mg kg^{-1} diet).

Table 6. Effects of ME, Enzyme and their interactions on efficiency of metabolizable energy (ME) use for tissue energy deposition.

Treatments	Efficiencies of ME use for energy retention					
	K_{REf}		K_{Rep}		K_{RE}	
	Period		Period		Period	
	0-10d	0-21d	0-10d	0-21d	0-10d	0-21d
T_1 (A_1B_1)	0.177[a]	0.170[ab]	0.199 [bc]	0.198[ab]	0.398[b]	0.377[b]
T_1 (A_1B_2)	0.193[a]	0.196[a]	0.246[a]	0.227[a]	0.532[a]	0.459[a]
T_2 (A_2B_1)	0.177[a]	0.168[b]	0.193[c]	0.193[b]	0.413[b]	0.382[b]
T_2 (A_2B_2)	0.185[a]	0.177[ab]	0.235[ab]	0.210[ab]	0.486[a]	0.429[ab]
SEM	0.015	0.006	0.009	0.008	0.017	0.012
ME	0.808	0.134	0.430	0.181	0.394	<0.001
Enzyme	0.440	0.019	<0.001	0.0122	<0.001	0.2778
ME × Enzyme	0.822	0.199	0.812	0.439	0.094	0.124

K_{RE}, efficiency of ME use for carcass energy retention; K_{REf}, efficiency of ME use for energy retained as fat; K_{REp}, efficiency of ME use for energy retained as protein. Note: means with different superscripts on same column differ significantly ($p < 0.05$); A: ME concentration (A_1: 13.81; A_2: 11.51); B: Enzyme inclusion rate (B_1: 0; B_2: 150 mg kg^{-1} diet).

Discussion

Growth Performance and ME

The use of wheat instead of corn has become more popular in poultry industry since it contains greater amounts of crude protein, lysine, methionine, arginine, phenylalanine, tryptophan, threonine and valine (Esmaeilipour et al., 2012) and improve quality of pellet feeds (Zimonja & Svihus, 2009). However, the presence of NSP, such as arabinoxylans and β-glucans in wheat has limited use of this feed ingredient in broilers. It has been reported that the soluble arabinoxylan of wheat is responsible for intestinal viscosity and depressed performance of poultry (Kiarie et al., 2014; Rodríguez et al., 2012). This condition would alter the transport of the nutrients at the mucosal surface and limit the capacity to produce the digestive enzymes, especially at early ages (Ribeiro et al., 2011). Application of NSP-degrading enzymes to feeds may unlock the encapsulated nutrients from the cell wall structure and increasing accessibility to digestive enzymes, thus reducing the energy needs for producing some of the digestive enzyme and enhance the performance of broilers (Ravn, Martens, Pettersson, & Pedersen, 2016; Wu et al., 2017).

Furthermore, responses of birds to enzyme were greater especially when lower density of nutrients were used for diet formulation (Cowieson, 2010; Gitoee et al., 2015; Zanella, Sakomura, Silversides, Fiqueirdo, & Pack, 1999). Kocher, Choct, Ross, Broz, and Chung (2003) reported an improvement in nutrient utilization, growth, and FCR of broilers fed a low-energy diet supplemented with enzyme. Zhou et al. (2009) found that xylanase, amylase, and protease was able to increase ME available from corn-soy diets with lower energy contents compared to diets with higher energy levels fed to broiler chickens. In current study, the experimental diets were designed to provide high and low energy levels diets in order to investigate in which certain energy level enzyme supplementation is more effective at the early growth stage for broilers. In current study, the effect of enzyme in terms of growth performance and ME indicated that improvement was more significant at lower energy concentration, which is reported in the literatures (O'Neill, Liu, Wang, Diallo, & Hill, 2012; Zanella et al., 1999).

Digestibility of DM and OM

Digestibility of DM and OM of diet significantly improved with enzyme supplementation in 0-10 d and 0-21d of age. This is in agreement with the observation of Wu et al. (2017), where they investigate carbohydrase effect on broilers fed nutritionally adequate and marginal wheat-based diet. Moreover, Almirall et al. (1995) stated that NSP-degrading enzyme supplementation increased amylase and lipase activities in small intestine contents, alongside with decreasing of intestinal viscosity in early-age broiler chicks. It is proposed that dietary NSP contents reduce feed DM and OM availability probably due to its chemical structure and low digestibility in broilers (Adeola, Jendza, Southern, Powell, & Owusu-Asiedu, 2010). Indeed, in the present study, addition of β-glucanase and xylanase to wheat-based diet result in higher nutrient digestion at both energy levels of diets. It seems exogenous enzyme may have reduced digesta viscosity caused by soluble NSP from wheat and enhanced the accessibility of gastrointestinal enzymes to the cell contents of the grain.

Energy utilization

Based on table 3, there was no significant effect of enzyme on MEI in 1-10 d and in 1-21 d. In contrast with our results, Nian et al. (2011) observed that exogenous xylanase enhanced MEI of broilers fed wheat-based diet. However, supplementation of diets with enzyme in current experiment trend to increase MEI of broiler chickens. According to the formula (F), determination of MEI as a measure of energy utilization response to enzyme addition is based on the amounts of feed consumed and ME of diet. The present study showed that the improvement in ME and feed intake values induced by enzyme at 11.51 MJ kg^{-1} energy was greater compared to 13.81 MJ kg^{-1} energy concentrations, possibly as a result of greater feed consumption and ME digestibility of those diet in broilers. However, when MEI for birds was calculated, the significant difference disappeared. On the other hand, the NEp amount, is depended on gross energy of the body content as well as body weight of chicken (D). An improvement in NSP degradation and nutrient digestion may be responsible for the enhancement in energy utilization. Barekatain et al. (2013) observed that exogenous xylanase improved NEp of birds fed sorghum distillers dried grains using respiratory chamber and comparative slaughter methods. Comparing to MEI, the greater improvement occurred in NEp by enzyme supplementation at both energy levels suggest more reliability of body weight and its gross energy than ME and feed intake to predict true amount of energy released when enzyme is added to diet. The greater NEp for birds received low energy content diet with enzyme could account for higher body weight and available energy of those chickens. It is proposed that extra energy released from nutrients retention would be deposited in the carcass and enhances body weight and tissue energy concentration; as consequence, this energy would be deposited as either fat or protein (Olukosi et al., 2008).

Heat production

In the current study, there was only significant difference in HP among treatments in 0-10 days and enzyme supplementation significantly reduced HP only for birds fed 11.51 MJ kg^{-1}. These results are consistent with Olukosi et al. (2008) who observed addition of xylanase and amylase cocktail decreased heat production of chickens fed diet containing wheat and corn. Interestingly, in their study addition of phytase as only enzyme increased heat loss of broilers. While viscous grains such as wheat or barley increase the relative size and length of gastrointestinal tract of broiler chickens (Fan, Han, Xu, Wang, & Shi, 2009; Moftakharzadeh et al., 2017), addition of enzyme to NSP containing diets in many studies has reduced relative weight of these energetically active organs (Rodríguez et al., 2012; Wang, Qiao, Lu, & Li, 2005). One possible reason for lower HP in enzyme treatments would be lower maintenance of digestive active organs induced by enzyme supplementation. In addition, it seems exogenous enzyme enhanced NEp of chickens especially at lower energy concentration, which could partly explain decreased expenditure of energy consumed by birds. Although heat production was not significantly affected by treatment, the enzyme supplementation trend to reduce HP of birds fed two certain ME levels diet during the whole period. The results reported here are in agreement with Barekatain et al. (2013), who hypothesized that the ME level of diets containing high fiber cereal may overestimate the energy available for productive uses and may result in higher heat production and enzyme could alleviate this negative effect of these indigestible components. However, the reduction was numerically higher at 11.51 MJ kg^{-1} ME, which is probably related to greater improvement that occurred in NEp when enzyme was added to diet (formula E).

Body Composition and ME efficiency

As it presented, in 0-10 d, addition of enzyme at both energy levels only improved protein retention, while in 0-21 d effects of enzyme supplementation at lower energy concentration diet was more apparent and caused higher quantity of nutrients and energy release from diet, possibly, resulting in higher carcass fat and protein. Many researchers report increase in abdominal fat of birds when wheat diet supplemented with carbohydrase especially at earlier ages. However, energy deposited as protein was more than fat in all periods. Similarly, Bregendahl, Sell, and Zimmerman (2002) observed higher protein deposition in carcass than fat in broilers at 21 d fed a common corn-soybean meal diet. In accordance with current experiment, Olukosi et al. (2008) observed that xylanase addition improved the efficiency of ME intake utilization for protein deposition, while ME efficiency for fat and energy retention had not changed. They concluded that the reason for higher efficiency of ME on protein compared to fat is possibly because the broiler chicks at that age 0–21 d were still actively growing and have not reached the stage at which fat deposition can overtake protein deposition as it confirmed by Hellwing, Tauson, and Skrede (2006).

Conclusion

In conclusion, the current study demonstrates that dietary supplementation of enzyme at 11.51 MJ kg^{-1} and 13.81 MJ kg^{-1} affects energy partitioning resulting in higher of NEp compared to basal diets, while MEI and HP was not changed during the whole study period. Enzyme supplementation only at 11.51 MJ kg^{-1} increases fat and protein retained in the carcass. According to the results, it seems NEp is more sensitive measure to evaluate the value of β-glucanase and xylanase for broilers fed wheat-soybean diet. In addition, determination of NEp by comparative slaughter method allows the partitioning of energy deposition and hence allows the evaluation of the effect of exogenous enzyme use on efficiency of energy utilization.

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