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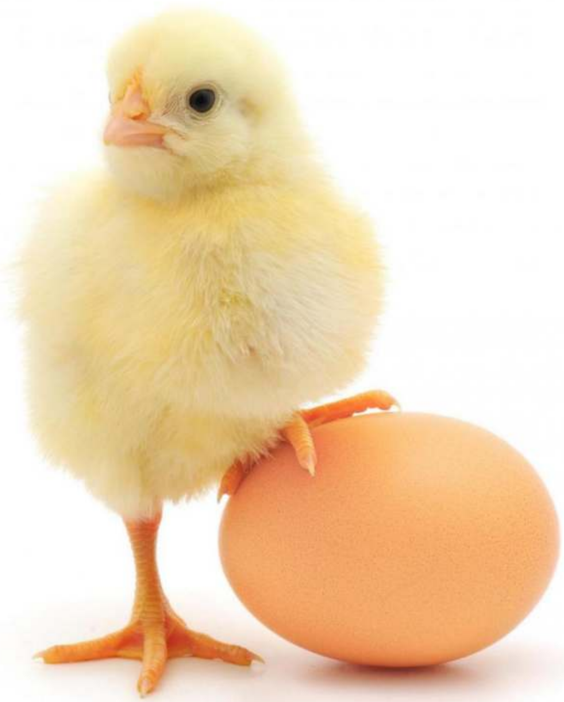

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Effects of Thermostable Phytase Supplementation on the Growth Performance and Nutrient Digestibility of Broilers

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Abstract: A study was conducted to investigate the effects of thermostable phytase supplementation on the growth performance and nutrient digestibility of broilers for 42 d. 1-day-old Arbor Acres chicks with similar initial body weight were randomly divided into 3 treatments consisting 6 pens of 20 chicks per pen. The control group was given maize-soybean meal basal diet and the other two groups were fed the basal diet including either 500 U ordinary phytase/kg diet or 500 U thermostable phytase/kg diet. The results indicated that broilers given diets supplemented with thermostable phytase have increased body weight, body weight gain and feed intake than these fed basal diet ($p<0.05$). However, the same effect was only noted for body weight in ordinary phytase-supplemented group ($p<0.05$). Birds fed diet supplemented with thermostable phytase had enhanced crude protein utilization when compared with these given basal diet ($p<0.05$) but the same effect was not observed for broilers fed ordinary phytase-supplemented diet. As expected, dietary phytase supplementation significantly enhanced nitrogen-corrected apparent metabolizable energy of broilers ($p<0.05$) and this effect was more pronounced in the thermostable phytase-supplemented group. The results suggested that phytase supplementation, especially thermostable phytase supplementation, can improve the growth performance and nutrient utilization of broilers.

Key words: Thermostable phytase, growth performance, nutrient digestibility, broilers

INTRODUCTION

Poultry diets are based primarily on cereals, legumes and oilseed crops and their by-products. However, the major form of phosphorus present in these feed ingredients is phytate which is poorly available to poultry since monogastric animals have inadequate endogenous mucosal phytase to effectively digest dietary phytate (Nelson, 1967). Besides, phytate can chelate minerals, react with dietary protein and other nutrients, thus reducing the availability of many nutrients (Selle and Ravindran, 2007). One way to overcome these problems is the introduction of microbial phytase as feed additive. Addition of microbial phytase has been shown to release phytate-bound P, reduce P load on the environment and improve energy, protein and amino acid utilization (Selle and Ravindran, 2007; Pirgozliev *et al*., 2009). The efficacy of phytase preparation depends not only on type, inclusion rate and the level of activity present but also on the ability of enzyme to maintain its activity in different conditions, especially the conditions used for the pre-treatment of a diet as pelleting. The stability of added enzyme to pelleting process is a major concern of feed manufactures (Silversides and Bedford, 1999). Ordinary phytase feed enzymes can not withstand high pelleting temperature up to 85°C and then the residual activity of exogenous phytase present in the finished pellets might become low and can not play the expected role. However, phytase feed enzymes can be included in animal rations in stable form to avoid thermostability problems at high pelleting temperatures. Therefore, the present study was conducted to study the effect of thermostable phytase supplementation on the growth performance and nutrient digestibility of broilers in comparison with ordinary phytase.

MATERIALS AND METHODS

Experimental design, animals and diets: The animal care and use protocol were approved by Nanjing Agricultural University Institutional Animal Care and Use Committee. The study was conducted to study the effects of thermostable phytase supplementation on the growth performance and nutrient digestibility of broilers for 42 d. 360 1-day-old Arbor Acres chicks with similar initial body weight (47.02±0.04 g) were randomly divided into 3 treatments consisting 6 replicates of 20 chicks per replicate (1 replicate per cage). The control group (CO group) was given maize-soybean meal basal diet (Table 1), the other two groups were fed basal diets including either 500 U ordinary phytase/kg diet (OP group) or 500 U thermostable phytase U/kg diet (TP group). The two phytase-supplemented diets were formulated by adding 100 g ordinary phytase/kg of diet (provided by Weifang

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Table 1: Ingredients and nutrient composition of basal diet (g/kg, as-fed basis unless otherwise stated)

Ingredients	1-21 d	22-42 d
Maize	591	643
Soybean meal	306	243
Corn gluten meal	38	45
Lard	17	25
Dicalcium phosphate	18	16
Limestone	13	12
L-Lysine	1.5	1.6
DL-Methionine (99%)	1.5	1.0
Sodium chloride	4.2	3.3
Premix[a]	10	10
Calculated composition		
ME (MJ/kg)	12.3	12.8
CP[b]	212	193
Calcium	10	9.1
Available phosphorus	4.3	3.8
Lysine[b]	11.0	9.5
Methionine[b]	5.0	4.3
Methionine + cystine	8.2	7.3

[a]Premix provided per kilogram of diet: transretinyl acetate, 24 mg; cholecalciferol, 6 mg; all-rac-α-tocopherol acetate, 20 mg; menadione, 1.3 mg; thiamin, 2.2 mg; riboflavin, 8 mg; nicotinamide, 40 mg; choline chloride, 400 mg; calcium pantothenate, 10 mg; pyridoxine. HCl, 4 mg; biotin, 0.04 mg; folic acid, 1 mg; vitamin B12 (cobalamin), 0.013 mg; Fe (from ferrous sulfate), 80 mg; Cu (from copper sulphate), 7.5 mg; Mn (from manganese sulphate), 110 mg; Zn (from zinc oxide), 65 mg; I (from calcium iodate), 1.1 mg; Se (from sodium selenite), 0.3 mg; bacitracin zinc, 30 mg

[b]Values calculated based on the Table of Feed Composition and Nutritive Values in China

Table 2: Effects of thermostable phytase supplementation on growth performance of broilers

Item	BW (g)	BWG (g/d)	FI (g/d)	FGR
CO	2683[c]	62.9[b]	109[b]	1.74
OP	2709[b]	63.4[b]	113[ab]	1.78
TP	2830[a]	66.3[a]	118[a]	1.77
SEM[1]	25	0.6	0.9	0.02

[1]SEM = Standard error of means (n = 8). BW: Body weight, BWG: Body weight gain, FI: Feed intake, FGR: Feed gain ratio.

[a-c]Means within a column with no common superscript differ significantly (p<0.05)

Table 3: Effects of thermostable phytase supplementation on coefficient of total tract apparent retention and nitrogen-corrected apparent metabolizable energy (AMEn) of broilers

Item	DM	CP	EE	AMEn, MJ/kg
CO	0.69	0.50[b]	0.72	11.4[c]
OP	0.66	0.47[b]	0.69	11.9[b]
TP	0.71	0.59[a]	0.74	12.5[a]
SEM[1]	0.01	0.01	0.02	0.06

[1]SEM = Standard error of means (n = 8). DM: Dry matter, CP: Crude protein, EE: Ether extract.

[a-c]Means within a column with no common superscript differ significantly (p<0.05)

Bio-Tech Co., Ltd., Weifang, China, Trade name is KINGPHOS®) and 100 g thermostable phytase/kg of diet (provided by the same company mentioned above, Trade name is KINGPHOS HS®), respectively. The declared activity was both 5000 U/g of product for the two phytase enzymes. One unit of enzyme is defined as the amount of enzyme that liberates 1 µmol inorganic orthophosphate per min from 5 mmol/L sodium phytate at pH 5.5 and a temperature of 37°C. Diets were given in the pellet form (Pelleting temperature 83°C). Feed and fresh water were available *ad libitum* at all time. Body weight and feed intake on a cage basis were recorded from d 1-42 for the calculation of the average daily gain, average daily feed intake and Feed/gain ratio. Feed was withdrawn for 12 h, with water being provided *ad libitum*, before the chicks were weighed at d 42. At the end of the experimental period, 36-poult sample (two chicks per replicate) was randomly selected and allocated into individual cages. The birds were fed their respective experimental diet for a 3 d period followed by a 72 h complete excreta collection. Excreta were collected twice per day from a plastic tray placed under the cages, stored at -20°C and finally pooled for each cage. The determination of coefficient of total tract apparent retention of nutrients and the nitrogen-corrected Apparent Metabolizable Energy (AMEn) were calculated according to Wen *et al*. (2012). Briefly, The excreta samples were then dried for 48 h in an oven at 65°C. The dried excreta were allowed to equilibrate to atmospheric conditions for 24 h before being weighed. Feed and excreta samples were then ground through a 0.45-mm screen. The samples were analyzed (AOAC, 1990) for dry matter (DM, 934.01), ether extract after HCl treatment (EE, 920.39), Gross energy was determined using an adiabatic calorimeter (SXHW-III, Tianyu, Hebi, China). The coefficient of total tract apparent retention (CTTAR) of nutrients and the nitrogen-corrected apparent metabolizable energy (AMEn) were calculated according to Adeola *et al*. (2008).

Statistical analysis: All data were analyzed as a completely randomized design using one-way ANOVA (SPSS, 2008). The differences were considered to be significant at p<0.05.

RESULTS

Growth performance: Broilers given diet supplemented with thermostable phytase had increased growth performance in terms of body weight, body weight gain and feed take, rather than feed conversion ratio, than these fed the basal diet (p<0.05). The same effect was only noted for body weight in ordinary phytase-supplemented group (p<0.05), though feed intake and body weight gain in this group were numerically higher when compared with control. Besides, it is worth noting that the effect of thermostable phytase in improving growth performance of broiler chickens is more efficient when compared to the ordinary one as evidenced by increased body weight and body weight gain (p<0.05).

Nutrient digestibility: Birds fed diet supplemented with thermostable phytase had enhanced crude protein utilization when compared with these given basal diet ($p<0.05$), however, the same effect was not observed for broilers fed ordinary phytase-supplemented diet. As expected, dietary phytase supplementation significantly enhanced AMEn of broilers ($p<0.05$) and this effect was more pronounced in the thermostable phytase-supplemented group. Dry matter and ether extract retention were uninfluenced by phytase supplementation ($p>0.05$).

DISCUSSION

In our experiment, the effect in improving growth performance is more effective in thermostable phytase-supplemented group which may attribute to the thermostability of this kind of phytase since ordinary phytase can not withstand high pelleting temperature and therefore residual activity present in the finished pellets is relative lower. It is generally thought that responses to exogenous phytase supplementation in feed intake and body weight gain are more consistent than feed efficiency responses (Selle and Ravindran, 2007). The results in present study confirmed the beneficial influence of dietary phytase on the growth performance of broilers in terms of body weight, weight gain and feed intake rather than feed conversion rate which was in accordance with the previous repots by Ahmad *et al.* (2000) and Viveros *et al.* (2002). This effect is probably as result of simultaneous increases in weight gain and feed consumption or declining feed efficiency responses to phytase with time (Rosen, 2003). The ability of phytase to improve AMEn and protein utilization has been well documented. Shirley and Edwards (2003) demonstrated an improvement in AMEn when feeding broilers a corn and soy-based diet supplemented with phytase. Camden *et al.* (2001) evaluated two phytase feed enzymes in broilers offered maize-soy diets. Overall, phytase increased ileal digestibility coefficients of fat by 3.5%, protein by 2.6% and starch by 1.4% which was associated with phytase-induced increases in AME of 0.17 MJ. Alternatively, Tejedor *et al.* (2001) reported that phytase supplementation of maize-soy diets did not enhance N-corrected AME in broiler chicks and the similar result was also reported by Liebert *et al.* (2005) in layers. The discrepancy in the aforementioned reports may attribute to avian species, diet compositions, efficacy of phytase enzyme preparation mainly depending on type, inclusion rate, the level of activity present and the ability of enzyme to maintain its activity *in vivo* and *in vitro*. The results obtained form present study confirmed the beneficial influence on crude protein digestibility and energy utilization of broilers by phytase supplementation which may result from the ability of phytase to disrupt the various interactions between phytate, minerals, starch and protein, thereby allowing a greater utilization of nutrients from the diet. It is worth noting that the ability to improve nutrient retention and energy is more pronounced with thermostable phytase-supplemented diet in present study, as evidenced by various parameters measured above. Most of the added feed enzymes inactivation takes place during conditioning, when the feed is heated with steam, especially the pelleting process (Eeckhout *et al.*, 1995). Thus, by adding the thermostable enzymes to the formulation partially overcomes this problems and the result obtained in our study may because of the high residual activity of the thermostable phytase in the finished pellets after high temperature pelleting process.

Conclusion: The effect of dietary thermostable phytase supplementation in enhancing the growth performance, crude protein utilization and AMEn of broilers is more efficient when compared with ordinary one.

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