Pakistan J. Zool., vol. 50(1), pp 347-351, 2018. DOI: http://dx.doi.org/10.17582/journal.pjz/2018.50.1.347.351

Effect of Supplementing Microbial Phytase on Broiler Chicks Fed Low Di-calcium Phosphate Diets

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Article Information
Received 03 June 2016
Revised 23 August 2016
Accepted 27 November 2017
Available online 18 January 2018

Authors' Contribution
MIA conceived and designed the study, executed the experimental trial and wrote the article. SJ analyzed the data, assisted in experimental trial and article writing. MAN helped in experimental feed preparation and chicks supply.

Key words
Microbial phytase, Broiler chicks, Ca and P concentration in serum and tibia

ABSTRACT

For this study 195 day-old broiler chicks were randomly divided into three treatment groups with five replicates in each treatment group having 13 chicks per replicate per pen. Three iso-nitrogenous and iso-caloric rations were prepared as: R-I served as control with 0.83% di-calcium phosphate (DCP); R-II contained 50% less DCP (0.41%) than R-I and R-III contained R-II plus phytase Quantum® Blue @ 100 g/ton feed. Daily feed intake and weekly body weight gain per replicate were recorded for 35 days. On 35th day, blood and tibia bone samples from 5 birds per treatment were collected. Total weight gain and dry matter intake in chicks fed rations I, II and III were 1861, 1736 and 2023 gm/bird and 3242, 3284 and 3265 gm/bird, respectively. The chicks fed R-III got 16.00 and 8.00% higher weight gain compared to chicks fed R-II and R-I, respectively. Feed: gain ratio (gm/gm) was found best ($P<0.05$) in chicks fed R-III (1.61) and poorer in chicks on R-II (1.89). The concentration of Ca and P were 10.60, 9.47 and 11.43 mg/dL and 5.76, 4.91 and 7.11 mg/dL in blood serum and 32.61, 31.36 and 32.83% and 16.57, 15.92 and 16.60% in tibia bone of chicks fed rations I, II and III, respectively. The total feed cost per unit weight gain of broilers fed R-III diet was numerically 14% and 9% less than the broilers fed R-II and R-I, respectively. Results suggested that exogenous microbial phytase supplementation to rations having low DCP had positive effects on weight gain, feed: gain ratio and economic efficiency in broiler chicks.

INTRODUCTION

After calcium (Ca) the second abundant mineral in animal body is phosphorus (P) and majority (80%) of the P is found in bones and teeth (NRC, 1994). The P is not only essential for bone development, growth, health, production and reproduction but it also aids in energy metabolism, synthesis of sugars and maintenance of acid-base balance in the body. However, the quantity and availability of dietary P is vital for growing animals. It is well established that phytate is the major source of P in plant based feedstuffs. Since birds are unable to produce any endogenous phytase, therefore their capability to utilize P from phytate is very limited (Woyengo *et al*., 2010).

Phytate is a ubiquitous component of plant sourced feed ingredients which encompasses approximately two-thirds of total plant P (Hughes *et al*., 2009). In poultry, phytate P is normally utilized with availability from 0 to 50%, depending on age and metabolic adaptation in critical circumstances. Therefore, to meet the P requirement, generally expensive inorganic P sources are added to poultry diets. This practice leads to non-utilization of a large portion of dietary P from feedstuff and its excretion in faeces (Hughes *et al*., 2008; Woyengo *et al*., 2010) which ultimately pollutes environment. Recently, microbial phytase supplementation in poultry diets has got remarkable attention to reduce negative impact of phytate P on environment and performance of birds (Ceylan *et al*., 2003; Francesch and Geraert, 2009). Since phytase efficiency in the digestive tract is influenced by various factors like phytase origin, type of birds and ambient temperature etc. Therefore, each phytase preparation for poultry must be tested on broilers/layers to ensure its efficacy (Hughes *et al*., 2008; Onyango *et al*., 2005). Information regarding effect of microbial phytase addition in broiler chicks' diet having low DCP is limited. Therefore, current study was designed to asses the effect of microbial phytase supplementation in broiler chicks fed low DCP diets on growth, economic efficiency and P contents in serum and tibia bones.

MATERIALS AND METHODS

Birds and management

The research work was carried out at Animal Nutrition Program, Animal Sciences Institute, National Agricultural Research Centre, Islamabad, Pakistan. One hundred ninety

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five (195) one day old mixed sex broiler chicks (initial mean body weight 43 ± 2 gm) were allocated randomly to three dietary treatments, each having five replicates with 13 birds per replicate. These birds were placed in separate battery cages (measuring 120 cm long × 94 cm wide × 73 cm height). Each cage was equipped with drinker and feeder adjusted inside of the cage. Water and experimental rations (in mash form) were offered *ad libitum*. During the trail all of the birds were kept under 24 h lighting regimens. All chicks were vaccinated against Newcastle disease, infectious bursal disease, infectious bronchitis, as per recommended schedule in Pakistan.

Table I.- Ingredient and chemical composition (%, DM) of experimental diets

	Experimental diets (0-35 day)[1]		
Ingredients (%)	R-I	R-II	R-III
Constant ingredients †	98.73	98.73	98.73
Di-calcium phosphate	0.83	0.41	0.41
Phytase (Quantum Blue)[2]	-	-	0.01
Premix	0.45	0.86	0.85
Total	100	100	100
Nutrient composition (as such basis), %			
Metabolizable energy, Kcal/Kg	2750.61	2750.61	2750.61
Crude protein	20.00	20.00	20.00
Crude fat	3.39	3.39	3.39
Crude fibre	4.33	4.33	4.33
Total ash	5.73	5.73	5.73
Calcium	0.78	0.59	0.59
Total phosphorus	0.46	0.39	0.39
Phytate phosphorus	0.21	0.21	0.21
Common salt	0.39	0.39	0.39
Aflatoxin, ppb	11.44	11.44	11.44

[1]R-I, positive control diet containing 0.83% di-calcium phosphate (DCP); R-II contained 50% less DCP (0.41%) than R-I without phytase and R-III contained R-II plus phytase Quantum® Blue.
[2]According to the producer's declaration Quantum® Blue contains solid forms 5000 FTU/g and it was added to broiler rations @ 100 g/ton.
†Constant ingredients included corn ground 65%; corn gluten meal (60%) 1%; soybean meal 22%; fish meal 8%; vegetable oil 0.55%; lysine sulphate 0.50%; liquid-methionine 0.30%; threonine 0.08%; marble chips 1%; sodium chloride 0.15% and soda bicarb 0.15%.

Dietary treatments and laboratory analysis

Three iso-nitrogenous and iso-caloric rations were prepared at Islamabad Poultry Feeds, Islamabad as: Commercial ration with 0.83% DCP served as control (R-I) whereas similar as R-I was formulated but with 50% less DCP (0.41%) without (R-II) or with Quantum® Blue (6- phytase) supplementation (R-III). The composition of rations used during experimentation is given in Table I. Phytate P content in all rations was similar i.e., 0.21%. According to the producer's declaration, Quantum® Blue contains solid forms 5000 FTU/g and it was added to broiler rations @ 100 g/ton. Phytate P of corn grains, corn gluten meal 60% and soybean meal were determined by Sibbald (1986) technique and diets were also analyzed for CP, CF, EE, total ash, Ca and total P according to AOAC (1990). Daily feed intake and weekly body weight gain (BWG) per replicate per pen were recorded. On 35th day, blood and tibia bone samples from 5 birds per treatment were collected. The left tibia from each bird was excised and stored in sealed plastic bagsat -20ºC until further analysis. At the time of analysis, meat and fat were gently removed from tibia bones. After overnight drying at 100ºC, the bones were extracted in ether for 6 h and burnt to ash in a Muffale furnace at 600ºC. The ash from each tibia was used for Ca and P analysis according to AOAC (1990).

Statistical analysis

All data were presented as means±SE. The data were analyzed statistically using the standard procedure of analysis of variance technique in completely randomized design as described by Steel *et al*. (1997) by using Minitab 15 software. For significant differences, means were compared using Duncan's multiple range test at 5% level of probability.

RESULTS AND DISCUSSION

Growth performance

Results regarding effects of microbial phytase (Quantum® Blue) supplementation to diets having low di-calcium phosphate (DCP) on growth performance in broiler chicks are given in Table II. The results showed that dietary DCP levels (0.83% and 0.41%) with or without Quantum® Blue supplementation did not influence feed intake and feed: gain ratio in chicks however, body weight gain (BWG) reduced significantly (P<0.05) on diet with 0.41% DCP without Quantum® Blue (R-II) compared to those fed diet with 0.83% DCP (R-I) or diet with 0.41 % DCP plus Quantum® Blue (R-III) up to 21 days of age. Whereas, feed intake in chicks fed diets having 0.41 % DCP without (R-II) or with Quantum® Blue (R-III) improved significantly (P<0.05) during 22-35 days of age and overall feeding period (1-35 days) compared to those fed diet having 0.83% DCP (R-I) however, difference between R-II and R-III was non-significant (P>0.05). Our results are partially supported by Boling-Frankenbach *et al*. (2001) who reported that dietary

Table II.- Effects of phytase supplementation on growth performance of broiler chicken

Treatment	BWG, g/bird/period (days)			FI, g/bird/period (days)			FCR, g Feed/g BWG (days)		
	1-21	22-35	1-35	1-21	22-35	1-35	1-21	22-35	1-35
R-I	650[a]	1211[ab]	1861[ab]	1216	2026[b]	3242[b]	1.87	1.67[a]	1.74[a]
R-II	625[b]	1111[b]	1736[b]	1219	2065[a]	3284[a]	1.95	1.86[b]	1.89[b]
R-III	665[a]	1358[a]	2023[a]	1209	2056[a]	3265[a]	1.82	1.51[a]	1.61[a]
SEM	24	75	119	22	27	33	0.18	0.16	0.15
P level	0.05	0.04	0.03	0.38	0.04	0.04	0.08	0.03	0.04

[a, b]Means with different superscript letters within rows are significantly different (P<0.05), BWG, body weight gain; FI, feed intake; SEM, standard error of mean. For detail of treatments, see Table I.

phytase supplementation did not affect feed intake during first 21 days of age whereas others reported improvement in feed intake (Cabahug *et al*., 1999; Denbow *et al*., 1995). The reason for these contrasting results might be due to number of factors i.e. phytase source (type and phytate content), and dietary characteristics (processing, Ca: P ratio) etc. (Ravindran *et al*., 1995).

Quantum® Blue supplementation to low DCP (0.41%) diet (R-III) resulted in improved (P<0.05) BWG as well as best feed: gain ratio in chicks during 22-35 days of age and overall feeding period (1-35 day) compared to chicks fed R-II, however, both rations did not differ from R-I diet. The lowest growth rate in broiler chicks fed diet R-II was may be due to lower availability of dietary P from phytic acid molecules of feedstuffs. But when Quantum® Blue added to diet with low DCP (R-III), presumably it liberated more P from phytic acid molecules of feedstuffs to satisfy the chicks requirement and ultimately improved BWG and feed: gain ratio that were comparable to those fed positive control diet (R-I) having 0.83% DCP. Similar findings were observed by Bozkurt *et al*. (2006) who reported comparable or even better growth rate and FCR in broilers fed diets with low-P plus microbial phytase than those fed the standard P diets. Viveros *et al*. (2002) explained that microbial phytase supplementation in poultry diets increases P availability, consequently improves growth performance and minerals (Ca, P, Mg and Zn) utilization in chicks. Better growth in phytase supplemented chicks may also be due to some positive effects of enzyme on utilization of metabolizable energy in chicks (Ravindran *et al*., 2001).

Ca and P contents in serum and tibia

The concentration of Ca and P in serum and tibia bones of experimental birds are given in Table III. In blood serum and tibia bones, Ca and P contents were comparable between R-I and R-III but both were higher (P<0.05) than in R-II, presumably due to the added phytase that liberated P from phytic acid molecules to satisfy the broilers' requirement for deposition of Ca and P in bones. This result is consistent with previous reports that phytase supplementation improved bone strength and mineralization in chicks fed low P diets (Dilger *et al*., 2004; Woyengo *et al*., 2010). Supplementation of diets with microbial phytase derived from *Aspergillus niger* increases availability and retention of phytate P and Zn in chicks as measured by ash content of bone (Ahmad *et al*., 2000; Qian *et al*., 1995; Sebastian *et al*., 1996).

Table III.- Effect of phytase supplementation on calcium and phosphorus concentrations in blood serum and tibia bones

Treatment	Blood serum		Tibia bone, % of DM	
	Ca (mg/dL)	P (mg/dL)	Ca % of ash	P % of ash
R-I	10.60[ab]	5.76[b]	32.61[a]	16.57[a]
R-II	9.47[b]	4.91[b]	31.36[b]	15.92[b]
R-III	11.43[a]	7.11[a]	32.83[a]	16.60[a]
SEM	0.81	0.53	0.53	0.34
P level	0.01	0.02	0.04	0.04

For statistical details and treatment groups, see Table II.

Economic efficiency

Feed cost of live weight gain mainly relies on cost of feed and efficiency of feed utilization by animals (Nair *et al*., 2004). In this study, rate of one Kg of commercial diet was Rs. 46; Quantum® Blue Rs. 3000 and of DCP was Rs. 80. The maximum feed cost incurred per unit weight gain was recorded in broilers fed R-II diet (Rs. 84.10) followed by chicks on R-I (Rs. 80.04) and R-III (72.13) diets indicating that chicks on R-I and R-II ate more and gain less weights (Table IV). The total feed cost per unit weight gain of broilers fed R-III diet was numerically 14% and 9% less than the broilers fed R-II and R-I, respectively.

Table IV.- Feed cost of live body weight gain in broilers.

Parameters	Treatments[1]		
	R-I	R-II	R-III
Feed conversion ratio	1.74	1.89	1.61
Cost of per kg feed (Rs.)	46.00	44.50	44.50
Cost of phytase (Rs.)	-	-	0.30
Cost of per kg feed + phytase (Rs.)	46.00	44.50	44.80
Cost of feed per kg live weight gain (Rs.)	80.04	84.10	72.13

For details of treatment groups, see Table II.
Per kg cost of commercial diet was Rs. 46; Quantum® Blue Rs. 3000; Di-calcium phosphate Rs. 80.

CONCLUSION

It is concluded from results that supplementation of phytase (Quantum® Blue) to diet containing low DCP had positive effects on body weight gain; feed: gain ratio and economic efficiency as compared to chicks fed non-phytase supplemented diets.

ACKNOWLEDGEMENT

The authors would like to acknowledge the Islamabad Poultry Co. Pvt. Ltd. Rawalpindi for preparation of experimental diets for this study.

Statement of conflict of interest

The authors have no conflict of interest.

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