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Full Length Research Paper

Effects of supplemental microbial phytase enzyme on performance and phytate phosphorus digestibility of a corn-wheat-soybean meal diet in broiler chicks

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This experiment was conducted to investigate the effects of supplemental phytase in a corn-wheat-soybean meal basal diet on phosphorus (P) digestibility and performance of broiler chicks. 378 one-day old broiler chicks (Ross 308) were allocated to 3×3 factorial arrangements with three levels of phytase enzyme (0, 500 and 1000 FTU/kg) and three levels of non-phytate P (100, 80 and 60% of NRC requirements). Broiler chicks received experimental diets from 7 to 49 days of age. Phytase significantly (P < 0.05) improved body weight gain and feed intake. Tibial ash and P contents increased significantly by phytase supplementation. Ileal P digestibility increased and P excretion reduced by added phytase. The greatest response due to supplemental phytase regarding P digestibility and utilization was obtained at the lowest dietary non-phytate P (NPP) level (60% of NRC requirements). Difference between various levels of added phytase (500 and 1000 FTU/kg) regarding P excretion and utilization was not significant. The interactions between supplemental phytase and dietary NPP for P utilization, tibial ash and P contents were significant (P < 0.05). The results indicate that, supplemental microbial phytase (500 FTU/kg of diet) added to diet containing NPP lower than NRC requirements (60%) can improve growth performance, tibial ash and phytate P utilization in broiler chickens.

Key words: Broiler, phytase, phosphorus digestibility, growth performance, tibia.

INTRODUCTION

Plant materials are the major constituents of poultry diets. Unfortunately, about two-third of the phosphorus (P) in cereal grains, oilseed meals and plant by- products is present in the form of P bound to phytic acid (phytate P), which is not available to poultry and most of it is excreted in the manure. This is due to the lack of phytase, the enzyme that hydrolyses phytic acid into inositol and ortophosphate. Poultry manure is often used as fertilizer on pastures and other croplands. In areas of intensive poultry production, however, the P content of the manure often exceeds the requirements for plant growth. If this excess occurs, the additional P can contribute to a significant environmental problem (Li et al., 2000; Singh, 2008; Kaya et al., 2009).

Many researchers have shown that, supplemental microbial phytase improves the bioavailability of phytate P and reduces the need to inorganic P and decreasing P excretion (Huff et al., 1998; Ravindran et al., 2000; Yan et al., 2000; Yan et al., 2003; Angel et al., 2005; Singh., 2008; Plumstead et al., 2008).

Some reports have shown that microbial phytase added to poultry feeds, by phytate dephosphorylation, increases P digestibility from 35 to about 60% (Kornegay et al., 1996) and reduces P content of excreta by 42% (Simons et al., 1990). Denbow et al. (1995) observed that, in soybean meal diets fed 0 to 3 week-old male broilers, added phytase improved body weight gain and feed intake in all the non-phytate P levels. Qian et al. (1997) reported that, phytase increased body weight gain, feed intake, toe ash content and P and Ca retention

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Abbreviations: NPP, Non-phytate phosphorus; **BWG,** body weight gain; **FCR,** feed conversion ratio**; FI,** feed intake.

Table 1. Composition of basal diet[1] (g/kg).

NPP (% of NRC)	7 to 21 days (starter)			21 to 42 days (grower)			42 to 49 days (finisher)		
	100	80	60	100	80	60	100	80	60
Composition									
Corn	265	266	266	335	335.7	336	368	369	369
Wheat	300	300	300	300	300	300	300	300	300
Soybean meal (37% CP)	388	387.4	387.8	317.2	317	317	275.7	275	275.3
Soybean oil	12	11.8	11.7	14.5	14.3	14.1	22.7	22.6	22.5
Oyster shell	9.4	14.9	20.3	12.8	16.8	21.6	12.8	16.9	20.3
Dicalcium phosphate[2]	16.4	10.7	5	12.1	7.8	2.9	10	5.7	2.1
Salt	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5	2.5
Dl-methionine	1.7	1.7	1.7	0.9	0.9	0.9	0.3	0.3	0.3
Vitamin-trace-mineral-premix[3]	5	5	5	5	5	5	5	5	5
Cr_2O_3	-	-	-	-	-	-	3	3	3
Calculated analysis									
ME (MJ/kg)	11.1	11.1	11.1	11.5	11.5	11.5	11.9	11.9	11.9
Crude protein (g/kg)	201.3	201.3	201.3	181.3	181.3	181.3	168.8	168.8	168.8
Methionine+cystine (g/kg)	7.9	7.9	7.9	6.5	6.5	6.5	5.6	5.6	5.6
Lysine (g/kg)	9.6	9.6	9.6	9.1	9.1	9.1	8	8	8
Non-phytate phosphorus (g/kg)	3.9	3.1	2.3	3.2	2.6	1.9	2.8	2.2	1.7
Total phosphorus (g/kg)	6.3	5.5	4.7	5.4	4.9	4.2	5.1	4.5	4
Calcium (g/kg)	8.6	8.6	8.6	8.2	8.2	8.2	7.5	7.5	7.5
Calcium: Total phosphorus	1.4	1.6	1.8	1.5	1.7	1.9	1.5	1.7	1.9

[1]Phytase enzyme (0, 500, and 1000 FTU/kg of diet) were added to each of the basal diets; [2]dicalcium phosphate 280 g/kg Ca and 140 g/kg P; [3]supplied per kilogram of diet: vitamin A, 11000 IU; vitamin D, 2200 IU; vitamin E, 30 IU; vitamin B12, 0.02 mg; menadion, 2.0 mg; thiamin, 1.5 mg; riboflavin, 6.0 mg; niacin, 60.0 mg; pyridoxine, 4.0 mg; folic acid 0.6 mg; pantothenic acid, 10.0 mg; biotin, 0.15 mg; zinc, 80.0 mg; copper, 10.0 mg; iron, 80.0 mg; manganese, 80.0 mg; selenium, 0.3 mg; and iodine, 0.8 mg.

linearly. They suggested that dietary Ca to total P ratio and the level of cholecalciferol influence utilization of phytate P and Ca.

The aim of this study was to evaluate the effects of different levels of dietary supplemental phytase, non-phytate phosphorus and their interaction on P digestibility and performance parameters of broiler chickens.

MATERIALS AND METHODS

Experimental design

Three hundred and seventy eight (378) day-old broiler chicks (Ross 308) were used in a completely randomized design with a 3×3 factorial arrangement with three levels of phytase enzyme (0, 500 and 1000 FTU/kg) and three levels of non-phytate P (100, 80 and 60% of NRC requirements) of three replicates.

The phytase enzyme was Natuphos® 5000 FTU/g (BASF Corp., 3000# Continental Drive North, Mount Olive, NJ, 07828-1234). Dietary P levels were formulated at 100, 80 and 60% NRC requirements. Non-phytate phosphorus and each level of NPP were supplemented with 0, 500 and 1000 FTU/kg diet. One unit of phytase is defined as the quantity of enzyme which releases 1 mmol of inorganic P/min from 0.00015 mol/l sodium phytate at pH 5.5 at 37°C (Ravindran et al., 2000).

All birds were fed a common commercial starter ration until 7 days of age at which time 378 mixed broiler chickens were randomly placed in floor pens (14 birds in a pen) and each experimental diet was given to three pens. Room temperature was kept at 34°C during the first 3 days of the trail and was then, reduced gradually according to age until reaching 22°C at 21 days. The light was continuous during the first 3 days and then the lighting regimen was 23 h/days. The composition of experimental diets was formulated to meet the requirements of broiler chicks as established by the NRC (1994) and is presented in Table 1.

Broiler chicks received dietary treatments in mash form at three stages: starter diet (7 to 21 days), grower diet (21 to 42 days) and finisher diet (42 to 49 days). Chicks had free access to feed and water during experimental period. Body weight gain, feed intake and feed conversion ratio (FCR) were recorded for the same periods.

Samples collection and chemical analysis

At 49 days of age, four birds per pen (2 males and 2 females) with a body weight close to the mean body weight of the pen were selected, slaughtered and each bird was immediately dissected, the left tibia removed and stored at 4°C. The ileum location defined as extending from Meckels diverticulum to a point 40 mm proximal to the ileo-cecal junction, excised and this segment was bisected transversely and its contents were gently squeezed out into a plastic cap and stored at -20°C until chemical analysis. After thawing, the samples were dried in an oven 60°C. Chromic oxide in digesta and diets was determined according to Fenton and Fenton (1979) procedure.

The bones were fat extracted in a soxhlet extractor for 24 h with petroleum ether, then ashed at 600°C over night. P concentration in digesta, diets and tibial ash were determined according to AOAC (1994).

Calculations

Apparent ileal digestibility coefficient of P was estimated by using 0.3% chromic oxide (Cr_2O_3) as an indigestible marker. The following formula was used to calculate the digestibility coefficients:

$$AID_p = 100 - \left[100 \times \frac{[Cr]_{diet} \times [P]_{digesta}}{[Cr]_{digesta} \times [P]_{diet}} \right]$$

Where AID_p= apparent ileal digestibility of P (percentage); $[Cr]_{diet}$ = chromic oxide concentration in the diet; $[Cr]_{digesta}$= chromic oxide concentration in the digesta; $[P]_{digesta}$ = phosphorus concentration in the digesta; $[P]_{diet}$ = phosphorus concentration in the diet.

Statistical analysis

Data were analyzed using the General Linear Models procedure for analysis of variance (SAS Institute, 1990). Significant differences among treatment means were separated by Duncan's new multiple range test (Duncan, 1955) with a 5% level of probability. To evaluate the sensitivity of measurements, second order translog functions were derived for the 3×3 factorial arrangements of treatments with the following model:

$$LnY = \alpha_0 + \alpha_1 LnX_1 + \alpha_2 LnX_2 + \alpha_3 (LnX_1)^2 + \alpha_4 (LnX_2)^2 + \alpha_5 LnX_1X_2$$

Where Y= the response measurements; X_1 = NPP (percentage); X_2 = added phytase (units per kilogram of diet).

RESULTS

Performance

The effect of microbial supplemental phytase at different levels of dietary NPP is presented in Tables 2 and 3. The effect of phytase except at 42 to 49 days on body weight gain was significant (P < 0.05), body weight gain increased by 500 FTU/kg. There was no significant difference between 500 and 1000 FTU/kg regarding body weight gain. However, body weight gain was significantly (P < 0.05) affected by dietary non-phytate phosphorus level and reducing dietary P to 60% but not 80% of NRC requirements, significantly (P < 0.05) decreased body weight gain. Interaction between phytase and dietary NPP was significant (P < 0.05) and the highest body weight gain was obtained at the lowest dietary NPP (60% NRC) so that its difference with control group was significant (P < 0.05). Supplemental phytase significantly (P < 0.05) increased daily feed intake during grower and over the entire growth period (7 to 49 days). Feed intake was significantly (P < 0.05) affected by dietary non-phytate phosphorus levels and reducing dietary NPP caused a decrease in feed intake during over all growth period. In addition, interaction between phytase and dietary non-phytate phosphorus was significant (P < 0.05) and addition of phytase to the lowest dietary NPP (60% NRC) increased feed intake significantly (P < 0.05).

The effect of added phytase and interaction between dietary NPP and supplemental phytase on feed conversion ratio was not significant in any experimental periods, but the effect of dietary non-phytate phosphorus on FCR was significant (P < 0.05) over the entire growth period.

Dietary NPP had critical effects on chick's livability so that 40% reduction in NPP caused an increase in mortality and added supplemental phytase to these diets reduced total mortality during experimental period.

Tibial ash and phosphorus contents

The effects of phytase and NPP on tibial ash its P content at 49 days is presented in Table 4. Phytase and dietary P had significant (P < 0.05) effects on tibial ash and its P content P contents. Reducing dietary non-phytate phosphorus to 60% of NRC requirements significantly (P < 0.05) decreased tibial ash and its P content. Addition of phytase to the diets containing the lowest NPP increased tibial ash significantly (P < 0.05).

Ileal phosphorus digestibility and excretion

The effects of supplemental phytase and dietary NPP on apparent ileal digestibility and excretion of P are presented in Table 4. Effects of phytase and dietary NPP on apparent ileal digestibility and excretion of P were significant (P < 0.05). Reducing dietary non-phytate phosphorus from 100 to 80 and 60% of NRC recommendations significantly (P < 0.05) decreased P excretion and increased P digestibility. Added microbial phytase significantly (P < 0.05) improved P digestibility and reduced P excretion. Interaction between phytase and dietary NPP was significant (P < 0.05).

Second order translog functions

The second order translog for the performance, apparent ileal digestibility and excretion of P are shown in Table 5. Second order translog equation for apparent ileal digestibility and excretion of P, body weight at 42 days, body weight gain from 21 to 42 days and body weight at 49 day of age had high R^2 values, indicating a good fit of the equation to the data. The R^2 was relatively high for feed intake 21 to 42 days, body weight at 21 days and body weight gain from 7 to 21 days of age, but other measurements had low R^2 values (P < 0.5).

Table 2. Effect of phytase supplementation at different levels of non-phytate phosphorus (NPP) on body weight gain (BWG), feed intake (FI) and feed conversion ratio (FCR) of broiler chicks fed corn-wheat-soybean meal diet over 7 to 21 and 21 to 42 periods.

Treatment		7 to 21 days			21 to 42 days		
NPP	Phytase	BWG	FI	FCR	BWG	FI	FCR
(% of NRC)	(U/kg)	(g/day/bird)		(g/g)	(g/day/bird)		(g/g)
100	0	23.5	42.3	1.80	59.8 [ab]	114.8	1.92
100	500	23.6	42.9	1.82	60.9 [ab]	115.6	1.90
100	1000	24.3	43.7	1.80	61.2 [ab]	118.3	1.93
80	0	23.3	42.9	1.84	61.2 [ab]	117.8	1.92
80	500	23.5	42.5	1.81	65.3 [a]	124.1	1.90
80	1000	24.6	43	1.78	63.1 [ab]	121.2	1.92
60	0	20.7	39.2	1.89	48.9 [c]	95.9	1.96
60	500	23.1	48.2	1.83	57.9 [b]	107.8	1.86
60	1000	23.6	41.5	1.76	59.9 [ab]	112.1	1.87
SEM		2.97	0.82	0.04	1.66	3.34	0.01
Probabilities							
Treatments		NS	0.01	NS	0.001	0.001	NS
NPP		NS	0.008	NS	0.000	0.000	NS
Phytase		NS	NS	NS	0.003	0.025	NS
NPP × Phytase		NS	NS	NS	0.046	NS	NS
Main effects NPP							
100		23.8	42.9 [a]	1.80	60.7 [a]	116.2 [a]	1.91
80		23.8	43.1 [a]	1.81	63.2 [a]	121.0 [a]	1.91
60		22.5	40.9 [b]	1.82	55.6 [b]	105.2 [b]	1.89
Phytase							
0		22.5	41.5	1.84	56.6 [b]	109.5 [b]	1.93
500		23.4	42.5	1.82	61.4 [a]	115.8 [a]	1.89
1000		24.1	43.0	1.74	61.4 [a]	117.2 [a]	1.91

[a-c] Means within columns with no common superscript differ significantly ($P < 0.05$).

DISCUSSION

The results of this experiment indicated that, supplemental phytase was effective on improving apparent ileal phosphorus digestibility and reduced P excretion. It seems that lack of ability of broilers to use phytate, caused reduction in body weight gain (Tables 2 and 3) and tibial ash and P contents (Table 4). It was more pronounced with chicks consuming the diet containing the lowest level of NPP, because most of their P requirements were supplied by soybean meal, corn and wheat (sources of phytate). Reduced body weight gain and feed intake in chicks which consumed diet containing the lowest non-phytate phosphorus (60% of NRC) may be due to inadequate supply of P in the diet to meet their P requirements. Also, it seems that reduction of appetite as a result of the low level of dietary P and reduction in the solubility of minerals complex as a consequence of increased ileal pH by the relatively high dietary calcium level (Shafey et al., 1991).

20% reduction in dietary non-phytate phosphorus caused an improvement in feed intake and body weight gain, which indicated that reducing the dietary P to this marginal level, stimulate chicks to increase feed intake in order to meet their phosphorus requirements. Because the diets contained the same nutrients density birds consumed higher levels of dietary nutrients and gained more body weight (Tables 2 and 3).

Added microbial phytase improved the amounts of feed intake and body weight gain (7 to 49 days of age). Phytase can degrade the phytate and more P was available in the gut, consequently, the chicks' appetite resumed and their phosphorus requirements were met. Feed intake and body weight gain were increased by supplemental phytase; there was not significant improvement in non-phytate phosphorus (Tables 2 and 3). The

Table 3. Effect of phytase supplementation at different levels of non-phytate phosphorus (NPP) on body weight gain (BWG), feed intake (FI), feed conversion ratio (FCR) and mortality of broiler chicks fed corn-wheat-soybean meal diet over 42 to 49 and 7 to 49 periods.

Treatment		42 to 49 days			7 to 49 days			
NPP	Phytase	BWG	FI	FCR	BWG	FI	FCR	Mortality
(% of NRC)	(U/kg)	(g/day/bird)		(g/g)	(g/day/bird)		(g/g)	(%)
100	0	58.6	152.1 ^{bc}	2.59	47.5 ^{ab}	97.6 ^{ab}	2.05	7.14
100	500	66.0	158.4 ^{abc}	2.40	49.4 ^{ab}	98.7 ^{ab}	2.00	4.76
100	1000	61.3	158.4 ^{abc}	2.58	48.9 ^{ab}	99.9 ^{ab}	2.04	4.76
80	0	70.7	169.7 ^{a}	2.40	50.1 ^{ab}	100.2 ^{ab}	2.00	14.28
80	500	59.5	160.0 ^{abc}	2.69	50.4 ^{ab}	101.8 ^{a}	2.02	4.76
80	1000	63.7	160.7 ^{ab}	2.52	50.9 ^{a}	101.4 ^{a}	1.99	4.76
60	0	58.2	148.3 ^{c}	2.55	41.2 ^{c}	82.6 ^{d}	2.01	21.43
60	500	65.1	160.7 ^{ab}	2.47	47.4 ^{b}	92.9 ^{c}	1.96	9.52
60	1000	63.3	159.7 ^{abc}	2.52	48.4 ^{ab}	95.8 ^{bc}	1.98	9.52
SEM		0.75	8.82	0.10	1.06	1.62	0.02	2.74
Probabilities								
Treatments		NS	0.041	NS	0.001	0.000	0.046	0.011
NPP		NS	0.037	NS	0.000	0.000	0.049	0.014
Phytase		NS	NS	NS	0.004	0.001	NS	0.012
NPP × Phytase		NS	0.044	NS	0.033	0.009	NS	NS
Main effects								
NPP								
100		62.0	156.3 ^{b}	2.52	48.6 ^{b}	98.7 ^{a}	2.03 ^{a}	5.54 ^{b}
80		64.0	163.5 ^{a}	2.53	50.5 ^{a}	101.1 ^{a}	2.00 ^{ab}	7.93 ^{b}
60		62.2	156.3 ^{b}	2.51	45.6 ^{c}	90.5 ^{b}	1.98 ^{b}	13.49 ^{a}
Phytase								
0		62.5	156.7	2.51	46.3 ^{b}	93.5 ^{b}	2.02	14.27 ^{a}
500		63.5	159.7	2.51	49.0 ^{a}	97.0 ^{a}	1.99	6.35 ^{b}
1000		62.8	159.6	2.54	49.4 ^{a}	99.0 ^{a}	2.00	6.35 ^{b}

$^{a-c}$ Means within columns with no common superscript differ significantly ($P < 0.05$).

greatest response to added phytase belonged to chicks fed diets containing the lowest non-phytate phosphorus non-phytate phosphorus (60% NRC), which indicate that the level of dietary P has direct effect on supplemental phytase effectiveness. Results obtained in this experiment confirm the findings of other researchers (Simons et al., 1990; Perney et al., 1993; Denbow et al., 1995; Sebastian et al., 1996; Cabahug et al., 1999; Bozkurt et al., 2006; Plumstead et al., 2008). Addition 500 FTU/kg of microbial phytase to the diets containing different levels of non-phytate phosphorus (100, 80 and 60% of NRC requirements), increased 4, 6 and 15% body weight gain and 1.1, 1.6 and 12.5% feed intake, respectively. These results are in agreement with the findings of Ravindran et al. (1995) and Denbow et al. (1995).

Added microbial phytase to the diets significantly improved tibial ash (Table 4), confirming the findings of Broz et al. (1994), Sebastian et al. (1996), Yan et al. (2000, 2003), Angel et al. (2006) and Bozkurt et al. (2006). Increasing tibial ash may be due in partly to increase tibial P content. Sebastian et al. (1996) found an improvement in tibial ash and they suggested that, tibial ash is a good indication of increased bone mineralization due to an increasing apparent availability of P, Ca, Zn and Cu from the phytate minerals complex by the action of phytase.

Table 4. The effect of phytase supplementation at different levels of non-phytate phosphorus (NPP) on tibial ash, tibial ash P, apparent ileal digestibility and excretion of P in broiler chicks fed corn-wheat-soybean meal diet at 49.

Treatment		Tibia		Ileal	
NPP	Phytase	Ash	Ash P	Digestibility	Excretion
(% of NRC)	(U/kg)	(% DM)			(g/kg DM intake)
100	0	56.54 [a]	15.31	50.00 [b]	2.86 [a]
100	500	56.80 [a]	15.70	55.00 [b]	2.50 [abc]
100	1000	56.18 [a]	15.70	55.00 [b]	2.57 [ab]
80	0	55.00 [a]	14.55	51.00 [b]	2.40 [bc]
80	500	56.10 [a]	15.27	61.00 [a]	1.87 [de]
80	1000	56.80 [a]	15.72	64.35 [a]	1.82 [def]
60	0	47.12 [c]	13.23	48.88 [b]	2.13 [cd]
60	500	53.70 [b]	14.75	63.10 [a]	1.57 [ef]
60	1000	54.76 [ab]	14.82	65.11 [a]	1.45 [f]
SEM		0.63	0.40	1.95	0.12
				Probabilities	
Treatments		0.001	0.043	0.001	0.001
NPP		0.000	0.001	0.003	0.000
Phytase		0.000	0.006	0.000	0.000
NPP × Phytase		0.000	NS	0.050	0.051
Main effects NPP					
100		56.50 [a]	15.57 [a]	53.30 [b]	2.64 [a]
80		56.13 [a]	15.18 [a]	58.77 [a]	2.03 [b]
60		51.58 [b]	14.27 [b]	59.02 [a]	1.72 [c]
Phytase					
0		53.05 [b]	14.36 [b]	49.94 [b]	2.46 [a]
500		55.52 [a]	15.24 [a]	59.69 [a]	1.98 [b]
1000		55.91 [a]	15.41 [a]	61.47 [a]	1.94 [b]

[a-f] Means within columns with no common superscript differ significantly (P<0.05).

As shown in Table 4, apparent ileal phosphorus digestibility increased by phytase, but this improvement was significant with diet containing the lowest NPP level. These results showed that, dietary level of non-phytate phosphorus had significant effect on apparent ileal P digestibility and microbial phytase activity. Addition of 500 FTU/kg to diets containing 100, 80 and 60% of NRC requirements for NPP, increased by 10, 19.7 and 29.1% in apparent ileal P digestibility, respectively. In this regard, Irving and Cosgrove (1974) suggested that high concentration of inorganic phosphorus may have inhibitory effects on phytase activity because of a negative feed back mechanism.

Results of phosphorus excretion are consistent with apparent ileal P digestibility, because as apparent ileal P digestibility increased due to added phytase, excretion of P reduced. Addition of 500 FTU/kg to diets containing 100, 80 and 60% of NRC requirements increased 10, 19.7 and 29.1% of apparent ileal P digestibility and reduced 12.5, 22 and 26.5% of P excretion, respectively. This reduction in phosphorus excretion is an effective factor in reducing environmental pollution. These results are in agreement with the findings of Selle et al. (2009), Angel et al. (2005), Ravindran et al. (2000), Denbow et al. (1998) and Yi et al. (1996).

Based on the R^2 of second order translog equations, the percentage of apparent ileal digestibility and excretion of P, body weight gain of 21 to 42 days, body weight at 42 and 49 days, were the most sensitive indicators and feed intake of 21 to 42 days, body weight of 21 days and body

Table 5. Second order translog functions for performance, apparent ileal digestibility and excretion of P in broiler chicks fed corn-wheat- soybean meal diets with three phytase and non-phytate P levels from 7 to 21 d, 21 to 42 d, 42 to 49 d, and 7 to 49 d.

	Coefficients of second order translog equations[1]							
Item	α_0	α_1	α_2	α_3	α_4	α_5	P value	R^2
Performance								
Body weight 21d (g)	5.8902	-0.2919	-0.2919	-0.0529	0.0053	-0.0197	0.007	0.51
Feed conversion Ratio 7-21 d	0.5206	-.00451	-0.2919	-0.0529	0.0053	-00197	0.07	0.22
Feed intake 7-21d (g/day/bird)	3.3854	-0.7263	-0.3593	-0.0220	0.0020	-0.0115	0.010	0.49
Body weight gain 7-21d (g/day/ bird)	2.8983	-06252	-0.3619	-0.0634	0.0053	-0.0307	0.009	0.51
Body weight 42d (g)	6.0702	-2.2936	-0.9291	-0.0459	0.00001	-0.0408	0.000	0.77
Feed conversion ratio 21-42d	0.4924	-0.2017	-0.0571	-0.0036	0.0031	0.0144	0.036	0.41
Feed intake 21-75d (g/day/bird)	2.7999	-3.1379	-1.2512	-0.0501	0.0012	-0.0378	0.000	0.69
Body weight gain 21-42d (g/day/ bird)	2.3278	-2.9029	-1.1810	-0.0458	-0.0019	-0.0518	0.000	0.74
Body weight 49d (g)	5.6365	-2.9541	-1.0606	-0.0449	0.0002	-0.0349	0.000	0.70
Feed conversion ratio 42-49d	0.8301	-0.1424	-0.0525	-0.0206	0.0030	-0.0012	0.998	0.01
Feed intake 42-49d (g/day/bird)	3.4565	-2.1835	-.07340	-0.0103	-0.0007	-0.0118	0.230	0.26
Body weight gain 42-49d (g/day/bird)	2.6469	-2.0169	-0.6736	0.0084	-.0034	-0.0105	0.918	0.06
Digestibility of phosphorus[2](%)	2.2891	-2.2140	-0.7418	-0.0629	0.0015	-0.0539	0.000	0.83
Excretion of phosphorus[2](%)	4.2919	3.9462	1.1015	0.0794	-0.0012	0.0707	0.000	0.86

[1] Model: $LnY=\alpha_0 + \alpha_1 LnX_1 + \alpha_2 LnX_2 + \alpha_3(Ln\ X_1)^2 + \alpha_4(Ln\ X_2)^2 + \alpha_5 Ln\ X_1 LnX_2$; Y= response measurements, X_1 =non-phytate phosphorus percentage; X_2 = phytase added, FTU/kg of diet.
[2]These parameters were determined at 49 day of age.

weight gain of 7 to 21 days were also good indicators in order to assess the digestibility of P for broilers fed corn-wheat-soybean meal diets. FCR and body weight gain of 42 to 49 days were less sensitive. Similar results were observed in turkey poultry (1 to 21 days) fed soybean meal based semi-purified diets (Ravindran et al., 1995). Simons et al. (1990) and Denbow et al. (1995) also found that, growth of broilers was sensitive response for evaluating P availability in birds.

In conclusion, these results indicated that supplementation microbial phytase (500 FTU/kg of diet) can enhance the growth performance, tibial ash, apparent ileal digestibility and reduced excretion of P in broiler chicks fed corn-wheat-soybean meal diets. The magnitude of the improvements at the lower non-phytate phosphorus level was greater than NRC level of NPP. The results of this study showed that, the use of phytase enzyme in broilers' diets improved P availability which may reduce the amount of P that would be excreted in the manure, thus, reducing the environmental pollution potential.

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