

**MICROTECH 5000 (mini-granular)Phytase
for Broiler Ration**

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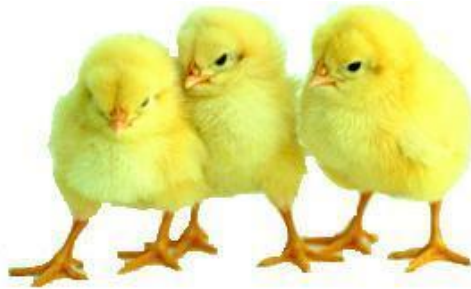
FEEDING TRIAL REPORT

MICROTECH 5000 (mini-granular) phytase

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for

GUANGDONG VTR BIO-TECH Co. Ltd.



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ABSTRACT

This experiment was conducted to investigate the effects of MICROTECH 5000 (mini-granular) phytase on growth performance, blood profiles and tibial breaking strength in broiler chickens fed diets with varying levels of available phosphorus. A total seven hundred-fifty chicks randomly assigned into six groups with five replicates and fed one of six corn-wheat-SBM based test diets containing three levels of available phosphorus (0.45, 0.35 or 0.25% for starter period; 0.35, 0.25 or 0.15% for finisher period) and two levels of phytase [MICROTECH 5000 (mini-granular), 0 or 600 units/kg diets] for 33-d. There were linear decrease in final body weight, daily weight gain and feed intake with decreasing dietary phosphorus levels. The addition of MICROTECH 5000 (mini-granular) phytase to low phosphorus or phosphorus-deficient diets improved each growth criteria during starter and finisher periods. The mean value of tibial breaking strength in groups fed diets with phytase was significantly higher than groups fed diets without phytase. The concentrations of blood P in groups fed diets with phytase were significantly higher than groups fed diets without phytase. These results indicated that the use of MICROTECH 5000 (mini-granular) phytase to broiler diets elicited improved growth performance and tibial strength. The normal growth and bone breaking strength were maintained in chicks fed low phosphorus diet or phosphorus-deficient diet with MICROTECH 5000 (mini-granular) phytase at 600 unit/kg diets.

Key Words: Available phosphorus levels, MICROTECH 5000 (mini-granular) phytase, Growth performance, Tibial breaking strength, blood phosphorus, broiler chickens

1. Background of phytase research

Phytate is a predominant form of phosphorus (about 61 ~ 80%) found in cereals, oilseed and plant protein sources in broiler diets (Ravindran et al., 1995). The phytate can bind amino acids and essential minerals, and thus reducing their bioavailability (Singh, 2008). The mono-gastric animals like broiler chicks are largely unable to use the phytate phosphorus because they lack the endogenous phytase (Simons et al., 1990; Singh, 2008).

Phosphorus (P) is essential nutrients and plays an important part in cell membrane and as a component of bone, and resulting in an increased requirement for P that is met by addition of inorganic P sources (Korean feeding standard for poultry, 2012). The addition of inorganic phosphates not only increase the feed and production costs (Dilger et al., 2004), but may lead to an burden of soluble P in the excreta (Ravindran et al., 1995; Ravindran et al., 1999; Simons et al., 1990). The excessive excretion of P in animal manure has received the greater attention as potential environmental pollutants (Correll, 1999), thus exogenous phytase has merit as a effective tool for minimizing P excretion (Yu et al., 2004). Saylor et al. (1991) reported that total P was decreased in the excreta of broiler chickens fed diet with exogenous phytase.

The use of novel exogenous phytase can improve dietary energy, minerals and amino acid digestibility (Selle and Ravindran, 2007) and consistently reduce the need to supplement of inorganic phosphate (Singh, 2008). The improved weight gain and feed efficiency by addition of dietary phytase are caused through phytate hydrolysis and alleviation of the anti-nutritional property of phytate (Walk et al., 2013).

Some criteria have been used for the estimation of P bioavailability by broilers, many researchers prefer growth performance and physico-chemical properties of bones (Qlan et al., 1996; Tang et al., 2012). Nelson et al. (1971) suggested that addition of phytase to broiler diets resulted in an improvement of utilization of phytate P as measured by ash contents of bone. The tibial breaking strength in chicks fed P-deficient diet with phytase was comparable with that of chicks fed P-adequate

control diet (Shaw et al., 2010).

The objectives of this experiment was to investigate the effects of Mini-Granular phytase on growth performance, blood profiles and tibial breaking strength of broiler chickens fed diets with varying levels of available phosphorus.

2. Materials and Methods

2.1. Experimental design

The male broiler chicks (Ross 308) were received on the day of hatch from a local hatchery and individually weighed. A total seven hundred-fifty chicks randomly assigned into six groups with five replicates and fed one of six corn-wheat-SBM based test diets containing three levels of available phosphorus (0.45, 0.35 or 0.25% for starter period; 0.35, 0.25 or 0.15% for finisher period) and two levels of phytase [MICROTECH 5000 (mini-granular), 0 or 600 units/kg diets] for 33-d (Table 1). The phytase used in this study was provided by GUANGDONG VTR BIO-TECH Co. Ltd. All test diets were formulated to be equal in the contents of TME_n, CP and limiting amino acids and processed as crumble and pellet forms for starter and finisher periods, respectively (Table 2 ~ 3). All test diets were formulated to be equal in the contents of TME_n, CP and limiting amino acids (NRC, 1994; Korean feeding standard for poultry, 2012).

Table 1. Experimental design

Treatments		Avail. P (%)	MICROTECH 5000 (mini-granular) (FTU/kg)
Starter	A	0.45	-
	B	0.45	600
	C	0.35	-
	D	0.35	600
	E	0.25	-
	F	0.25	600
Finisher	A	0.35	-
	B	0.35	600
	C	0.25	-
	D	0.25	600
	E	0.15	-

	F	0.15	600
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2.2. Management

The chicks were initially reared at 33°C; the room temperature was gradually decreased by 4°C weekly until a final temperature of 23°C was reached. Lighting was kept at 23/1 light/dark cycle throughout the experimental period. The chicks were allowed to have free access to diet and water. The experimental diets were freshly added everyday and feed intake of each replicate was recorded weekly. All animal care procedures were approved by Institutional Animal Care and Use Committee in Konkuk University.

2.3. Measurements

The chicks and feed were weighed at initiation, during phase changes, and at termination of experiment to calculate weight gain, feed intake and FCR on a pen basis. At the end of the experimental period, 10 chicks per each treatment were selected and weighed individually. The blood was drawn from wing vein using sterilized syringes for determination of the blood profiles. The concentration of serum Ca and P were measured according to the colorimetric method using biochemical analyzer (Hitachi modular system). At necropsy, right legs were immediately removed and weighed, and obtained tibia was frozen for subsequent determination of bone breaking strength. Tibial breaking strength was measured an Instron (Moel 3342, Instron Universal Testing Machine, Instron Corp., MA) with 50-kg-load cell as 50-kg load range with a crosshead speed of 50 mm/min with tibia supported on a 3.35 cm span.

2.4. Statistical analysis

Data were analyzed by the GLM procedure of the SAS (SAS, 2002) with pen means as the experimental units for evaluating growth performances, and individual birds as the experimental unit for the other criteria. Contrast statement were included to examine phosphorus, phytase, and phosphorus × phytase as a 3 × 2 factorial arrangement of treatments.

Table 2. Formula and chemical compositions of experimental diets (Starter)¹

	Starter					
	A	B	C	D	E	F
Corn	46.12	46.06	45.95	45.89	45.77	45.71
Wheat	10.00	10.00	10.00	10.00	10.00	10.00
Corn gluten meal	1.72	1.72	1.72	1.72	1.73	1.73
Wheat bran	3.19	3.19	3.74	3.74	4.28	4.28
Soybean oil	4.00	4.00	4.00	4.00	4.00	4.00
Soybean meal	20.75	20.75	20.58	20.58	20.41	20.41
Soybean meal (domestic)	10.00	10.00	10.00	10.00	10.00	10.00
Syn. lysine-HCl, 78%	0.16	0.16	0.16	0.16	0.17	0.17
Dicalcium phosphate	1.93	1.93	1.37	1.37	0.81	0.81
Syn. DL-methionine, 98%	0.25	0.25	0.25	0.25	0.25	0.25
Limestone	1.20	1.20	1.55	1.55	1.90	1.90
Choline-Cl, 50%	0.08	0.08	0.08	0.08	0.08	0.08
Salt	0.30	0.30	0.30	0.30	0.30	0.30
Poultry vitamin mix.	0.12	0.12	0.12	0.12	0.12	0.12
Poultry mineral mix.	0.15	0.15	0.15	0.15	0.15	0.15
NaHCO ₃	0.01	0.01	0.01	0.01	0.01	0.01
Syn. L-threonine, 98%	0.02	0.02	0.02	0.02	0.02	0.02
MICROTECH 5000 (mini-granular)	-	0.06	-	0.06	-	0.06
Crude protein, %	20.50	20.50	20.50	20.50	20.50	20.50
Ca, %	1.00	1.00	1.00	1.00	1.00	1.00
Available P, %	0.45	0.45	0.35	0.35	0.25	0.25
Lysine, %	1.20	1.20	1.20	1.20	1.20	1.20
Methionine, %	0.57	0.57	0.57	0.57	0.57	0.57
TME _n , kcal/kg	3,080	3,080	3,080	3,080	3,080	3,080

¹Vit. and Min. mixture provided the following nutrients per kg of diet: vitamin A, 40,000 IU; vitamin D3, 8,000 IU; vitamin E, 10 IU; vitamin K3, 4.0 mg; vitamin B1, 4.0 mg; vitamin B2, 12.0 mg; vitamin B6, 6.0 mg; vitamin B12, 0.02 mg; niacin, 60.0 mg; pantothenic acid, 20 mg; folic acid, 2.0 mg; biotin, 0.02 mg; Fe, 30.0 mg; Zn, 25.0 mg; Mn, 20.0 mg; Cu, 5.0 mg; Se, 0.1 mg.

Table 3. Formula and chemical compositions of experimental diets (Finisher)¹

	Finisher					
	A	B	C	D	E	F
Corn	50.46	50.40	50.29	50.23	50.12	50.06
Wheat	10.00	10.00	10.00	10.00	10.00	10.00
Corn gluten meal	1.52	1.52	1.52	1.52	1.53	1.53
Wheat bran	1.15	1.15	1.70	1.70	2.24	2.24
Soybean oil	4.00	4.00	4.00	4.00	4.00	4.00
Soybean meal	19.11	19.11	18.94	18.94	18.77	18.77
Soybean meal (domestic)	10.00	10.00	10.00	10.00	10.00	10.00
Syn. lysine-HCl, 78%	0.09	0.09	0.09	0.09	0.09	0.09
Dicalcium phosphate	1.41	1.41	0.86	0.86	0.30	0.30
Syn. DL-methionine, 98%	0.19	0.19	0.19	0.19	0.19	0.19
Limestone	1.40	1.40	1.74	1.74	2.09	2.09
Choline-Cl, 50%	0.07	0.07	0.07	0.07	0.07	0.07
Salt	0.30	0.30	0.30	0.30	0.30	0.30
Poultry vitamin mix.	0.12	0.12	0.12	0.12	0.12	0.12
Poultry mineral mix.	0.15	0.15	0.15	0.15	0.15	0.15
NaHCO ₃	0.01	0.01	0.01	0.01	0.01	0.01
Syn. L-threonine, 98%	0.02	0.02	0.02	0.02	0.02	0.02
MICROTECH 5000 (mini-granular)	-	0.06	-	0.06	-	0.06
Crude protein, %	19.50	19.50	19.50	19.50	19.50	19.50
Ca, %	0.95	0.95	0.95	0.95	0.95	0.95
Available P, %	0.35	0.35	0.25	0.25	0.15	0.15
Lysine, %	0.95	0.95	0.95	0.95	0.95	0.95
Methionine, %	0.48	0.48	0.48	0.48	0.48	0.48
TMEn, kcal/kg	3,150	3,150	3,150	3,150	3,150	3,150

¹Vit. and Min. mixture provided the following nutrients per kg of diet: vitamin A, 40,000 IU; vitamin D3, 8,000 IU; vitamin E, 10 IU; vitamin K3, 4.0 mg; vitamin B1, 4.0 mg; vitamin B2, 12.0 mg; vitamin B6, 6.0 mg; vitamin B12, 0.02 mg; niacin, 60.0 mg; pantothenic acid, 20 mg; folic acid, 2.0 mg; biotin, 0.02 mg; Fe, 30.0 mg; Zn, 25.0 mg; Mn, 20.0 mg; Cu, 5.0 mg; Se, 0.1 mg.

3. Results

3.1. Growth performance

The growth performance of broiler chickens fed diets varying levels of available phosphorus with or without MICROTECH 5000 (mini-granular) phytase were shown in Table 4 ~ 7. The final body weights of chicks fed phosphorus-adequate diets with or without phytase (A and B) and low phosphorus diet with phytase (D) were higher than those of other diets. The body weight of E group (phosphorus-deficient diet without phytase) had the lowest value. The final body weight of F group (phosphorus-deficient diet with phytase) was comparable with that of C group (low phosphorus diet without phytase). With decreasing dietary phosphorus levels, the body weights were significantly decreased. The addition of MICROTECH 5000 (mini-granular) phytase to low phosphorus or phosphorus-deficient diets improved the body weights in the starter and finisher periods. There was a significant phosphorus \times phytase interaction for body weights during starter and finisher periods.

The change in daily body weight gain was similar to that of final body weight (Table 5). The daily body weight gains of A, B and D (low phosphorus diet with phytase) groups were relatively higher, and that of E group (phosphorus-deficient diet without phytase) had the lowest value. The body weight gains were linearly decreased with decreasing dietary phosphorus levels. The addition of MICROTECH 5000 (mini-granular) phytase to low phosphorus or phosphorus-deficient diets increased in daily body weight gains in both periods. There was a significant phosphorus \times phytase interaction for body weights during starter and finisher periods.

The feed intake was tended to be reduced by decreasing dietary phosphorus levels (Table 6). The average feed intake of chicks fed phosphorus-adequate diets (A and B) were 81.71g, and those of low phosphorus diets (C and D) and phosphorus-deficient diets (E and F) were 77.07 and 68.54g, respectively. The average feed intake of chicks fed diets with phytase (B, D and F) was significantly higher than those of chicks fed diets without phytase (A, C and E). There were significant trends of dietary phosphorus levels, phytase and phosphorus \times phytase interaction affecting the average feed intake. Feed conversion ratio was not influenced by dietary phosphorus levels and phytase addition

(Table 7).

Table 4. Effects of diets varying levels of available phosphorus with or without phytase on body weight¹

Groups	Treatments		Body weight (g/bird)		
	Phosphorus (%)	Phytase (FTU)	1 d	21 d	33 d
A	0.45/0.35	0	41.70	856.5	1897.2
B	0.45/0.35	600	41.76	839.6	1887.5
C	0.35/0.25	0	41.74	794.0	1800.3
D	0.35/0.25	600	41.76	814.5	1908.1
E	0.25/0.15	0	41.75	644.3	1483.1
F	0.25/0.15	600	41.72	762.9	1800.1
	SEM ¹		0.0250	23.697	32.219
Main effects					
	0.45/0.35		41.73	848.1	1892.3
	0.35/0.25		41.75	804.2	1854.2
	0.25/0.15		41.74	703.6	1641.6
	SEM		0.019	17.773	24.164
		0	41.73	764.9	1726.9
		600	41.75	805.7	1865.2
		SEM	0.0150	14.240	19.361
	Statistical effects			<i>p</i> > F	
	Model		0.5587	<0.0001	<0.0001
	Phytase		0.4508	0.0654	<0.0001
	Phosphorus		0.6964	<0.0001	<0.0001
	Phosphorus × Phytase		0.3119	0.0381	0.0004

¹SEM : Standard error of the means.

Table 5. Effects of diets varying levels of available phosphorus with or without phytase on weight gain¹

Groups	Treatments		Weight gain (g/bird)		
	Phosphorus (%)	Phytase (FTU)	1-21d	22-33d	1-33d
A	0.45/0.35	0	38.80	86.72	56.23
B	0.45/0.35	600	37.99	87.32	55.93
C	0.35/0.25	0	35.82	83.86	53.29
D	0.35/0.25	600	36.80	91.13	56.56
E	0.25/0.15	0	28.70	69.91	43.68
F	0.25/0.15	600	34.35	86.43	53.29
	SEM ¹		1.261	2.197	1.091
Main effects					
	0.45/0.35		38.40	87.02	56.08
	0.35/0.25		36.31	87.50	54.92
	0.25/0.15		31.52	78.16	48.48
	SEM		0.8458	1.6478	0.7320
		0	34.44	80.16	51.07
		600	36.38	88.30	55.26
		SEM	0.728	1.320	0.587
	Statistical effects			<i>p</i> > F	
	Model		<0.0001	<0.0001	<0.0001
	Phytase		0.0654	0.0004	<0.0001
	Phosphorus		<0.0001	0.0010	<0.0001
	Phosphorus × Phytase		0.0380	0.0098	0.0004

¹SEM : Standard error of the means.

Table 6. Effects of diets varying levels of available phosphorus with or without phytase on feed intake¹

Groups	Treatments		Feed intake (g/bird)		
	Phosphorus (%)	Phytase (FTU)	1-21d	22-33d	1-33d
A	0.45/0.35	0	52.6	135.2	82.1
B	0.45/0.35	600	53.8	129.8	81.3
C	0.35/0.25	0	49.0	122.5	75.0
D	0.35/0.25	600	50.3	131.1	79.1
E	0.25/0.15	0	40.9	106.0	61.6
F	0.25/0.15	600	48.5	122.6	75.4
	SEM ¹		1.691	2.506	2.343
Main effects					
	0.45/0.35		53.2	132.8	81.7
	0.35/0.25		49.7	126.8	77.1
	0.25/0.15		44.3	113.4	68.5
	SEM		1.135	1.880	1.572
		0	47.4	121.2	72.9
		600	50.9	127.8	78.6
		SEM	0.936	1.561	1.301
	Statistical effects			<i>p</i> > F	
	Model		0.0002	<0.0001	<0.0001
	Phytase		0.0193	0.0073	0.0059
	Phosphorus		0.0002	<0.0001	<0.0001
	Phosphorus × Phytase		0.1035	0.0018	0.0127

¹SEM : Standard error of the means.

Table 7. Effects of diets varying levels of available phosphorus with or without phytase on feed conversion ratio¹

Groups	Treatments		FCR (feed/gain)		
	Phosphorus (%)	Phytase (FTU)	1-21d	22-33d	1-33d
A	0.45/0.35	0	1.36	1.57	1.46
B	0.45/0.35	600	1.42	1.49	1.45
C	0.35/0.25	0	1.37	1.46	1.41
D	0.35/0.25	600	1.37	1.44	1.40
E	0.25/0.15	0	1.43	1.51	1.41
F	0.25/0.15	600	1.42	1.43	1.42
	SEM ¹		0.015	0.038	0.026
Main effects					
	0.45/0.35		1.39	1.53	1.46
	0.35/0.25		1.37	1.45	1.40
	0.25/0.15		1.42	1.47	1.41
	SEM		0.012	0.030	0.019
		0	1.38	1.51	1.43
		600	1.40	1.45	1.42
		SEM	0.010	0.024	0.017
Statistical effects				<i>p</i> > F	
	Model		0.0169	0.1663	0.4770
	Phytase		0.2233	0.0674	0.9083
	Phosphorus		0.0139	0.1710	0.1381
	Phosphorus × Phytase		0.1033	0.7124	0.9351

¹SEM : Standard error of the means.

3.2. Tibial breaking strength

The tibial breaking strength of broiler chickens fed diets varying levels of available phosphorus with or without MICROTECH 5000 (mini-granular) phytase were shown in Table 8. The tibial breaking strength of chicks fed phosphorus-adequate diets with or without phytase (A and B) and low phosphorus diet with phytase (D) were higher than those of other diets, and that of E group (phosphorus-deficient diet without phytase) had the lowest value. The average tibial breaking strength of chicks fed diets with 0.25/0.15% available phosphorus (E and F) was markedly lower than that of other groups. The mean value in groups fed diets with phytase (B, D and F) was significantly higher than groups fed diets without phytase (A, C and E). There were significant trends of dietary phosphorus levels, phytase and phosphorus×phytase interaction affecting the tibial breaking strength.

Table 8. Effects of diets varying levels of available phosphorus with or without phytase on tibial breaking strength ¹

Treatments			Breaking strength, Kgf
Groups	Phosphorus (%)	Phytase (FTU)	
A	0.45/0.35	0	32.49
B	0.45/0.35	600	33.41
C	0.35/0.25	0	30.99
D	0.35/0.25	600	33.48
E	0.25/0.15	0	19.22
F	0.25/0.15	600	28.16
SEM ¹			1.5238
Main effects			
0.45/0.35			32.95
0.35/0.25			32.23
0.25/0.15			23.70
SEM			1.077
			0
			600
			SEM
			0.8798
Statistical effects			<i>p</i> > F
Model			<0.0001
Phytase			0.0022
Phosphorus			<0.0001
Phosphorus × Phytase			0.0295

¹SEM : Standard error of the means.

3.3. Blood profiles

The concentrations of blood calcium and phosphorus in chicks fed diets varying levels of available phosphorus with or without MICROTECH 5000 (mini-granular) phytase were shown in Table 9. Neither the phosphorus level fed nor phytase addition affected blood calcium concentrations. On the other hand, the mean concentration of blood phosphorus in chicks fed diets with 0.35/0.25% available phosphorus (C and D) was comparable with those of chicks fed diets with 0.45/0.35% available phosphorus (A and B). The concentration of blood phosphorus in E group (phosphorus-deficient diet without phytase) had the lowest value. The mean concentration of blood phosphorus in groups fed diets with phytase (B, D and F) was significantly higher than groups fed diets without phytase. There were significant trends of dietary phosphorus levels, phytase and phosphorus \times phytase interaction affecting the levels of blood phosphorus.

Table 9. Effects of diets varying levels of available phosphorus with or without phytase on blood profiles ¹

Treatments			Ca, mg/dl	P, mg/dl
Groups	Phosphorus (%)	Phytase (FTU)		
A	0.45/0.35	0	10.59	7.73
B	0.45/0.35	600	10.37	8.10
C	0.35/0.25	0	10.40	8.01
D	0.35/0.25	600	10.44	8.09
E	0.25/0.15	0	10.65	4.32
F	0.25/0.15	600	10.21	8.10
	SEM ¹		0.263	0.367
Main effects				
	0.45/0.35		10.48	7.91
	0.35/0.25		10.42	8.05
	0.25/0.15		10.43	6.21
	SEM		0.186	0.259
		0	10.54	6.69
		600	10.34	8.09
		SEM	0.152	0.212
Statistical effects			<i>p</i> > F	
	Model		0.8751	<0.0001
	Phytase		0.3473	<0.0001
	Phosphorus		0.9689	<0.0001
	Phosphorus × Phytase		0.6676	<0.0001

¹SEM : Standard error of the means.

4. Conclusion

- This experiment was conducted to investigate the effects of MICROTECH 5000 (mini-granular) phytase on growth performance, blood profiles and tibial breaking strength in broiler chickens fed diets with varying levels of available phosphorus.
- There were linear decrease in growth performance (final body weight, daily weight gain and feed intake) with decreasing dietary phosphorus levels.
- The addition of MICROTECH 5000 (mini-granular) phytase to low phosphorus or phosphorus-deficient diets improved each growth criteria during starter and finisher periods.
- The mean value of tibial breaking strength in groups fed diets with phytase was significantly higher than groups fed diets without phytase.
- These results indicated that the adding MICROTECH 5000 (mini-granular) phytase could improve the growth performance and tibial strength of broiler chickens fed available phosphorus restricted diets. The normal growth and bone breaking strength were maintained in chicks fed low phosphorus diet or phosphorus-deficient diet with MICROTECH 5000 (mini-granular) phytase at 600 unit/kg diets.

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