Replacement of inorganic phosphorus by microbial phytase for young pigs fed on a maize–soyabean-meal diet

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(*Received 13 April 1995 – Revised 23 November 1995 – Accepted 16 January 1996*)

Ninety-six crossbred young pigs (body weight 7·8 kg) were used in a 5-week trial to determine the effectiveness of microbial phytase (*EC* 3.1.3.26) in improving the bioavailabilities of P and other nutrients in maize–soyabean-meal diets and, thus, replacing inorganic P with phytase. A 2 × 5 factorial arrangement of treatments was employed with two available P (aP) levels (0·7 and 1·6 g/kg) and five phytase levels (0, 350, 700, 1050, 1400 U (the quantity of enzyme that liberates 1 μmol inorganic phosphate/min from 5·1 mm-sodium phytate at pH 5·5 and 37°)/kg diet). In addition, two extra diets were formulated to supply the National Research Council (1988) recommended level of aP (3·2 g/kg) with 0 or 1400 U phytase. The addition of graded levels of phytase resulted in linear increases in average daily weight gain, average daily feed intake and weight gain: feed intake for pigs fed on diets containing 0·7 or 1·6 g aP/kg ($P < 0·04$). Also, the addition of phytase linearly increased apparent digestibilities of P and Ca ($P < 0·01$), whereas faecal P excretion was linearly decreased ($P < 0·01$). Linear increases in shear force, shear energy and ash content of both the metacarpal and tenth rib, and shear stress of the metacarpal were found to respond to added phytase ($P < 0·01$). These improvements in performance, apparent P absorption and bone measurements by phytase were also observed by increasing dietary aP levels for most measurements. Adding 1400 U phytase to the 3·2 g aP/kg diet further increased average daily weight gain, average daily feed intake, apparent absorption of P, Ca and N and metatarsal shear force and ash content ($P < 0·01$ to 0·05). Generally, maximum responses occurred at a phytase level of 1050 U/kg diet for the 0·7 g aP/kg diets and 700 U for the 1·6 g aP/kg diets. Based on non-linear and linear response equations generated for the phytase and aP levels, the average function of the equivalency of P (Y, g/kg) *v*. microbial phytase (X, U/kg) was developed across aP levels of 0·7 and 1·6 g/kg for average daily weight gain and apparent digestibility of P: $Y = 2·622 - 2·559e^{-0·00185X}$. The replacement of 1 g inorganic P as defluorinated phosphate would require about 246 U microbial phytase. This represents 41% of released P from phytate.

Phosphorus: Phytase: Maize: Soyabean

Interest in the addition of phytase (*EC* 3.1.3.26) to pig and poultry diets has occurred because of a general need to reduce the amount of P excreted because of the potential for environmental pollution, particularly in areas where large numbers of pig and poultry are produced. P is a key element in swine nutrition, and adequate amounts are necessary for optimal growth, reproduction and bone development (National Research Council, 1988). However, much of the P in swine diets (composed primarily of maize and soyabean meal in the United States) cannot be utilized because it occurs bound as phytate-P (Cromwell, 1992). The poor availability of this bound P leads to large amounts of P present in pig manure (Cromwell & Coffey, 1991) because inorganic P must be added to meet the P requirement. Microbial phytase supplementation has been shown to be very effective in releasing a significant portion of the bound P and, thus, improves the availability of P (Simons *et al.* 1990; Cromwell *et al.* 1993*b*; Hoppe *et al.* 1993). P excretion can be reduced, with estimates ranging from 25 to 50%, by the addition of phytase to the diet (Simons *et al.* 1990; Jongbloed *et al.* 1992; Cromwell *et al.* 1993*a*; Lei *et al.* 1993*b*). An estimated equivalency value of phytase for inorganic P was reported by Hoppe *et al.* (1993) but only a single level of P was fed. The objective of the present study was to determine the effectiveness of microbial phytase for increasing the absorption of Ca, P and N, increasing bone mineralization and improving performance of young pigs fed on a maize–soyabean-meal basal diet with varying levels of phytase in combination with two deficient levels of available P (aP). Response equations were generated and evaluated; the most sensitive indicators were used to calculate equivalency values of phytase for inorganic P.

MATERIALS AND METHODS

Animal and feeding management. A total of ninety-six crossbred pigs (equal number of males and females) were utilized. The pigs were weaned between 28 and 35 d of age and given a 7 d adjustment period before dietary treatments were started. During the adjustment period they were fed on a pre-starter formula containing 220 g crude protein (N × 6·25; CP)/kg for 4 d (Maximum Wean 10–15; Southern States Cooperative, Richmond, VA, USA) and then fed on a 200 g CP/kg maize–soyabean-meal diet containing 100 g dried whey/kg for the remaining 3 d. After the adjustment period, pigs were weighed (average weight 7·8 (SE 0·13) kg) and randomly placed on treatments within outcome groups based on sex and weight. Littermates were balanced across treatments as far as possible.

The pigs were housed two per pen (0·6 m × 0·9 m) in environmentally-controlled rooms with expanded metal floors. Each pen had a plastic-coated welded wire floor and was equipped with a nipple waterer and a stainless-steel feeder. Room temperatures were initially set at 29° and were lowered about 2° per week after the second week. A continuous lighting regimen and recommended air ventilation rates were maintained (Murphy *et al.* 1990). Feed and water were available *ad libitum* at all times. Pigs were weighed individually at weekly intervals during the 5-week study period. Pen feed intakes were recorded. The care and treatment of pigs followed published guidelines (Consortium, 1988).

Treatments and diets. A 2 × 5 factorial arrangement of treatments was employed to evaluate the response of weanling pigs to graded levels of phytase added to diets containing two levels of aP that were below the pig estimated requirements (National Research Council, 1988). Dietary aP levels were formulated at 0·7 and 1·6 g aP/kg, or 3·6 and 4·5 g total P (tP)/kg respectively, and each level of aP was supplemented with 0, 350, 700, 1050 and 1400 U phytase/kg diet. One Unit is defined as the quantity of enzyme which liberates 1 μmol inorganic P/min from 5·1 mM-sodium phytate at pH 5·5 and 37°. These P levels were formulated below the current National Research Council (1988) recommendations to ensure maximum response to phytase additions. In addition to the ten diets described previously, two additional diets were formulated to supply the recommended level of P (3·2 g aP/kg or 6·1 g tP/kg); one diet was fed without phytase and one was fed with 1400 U phytase/kg diet. The diet without the addition of phytase served as the positive control. Each of the twelve dietary treatments was fed to four replicate pens of two pigs each (one barrow and one female).

The basal diets were based on maize and soyabean meal as the protein sources (Table 1). Maize and soyabean meal supplied all the P (0·7 g aP/kg or 3·6 g tP/kg contained in the lowest P basal diet. The desired levels of aP in the other basal diets were achieved by the addition of defluorinated phosphate (CDP®; Consolidated Minerals Inc., Feed Supplemental Division, Plant City, FL, USA). The Ca:tP ratio was maintained at 2:1 in all basal diets. Defluorinated phosphate and limestone were added to the diets at the expense of maize starch. Since phytate was supplied from maize and soyabean meal, the calculated

Table 1. *Composition and calculated analysis (g/kg) of the basal diets*

Available P (g/kg)...	0·7*	1·6*	3·2†
Ingredients			
Ground yellow maize	726·2	720·7	724·5
Soyabean meal (485 g CP/kg)	245·0	245·0	245·0
Limestone	16·8	17·3	4·7
Defluorinated phosphate‡	—	5·0	13·8
Salt	3·0	3·0	3·0
Vitamin premix§	2·5	2·5	2·5
Trace mineral premix‖	1·0	1·0	1·0
Se premix¶	0·5	0·5	0·5
Cr_2O_3–starch mixture	4·0	4·0	4·0
Lysine	1·0	1·0	1·0
Calculated analysis			
CP	183·0	182·5	182·9
Lysine	10·5	10·5	10·5
Methionine and cysteine	6·1	6·1	6·1
Ca	7·2	9·0	7·0
Total P	3·6	4·5	6·1
Available P	0·7	1·6	3·2

CP, crude protein (N × 6·25).
* 0, 350, 700, 1050 and 1400 FTU phytase/kg diet were added. Phytase (Natuphos-5000 FTU/g) was supplied by BASF Corp., 3000 Continental Drive North, Mount Olive, NJ 07828-1234, USA.
† 0 and 1400 U phytase/kg diet were added.
‡ Fine CDP; Southern Bag Corp., Valdosta, GA 31803, USA.
§ Supplied (/kg diet): retinyl acetate 1514 μg, cholecalciferol 110 μg, DL-α-tocopheryl acetate 22 mg, riboflavin 4·4 mg, niacin 22 mg, choline 440 mg, D-pantothenic acid 22 mg, D-biotin 0·44 mg, cyanocobalamin 22 μg, vitamin K (as menadione dimethylprimidinol bisulphite) 2·2 mg.
‖ Supplied (/kg diet): Zn 150 mg, Fe 175 mg, Mn 60 mg, Cu 17·5 mg, I 2 mg.
¶ Supplied 0·3 mg Se/kg diet.

(National Research Council, 1988) dietary content of phytate-P, 2·44 g/kg, was similar in all diets. A Cr_2O_3 mixture was included at a level of 4·0 g/kg in diets as an indigestible indicator for digestible measurements. To obtain a homogeneous distribution of the indicator in the diet, Cr_2O_3 was first mixed in a small mixer with maize starch at a ratio of 1:3 (w/w) and then ground in a laboratory mill to pass through 1 mm sieve (Dellaert *et al.* 1990).

Sampling and analysis. Grab faecal samples of approximately the same amount were collected from each pen during the fourth and fifth weeks. During each of these weeks, faeces were collected twice daily (morning and evening) on three alternate days. Collections from each of the 3 d within 1 week were pooled and frozen at −20° in airtight plastic bags for subsequent analysis. After thawing, faecal samples were dried in an oven at 70°. The dried samples, along with representative samples of diets, were ground to pass through a 1 mm sieve. DM was determined according to standard Association of Official Analytical Chemists (1990) methods. After wet digestion of samples using nitric acid–perchloric acid (5:3, v/v), P concentrations were assayed photometrically (Association of Official Analytical Chemists, 1990), and Ca and Cr contents were determined using an atomic absorption spectrophotometer (Perkin Elmer 5100, Norwalk, CT, USA) according to the manufacturer's recommendations. N content was determined using the Kjeldahl method. The apparent digestion or absorption coefficients of Ca, P and N over the total gastrointestinal tract were calculated (Dellaert *et al.* 1990).

At the end of week 5 the barrow in each replicate pen (four per treatment) was slaughtered for the collection of bone samples. The front foot and the tenth rib on the left side were removed and frozen in air-tight plastic bags. The foot samples were later thawed, extraneous tissue was removed and the fourth metacarpal was retained. The rib samples were also defleshed. Bones were then refrozen in air-tight plastic bags until required for shear force determination as described by Combs *et al.* (1991).

The shear force of the fourth metacarpal and the tenth rib were determined using an Instron Universal Testing Machine (Model 1123; Instron Corp., Canton, MA, USA). Bones were thawed in air-tight plastic bags immediately before testing to prevent desiccation. After the shear failure test, wall thickness was measured near the sheared section (after cutting the sheared end off with a band saw) using dial calipers. Shear stress values were calculated according to the formula of Combs *et al.* (1991) for metacarpals and of Wilson (1991) for ribs. After the shear test the bones were oven-dried at 100° for 24 h and ashed in a muffle furnace at 600° for 24 h. Bone ash was expressed on a dry weight basis.

Statistical analysis and calculation of P-equivalency values. The data were analysed by the GLM procedure of Statistical Analysis Systems (1990). Performance and absorption data were analysed using the pen as the experimental unit, whereas bone measurements were analysed using individual pig values as the experimental unit. The digestibility of the DM was used as a covariate for the digestibility of Ca, P and N as suggested by Dellaert *et al.* (1990), but only very small differences were observed and adjusted coefficients are reported. Linear and quadratic effects of supplemental phytase within each aP level, and within dietary aP levels without phytase were tested using orthogonal polynomials. A comparison between the positive control diet with and without phytase was made using non-orthogonal contrasts. $P < 0{\cdot}05$–$0{\cdot}10$ was considered important and was generally referred to as a trend. Second-order translog functions were derived for the 2×5 factorial with the following model: $\ln Y = \alpha_0 + \alpha_1 D_1 + \alpha_2 \ln X + \alpha_3 (\ln X)^2 + \alpha_4 D_1 \ln X$, where, Y is response measurement such as average daily weight gain (ADG; g) or P absorption (%), X is phytase added (U/kg diet), D_1 is aP in the diet (when aP = 0·7 g/kg, $D_1 = 0$, when aP = 1·6 g/kg, $D_1 = 1$).

Non-linear (Proc NLIN) functions were derived using treatment means for phytase levels at each P level and linear functions for the three aP levels without phytase added. The non-linear regression model used was $Y = a(1 - be^{-kX})$ and the linear regression model was $Y = a + bX$, where Y is response measurements, X is phytase levels added (U/kg diet) or aP (g/kg) levels of diet. The sensitivity of various measurements was determined by examining the r^2 values of the second-order translog equations for all measurements and the non-linear and linear equations. The functions generated from all measurements with the higher r^2 values for aP levels and phytase levels were used to generate P-equivalency equations of phytase. The equation for aP and the equation for added phytase at each of the two levels of aP were set equal. For example, the equation for ADG at 0·7 g aP/kg was as follows (Tables 6 and 7):

$$212{\cdot}4 + 34{\cdot}5\, X_1 = 376{\cdot}7 - 133{\cdot}4 e^{-0{\cdot}00095 X_2},$$
$$X_1 = 4{\cdot}762 - 3{\cdot}867 e^{-0{\cdot}00095 X_2},$$

where, X_1 is available P (g/kg), X_2 is phytase added (U/kg of diet). The resulting equations were used to calculate the equivalent aP (g/kg) at 250, 500, 750 and 1000 U phytase/kg diet. Because of their sensitivity, economic importance and relative ease of determination, values for ADG and apparent absorption of P were then used to determine the amount of P released. The released P was expressed as a percentage of phytate-P. The amount of P released per 100 U phytase was also calculated. The average function for amount of P released (Y, g/kg) by microbial phytase (X, units/kg) was developed for the average of the aP levels of 0·7 and 1·6 g/kg.

RESULTS

Growth performance. Cumulative ADG and average daily feed intake (ADFI) of young pigs increased linearly ($P < 0.01$) and weight gain:feed intake (G:F) tended ($P < 0.08$) to be increased as the level of aP increased (only diets without phytase; Tables 2 and 3). The addition of graded levels of phytase resulted in linear increases of ADG ($P < 0.01$), ADFI ($P < 0.04$) and G:F ($P < 0.03$) for pigs fed on diets containing 0·7 and 1·6 g aP/kg. This linear increase in ADG reached a plateau at 1050 U phytase/kg diet for the 0·7 g aP/kg diets, and at 700 U phytase for the 1·6 g aP/kg diets. ADFI continued to increase to the highest level of phytase (1400 U) for the 0·7 g aP/kg diets, but appeared to reach a plateau at 700 U phytase for the 1·6 g aP/kg diets. G:F appeared to reach a plateau at 1050 U phytase for the 0·7 g aP/kg diets, and continued to increase linearly ($P < 0.03$) to 1400 U phytase for 1·6 g aP/kg diets. The addition of 1400 U phytase to the diet containing a National Research Council (1988) recommended level of 3·2 g aP/kg (6·1 g tP/kg) increased ADG ($P < 0.05$) and tended to increase ADFI ($P < 0.07$), with no effect on G:F.

Apparent digestibility and absorption coefficients. For diets without added phytase, increasing the aP level linearly increased ($P < 0.01$) apparent absorption of P, but linearly decreased the digestibility of DM and N ($P < 0.05$ and 0·01 respectively; Tables 2 and 3). The effect of aP level on Ca absorption was not significant. The apparent absorption of P continued to increase to the highest level of phytase (1400 U/kg) for pigs fed on the 0·7 g aP/kg diets with both linear ($P < 0.01$) and quadratic ($P < 0.02$) effects. For pigs fed on the 1·6 g aP/kg diets, P absorption reached a plateau at 1050 U phytase with both linear ($P < 0.01$) and quadratic ($P < 0.01$) effects. DM digestibility was only decreased ($P < 0.05$) by phytase addition for pigs fed on 1·6 g aP/kg diets. The apparent absorption of Ca increased linearly ($P < 0.01$) as phytase was added, with the magnitude of the increase greater for the 0·7 g aP/kg diets than for the 1·6 g aP/kg diets ($P < 0.01$ and 0·03 respectively). Adding 1400 U phytase/kg to the diet containing 3·2 g aP/kg increased the absorption of P ($P < 0.01$), Ca ($P < 0.05$) and N ($P < 0.05$), but did not change the absorption of DM. Differences for P and Ca absorption were slightly greater when adjusted for DM digestibility, whereas, the effect on N digestibility was very limited.

Faecal P excretion tended to be increased (linear, $P < 0.06$) as the level of aP (without phytase) was increased. Addition of graded levels of phytase decreased faecal P excretion; both linear ($P < 0.01$) and quadratic ($P < 0.01$) effects were observed. The addition of 1400 U phytase to the 3·2 g aP/kg diet decreased faecal P excretion ($P < 0.01$).

Bone characteristics. Bone measurements, shear force, shear energy, and ash content of the metacarpal and tenth rib for pigs fed on diets without added phytase increased linearly as the level of aP increased ($P < 0.05$ and 0·01 respectively; Tables 2 and 3). The addition of graded levels of phytase resulted in linear increases ($P < 0.01$) in shear force, shear energy and ash content for both the metacarpal and tenth rib at both aP levels. Also, shear stress increased linearly ($P < 0.01$) for the metacarpal as phytase was added, but the effect of phytase on shear stress of the tenth rib was not significant. The aP × phytase interactions generally were not significant, except for shear force ($P < 0.01$) and energy ($P < 0.05$) for the tenth rib. When pigs were fed on 3·2 g aP/kg the addition of 1400 U phytase increased shear force and ash content of the metacarpal ($P < 0.02$), and shear force and energy of the tenth rib ($P < 0.07$ and 0·01 respectively).

Response curves. Second-order translog functions were generated from data for the five phytase and two aP levels on performance, apparent digestibility, faecal P excretion, and bone characteristics of young pigs fed on maize–soyabean-meal diets (Table 4). Response

Table 2. *Performance, apparent digestibility coefficients of DM and phosphorus, calcium and nitrogen, faecal P excretion and bone characteristics of young weanling pigs fed on a soyabean-meal-based diet containing varying levels of available phosphorus (aP) and supplemental phytase* (EC *3.1.3.26; PY*)*

Diet no. ...	1	2	3	4	5	6	7	8	9	10	11	12	
aP (g/kg)...	0·7					1·6					3·2		
PY (U/kg diet)...	0	350	700	1050	1400	0	350	700	1050	1400	0	1400	SEM
Performance for weeks 1–5†													
Average daily wt gain (g)	244	282	300	338	337	256	328	387	357	386	327	368	24·1
Average daily feed intake (g)	528	569	621	645	675	554	628	706	633	621	596	658	51·6
Wt gain:feed intake (g/kg)	463	501	491	529	517	469	523	549	580	625	550	563	50·3
Apparent digestibility coefficients‡													
DM (%)	87·9	86·8	87·1	87·2	87·4	87·6	86·7	86·5	86·8	86·2	86·1	87·1	0·43
P (%)	27·6	40·2	42·1	47·7	52·5	35·3	47·6	53·9	56·2	54·4	57·2	68·3	2·35
Ca (%)	72·1	77·3	77·5	79·6	81·0	72·9	74·8	78·1	76·9	77·8	74·6	76·8	1·68
N (%)	85·4	83·2	83·4	83·4	83·9	83·4	82·8	81·8	83·1	83·4	81·0	83·0	0·73
Faecal excretion of P (g/d)	1·38	1·22	1·29	1·21	1·15	1·61	1·48	1·46	1·25	1·27	1·45	1·23	
Bone characteristics§													
Metacarpal													
Shear force (N)	0·65	0·67	0·75	0·82	0·87	0·71	0·86	0·89	0·91	0·97	0·77	1·01	0·06
Shear stress (N/cm²)	1·15	1·10	1·17	1·30	1·42	1·15	1·27	1·31	1·45	1·43	1·30	1·45	0·11
Shear energy (N.mm)	1·33	1·61	2·32	2·31	2·67	1·96	2·30	2·28	2·61	2·79	2·25	2·42	0·21
Ash content (g/kg)	295	319	329	328	338	307	337	354	342	377	332	361	7
Tenth rib													
Shear force (N)	0·65	0·74	0·83	0·75	0·83	0·67	0·79	1·12	0·85	1·09	0·74	0·85	0·05
Shear stress (N/cm²)	3·30	3·39	3·24	3·05	3·55	2·92	3·32	3·59	3·00	3·73	3·86	3·37	0·37
Shear energy (N.mm)	0·60	0·68	0·81	0·78	0·88	0·65	0·84	1·14	0·85	1·19	0·72	1·09	0·06
Ash content (g/kg)	370	442	443	457	462	407	459	497	480	483	480	482	9

* For statistical analysis see Table 3, and for details of diets and procedures, see Table 1 and pp. 564–567.
† Four pens (two pigs per pen) per treatment mean. Average initial wt 7·8 kg.
‡ Eight pens (two pigs per pen for week 4 and 5) per treatment mean.
§ Four pigs per treatment mean.

Table 3. *Probability values of main effects, interactive effects, linear (Lin) and quadratic (Quad) effects, and contrast of pigs fed on a maize–soyabean-meal diet containing varying levels of available phosphorus (aP) and supplemental phytase* (EC *3.1.3.26; PY*)*

	PY	aP	PY × aP	Lin	Lin 1	Lin 2	Quad	Quad 1	Quad 2	Con 1	Con 2	Con 3
Performance for weeks 1–5												
ADG	0·01	0·01	0·58	0·01	0·01	0·01	0·08	0·42	0·02	0·05	0·01	0·46
ADFI	0·29	0·21	0·68	0·04	0·03	0·42	0·24	0·83	0·16	0·07	0·08	0·44
Wt gain:feed intake	0·09	0·28	0·87	0·03	0·40	0·03	0·13	0·36	0·84	0·74	0·08	0·49
Apparent digestibility coefficients												
DM	0·14	0·12	0·21	0·11	0·12	0·05	0·16	0·15	0·15	0·10	0·05	0·11
P	0·01	0·01	0·06	0·01	0·01	0·01	0·01	0·02	0·01	0·02	0·01	0·01
Ca	0·01	0·19	0·12	0·01	0·01	0·03	0·31	0·25	0·13	0·05	0·25	0·13
N	0·18	0·02	0·12	0·12	0·24	0·20	0·06	0·05	0·10	0·05	0·01	0·11
Faecal P excretion	0·01	0·07	0·01	0·01	0·02	0·01	0·01	0·01	0·01	0·01	0·06	0·10
Bone characteristics												
Metacarpal												
Shear force	0·01	0·01	0·87	0·01	0·01	0·01	0·64	0·93	0·47	0·02	0·04	0·72
Shear stress	0·04	0·15	0·88	0·01	0·03	0·03	0·68	0·31	0·67	0·35	0·32	0·56
Shear energy	0·01	0·09	0·38	0·01	0·01	0·01	0·64	0·51	0·99	0·55	0·01	0·45
Ash content	0·01	0·01	0·69	0·01	0·01	0·01	0·10	0·10	0·48	0·01	0·01	0·46
Tenth rib												
Shear force	0·01	0·01	0·01	0·01	0·01	0·01	0·01	0·31	0·01	0·07	0·05	0·57
Shear stress	0·18	0·95	0·71	0·22	0·85	0·12	0·64	0·44	0·92	0·47	0·44	0·30
Shear energy	0·01	0·01	0·05	0·01	0·01	0·01	0·23	0·72	0·19	0·01	0·02	0·84
Ash content	0·01	0·01	0·12	0·01	0·01	0·01	0·01	0·01	0·01	0·86	0·01	0·11

* For details of diets, see Table 1, and for data, see Table 2.

ADG, average daily wt gain; ADFI, average daily feed intake; Con 1, contrast for diets 11 *v.* 12 (3·2 g aP/kg, with 0 and 1400 U PY/kg respectively; Con 2, contrast for diets 1, 6 and 11 (0·7, 1·6 and 3·2 g aP/kg, without PY respectively), linear; Con 3, contrast for diet 1, 6 and 11, quadratic.

Table 4. *Second-order translog functions for performance, apparent digestibility coefficients and bone characteristics of young pigs fed on maize–soyabean-meal diets containing two levels of available phosphorus (aP; 0·7 and 1·6 g/kg) and five levels of phytase* (EC *3.1.3.26*)*; 0, 350, 700, 1050 and 1400 U/kg)**

	Coefficients of equations†					Statistical significance: P	r^2
	a_0	a_1	a_2	a_3	a_4		
Performance for weeks 1–5							
Average daily wt gain	5·2900	0·1280	0·0190	0·0070	0·0036	0·01	0·43
Average daily feed intake (kg)	−0·7480	0·0719	0·0120	0·0039	−0·0050	0·44	0·11
Wt gain:feed intake (g/kg)	6·0380	0·0560	0·0070	0·0030	0·0087	0·02	0·28
Apparent digestibility coefficients (%)							
DM	4·4640	−0·0052	−0·0003	0·0001	−0·0002	0·03	0·85
P	3·1320	0·1940	0·0511	0·0077	−0·0062	0·01	0·98
Ca	4·2320	−0·0098	0·0097	0·0016	−0·0023	0·01	0·94
N	4·4078	−0·0148	−0·0004	0·0004	0·0010	0·04	0·83
Faecal P excretion (g/d)	1·2800	−0·7500	−0·0198	−0·0049	−0·0035	0·01	0·99
Bone characteristics							
Fourth metacarpal							
Shear force (N)	−0·9650	0·1460	0·0370	0·0096	0·0010	0·02	0·26
Shear stress (N/cm²)	−0·5380	0·0820	0·0301	0·0090	0·0050	0·05	0·15
Shear energy (N.mm)	−0·5910	0·2660	0·0770	0·0170	−0·0190	0·01	0·31
Ash content (g/kg)	−1·3460	0·0460	0·0138	0·0032	0·0029	0·01	0·48
Tenth rib							
Shear force (N)	−0·7890	0·1630	0·0350	0·0067	0·0077	0·01	0·55
Shear stress (N/cm²)	1·0680	0·0465	0·0135	0·0007	0·0012	0·20	0·09
Shear energy (N.mm)	−0·9800	0·1500	0·0380	0·0100	0·0140	0·01	0·38
Ash content (g/kg)	−0·9500	0·0810	0·0156	0·0010	−0·0030	0·01	0·69

* For details of diets and procedures, see Table 1 and pp. 564–567.

† $\ln Y = a_0 + a_1 D_1 + a_2 \ln X + a_3(\ln X)^2 + a_4 D_1 \ln X$, where D_1 is aP (g/kg); when aP = 0·7 g/kg, $D_1 = 0$; when aP = 1·6 g/kg, $D_1 = 1$ and X is added phytase (U/kg diet).

Table 5. *Non-linear functions of supplemental phytase* (EC *3.1.3.26*) *for performance, apparent digestibility coefficients and bone characteristics of young pigs fed on maize–soyabean-meal diets containing two levels of available phosphorus* (*aP; 0·7 and 1·6 g/kg*) *and five levels of phytase* (*0, 350, 700, 1050 and 1400 U/kg*)*

	0·7 g aP/kg		1·6 g aP/kg	
	Equation	r^2	Equation	r^2
Performance for weeks 1–5				
Average daily wt gain (g)	$Y = 376{\cdot}71\,(1-0{\cdot}354e^{-0{\cdot}00095X})$	0·97	$Y = 383{\cdot}6\,(1-0{\cdot}304e^{-0{\cdot}00266X})$	0·91
Average daily feed intake	$Y = 0{\cdot}859\,(1-0{\cdot}387e^{-0{\cdot}0004X})$	0·99	$Y = 0{\cdot}648\,(1-0{\cdot}105e^{-0{\cdot}0045X})$	0·99
Wt gain:feed intake (g/kg)	$Y = 520\,(1-0{\cdot}107e^{-0{\cdot}0021X})$	0·88	$Y = 718\,(1-0{\cdot}344e^{-0{\cdot}0006X})$	0·94
Apparent digestibility coefficients (%)				
P	$Y = 58{\cdot}08\,(1-0{\cdot}51e^{-0{\cdot}00107X})$	0·97	$Y = 56{\cdot}0715\,(1-0{\cdot}373e^{-0{\cdot}00284X})$	0·99
Ca	$Y = 81{\cdot}508\,(1-0{\cdot}1122e^{-0{\cdot}0016X})$	0·95	$Y = 78{\cdot}0877\,(1-0{\cdot}068e^{-0{\cdot}00212X})$	0·88
Faecal P excretion (g/d)	$Y = 1{\cdot}3461e^{-0{\cdot}00011X}$	0·84	$Y = 1{\cdot}6028e^{-0{\cdot}00019X}$	
Bone characteristics				
Fourth metacarpal				
Shear force (N)	$Y = 2{\cdot}97\,(1-0{\cdot}794e^{-0{\cdot}00008X})$	0·99	$Y = 0{\cdot}95\,(1-0{\cdot}25e^{-0{\cdot}00188X})$	0·99
Shear stress (N/cm²)	$Y = 2{\cdot}10\,(1-0{\cdot}541e^{-0{\cdot}00013X})$	0·68	$Y = 1{\cdot}475\,(1-0{\cdot}309e^{-0{\cdot}0006X})$	0·95
Shear energy (N.mm)	$Y = 3{\cdot}575\,(1-0{\cdot}641e^{-0{\cdot}00067X})$	0·95	$Y = 6{\cdot}752\,(1-0{\cdot}7118e^{-0{\cdot}00009X})$	0·71
Ash content (g/kg)	$Y = 33{\cdot}06\,(1-0{\cdot}132e^{-0{\cdot}0025X})$	0·97	$Y = 39{\cdot}06\,(1-0{\cdot}219e^{-0{\cdot}0008X})$	0·83
Tenth rib				
Shear force (N)	$Y = 0{\cdot}837\,(1-0{\cdot}276e^{-0{\cdot}00197X})$	0·98	$Y = 1{\cdot}064\,(1-0{\cdot}403e^{-0{\cdot}00197X})$	0·65
Shear stress (N/cm²)	$Y = 3{\cdot}4345\,(1-0{\cdot}1506e^{-0{\cdot}00569X})$	0·39	$Y = 3{\cdot}435\,(1-0{\cdot}1506e^{-0{\cdot}00572X})$	0·40
Shear energy (N.mm)	$Y = 1{\cdot}076\,(1-0{\cdot}443e^{-0{\cdot}0006X})$	0·96	$Y = 1{\cdot}206\,(1-0{\cdot}464e^{-0{\cdot}0012X})$	0·67
Ash content (g/kg)	$Y = 45{\cdot}06\,(1-0{\cdot}1889e^{-0{\cdot}0045X})$	0·97	$Y = 48{\cdot}07\,(1-0{\cdot}166e^{-0{\cdot}0036X})$	0·93

X, added phytase, U/kg diet; *Y*, response obtained.

* For details of diets and procedures, see Table 1 and pp. 564–567.

Table 6. *Linear functions of available phosphorus (aP) for performance, apparent digestibility coefficients and bone characteristics of young pigs fed on maize–soyabean-meal diets containing two levels of aP (0·7 and 1·6 g/kg) and five levels of phytase* (EC *3.1.3.26; 0, 350, 700, 1050 and 1400 U/kg)**

	Equations	r^2	Statistical significance: *P*
Performance for weeks 1–5			
Average daily wt gain (g)	$Y = 212{\cdot}4 + 34{\cdot}5X$	0·99	0·04
Average daily feed intake (kg)	$Y = 0{\cdot}5097 + 0{\cdot}02709X$	0·99	0·11
Wt gain: feed intake (g/kg)	$Y = 366{\cdot}4 + 42{\cdot}78X$	0·91	0·16
Apparent digestibility coefficients (%)			
P	$Y = 17{\cdot}9323 + 12{\cdot}055X$	0·99	0·04
Ca	$Y = 71{\cdot}35 + 1{\cdot}007X$	0·99	0·02
Faecal P excretion (g/d)	$Y = 1{\cdot}4500 + 0{\cdot}013X$	0·12	0·77
Bone characteristics			
Fourth metacarpal			
Shear force (N)	$Y = 0{\cdot}6242 + 0{\cdot}0468X$	0·98	0·09
Shear stress (N/cm²)	$Y = 1{\cdot}0828 + 0{\cdot}0639X$	0·87	0·21
Shear energy (N.mm)	$Y = 1{\cdot}2119 + 0{\cdot}3463X$	0·87	0·26
Ash content (g/kg)	$Y = 28{\cdot}40 + 1{\cdot}490X$	0·99	0·01
Tenth rib			
Shear force (N)	$Y = 0{\cdot}6190 + 0{\cdot}0369X$	0·98	0·03
Shear stress (N/cm²)	$Y = 2{\cdot}8717 + 0{\cdot}26632X$	0·51	0·32
Shear energy (N.mm)	$Y = 0{\cdot}5696 + 0{\cdot}0475X$	0·99	0·01
Ash content (g/kg)	$Y = 33{\cdot}80 + 4{\cdot}4190X$	0·99	0·02

X, aP levels (g/kg); *Y*, response obtained.
* For details of diets and procedures, see Table 1 and pp. 564–567.

equations of apparent digestibility coefficients of P and Ca, and faecal P excretion indicated high correlation ($r^2 > 0{\cdot}9$) and low significance levels ($P < 0{\cdot}01$). Equations for ADG, apparent digestibility of DM and N, ash content of the fourth metacarpal, and shear force and ash content of the tenth rib had r^2 values ranging from 0·43 to 0·85 ($P < 0{\cdot}04$); whereas, r^2 values for the remainder of the response equations were low ($< 0{\cdot}38$) with significance levels of $P > 0{\cdot}10$.

The non-linear response equations of graded levels of phytase at each of the two aP levels were developed for performance, apparent digestibility coefficients, and bone characteristics (Table 5). High r^2 values ($> 0{\cdot}8$) were obtained for all response equations except shear force and shear stress and energy for both the metacarpal and tenth rib. The r^2 values for the linear equations generated for the effects of only aP on ADG, Ca and P digestibility, shear force and ash content for both the metacarpal and tenth rib, and shear energy for the tenth rib were 0·95 or greater (Table 6). Other measurements had lower r^2 values.

Replacement of available phosphorus by phytase. To evaluate microbial phytase efficacy, P-equivalency equations were developed using non-linear and linear functions of phytase and aP (Table 7). Equivalency equations with high r^2 values ($> 0{\cdot}9$) were found for ADG, ADFI, apparent digestibility of P, shear force for the metacarpal and ash content for the tenth rib. The equations for ADG and apparent digestibility of P were used for determining P-equivalency values of phytase shown in Table 8. Both these measurements are non-destructive and practical to use. Averaged across the two aP levels, added microbial phytase of 250, 500, 750 and 1000 U/kg diet could release 26·7, 52·0, 75·1 and 91·1% of phytate-P in the diet. The amount of P released per 100 U phytase decreased as the total

Table 7. *Replacement functions of available phosphorus (aP) by microbial phytase* (EC *3.1.3.26*) *for young pigs fed on maize–soyabean-meal diets containing two levels of aP (0·7 and 1·6 g/kg) and five levels of phytase (0, 350, 700, 1050 and 1400 U/kg)**

	0·7 g aP/kg		1·6 g aP/kg	
	Equation†	r^2	Equation†	r^2
Performance for weeks 1–5				
Average daily wt gain	$Y = 4{\cdot}762 - 3{\cdot}865e^{-0{\cdot}00095X}$	0·96	$Y = 4{\cdot}962 - 3{\cdot}380e^{-0{\cdot}00266X}$	0·90
Average daily feed intake	$Y = 12{\cdot}894 - 12{\cdot}271e^{-0{\cdot}004X}$	0·99	$Y = 5{\cdot}105 - 2{\cdot}512e^{-0{\cdot}0045X}$	0·99
Wt gain:feed intake	$Y = 3{\cdot}599 - 1{\cdot}304e^{-0{\cdot}0021X}$	0·80	$Y = 8{\cdot}238 - 5{\cdot}787e^{-0{\cdot}00063X}$	0·86
Apparent digestibility coefficients				
P	$Y = 3{\cdot}331 - 2{\cdot}965e^{-0{\cdot}00108X}$	0·96	$Y = 3{\cdot}164 - 1{\cdot}735e^{-0{\cdot}00284X}$	0·98
Ca	$Y = 10{\cdot}087 - 9{\cdot}082e^{-0{\cdot}0016X}$	0·95	$Y = 6{\cdot}691 - 5{\cdot}273e^{-0{\cdot}00212X}$	0·87
Bone characteristics				
Fourth metacarpal				
Shear force	$Y = 50{\cdot}145 - 50{\cdot}410e^{-0{\cdot}0008X}$	0·99	$Y = 6{\cdot}965 - 5{\cdot}077e^{-0{\cdot}00188X}$	0·97
Shear stress	$Y = 15{\cdot}911 - 17{\cdot}771e^{-0{\cdot}00013X}$	0·59	$Y = 6{\cdot}135 - 7{\cdot}129e^{-0{\cdot}0006X}$	0·83
Shear energy	$Y = 6{\cdot}825 - 6{\cdot}618e^{-0{\cdot}00067X}$	0·83	$Y = 16{\cdot}000 - 13{\cdot}880e^{-0{\cdot}00009X}$	0·62
Ash content	$Y = 3{\cdot}128 - 2{\cdot}929e^{-0{\cdot}0025X}$	0·97	$Y = 7{\cdot}154 - 5{\cdot}741e^{-0{\cdot}0008X}$	0·83
Tenth rib				
Shear force	$Y = 5{\cdot}908 - 6{\cdot}260e^{-0{\cdot}00197X}$	0·96	$Y = 12{\cdot}060 - 11{\cdot}620e^{-0{\cdot}00197X}$	0·64
Shear stress	$Y = 2{\cdot}113 - 1{\cdot}942e^{-0{\cdot}00569X}$	0·20	$Y = 2{\cdot}115 - 1{\cdot}942e^{-0{\cdot}0057X}$	0·20
Shear energy	$Y = 10{\cdot}659 - 10{\cdot}033e^{-0{\cdot}00061X}$	0·96	$Y = 13{\cdot}395 - 11{\cdot}778e^{-0{\cdot}00123X}$	0·67
Ash content	$Y = 2{\cdot}548 - 1{\cdot}926e^{-0{\cdot}0045X}$	0·97	$Y = 3{\cdot}229 - 1{\cdot}806e^{-0{\cdot}0036X}$	0·93

X, added phytase, (U/kg diet); Y, replaced aP (g/kg).
* For details of diets and procedures, see Table 1 and pp. 564–567.
† Created using non-linear and linear functions of supplemental phytase and inorganic P in Tables 5 and 6.

Table 8. *Phosphorus equivalency of inorganic P by microbial phytase*, (EC *3.1.3.26*) *for young pigs fed on maize–soyabean-meal diets containing varying levels of available P* (*aP*) *and phytase**

aP (g/kg)...	0·7				1·6			
Added phytase (U/kg diet)...	250	500	750	1000	250	500	750	1000
Equivalent of aP (g/kg)								
Average daily wt gain	1·71	2·36	2·87	3·26	3·22	4·07	4·50	4·73
Apparent P digestibility coefficient	1·45	1·89	2·23	2·49	2·31	2·74	2·96	3·06
Metacarpal ash content	1·56	2·29	2·68	2·89	2·45	3·31	4·00	4·57
Rib ash content	1·92	2·35	2·48	2·53	2·49	2·93	3·11	3·18
Mean equivalent†								
aP (g/kg)	1·58	2·12	2·55	2·85	2·76	3·40	3·73	3·89
Released P (g/kg)‡	0·88	1·42	1·85	2·15	1·16	1·80	2·13	2·29
% of phytate P§	36·0	58·4	75·8	88·3	47·7	73·9	87·2	94·0
Mean equivalents of the two aP levels								
Released P (g/kg)‖					1·02	1·61	1·99	2·22
% of phytate-P					41·9	66·2	81·5	91·1
Released P (g/100 U phytase)					0·40	0·32	0·27	0·22

† Data of average daily wt gain and apparent P digestibility coefficients were used to calculate the mean equivalent.

‡ Released P = equivalent of aP − 0·7 (or 1·6).

§ Plant P in diets 3·56 g/kg, phytate-P 2·44 g/kg and non-phytate P 1·10 g/kg; released phytate-P = (released P/phytate-P) × 100.

‖ The function of released P (Y) *v.* microbial phytase (X) was created using mean released P for pigs fed on diets containing aP levels from 0·7 to 1·6 g/kg; $Y = 2{\cdot}622 - 2{\cdot}559e^{-0{\cdot}00185X}$.

level of phytase increased. Using the P-equivalency equation developed for the average of two aP levels and two response measurements (ADG and P absorption), $Y = 2{\cdot}622 - 2{\cdot}559e^{-0{\cdot}00185X}$ (Table 8), 246 U microbial phytase would equal 1 g inorganic P added as defluorinated phosphate.

DISCUSSION

The present study indicates that supplemental microbial phytase is effective in improving performance, P digestibility and bone mineralization by enhancing hydrolysis of phytate-P for young pigs fed on diets of maize–soyabean meal, which has been reported also by other researchers (Simons *et al.* 1990; Cromwell *et al.* 1993*b*; Hoppe *et al.* 1993). The addition of 1050 U phytase/kg diet at 0·7 or 1·6 g aP/kg level increased ADG by 38·5 and 39·5%, ADFI by 22 and 14·3%, and G:F by 14·3 and 24% respectively (Table 2). ADG was found to be a good indicator of the response to graded supplemental levels of phytase and aP. These findings were consistent with other studies in pigs (Cromwell *et al.* 1993*b*; Lei *et al.* 1993*a*; Yi *et al.* 1994).

Recently, much attention has been directed to P pollution from manure. Microbial phytase has been shown to be effective in reducing P excretion by increasing the utilization of phytate-P (Simons *et al.* 1990; Jongbloed *et al.* 1992; Yi *et al.* 1994). The data from the present study further supported these findings. Adding 1050 U phytase/kg diet decreased faecal P excretion by 12 and 22% respectively for 0·7 and 1·6 g aP/kg diets, which resulted from increases in apparent P absorption by 72 and 59% respectively.

The addition of 1400 U phytase to diets containing a recommended level of 3·2 g aP/kg further increased ADG, ADFI, apparent absorption of P, Ca and N, and bone strength and ash contents, which suggest that microbial phytase could be beneficial in the utilization of macro and trace minerals, protein and amino acids. Similar results were also found in young pigs fed on a soyabean-meal-based semi-purified diet (Yi *et al.* 1994) or rapeseed–grain-sorghum basal diets (Veum *et al.* 1994). Supporting evidence for improved utilization of minerals and N has been shown also in the studies of Pallauf *et al.* (1992, 1994), Lei *et al.* (1993*b*), and Mroz *et al.* (1994). Ketaren *et al.* (1993) found that the addition of phytase increased ADG, protein deposition and retention, and energy retention, with no effects on the apparent digestibility of DM and CP. Mroz *et al.* (1994) reported that phytase supplementation enhanced the utilization of DM, OM, CP, amino acids and Ca. Further research, however, is needed to assess the effect of microbial phytase on the bioavailability of Ca, trace minerals and amino acids. The effect of phytase on feed intake needs to be considered. The increased feed intake could reduce the apparent absorption of some nutrients such as OM, N and minerals, and mask the effect of phytase on their responses.

Generally, the response curves for performance, apparent P absorption, and bone measurements indicated the breaking points were at phytase levels of 700–1050 U/kg diet for pigs fed on maize–soyabean-meal diets with 0·7 and 1·6 g aP/kg, which suggested the maximum responses occurred at 700 or 1050 U phytase respectively at 0·7 or 1·6 g aP/kg levels. These results agreed with other phytase studies. The study of Yi *et al.* (1994) achieved the same conclusion using the same semi-purified diets of maize–soyabean meal. Lei *et al.* (1993*b*) found that the stationary point of added Finase® phytase was at approximately 1200 U/kg diet in weanling pigs fed on maize–soyabean-meal diet with 0·5 g aP/kg. For growing pigs, the maximum response of graded Natuphos® phytase ranged between 800 and 1200 U/kg diet (Veum *et al.* 1994). Thus the maximum effect of phytase on pigs appeared to occur at the supplemental level within the range 700 and 1200 U/kg diet. When compared with the 3·2 g aP/kg diet (National Research Council (1988) recommended level), the maximum responses in performance, digestibilities and bone measurements of pigs receiving phytase supplementation in 0·7 or 1·6 g aP/kg diets were found close to, or even better than, with defluorinated phosphate addition.

Evaluation of non-linear and linear functions for the various performance, digestibility and bone measurements suggested that ADG, apparent P digestibility and shear force and ash content of bone were sensitive indicators, which was consistent with other studies (Cromwell *et al.* 1993*a*, *b*; Yi *et al.* 1994). Second-order translog functions for these indicators gave similar results with relatively high R^2 and low level of significance (Table 4). The apparent P digestibility was considered the best indicator to determine the effectiveness of feed phosphates and phytase (Dellaert *et al.* 1990; Qian *et al.* 1996). The data for apparent P digestibility in the present study further confirm this finding (Tables 4, 5 and 6). In addition, faecal P excretion also appeared highly sensitive to dietary supplemental phytase and P. In comparison with the 3·2 g aP/kg diet (National Research Council (1988) recommended level), faecal P excretion was decreased by up to 25% by adding phytase in young pigs fed on maize–soyabean-meal diets, which was consistent with other findings (Jongbloed *et al.* 1992; Cromwell *et al.* 1993*a*; Yi *et al.* 1994).

Bone indicators are often used to evaluate P and Ca status in pigs (Koch & Mahan, 1985; National Research Council, 1988; Combs *et al.* 1991). Generally, the metacarpal, metatarsal and femur of pigs were used. In this experiment, shear force and ash content of the metacarpal and tenth rib were sensitive. Shear energy is the energy required to deform a bone to the point of fracture, which represents the area under the force *v.* deformation curve up to the point of fracture. Bone shear energy was less sensitive after shear force and ash content. In the present study, both metacarpal and tenth rib shear stress showed low sensitivity.

Using the P-equivalency function shown in Table 8, the replacement of 1 g inorganic P from defluorinated phosphate would require about 246 U phytase. The P-equivalency values are lower than those calculated from published data using the same type of diets, or in a soyabean-meal semi-purified diets for pigs (Yi *et al.* 1994). Hoppe *et al.* (1993) reported 1 g P from monocalcium phosphate (MCP) was equivalent to 380 U phytase when based on P retention and 403 U phytase when based on phalanx crude ash, when pigs were fed on a maize, oat and soyabean-meal-based diet. In a review paper, Hoppe & Schwarz (1993) concluded that for diets based mainly on maize and soyabean meal, 500 U phytase was equivalent to 1 g P from MCP (0·8 g digestible P).

Using the P-equivalency equation generated in the present study and based on additions of 250, 500, 750 and 1000 U phytase/kg diet, 42, 66, 82 and 91% of the phytate-P in maize–soyabean meal was released. In a similar study using a soyabean-meal-based diet, Yi *et al.* (1994) reported 40, 64, 83 and 93% of the phytate-P was released, respectively, based on 250, 500, 750 and 1000 U phytase/kg diet. Cromwell *et al.* (1993*b*) observed that adding 250, 500 and 1000 U phytase/kg diet would release 14, 22 and 43% of the phytate-P in a soyabean-meal-based semi-purified diet fed to growing pigs, in terms of the measurements of bone shear force. Hoppe *et al.* (1993) in a study using pigs fed on a grain and soyabean-meal diet reported that adding 250, 500 and 1000 U phytase/kg diet released 15, 42 and 57% of the phytate-P, based on bone ash content, and P and Ca retention. Our results are supported also by the observation of Jongbloed *et al.* (1992) who reported that the ileal digestibility of phytic acid increased from 9 to 59% by adding 1500 U phytase/kg diet to a maize and soyabean-meal diet fed to growing pigs.

Increasing the Ca:tP ratio lowers P absorption, which results in decreased growth and bone calcification (Koch *et al.* 1984; Reinhart & Mahan, 1986; Pointillart *et al.* 1989). The high Ca:tP value (2:1) used in the present experiment probably reduced the overall response to phytase and inorganic P. Detrimental effects of Ca:tP ratio on performance, bone characteristics and serum variables were generally observed when the Ca:tP ratio exceeded 2·0:1; however, no significant effects were observed when the Ca:tP ratio was under 2·0:1, especially at the range 1·0–1·6:1 (Koch *et al.* 1984; Reinhart & Mahan, 1986; Pointillart *et al.* 1989; Ketaren *et al.* 1993). In some studies relating to Ca:tP ratio, performance, bone measurements and serum variables were not influenced by the Ca:tP, ratio, even at a larger range than 2·0:1, such as 3:1 used by Koch *et al.* (1984). The adverse effects of a high Ca:tP ratio may have reduced the efficacy of supplemental phytase in the present study when the dietary Ca:tP ratio increased from 1·2 to 2·0:1. Based on the study of Qian *et al.* (1995), the response of most measurements in young pigs was reduced more than 10% when Ca:tP ratios were increased from 1·2:1 to 2·0:1. The influence that Ca:tP ratio may have had was not taken into account in most recent phytase studies (Nasi, 1990; Jongbloed *et al.* 1992; Lei *et al.* 1994). In most studies, Ca:tP ratios ranged from 1·7 to 2·5:1. The effect of phytase on Ca release did not appear to have been given much attention.

In summary, microbial phytase is effective in improving performance, P utilization and bone mineralization, and decreasing faecal P excretion of pigs by enhancing the bioavailability of phytate-P in maize and soyabean meal. One gram of inorganic P as defluorinated phosphate could be released by 246 U microbial phytase. In addition, phytase also appeared effective in improving the utilization of other nutrients such as Ca, N, and trace minerals.

Appreciation is expressed to the United Soybean Board and the John Lee Pratt Animal Nutrition Program for financial support, to Donald Conner Jr for lab technical assistance, to Zhixong Yi for help with data collection and calculation of P-equivalency, to Rob French for help with data collection, to Dave Notter for advice with statistical treatments of data, and to Cindy Hixon for manuscript preparation. This material is based on work supported in part by the Cooperative State Research Service, USDA, under projection no. 6129880.

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Printed in Great Britain