Body weight gain and nutrient utilization in starter pigs that are liquid-fed high-moisture corn-based diets supplemented with phytase

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Received 13 July 2009, accepted 22 October 2009.

Columbus, D., Zhu, C. L., Pluske, J. R. and de Lange, C. F. M. 2010. **Body weight gain and nutrient utilization in starter pigs that are liquid-fed high-moisture corn-based diets supplemented with phytase.** Can. J. Anim. Sci. **90:** 45–55. A total of 384 starter pigs were used to examine the application of exogenous phytase in high-moisture corn (HMC)-based liquid diets. Pigs were randomly assigned to 24 pens in six blocks. Pens were randomly assigned one of four HMC-based diets formulated to vary in total phosphorus (P) content (Low, Medium and High), with phytase added to only the Low P diet (Phy). Body weight gain and feed intake were monitored until body weight exceeded 20 kg. Apparent total tract digestibility of crude protein and P were measured on day 21 (Phase II) and day 42 (Phase III). At the end of the trial, two pigs from each pen were sacrificed for analysis of carcass composition and evaluation of metacarpals. Pigs fed the Phy treatment had increased digestibility of crude protein ($P < 0.05$) and P ($P = 0.062$) in Phase III, and increased metacarpal breaking strength ($P < 0.01$) and P content ($P < 0.05$). Average daily gain, feed intake, and carcass composition were not affected by treatment ($P > 0.05$). In conclusion, performance of starter pigs fed liquid HMC-based diets was maintained at dietary P levels below established requirements, but addition of phytase improved bone strength and mineralization. This study provides evidence for the effectiveness of phytase, and that P requirement for maximum rate of weight gain in pigs is not sufficient for maximum skeletal development.

Key words: High-moisture corn, liquid feed, phosphorus, phytase, starter pigs

Columbus, D., Zhu, C. L., Pluske, J. R. et de Lange, C. F. M. 2010. **Gain de poids et assimilation des éléments nutritifs par les porcs de démarrage nourris avec une ration liquide de maïs à haute teneur en eau enrichie de phytase**. Can. J. Anim. Sci. **90**: 45–55. Les auteurs ont recouru à 384 porcs de démarrage pour étudier l'utilité de la phytase exogène dans une ration liquide à base de maïs à haute teneur en eau (MHTE). Les animaux ont été répartis au hasard entre 24 enclos, en six blocs. Quatre rations de MHTE ont ensuite été attribuées au hasard aux enclos. Ces rations avaient été préparées pour faire varier la concentration de phosphore (P) (faible, moyenne et élevée) et la phytase n'a été ajoutée qu'à la ration contenant le moins de P (Phy). Les auteurs ont surveillé le poids corporel et l'ingestion d'aliments jusqu'à ce que le poids des animaux dépasse 20 kg. Ils ont mesuré la digestibilité apparente totale des protéines brutes et du P dans le tube digestif le 21^{e} jour (Phase II) et le 42^{e} jour (Phase III). À la fin de l'expérience, deux sujets de chaque enclos ont été sacrifiés et on a analysé la composition de leur carcasse ainsi qu'évalué leurs métacarpiens. Les porcs recevant la ration Phy assimilaient mieux les protéines brutes ($P < 0{,}05$) et le P ($P = 0{,}062$) à la Phase III, et leurs métacarpiens étaient plus solides ($P < 0{,}01$), tout en contenant plus de P ($P < 0{,}05$). Le traitement n'affecte pas le gain quotidien moyen, l'ingestion des aliments ni la composition de la carcasse ($P > 0{,}05$). En conclusion, les porcs de démarrage nourri avec une ration liquide à base de MHTE maintiennent leur performance même si la concentration de P dans les aliments est inférieure à celle requise, mais l'addition de phytase accroît la solidité et la minéralisation des os. L'étude prouve l'efficacité de la phytase et le fait que les besoins en P associés au gain de poids maximal chez le porc ne suffisent pas pour un développement optimal du squelette.

Mots clés: Maïs à haute teneur en eau, aliment liquide, phosphore, phytase, porcs de démarrage

Most of the phosphorus content (P) in grains is bound in the form of phytate, ranging from 50% in wheat to 85% in corn [National Research Council (NRC) 1998], which reduces the availability of P to pigs. To address concerns about the environmental impact of excreting P with pig manure, feeding strategies should be developed to improve P availability and utilization by pigs. The addition of supplemental phytase to conventional dry diets has been shown to improve P digestibility and utilization by pigs. However, apparent total tract P digestibility rarely exceeds 55% in corn- and soybean-meal-based diets that are supplemented with phytase

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Abbreviations: **ADG**, average daily gain; **BW**, body weight; **CP**, crude protein; **DE**, digestible energy; **DM**, dry matter; **FTU**, phytase units; **HMC**, high-moisture corn; **SID**, standardized ileal digestible

(Kornegay and Verstegen 2001). The effectiveness of exogenous phytase may be enhanced by steeping ingredients with phytase prior to feeding. To our knowledge, no previous trials have examined the impact of phytase supplementation in liquid corn- and soybean meal-based diets on weight gain and nutrient digestibility in pigs.

With the development of liquid feeding systems, high-moisture corn (HMC) has become a more common feed ingredient in swine production. According to Niven et al. (2007), freshly harvested HMC has a total P content of approximately 2.88 g kg^{-1} dry matter (DM) and a soluble P content of approximately 0.27 g kg^{-1} DM. During storage, however, the soluble P content in HMC can increase to 44% of total P. Moreover, it was observed that steeping HMC in water for 24 h with 500, 750, or 1000 phytase units (FTU) kg^{-1} DM released nearly all phytate-P at 37°C and approximately 85% of phytate-P at 21°C (Niven et al. 2007). It has also been shown that soaking of a dry diet in water improves phytase activity (Skoglund et al. 1997; Carlson and Poulsen 2003).

The objective of this trial was to determine the effects of dietary P level and supplemental phytase in a low-P, HMC-based diet on body weight (BW) gain, feed intake, feed efficiency, nutrient digestibility, carcass nutrient composition, and bone characteristics in liquid-fed starter pigs. It was hypothesized that supplemental fungal phytase would increase P availability in the HMC-based diets compared with HMC-based diets devoid of additional phytase.

MATERIALS AND METHODS

Animals, Housing, Diets and General Conduct of Study

A total of 384 purebred Yorkshire pigs were weaned at 19 to 23 d of age at a BW of 5.8 ± 0.45 kg. They were housed in 24 wean-to-finish pens (396×198 cm) of 16 pigs each (eight barrows and eight gilts, balanced for initial BW and litter mates across treatments) at the University of Guelph's Arkell Swine Research Station (Guelph, ON, Canada) and were cared for according to the guidelines of the Animal Care Committee at the University of Guelph and Canadian Council on Animal Care (1993). The experimental protocol was reviewed and approved by the University's Animal Care Committee. Pens within blocks were randomly assigned one of four HMC-based dietary treatments for a total of six pens per treatment. Pigs were put on trial in six blocks, spaced 1 wk apart, with three (one block), four (four blocks), or five (one block) pens each. In one block, only three dietary treatments were included due to pig availability. This treatment was included in the subsequent block. Pigs were kept on the experimental treatments until the average block BW exceeded 20 kg, which was a minimum of 6 wk.

Across three treatments, diets were formulated to vary in P contents (High, Medium, and Low) and the fourth treatment (Phy) represented the Low treatment supplemented with 500 FTU kg^{-1} of HMC on a DM basis. A typical HMC and soybean meal-based, three-phase feeding program was designed and a gradual transition between phases occurred over a 3-d period from days 7 to 9 and 21 to 23 after weaning. Within each phase, diets were formulated to meet or exceed NRC (1998) nutrient requirements for starter pigs and to be similar in DM, digestible energy (DE), and nutrient content, except for P and calcium (Ca) contents. Diets were formulated on an available P basis to exceed NRC (1998) requirements by 10% (High) and to be deficient by 30% (Low). Diets did not contain growth-promoting feed additives. Diets were formulated using published nutrient contents of ingredients (NRC 1998), except for total P content in stored HMC, which was determined previously to be 0.25% (88% DM basis; Braun and de Lange 2004). Available P content in HMC with or without addition of phytase was estimated to be 96 or 27% of total P, respectively. These values were based on soluble P content of corn, as an indicator of available P content, measured in an in vitro trial (Niven et al. 2007). All diets were formulated to contain 0.10% titanium dioxide (Sigma-Aldrich Corporation, St. Louis, MO) as an indigestible marker for calculation of apparent total tract nutrient digestibility. The dry feed supplements for the High and Low treatments (Table 1), which contained all dietary components except HMC and phytase, were mixed, pelleted, and crumbled at the University of Guelph's Arkell Feed Mill (Guelph, ON). The HMC was stored in a sealed silo at the Arkell Feed Mill until being ground (4.8 mm screen) and transferred in batches of 1000 kg, mixed with 1500 kg water, and stored in fermentation tanks for at least 24 h before feeding. For the Phy treatment, HMC was steeped with 500 FTU kg^{-1} DM of phytase [Ronozyme P (CT) 2500; DSM Nutritional Products Canada Inc., Ayr, ON] for a minimum of 24 h prior to feeding. One fermentation tank was dedicated to the preparation and feeding of HMC steeped with phytase. During storage, the HMC and water mixtures were agitated for 10 min every hour to keep the HMC from settling in the tanks and to facilitate action of the phytase. The maximum storage time for the HMC and water mixture was 1 wk, after which any remaining HMC was discarded and a new batch received.

The complete liquid diets were automatically prepared for each pen at each feeding time using the Big Dutchman HydroJet^{TM} liquid feeding system (Big Dutchman Int., Vechta, Germany) by mixing dry supplements (Table 1), HMC, and water to achieve a DM to water ratio of 1:2.5. The High, Medium, and Low dietary treatments were achieved by mixing the appropriate feed supplements with HMC without added phytase. The Medium diets were achieved by mixing a 50:50 blend of the High and Low diets. All Phase II diets were achieved by mixing a 50:50 blend of the appropriate Phase I and Phase III diets. For the Phy

Table 1. Ingredient composition (as-fed basis) and calculated nutrient content (adjusted to 88% DM basis) of starter pig Phase I and III diet supplements formulated to vary in total dietary P content (High vs. Low)

	Phase I		Phase III	
Ingredient (%)	High	Low	High	Low
Wheat	16.35	16.77	21.59	21.93
Whey, dried[z]	33.09	33.52	–	–
Fat blend, animal and vegetable[z]	3.31	3.35	4.32	4.30
Fishmeal, herring[z]	6.62	6.70	–	–
Blood plasma[z]	6.62	6.70	–	–
Soybean meal, dehulled	28.31	28.49	66.26	67.07
Lysine, HCl	0.41	0.40	0.41	0.39
Methionine	0.25	0.25	0.17	0.17
Threonine	0.17	0.17	0.17	0.17
Tryptophan	0.05	0.05	–	–
Limestone	2.13	2.09	2.22	2.25
Dicalcium phosphate	1.19	–	2.91	1.75
Salt	0.50	0.50	0.65	0.66
Vitamin/mineral premix[y]	0.83	0.84	1.08	1.09
Titanium dioxide	0.17	0.17	0.22	0.22
Calculated nutrient content[x]				
DE (MJ kg^{-1})	14.1	14.3	14.5	14.7
CP (%)	28.5	28.5	33.6	34.0
Ca (%)	1.51	1.25	1.68	1.45
Total P (%)	0.85	0.65	1.04	0.84
Available P (%)	0.65	0.45	0.65	0.45
Total lysine (%)	2.23	2.24	2.34	2.35
SID[w] lysine (%)	2.02	2.03	2.13	2.14
SID methionine (%)	0.62	0.62	0.60	0.60
SID methionine+cysteine *(%)	1.06	1.07	1.05	1.06
SID threonine (%)	1.25	1.27	1.27	1.28
SID tryptophan (%)	0.45	0.46	0.47	0.48
SID isoleucine (%)	1.07	1.08	1.32	1.34

[z]Whey, fat, fishmeal, and blood plasma obtained from Saputo (11982409), Kenpal Farm Products, Inc. (Lacta-Fat), Swimco (CO1090F), and American Protein Corporation (AP920), respectively.
[y]Premix contains per kg: vitamin A, 2.0 MU; vitamin D3, 0.2 MU; vitamin E, 8000 IU; vitamin K, 0.5 g; thiamine, 0.3 g; vitamin K, 0.5 g; pantothenic acid, 3.0 g; riboflavin, 1.0 g; folic acid, 0.4 g; niacin, 5.0 g; pyridoxine, 0.3 g; vitamin B_{12}, 5.0 mg; biotin, 40 mg; choline, 100.0 g; Cu, 3.0 g; I, 0.1 g; Fe, 20.0 g; Mn, 4.0 g; Zn, 21.0 g; and Se, 60.0 mg.
[x]Nutrient content of diet supplements were estimated based on nutrient content of feed ingredients according to NRC (1998).
[w]Standardized ileal digestible.

treatment, the low P supplement was mixed with HMC steeped with phytase. The ingredient composition of the complete diets is given in Table 2. All diet components were mixed and agitated for 2 min in the liquid feeding system central mixing tank just prior to feeding. The feed system was programmed to rinse the feed mixing tank and feed lines with water between diet treatments to prevent cross contamination. This rinse water was discarded into the manure pit.

Feed was offered six times per day in equal meals starting at 0600 and at three hour intervals. Pigs were fed on a pen basis and feed was delivered to liquid feeding troughs with 16 cm of feeding space per pig. Delivery to individual pens was controlled by sensors located 10 mm from the bottom of the troughs that detected leftover feed from the previous meal. If any feed remained in the trough, the subsequent feeding was skipped. Daily feed allowance was adjusted automatically by the feeding system computer based on a reference feed intake curve (NRC 1998). However, feed allowance for each pen was manually increased or decreased relative to the reference feed intake curve in an attempt to maximize feed intake and to ensure that fresh feed was delivered to each pen at least four times per day. Pigs were given free access to water from bowl drinkers throughout the trial.

Observations and Sampling

Pigs were individually weighed each week for a minimum of 6 wk and until the average pen BW exceeded 20 kg. Per-pen feed usage was monitored at weekly intervals using the computerized liquid feeding system. The amount of feed weighed into the central mixing tank and delivered to the pens was monitored via load cells at the point of mixing and feed sensors in the troughs. Feeding system accuracy has been assessed in a number of previous trials (de Lange et al. 2006). All data were recorded by the liquid feeding system computer. Pig health was monitored daily and any health issues were recorded and pigs treated according to Arkell Swine Research Station standard operating procedures. Briefly, pigs were treated with Depocillin (procaine penicillin; 300 000 IU mL^{-1}; Intervet Canada Ltd., Whitby, ON) if not thrifty, or Excenel^{TM} RTU (ceftiofur; 50 mg mL^{-1}; Pfizer Animal Health, Kirkland, QC) if lame, for a maximum of 3 d, as per veterinary recommendations. Pigs were removed from the trial if they had not recovered after the treatment period.

Complete diet and faecal samples were obtained during weeks 3 and 6 (or 7, if the block of pigs had not reached an average BW of 20 kg by the end of week 6) after weaning for evaluation of accuracy of feed mixing and determination of nutrient digestibility. Meals representing the actual diet composition on these days were mixed by the feeding system and sent to a collecting tank at a valve dedicated to feed sampling where two representative samples were taken. A minimum of six samples was obtained for each dietary treatment. Random samples of uncontaminated faecal material that had accumulated over a 2-d period were collected from at least four locations in the slatted area of each pen and pooled. Diet and faecal samples were kept at –20°C until subsequent nutrient analyses.

At the end of the trial period, two pigs from each pen (one barrow and one gilt) with BW closest to the average pen BW were euthanized by injection of Euthansol (sodium pentobarbital; 340 mg mL^{-1}; Schering-Plough Animal Health, Pointe Claire, QC) via the intraorbital sinus. Immediately after euthanasia, all internal organs of each pig were removed and the digestive tract was emptied of all contents. The empty carcass and viscera were weighed and frozen. The front feet were removed at the joint immediately proximal to the metacarpals and frozen individually in plastic bags.

Table 2. Component composition (as-fed basis) and calculated nutrient content (adjusted to 88% DM basis) of Phase I, II, and III high-moisture corn (HMC) based starter pig diets formulated to vary in total dietary P content (High, Medium, and Low) or to have low P content with added phytase (Phy)

	Diets – Phase I[z]				Diets – Phase II[z]				Diets – Phase III[z]			
	High	Medium	Low	Phy	High	Medium	Low	Phy	High	Medium	Low	Phy
Supplement (%)												
Phase I High	60.40	30.20	–	–	26.69	13.26	–	–	–	–	–	–
Phase I Low	–	30.20	59.60	59.60	–	13.26	26.36	26.36	–	–	–	–
Phase III High	–	–	–	–	26.69	13.26	–	–	46.30	23.20	–	–
Phase III Low	–	–	–	–	–	13.26	26.36	26.36	–	23.20	45.80	45.80
HMC (%)												
Without phytase	39.60	39.60	40.40	–	46.62	46.96	47.28	–	53.70	53.60	54.20	–
With phytase[y]	–	–	–	40.40	–	–	–	47.28	–	–	–	54.20
Calculated nutrient content[x]												
DE (MJ kg^{-1})	14.4	14.5	14.5	14.5	14.5	14.6	14.6	14.6	14.6	14.7	14.7	14.7
CP (%)	20.5	20.6	20.6	20.6	20.4	20.4	20.4	20.4	20.2	20.2	20.2	20.2
Ca (%)	0.94	0.86	0.78	0.78	0.88	0.81	0.74	0.74	0.81	0.75	0.69	0.69
Total P (%)	0.62	0.56	0.49	0.49	0.62	0.57	0.51	0.51	0.62	0.57	0.52	0.52
Ca:P	1.53	1.56	1.58	1.58	1.42	1.44	1.45	1.45	1.30	1.31	1.32	1.32
Available P (%)[w]	0.43	0.37	0.30	0.37	0.39	0.34	0.28	0.31	0.34	0.30	0.25	0.25
Sodium (%)	0.45	0.45	0.45	0.45	0.30	0.30	0.30	0.30	0.14	0.14	0.14	0.14
Total lysine (%)	1.47	1.47	1.47	1.47	1.36	1.36	1.35	1.35	1.24	1.24	1.23	1.23
SID[v] lysine (%)	1.32	1.32	1.32	1.32	1.22	1.22	1.21	1.21	1.11	1.11	1.10	1.10
SID methionine (%)	0.44	0.44	0.44	0.44	0.40	0.40	0.40	0.40	0.36	0.36	0.36	0.36
SID methionine+cysteine (%)	0.77	0.77	0.77	0.77	0.71	0.72	0.72	0.72	0.65	0.66	0.66	0.66
SID threonine (%)	0.86	0.86	0.86	0.86	0.79	0.79	0.79	0.79	0.72	0.72	0.72	0.72
SID tryptophan (%)	0.30	0.30	0.30	0.30	0.28	0.28	0.28	0.28	0.25	0.25	0.25	0.25
SID isoleucine (%)	0.75	0.75	0.75	0.75	0.75	0.75	0.75	0.75	0.75	0.75	0.75	0.75

[z]Pigs were placed on a three-phase feeding program with diet transitions occurring from days 7 to 9 and days 21 to 23 post-weaning.
[y]Ronozyme P (CT) 2500, 500 FTU kg^{-1} DM inclusion.
[x]Nutrient contents of diets were estimated based on nutrient contents of feed ingredients according to NRC (1998). High-moisture corn phosphorus content (0.25%, 88% DM basis) according to Braun and de Lange (2004).
[w]Based on analysis of total (0.26%) and soluble P content in HMC with (0.25%) and without (0.07%) addition of phytase (88% DM basis) according to Niven et al. (2007).
[v]Standardized ileal digestible.

Sample Processing, Chemical and Statistical Analysis

To prepare complete diet and faecal samples for analysis, frozen samples were freeze dried and ground through a 1-mm mesh screen. Dry diet supplements (Table 1) and complete diets (Table 2) were analyzed for DM, crude protein (CP), Ca, and P content (Table 3) as well as ash and titanium dioxide content. Whole frozen carcasses including viscera were ground (Autio Meat Grinder #801, Autio Company, Astoria, OR) and two sub-samples were taken and immediately frozen for subsequent nutrient analyses (Tuitoek et al. 1997). The carcass sub-samples were freeze dried and then ground in liquid nitrogen to a powder. Carcass samples were analyzed for DM, CP, crude fat, ash, and P content (Tuitoek et al. 1997). The right feet were thawed and the third and fourth metacarpals were removed, manually cleaned of adhering tissue, and refrozen in plastic bags until subsequent analysis (Crenshaw 1986). Both metacarpals were analyzed for breaking strength and DM, ash, and P content. All analysis was performed in duplicate (faeces, carcass, metacarpal) or triplicate (diet).

All samples were analyzed for DM content by oven drying at 103°C to a constant weight for a minimum of 24 h [Association of Official Analytical Chemists (AOAC) 1997; method 950.46B]. Crude protein content was determined in feed, faces, and carcass samples by quantifying nitrogen content using a LECO-FP 428 automatic analyzer [Leco Instruments Ltd., Mississauga, ON; AOAC (1997), method 990.03] and multiplying the value by 6.25. Crude fat content in carcass samples was determined by extraction using an ANKOM XT20 fat analyzer (Ankom Technology Corp., Macedon, NY; Ankom Technology Method 2, 01-30-09). Phosphorus contents in diets, faeces, carcass and bones were determined as described by Heinoen and Lahti (1981). Titanium dioxide contents were analyzed in diet and faecal samples according to Myers et al. (2004). For determination of ash contents of diet, faeces, and carcass, samples were placed in a muffle furnace (Fisher Isotemp Programmable Muffle Furnace, Fisher Scientific, Ottawa, ON) with ramping rate of 2°C min^{-1} and held at 550°C for 12 h according to AOAC (1997) method 923.03. Apparent total tract digestibility values for DM, CP, ash, and P were calculated using titanium dioxide as an indigestible marker according to Zhu et al. (2005). Metacarpal length was taken at the longest point along the entire bone and widths were taken at the narrowest (Width 1) and widest (Width 2) points at the midpoint of the shaft (Digital Caliper #06-664–16, Fisher Scientific Company, Ottawa, ON). Breaking strength of bones was determined using an Instron three-point break test machine (Instron 4204, Norwood, MA). Force was applied by a load cell to the mid-point of the bone, which was supported at either end by two supports placed 3 cm apart. The load cell descended onto the bone at a rate of 10 mm min^{-1}. Kilograms of force applied was monitored by the Instron LabView data sampling program (250 samples/second) and the breaking force was determined as the highest recorded force (Combs et al. 1991).

Data were subjected to analysis of variance based on the mixed model procedure (PROC MIXED) of the SAS statistical program (SAS 9.1, SAS Institute Inc., Cary, NC). Pen was the experimental unit for all analyses. Block was considered a random effect and treatment was considered a fixed effect. Initial BW, final carcass plus viscera weight, and bone weight, which were not influenced by treatment ($P > 0.10$), were used as covariables for weight gain, carcass nutrient content, and bone data, respectively. Contrasts among the treatments containing no supplemental phytase were used to determine linear and quadratic effects of dietary P content. A contrast of the Low versus Phy treatments

Table 3. Analyzed nutrient content (adjusted to 88% DM basis) of Phase I, II, and III starter pig diets that were formulated to vary in total dietary P content (High, Medium, and Low) or to have low P content with added phytase (Phy)

	Phase I[z]				Phase II[z]				Phase III[z]			
	High	Medium	Low	Phy	High	Medium	Low	Phy	High	Medium	Low	Phy
Diet supplements												
DM (%)	94.7	–	94.0	–	–	–	–	–	92.0	–	91.7	–
CP (%)	26.6	–	27.8	–	–	–	–	–	33.6	–	33.3	–
Ca (%)	1.80	–	1.86	–	–	–	–	–	1.72	–	1.51	–
P (%)	1.00	–	0.84	–	–	–	–	–	1.08	–	0.90	–
Complete liquid diets												
DM (%)	19.6	25.5	24.7	24.6	25.1	21.9	24.4	23.6	25.0	24.9	23.8	25.0
CP (%)	19.5	20.6	21.2	21.0	19.1	18.2	18.8	18.0	18.5	17.9	17.0	17.2
Ca (%)	0.76	0.99	0.92	1.08	0.65	0.62	0.70	0.69	0.58	0.54	0.47	0.52
P (%)	0.70	0.70	0.65	0.68	0.66	0.61	0.60	0.59	0.64	0.62	0.54	0.55
Ca:P	1.09	1.41	1.42	1.59	0.98	1.02	1.17	1.17	0.91	0.87	0.87	0.95

[z]Pigs were placed on a three-phase feeding program with diet transitions occurring on days 7 and 21 post-weaning.

was used to determine the effects of supplemental phytase. Least squares means were determined for all treatments. Differences between the means were considered statistically significant at $P \leq 0.05$. A trend towards significance was also considered at $P < 0.10$.

RESULTS AND DISCUSSION

General Observations and Diet Analysis

During the trial, 20 pigs were not thrifty or became lame and were treated. Pigs that did not recover within 3d were removed from the trial. In total, 19 pigs were removed from the trial (3, 8, 5, and 3 pigs for the High, Medium, Low, and Phy treatments, respectively). All pigs removed from the trial were recorded and accounted for when calculating performance and feed intake data using the concept of pig days per pen. There did not appear to be any correlation between pig health or lameness and dietary treatment.

Slight differences between formulated and analyzed nutrient content in the feed supplements and complete diets were expected and represent variability in ingredient nutrient content, especially when considering calculated values were obtained using published ingredient nutrient contents (NRC 1998), except for HMC. The majority of analyzed nutrient contents were within $\pm$15% of formulated nutrient contents. In Phase I and II diets, the analyzed contents of Ca and P in the complete diets tended to be higher than formulated values and could be the result of higher than expected Ca and P content in fish meal, which was unique to diets in Phase I and II. A sample of fish meal was analyzed after the trial and found to contain 7.34% Ca and 3.88% P (88% DM basis), which was indeed higher than published values (NRC 1998). In addition, mineral analysis tends to have poor repeatability (Association of American Feed Control Officials 1998), which may further account for discrepancies between predicted and analyzed mineral content in the diets. Finally, accurate sampling of liquid feed can be difficult because some ingredients in liquid feeds tend to settle quickly, which further leads to inaccurate dietary analysis. The latter may explain the differences observed in diets in this experiment, especially between the Low and Phy diets that contained the same dry dietary supplement, with only the addition of phytase to HMC distinguishing the two treatments. Analyzed titanium dioxide contents did not differ between phases or between diets within phases ($P > 0.10$), so an average value for titanium dioxide of 0.099% (DM basis) was used across treatments for calculating nutrient digestibility.

Apparent Total Tract Digestibility

There was no influence of dietary treatment on DM, CP, ash, or P digestibility in Phase II of the trial ($P > 0.10$; Table 4). In Phase III, there was a significant linear decrease in DM, CP, and ash digestibility as dietary P content decreased ($P < 0.05$). In addition, there was a trend for a quadratic effect of dietary P content on both CP and ash digestibility ($P = 0.067$; Table 4). There were improvements in CP ($P < 0.05$) and ash ($P < 0.05$) digestibility as well as a trend for improvement in P digestibility ($P = 0.062$) with addition of phytase, in spite of the large variability of measured P digestibility.

Results in Phase III concur with a number of previous studies that examined the impact of phytase on nutrient digestibility in pigs fed dry diets (Han et al. 1997; Kim et al. 2005; Veum et al. 2006). Phosphorus bound in the form of phytate is largely unavailable to monogastric animals. Supplemental phytase has been shown to be effective in improving P digestibility when included in conventional dry swine diets, but P digestibility in corn- and soybean-meal-based diets near or below the P requirement with added phytase rarely exceeds 55% (Kornegay and Verstegen 2001). This is most likely due to the short retention time of feed in the stomach, as appropriate conditions for optimum

Table 4. Effect of dietary P level (High, Medium, and Low) and phytase supplementation of a low P diet (Phy) on apparent total tract digestibility of DM, CP, ash, and P during week 3 (Phase II) and week 6 or 7 (Phase III) in newly-weaned pigs fed HMC-based liquid diets

	Treatment					Contrasts (P values[z])			
	High	Medium	Low	Phy	SEM[y]	Main	Linear	Quad	Low vs. Phy
Phase II									
DM (%)	80.8	81.5	80.1	77.6	1.29	0.170	0.610	0.400	0.286
CP (%)	66.8	69.7	64.5	61.4	2.68	0.193	0.511	0.200	0.467
Ash (%)	42.5	37.4	48.2	48.1	5.67	0.535	0.390	0.175	0.947
P (%)	65.6	51.4	56.7	56.0	5.19	0.273	0.245	0.138	0.948
Phase III									
DM (%)	84.5	84.6	81.4	82.5	1.12	0.104	0.043	0.187	0.470
CP (%)	74.5	74.7	67.6	73.4	1.54	0.015	0.006	0.067	0.016
Ash (%)	51.3	53.7	42.7	51.6	2.79	0.061	0.046	0.067	0.036
P (%)	50.1	60.2	53.3	67.5	5.30	0.107	0.674	0.190	0.062

[z]Probability of main treatment effects, linear and quadratic (Quad) relationships between High, Medium, and Low diets, and simple contrast between Low and Phy treatments (Low vs. Phy).
[y]Based on six pens of 16 pigs per dietary treatment.

phytase activity are present only in the stomach. Upon entering the small intestine, phytase is quickly deactivated by rising pH or degraded by intestinal proteases (Kemme et al. 2006).

Values for P digestibility in the current trial are similar to those obtained in a previous trial that fed diets containing low-phytate corn to starter pigs (Sands et al. 2001). The lack of a significant response in P digestibility with the addition of phytase during Phase II in the current trial, however, conflicts with previous observations in studies where pigs were fed dry corn- and soybean-meal-based diets (Han et al. 1997; Zhang et al. 2000). Difference in diet form and ingredients may have accounted for the lack of influence of dietary treatment on P digestibility. High-moisture corn has been shown to have a higher soluble P content, an estimate of available P, than dry corn (Ross et al. 1986; Niven et al. 2007). Formulated dietary available P contents (Table 2) in Phase II and Phase III diets in the current trial were below published requirements (NRC 1998), but further restriction may have been required in order to observe a response to additional phytase with respect to P digestibility in Phase II. It has been shown that pigs reduce P absorption when dietary P levels exceed requirements in order to maintain whole body P homeostasis (Quamme 1985). However, it has also been demonstrated that P digestibility remains relatively constant over a wide range of dietary P concentration (Stein et al. 2008). The concept of P digestibility in relation to homeostatic mechanisms requires further exploration.

Phytate has been shown to affect the availability of nutrients other than P, including protein, carbohydrates, and other minerals because the binding capacity of phytate is reduced as phosphate groups are removed and the complex becomes more soluble (Harland and Morris 1995). This increase in solubility is confirmed in the current trial by the improvements in CP and ash digestibility with the addition of phytase to Phase III diets. The impact of phytase on apparent total tract digestibility in the current trial agrees with previous results in weaner pigs (Kornegay and Qian 1996; Han et al. 1997; Brana et al. 2006) and grower-finisher pigs (Johnston et al. 2004; Fan et al. 2005; Kim et al. 2005). The use of phytase in swine diets may reduce the need for mineral supplementation. These results demonstrate the importance of phytase supplementation, not only for improving P availability, but also the availability of other important nutrients in swine diets.

Animal Performance

There were no differences in initial or final BW across treatments ($P > 0.10$), which averaged 5.8 ± 0.45 and 22.7 ± 1.98 kg, respectively (Table 5). Dietary P content and phytase supplementation had no impact on average daily BW gain (ADG) in Phase I ($P > 0.10$). In Phase II, there was a trend for improved ADG ($P = 0.088$) and average daily feed intake ($P = 0.068$) when phytase was added to the low P diet. However, these differences were no longer apparent in Phase III

Table 5. Initial and final BW, and effect of dietary P level (High, Medium, and Low) and phytase supplementation of a low P diet (Phy) on average daily gain, average daily feed intake, and feed efficiency (gain:feed) in starter pigs fed liquid, high-moisture corn (HMC)-based diets

	Treatment					Contrasts (P values[z])			
	High	Medium	Low	Phy	SEM[y]	Main	Linear	Quad	Low vs. Phy
Initial BW (kg)	5.8	5.8	5.8	5.8	0.01	0.120	0.273	0.260	0.474
Final BW (kg)	23.0	22.4	22.0	23.2	0.70	0.534	0.356	0.940	0.228
Average daily gain (g d^{-1})									
Phase I[x]	29	34	41	43	21.4	0.948	0.648	0.957	0.939
Phase II[x]	263	253	238	283	18.9	0.380	0.355	0.912	0.088
Phase III[x]	484	483	457	462	18.9	0.643	0.305	0.579	0.836
Overall	359	363	349	367	14.2	0.790	0.613	0.600	0.354
Average daily feed intake (g DM pig^{-1} d^{-1})									
Phase I	120	112	123	148	10.6	0.171	0.826	0.475	0.133
Phase II	325	330	311	383	26.0	0.259	0.715	0.720	0.068
Phase III	858	810	811	884	52.6	0.694	0.528	0.706	0.335
Overall	559	561	533	556	24.9	0.825	0.468	0.611	0.499
Gain:Feed (kg kg–DM^{-1})									
Phase I	0.27	0.23	0.34	0.33	0.12	0.884	0.663	0.608	0.953
Phase II	0.83	0.75	0.76	0.78	0.03	0.391	0.194	0.261	0.772
Phase III	0.58	0.60	0.56	0.57	0.04	0.898	0.705	0.540	0.845
Overall	0.64	0.65	0.66	0.68	0.02	0.642	0.715	0.901	0.408

[z]Probability of main treatment effects, linear and quadratic (Quad) relationships between High, Medium, and Low diets, and simple contrast between Low and Phy treatments (Low vs. Phy).
[y]Based on six pens of 16 pigs per dietary treatment.
[x]Phase I, II, and III represent days 0 to 7, days s8 to 21, and days 22 to end of trial period, respectively. End of trial period was 49 d post-weaning if the average pen BW on day 42 was below 20 kg.

or when examined over the entire trial period. There was no effect of dietary P or phytase content on feed efficiency ($P > 0.10$).

Average daily BW gain in the current trial was lower than expected based on previous trials, especially during phase I (Brana et al. 2006; Veum et al. 2006), and may be primarily attributable to liquid feeding and management practices as well as the lack of growth-promoting antibiotics in the diets fed in this study. One of the main concerns with liquid feeding newly weaned pigs is balancing freshness of liquid feed in the trough with maximizing intake. Uneaten liquid feed in the trough may quickly spoil and become unpalatable to the pigs, reducing intake (Brooks et al. 2001). It is for this reason that sensor feeding was used in the current trial, which likely resulted in some degree of feed intake restriction. Poor growth performance in Phase I may have impacted subsequent performance. The observed improvement in ADG during Phase II when phytase was added to the low P diet was the result of increased feed intake as feed efficiency was not affected.

Improvement in ADG plateaus at higher inclusion levels of dietary phytase (Kornegay and Qian 1996), most likely because available P intake exceeds requirements for maximum ADG. The response to phytase supplementation also depends on the amount of P in the diet (Yi et al. 1996), and will be minimal when the available P level in the basal diet exceeds the pig's requirement. Consideration should be given to the fact that simply steeping HMC in water may have slightly increased P availability (Niven et al. 2007), and therefore, available P content in the diets may have been greater than estimated and may have reduced the effect of additional phytase. Previous work on the use of low-phytate corn, which has P availability similar to that of stored HMC, showed significant improvements in ADG in starter pigs when compared with dry corn (Sands et al. 2001). The analyzed total P dietary contents in these test diets were considerably lower than those achieved in the current study, with the highest total dietary P reported as 0.52% (88% DM basis; Sands et al. 2001).

Because ADG across the entire experimental period was maintained for all dietary treatments, the degree of P restriction in the current trial was not adequate for inducing depressions in ADG or a growth performance response to added phytase. Based on these observations, maximum ADG in pigs that are liquid-fed HMC-based diets is achieved when analyzed total dietary P levels are 0.65, 0.59, and 0.54% for Phase I, II, and III diets, respectively, which is below NRC (1998) requirements. These observations illustrate again that pig diets should be formulated based on available or digestible P contents and that P availability or digestibility should be accurately determined in pig feed ingredients. Considerations should also be made to account for increases in available P content in pig diets with the use of supplemental phytase. These observations support previous in vitro observations that available P in stored HMC is likely higher than in dry corn (Niven et al. 2007).

Carcass Composition

There were linear reductions ($P < 0.05$) in carcass dressing percentage, carcass weight, and carcass DM content as dietary P was reduced (Table 6); however, these response variables were not affected by added phytase. There was no effect of dietary P level or phytase supplementation on carcass content of CP, crude fat, ash, or P ($P > 0.10$).

Treatment effects on carcass weight and carcass dressing percentage can be partly attributed to differences in final BW. There were no differences in carcass nutrient composition despite treatment effects on nutrient digestibility

Table 6. Effect of dietary P level (High, Medium, and Low) and phytase supplementation of a low P diet (Phy) on carcass weight and CP, crude fat (CF), ash, and P content in carcass plus viscera (100% DM basis) of starter pigs fed HMC-based liquid diets

	Treatment					Contrasts (P values[z])			
	High	Medium	Low	Phy	SEM[y]	Main	Linear	Quad	Low vs. Phy
Final BW (kg)	22.3	23.2	23.1	24.0	0.42	0.045	0.190	0.362	0.075
Empty carcass (kg)	19.7	19.1	18.7	18.8	0.27	0.087	0.015	0.554	0.787
Empty viscera (kg)	1.95	2.00	1.93	1.91	0.07	0.711	0.799	0.360	0.837
Carcass dressing (%)	85.1	82.1	80.9	81.4	1.16	0.077	0.014	0.490	0.747
Composition of carcass plus viscera									
DM content (%)	34.9	34.2	33.7	34.6	0.38	0.091	0.025	0.754	0.090
CP content (%N × 6.25)[x]	53.4	54.3	54.2	53.9	1.69	0.968	0.723	0.780	0.872
CF content (%[x])	34.6	34.0	33.3	33.9	1.59	0.929	0.523	0.980	0.779
Ash content (%[x])	10.7	10.2	10.9	10.6	0.95	0.941	0.880	0.555	0.813
P content (%[x])	1.83	1.83	1.96	1.79	0.16	0.831	0.541	0.722	0.431

[z]Probability of main treatment effects, linear and quadratic (Quad) relationships between High, Medium, and Low diets, and simple contrast between Low and Phy treatments (Low vs. Phy).
[y]Based on six pens of 16 pigs per dietary treatment.
[x]Given as percent of dry carcass weight.

during Phase III of the trial, suggesting there was no influence of additional phytase and digestible P intake on retention of P, ash, protein and fat. The lack of treatment effects on whole body retention of P and other nutrients is consistent with the lack of treatment effects on overall ADG. Apparently, urinary excretion of excess P accounts for differences between digestible P intake and whole body P retention, which is consistent with previous observations by Fan et al. (2005).

Bone Characteristics

There was no effect of dietary P level or phytase supplementation on any of the physical characteristics of the third and fourth metacarpals ($P > 0.10$; Table 7), except for bone breaking strength, which was significantly improved when phytase was added to the Low diet ($P < 0.01$). There was a trend for bone P content (% of dry bone) to increase ($P = 0.089$) when comparing the Low and Phy treatments. Metacarpal P weight was also increased with phytase supplementation to the low P diets ($P < 0.05$).

Surprisingly, a significant increase in bone P content was observed despite the lack of a significant dietary effect on whole body P retention. This may be due to differences in sensitivity of P analyses in bone and carcass samples. The improvement in metacarpal P content and breaking strength as phytase is added to a low-P diet concurs with previous studies investigating effects of phytase supplementation in dry corn- and soybean meal-based diets on bone mineralization (Kornegay and Qian 1996; Omogbenigun et al. 2003; Brana et al. 2006). In the pig, P for use in soft tissue growth is prioritized over P for skeletal growth (Crenshaw 2001), therefore, inadequate dietary P or improvements in P availability would be more apparent in skeletal measures of P retention. Improvements in bone strength in response to phytase supplementation have been observed in the absence of any further response in ADG (Kornegay and Qian 1996; Brana et al. 2006). These observations confirm that measures of bone P content and bone breaking strength are more responsive to available supply of dietary P than measures of pig performance (Zhang et al. 2000), and that higher levels of available P are required for maximum skeletal development. The Phy treatment resulted in improvements in metacarpal strength greater than those observed on the Medium and High levels of phosphorus. This may be a result of extra-phosphoric effects of phytase, such as release of phytate-bound calcium, resulting in a more favourable nutrient balance than is achieved with the addition of inorganic P.

In conclusion, data from this experiment indicate that ADG and carcass nutrient composition are maintained in starter pigs fed HMC-based liquid diets with analyzed total P contents of 0.65, 0.59, and 0.54% (88% DM basis) in Phase I, II, and III diets, respectively. In addition, phytase supplementation to low P diets improves bone mineralization and breaking strength as well as digestibility of protein, ash, and P even though no improvements in BW gain were observed. Formulation of low P diets for starter pigs is therefore possible with the use of supplemental phytase. Measurements of P content in specific body tissues, such as bone, are better indicators of available dietary P content. The use of additional phytase in low P, HMC-based liquid diets will reduce the reliance on additional sources of inorganic P in swine diets.

ACKNOWLEDGEMENTS

The authors acknowledge the financial support provided by Ontario Pork, the Natural Sciences and Engineering Research Council of Canada, the Ontario Ministry of Agriculture, Food, and Rural Affairs, and industrial partners of the Swine Liquid Feeding Association: Big

Table 7. Effect of dietary P level (High, Medium, and Low) and phytase supplementation of a low P diet (Phy) on physical characteristics and P content of the third and fourth metacarpals obtained from starter pigs fed HMC-based liquid diets

	Treatment					Contrasts (P values[z])			
	High	Medium	Low	Phy	SEM[y]	Main	Linear	Quad	Low vs. Phy
Fresh weight (g)	9.7	9.8	9.4	9.5	0.26	0.791	0.398	0.573	0.723
Dry weight (g)	5.3	5.2	5.1	5.3	0.14	0.756	0.398	0.615	0.367
Length (mm)	51.8	52.4	51.4	51.4	0.62	0.650	0.683	0.275	0.994
Width 1 (mm)[x]	11.4	11.2	11.5	11.2	0.17	0.607	0.888	0.264	0.347
Width 2 (mm)[w]	14.5	14.2	14.4	14.4	0.16	0.434	0.540	0.150	0.971
P content (% of bone[v])	4.93	5.57	5.03	5.64	0.24	0.108	0.778	0.061	0.089
P weight (g bone^{-1})[v]	0.26	0.29	0.25	0.29	0.01	0.036	0.800	0.019	0.025
Strength (kg)[u]	36.6	37.2	33.3	40.3	1.57	0.041	0.171	0.244	0.006

[z]Probability of main treatment effects, linear and quadratic (Quad) relationships between High, Medium, and Low diets, and simple contrast between Low and Phy treatments (Low vs. Phy).
[y]Based on six pens of 16 pigs per dietary treatment.
[x]Measured as thinnest point at midpoint of metacarpal shaft.
[w]Measured as widest point at midpoint of metacarpal shaft.
[v]Given as unit per dry bone weight.
[u]Measured as maximum force applied at point of failure during three-point break test.

Dutchman, Daco Laboratories Inc., Grand Valley Fortifiers, B.S.C. Nutrition, Great Lakes Nutrition, Furst McNess, Chris Hansen Laboratories, Kenpal Farm Products Inc., Dwyer Manufacturing Ltd., Farmix/Ridley Inc., and Corn Products International. Technical assistance provided by Stewart Niven, Margaret Quinton, Manfred Hansel, Linda Trouten-Radford, Gord VanderVoort, and staff at the Arkell Swine Research Station is greatly appreciated.

Association of American Feed Control Officials Inc. 1997. Official methods of analysis of AOAC International. 16th ed. Volume II. AAFCO, Oxford, IN.

Association of Analytical Chemists. 1997. Official methods of analysis. 17th ed. AOAC, Washington, DC.

Brana, D. V., Ellis, M., Castaneda, E. O., Sands, J. S. and Baker, D. H. 2006. Effect of a novel phytase on growth performance, bone ash, and mineral digestibility in nursery and grower-finisher pigs. J. Anim. Sci. **84**: 1839–1849.

Braun, K. and de Lange, K. 2004. Liquid swine feed ingredients: Nutritional quality and contaminants. Proc. ANAC Eastern Nutrition Conference. Ottawa, ON.

Brooks, P. H., Moran, C. A., Beal, J. D., Demeckova, V. and Campbell, A. 2001. Liquid feeding for the young piglet. Pages 153–178 *in* M. A. Varley and J. Wiseman, eds. The weaner pig: Nutrition and management. CABI Publishing, Wallingford, UK.

Canadian Council on Animal Care. 1993. Guide to the care and use of experimental animals. Vol. 1. CCAC, Ottawa, ON.

Carlson, D. and Poulsen, H. D. 2003. Phytate degradation in soaked and fermented liquid feed – effect of diet, time of soaking, heat treatment, phytase activity, pH and temperature. Anim. Feed Sci. Technol. **103**: 141–154.

Combs, N. R., Kornegay, E. T., Lindeman, M. D., Notter, D. R., Wilson, J. H. and Mason, J. P. 1991. Calcium and phosphorus requirement of swine from weaning to market weight: 2. development of response curves for bone criteria and comparison of bending and shear bone testing. J. Anim. Sci. **69**: 682–693.

Crenshaw, T. D. 1986. Reliability of dietary Ca and P levels and bone mineral content as predictors of bone mechanical properties at various time periods in growing swine. J. Nutr. **116**: 2155–2170.

Crenshaw, T. D. 2001. Calcium, phosphorus, vitamin D, and vitamin K in swine nutrition. Pages 187–212 *in* A. J. Lewis and L. L. Southern, eds. Swine nutrition. 2nd ed, CRC Press LLC, Boca Raton, FL.

de Lange, C. F. M., Zhu, C. L., Niven, S. J., Columbus, D. and Wood, D. 2006. Swine liquid feeding: Nutritional considerations. Proceedings 27th Western Nutrition Conference. Department of Animal Science, University of Manitoba, Winnipeg, MB. pp. 37–50.

Fan, M. Z., Li, T. J., Yin, Y. L., Fang, R. J., Tang, Z. Y., Hou, Z. P., Huang, R. L., Deng, Z. Y., Zhong, H. Y., Zhang, R. G., Wang, B. and Schulze, H. 2005. Effect of phytase supplementation with two levels of phosphorus diets on ileal and faecal digestibilities of nutrients and phosphorus, calcium, nitrogen and energy balances in growing pigs. Anim. Sci. **81**: 67–75.

Han, Y. M., Yang, F., Zhou, A. G., Miller, E. R., Ku, P. K., Hogberg, M. G. and Lei, X. G. 1997. Supplemental phytases of microbial and cereal sources improve dietary phytate phosphorus utilization by pigs from weaning through finishing. J. Anim. Sci. **75**: 1017–1025.

Harland, B. F. and Morris, E. R. 1995. Phytate – A good or bad food component. Nutr. Res. **15**: 733–754.

Heinoen, J. K. and Lahti, R. J. 1981. A new and convenient colorimetric determination of inorganic orthophosphate and its application to the assay of inorganic pyrophosphate. Anal. Biochem. **113**: 313–317.

Johnston, S. L., Williams, S. B., Southern, L. L., Bidner, T. D., Bunting, L. D., Matthews, J. O. and Olcott, B. M. 2004. Effect of phytase addition and dietary calcium and phosphorus levels on plasma metabolites and ileal and total-tract nutrient digestibility in pigs. J. Anim. Sci. **82**: 705–714.

Kemme, P. A., Schlemmer, U., Mroz, Z. and Jongbloed, A. W. 2006. Monitoring the stepwise phytate degradation in the upper gastrointestinal tract of pigs. J. Sci. Food Agric. **86**: 612–622.

Kim, J. C., Simmins., P. H., Mullan, B. P. and Pluske, J. R. 2005. The effect of wheat phosphorus content and supplemental enzymes on digestibility and growth performance of weaner pigs. Anim. Feed. Sci. Tech. **118**: 139–152.

Kornegay, E. T. and Qian, H. 1996. Replacement of inorganic phosphorus by microbial phytase for young pigs fed on a maize-soyabean-meal diet. Br. J. Nutr. **76**: 563–578.

Kornegay, E. T. and Verstegen, M. W. A. 2001. Swine nutrition and environmental pollution and odor control. Pages 609–630 *in* A. J. Lewis and L. L. Southern, eds. Swine nutrition. 2nd ed, CRC Press LLC, Boca Raton, FL.

Myers, W. D., Ludden, P. A., Nayigihugu, V. and Hess, B. W. 2004. Technical note: A procedure for the preparation and quantitative analysis of samples for titanium dioxide. J. Anim. Sci. **82**: 179–183.

Niven, S. J., Zhu, C., Columbus, D., Pluske, J. R. and de Lange, C. F. M. 2007. Impact of controlled fermentation and steeping of high moisture corn on its nutritional value for pigs. Livest. Sci. **109**: 166–169.

National Research Council. 1998. Nutrient requirements of swine. 10th ed. National Academy Press,Washington, DC.

Omogbenigun, F. O., Nyachoti, C. M. and Slominski, B. A. 2003. The effect of supplementing microbial phytase and organic acids to a corn-soybean based diet fed to early-weaned pigs. J. Anim. Sci. **81**: 1806–1813.

Quamme, G. A. 1985. Phosphate-transport in intestinal brush-border membrane-vesicles – effect of pH and dietary phosphate. Am. J. Physiol. **249**: G168–G176.

Ross, R. D., Cromwell, G. L. and Stahly, T. S. 1986. Biological availability of the phosphorus in high-moisture and pelleted corn. J. Anim. Sci. *57* (Suppl. 1): 96 (Abstr.).

Sands, J. S., Ragland, D., Baxter, C., Joern, B. C., Sauber, T. E. and Adeola, O. 2001. Phosphorus bioavailability, growth performance, and nutrient balance in pigs fed high available corn and phytase. J. Anim. Sci. **79**: 2134–2142.

Skoglund, E., Larsen, T. and Sandberg, A. S. 1997. Comparison between steeping and pelleting a mixed diet at different calcium levels on phytate degradation in pigs. Can. J. Anim. Sci. **77**: 471–477.

Stein, H. H., Kadzere, C. T., Kim, S. W. and Miller, P. S. 2008. Influence of dietary phosphorus concentration on the digestibility of phosphorus in monocalcium phosphate by growing pigs. J. Anim. Sci. **86**: 1861–1867.

Tuitoek, K., Young, L. G., de Lange, C. F. and Kerr, B. J. 1997. The effect of reducing excess dietary amino acids on

growing-finishing pig performance: An elevation of the ideal protein concept. J. Anim. Sci. **75**: 1575–1583.

Veum, T. L., Bollinger, D. W., Buff, C. E. and Bedford, M. R. 2006. A genetically engineered *Escherichia coli* phytase improves nutrient utilization, growth performance, and bone strength of young swine fed diets deficient in available phosphorus. J. Anim. Sci. **84**: 1147–1158.

Yi, Z., Kornegay, E. T., Ravindran, V., Lindemann, M. D. and Wilson, J. H. 1996. Effectiveness of Natuphos® phytase in improving the bioavailabilities of phosphorus and other nutrients in soybean meal-based semipurified diets for young pigs. J. Anim. Sci. **74**: 1601–1611.

Zhang, Z. B., Kornegay, E. T., Radcliffe, J. S., Wilson, J. H. and Veit, H. P. 2000. Comparison of phytase from genetically engineered *Aspergillus* and canola in weanling pig diets. J. Anim. Sci. **78**: 2868–2878.

Zhu, C. L., Rademacher, M. and de Lange, C. F. M. 2005. Increasing dietary pectin level reduces utilization of digestible threonine intake, but not lysine intake, for body protein deposition in growing pigs. J. Anim. Sci. **83**: 1044–1053.