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Phosphorus utilization and characterization of excreta from swine fed diets containing a variety of cereal grains balanced for total phosphorus[1]

A. B. Leytem*[2] and P. A. Thacker†

*USDA-ARS, Northwest Irrigation and Soils Research Laboratory, Kimberly, ID 83341-5076;
and †Department of Animal and Poultry Science, University of Saskatchewan,
Saskatoon, Saskatchewan, Canada S7N 5A8

ABSTRACT: Intrinsic phytase in swine feeds may increase phytate utilization and alter the solubility of the excreted P. The objective of this experiment was to quantify changes in fecal P composition from swine fed a variety of cereal grains containing a range of phytate concentrations and endogenous phytase activities. Twenty-five crossbred barrows (89.3 ± 6.8 kg) were fed 1 of 5 dietary treatments that were based on wheat, corn, barley, low-phytate barley, or high-fat–low-lignin oats. Experimental diets were formulated to contain 75% of the test grain and were fed for a 7-d acclimation period followed by a 3-d fecal collection period. Total-tract apparent digestibility coefficients were determined for DM, P, and phytate using an indicator method. Fecal P was characterized using solution-state ^{31}P nuclear magnetic resonance spectroscopy. Water-soluble P (WSP) and WSP-to-total P (TP) ratio were determined in the feces. Apparent total-tract digestibility coefficients for P and phytate ranged from 0.33 (barley) to 0.45 (low-phytate barley) and from 0.20 (corn) to 0.79 (oats), respectively. The majority of P excreted in the feces was in the form of phosphate ($>47\%$ of TP), and phytate degradation was not related to the endogenous phytase activity in the diet. There was a positive linear relationship between dietary NDF and apparent total-tract phytate digestibility ($r^2 = 0.82$; $P = 0.03$), indicating that greater dietary fiber content may enhance microbial breakdown of phytate in the hindgut. There was a negative relationship between the fecal WSP-to-TP ratio and the percentage of TP that was in the form of phytate in the feces. In summary, our results indicate that the majority of P in the feces of pigs fed diets based on cereal grains is present in the form of phosphate and relatively small amounts of phytate were contained in the excreta. The exception to this was the corn diet, for which 45% of the total fecal P was in the form of phytate. Hydrolysis of phytate in the gut did not appear to be related to the content of either phytate or phytase in the grain, but was related to dietary fiber concentration.

Key words: cereal grain, endogenous phytase, nuclear magnetic resonance spectroscopy, phosphorus, phytate degradation, swine

INTRODUCTION

Phytate (*myo*-inositol hexakisphosphate) is the primary storage form of P found in cereal grains and constitutes a large portion of the total P (**TP**) contained in plant-based diets fed to swine (Ravindran et al., 1994). Pigs are inefficient in utilizing phytate P, which has been ascribed to inadequate secretion of the enzyme phytase that is required to hydrolyze the phytate molecule (Jongbloed et al., 1992; Pointillart, 1994; Yi and Kornegay, 1996). As a result, the bioavailability of P from cereal grains has been shown to be poor and varies from 14% in corn to 49% in wheat (Cromwell, 1992).

It is commonly believed that the poor bioavailability of phytate P in cereals results in substantial excretion of phytate P in swine feces. The amount of phytate excreted could alter the proportion of water-soluble P (**WSP**) in the feces, thereby affecting P losses after land application of swine manure and increasing the potential for P to contaminate surface and groundwaters (Kornegay and Harper, 1997).

There is very little published information on P speciation in feces of swine fed different cereal grains. Although Leytem and Thacker (2008) did elucidate the

P composition of feces from swine fed several cereal grains, the diets fed in that study were not adequate with respect to P and therefore may not truly represent the P composition of feces from swine fed balanced diets. Information is needed on the P composition of feces obtained from swine fed a wide range of diets to assess differences between cereal grains regarding the extent of phytate P hydrolysis in the gastrointestinal tract and its subsequent impact on the WSP content of manure. Therefore, the objective of this experiment was to quantify changes in fecal P composition from swine fed diets containing cereal grains that contained a range of phytate concentrations and endogenous phytase activities.

MATERIALS AND METHODS

Pigs used in this experiment were cared for following the guidelines of the Canadian Council on Animal Care (1993).

Origin of Cereal Grains

The test cereal grains used as the principal ingredients in 5 treatment diets fed in this experiment were commercial-grade corn and wheat obtained from a local feed mill, newly developed high-fat–low-lignin oats (Thacker and Rossnagel, 2005), Harrington barley (Harvey and Rossnagel, 1984), and low-phytate barley (mutant LP422), with approximately 50% less phytate P than Harrington barley (Dorsch et al., 2003).

Digestibility Trial

Twenty-five crossbred barrows (Camborough 15 Line female × Canabred sire, Pig Improvement Canada Ltd., Acme, Alberta, Canada), weighing an average of 89.3 ± 6.8 kg, were used in this experiment. Barrows were housed in a specially designed research facility with 36 concrete-floored pens (2.7 × 3.6 m). Each pen accommodated up to 4 pigs. Four individual feeding stations were located at the front of each pen. This design allowed for group housing of pigs during most of the day to facilitate social interaction, but allowed for individual DMI to be determined by locking the pigs into individual feed stations at feeding time. Barrows were randomly divided among 7 pens that were distributed throughout the facility. This resulted in 6 pens having 4 barrows/pen and 1 pen with a single barrow. Three additional barrows (same type but assigned to a different study) were added to the pen with the single barrow. Within each pen, barrows were randomly assigned a color code that corresponded to a color-coded feeder station. Throughout the study, barrows were locked in their corresponding feeding stations for 30 min twice daily (0700 and 1500 h) and were individually fed their treatment diets (see below). Water (0.00014% P) was supplied by a nipple waterer located in the back of each pen, allowing for ad libitum water intake.

Barrows were assigned randomly to 1 of 5 (n = 5) treatment diets (Table 1) in a completely randomized design. The experimental diets were formulated to contain 75% (as-fed basis) of one of the test grains (described above; Table 1). Treatment diets were supplemented with sufficient soybean meal and synthetic AA (Lys, Met, and Thr) to meet the AA requirements of pigs with a lean growth potential of 350 g/d (NRC, 1998). An attempt was made to ensure that all diets provided equal DE by varying the amount of canola oil and corn starch added to the diet. Diets were supplemented with sufficient vitamins and minerals (including dicalcium phosphate) to meet or exceed NRC concentrations (NRC, 1998). Chromic oxide (0.35%) was added as a digestibility indicator.

Treatment diets were fed for a 7-d acclimatization period, followed by a 3-d fecal collection period. Feces were obtained by bringing the barrows into a clean room immediately after feeding and recovering freshly voided feces. The feces from each day were immediately refrigerated after collection for the 3-d collection period, after which they were pooled by animal and then immediately frozen for storage. Before analysis, samples were dried in a forced-air oven dryer at 55°C for 60 h and subsequently ground through a 0.5-mm screen. Total-tract apparent digestibility coefficients for DM, P, and phytate were calculated using the equations for the indicator method described by Schneider and Flatt (1975).

Chemical Analysis of Feeds and Feces

Analyses were conducted for DM (method 930.15, AOAC, 1990), CP (method 984.13, AOAC, 1990), ash (method 942.05, AOAC, 1990), and ether extract (method 920.39, AOAC, 1990). Neutral detergent fiber was analyzed using the method of Van Soest et al. (1991). Diets were analyzed for Ca using atomic absorption spectroscopy (method 968.08, AOAC, 1990). Amino acid analysis of the diets was determined by HPLC (L-8800 Amino Acid Analyzer, Hitachi, Tokyo, Japan). All samples were hydrolyzed for 24 h at 110°C with 6 N HCl before analysis. Sulfur-containing AA were analyzed after cold formic acid oxidation for 16 h before acid hydrolysis.

Feed and fecal samples were wet-ashed using the nitric-perchloric acid method of Zasoski and Burau (1977), and TP was determined colorimetrically (Pharmacia Ultraspec III, LKB Biochrome, St Albans, UK) using a molybdovanadate reagent (method 965.17, AOAC, 1990). The ferric precipitation method (Raboy et al., 1990) was used to extract and precipitate the phytate P in the feed, and the resulting extracts were analyzed for phytate by the colorimetric assay of Chen et al. (1956). Phytase activity in the diet (samples as fed) was determined using a Danisco Animal Nutrition (Brabrand, Denmark) proprietary assay for complete feed samples. Chromic oxide was determined in feed and feces by the method of Fenton and Fenton (1979).

Table 1. Ingredient composition and chemical analysis of diets used to determine P utilization and excretion, and fecal P characterization of growing pigs fed various cereal grains

Item	Commercial-grade wheat	Harrington barley	Commercial-grade corn	High-fat–low-lignin oats	Low-phytate barley
Ingredient, % (as fed)					
Cereal grain	75.0	75.0	75.0	75.0	75.0
Soybean meal	14.0	14.0	14.6	14.0	14.0
Corn starch	6.7	1.5	6.3	1.5	1.5
Canola oil	0.71	5.9	—	5.9	5.9
Limestone	0.93	0.91	0.79	0.91	0.91
Dicalcium phosphate	0.77	0.77	1.1	0.77	0.77
Vitamin-mineral premix[1]	1.0	1.0	1.0	1.0	1.0
Salt	0.50	0.50	0.50	0.50	0.50
Chromic oxide	0.35	0.35	0.35	0.35	0.35
Thr	0.01	0.06	0.09	0.06	0.06
Met	0.01	0.05	0.13	0.05	0.05
Lys	—	0.05	0.13	0.05	0.05
Chemical analysis,[2] % (as fed)					
Moisture	10.5	10.3	9.6	9.8	8.6
CP	16.9	17.5	14.2	16.8	17.2
Lys	0.71	0.75	0.76	0.89	0.82
Met + Cys	0.64	0.53	0.61	0.72	0.67
Thr	0.61	0.57	0.59	0.70	0.65
Ether extract	3.0	8.5	3.6	10.2	8.1
NDF	11.7	16.3	8.6	17.7	14.1
Ash	4.8	5.7	4.7	6.0	4.8
Ca	0.64	0.65	0.69	0.68	0.64
Total P	0.56	0.52	0.55	0.59	0.56
Phytate P	0.27	0.23	0.20	0.27	0.13
Endogenous phytase activity, phytase units[3]/kg	384	133	<50	<50	318

[1]Supplied (as fed) per kilogram of diet: 8,250 IU of vitamin A; 825 IU of vitamin D_3; 40 IU of vitamin E; 4 mg of vitamin K; 1 mg of thiamine; 5 mg of riboflavin; 35 mg of niacin; 15 mg of pantothenic acid; 2 mg of folic acid; 12.5 µg of vitamin B_{12}; 0.2 mg of biotin; 80 mg of Fe; 25 mg of Mn; 100 mg of Zn; 50 mg of Cu; 0.5 mg of I; 0.1 mg of Se.

[2]All chemical composition data are the results of a chemical analysis conducted in duplicate.

[3]1 phytase unit is defined as the quantity of enzyme required to liberate 1 µM of P per minute from an excess of sodium phytate.

Fecal samples were analyzed for WSP by shaking 1 g of dry feces with 100 mL of deionized water for 1 h, filtering through a 0.45-µm membrane, and analyzing total WSP by inductively coupled plasma optical-emission spectrometry (Optima 4300 DV, Perkin-Elmer, Wellesley, MA). Total fecal P was determined by microwave-assisted digestion of a 0.5-g dried sample with 8 mL of concentrated HNO_3 and 2 mL of 30% H_2O_2 (vol/vol) with P quantified using inductively coupled plasma optical-emission spectrometry detection.

P Speciation of Feces

The P composition of feces was determined by solution ^{31}P nuclear magnetic resonance (**NMR**) spectroscopy as described by Turner (2004). Three animals were selected for each dietary treatment for P characterization because of the expense of conducting ^{31}P NMR spectroscopy (n = 3). Briefly, P was extracted in duplicate by shaking 2.00 ± 0.01 g of dried feces with 40 mL of a solution containing 0.5 M NaOH and 0.05 M EDTA for 4 h at 20°C. Extracts were centrifuged at 10,000 × g for 30 min at 20°C, and aliquots were analyzed for TP by inductively coupled plasma optical-emission spectrometry. The NaOH-EDTA extraction

recovered greater than 93% of the total manure P determined by digestion. The remaining solutions from the duplicate extracts were combined, rapidly frozen at −80°C, lyophilized, and then ground to a fine powder.

Freeze-dried extracts were redissolved in 0.1 mL of deuterium oxide (for signal lock) and 0.9 mL of a solution containing 1 M NaOH and 0.1 M EDTA, and were then transferred to a 5-mm NMR tube. Solution ^{31}P NMR spectra were obtained using a Bruker Avance DRX 500-MHz spectrometer (Bruker, Rhinestetten, Germany) operating at 202.456 MHz for [31]P. A 5-µs pulse (45°), a delay time of 5.0 s, an acquisition time of 0.8 s, and broadband proton decoupling were used for all samples. The number of scans varied between 3,331 and 10,580, and spectra were plotted with a line broadening of 1 Hz. Chemical shifts of signals were determined in parts per million relative to 85% H_3PO_4 and were assigned to individual P compounds or functional groups based on literature values (Turner et al., 2003). Signal areas were calculated by integration, and P concentrations were calculated by multiplying the proportion of the total spectral area assigned to a specific signal by the TP concentration (grams of P per kilogram of dry feces) in the original extract. This NMR procedure detects P compounds at concentrations of approximately 0.1 mg of P/kg of dried feces (Turner, 2004).

Table 2. Coefficients for total-tract apparent digestibility for DM, P, and phytate from diets composed of commercial-grade corn, Harrington barley, low-phytate barley, high-fat–low-lignin oats, or commercial-grade wheat fed to swine

Item	DM	P	Phytate
Diet			
Commercial-grade corn	0.89[a]	0.34[bc]	0.20[b]
Harrington barley	0.81[c]	0.33[c]	0.74[a]
Low-phytate barley	0.80[c]	0.45[a]	0.70[a]
High-fat–low-lignin oats	0.74[d]	0.39[b]	0.79[a]
Commercial-grade wheat	0.85[b]	0.35[bc]	0.66[a]
SEM	0.01	0.02	0.09
$P > F$	<0.0001	0.0007	0.0007

[a–d]Within a column, means without a common superscript differ ($P < 0.05$).

Statistical Analysis

All variables were tested for normality using the Shapiro-Wilk test with the UNIVARIATE procedure (SAS Inst. Inc., Cary, NC). To evaluate treatment effects of complete diets, data were analyzed with a one-way ANOVA with diet as the main effect, and means separation was conducted using Tukey's honestly significant differences test with $\alpha = 0.05$. Because the barrows were fed individually, the individual animal was considered the experimental unit for all statistical analyses.

RESULTS

Chemical Analysis of Diets

Chemical analyses of the diets are presented in Table 1. Dietary CP ranged from 14.2 to 17.5%. The AA analysis confirmed that all diets met or exceeded NRC (1998) recommendations for pigs with a lean growth potential of 350 g/d. Neutral detergent fiber ranged from a minimum of 8.6% in the corn diet to a maximum of 17.7% in the oat diet. Total P ranged from 0.52 to 0.59%, whereas phytate P ranged from 0.13% in the low-phytate barley diet to 0.27% in the wheat and oat diets. The phytase activity of the diets ranged from <50 (corn and oats) to 384 (wheat) phytase units/kg of endogenous phytase activity, where 1 phytase unit is defined as the quantity of enzyme required to liberate 1 μmol of P per minute from an excess of sodium phytate.

Apparent Total-Tract Digestibility Coefficients

The coefficients for the apparent total-tract digestibility of DM, P, and phytate are presented in Table 2. Dry matter digestibility coefficients ranged from 0.74 to 0.89 and followed the trend corn > wheat > Harrington barley = low-phytate barley > oats ($P < 0.0001$). Apparent P digestibility coefficients ranged from 0.33 to 0.45, with the low-phytate barley having the greatest coefficient ($P = 0.0007$) and the Harrington barley having the smallest coefficient. The apparent phytate digestibility coefficients ranged from 0.20 to 0.79, with corn having the smallest coefficient ($P = 0.0007$), whereas there were no significant differences between the remaining treatments. There was a positive linear correlation between the NDF content of the diets and the apparent total-tract phytate digestibility ($r^2 = 0.82$, data not shown).

Fecal P Characterization

The fecal P composition, expressed on a DM basis, is presented in Table 3. Total fecal P ranged from 13.8 to 31.4 g/kg, with corn having the greatest concentration and the low-phytate barley and oat diets having the least ($P < 0.001$). The majority of P was in the form of phosphate, with concentrations ranging from 10.3 to 15.8 g/kg constituting between 47 and 79% of the TP, with corn having the least percentage ($P = 0.006$). Phytate P concentrations ranged from 1.9 to 14.3 g/kg, constituting between 15 and 46% of the TP, with corn producing the greatest phytate concentration ($P = 0.002$), whereas the other treatments did not differ. There were very small concentrations of phosphate monoesters (typically fewer inositol phosphates), which ranged from 0.7 to 1.8 g of P/kg, constituting less than 11% of the TP. Low-phytate barley had the greatest percentage of phosphate monoesters, whereas corn and oats had the least ($P = 0.02$). There were small amounts of pyrophosphate in the feces, ranging from 0.22 to 0.48 g of P/kg, constituting less than 3% of the TP.

The WSP concentration of the feces ranged from 4.68 to 7.65 g/kg, with the normal barley diet producing significantly ($P = 0.0006$) greater concentrations than the corn, oat, and wheat diets (Figure 1). The WSP:TP ranged from 0.13 to 0.37 and followed the sequence Harrington barley = low-phytate barley = oats > wheat > corn ($P < 0.0001$, Figure 1). The WSP:TP of the feces was negatively correlated with the percentage of total fecal P in the form of phytate ($r = -0.61$; $P = 0.007$; Figure 2).

DISCUSSION

The apparent total-tract digestibility coefficients for DM were similar to other data reported in the literature. Leytem and Thacker (2008) reported DM digestibility coefficients for corn, barley, low-phytate barley, high-fat–low-lignin oat, and wheat diets of 0.81, 0.71, 0.76, 0.67, and 0.83, respectively. These coefficients are slightly less than those reported in the present study (0.74 to 0.89), likely because in the earlier study, 97% of the diet was grain compared with 75% grain in the present study. The inclusion of ingredients with greater digestibility (i.e., soybean meal) would account for these differences. Powers et al. (2006) reported that the DM digestibility of corn- and soy-based diets ranged from

0.78 to 0.85, whereas the DM digestibility coefficient for the corn diet was 0.89 in the present study. Thacker et al. (2003) reported DM digestibility coefficients of 0.77 and 0.74 when swine were fed normal- and low-phytate barley diets, respectively, and were similar to the values obtained for these treatments in the present study. Bruce and Sundstøl (1995) reported a DM digestibility coefficient for oat-based diets of 0.76, which is similar to the value of 0.74 obtained in the present study.

Leytem and Thacker (2008) reported apparent P digestibility coefficients of 0.11, 0.26, 0.35, 0.26, and 0.46 for corn, barley, low-phytate barley, high-fat–low-lignin oat, and wheat diets, respectively (cereal content was 97% of the diet). In all but one instance (wheat), the apparent P digestibility coefficients in the present study were greater than those reported by Leytem and Thacker (2008). This is likely due to the inclusion of supplemental dicalcium phosphate in the present study, which would have greater bioavailability than when P is supplied by grain alone. Thacker et al. (2003) reported a P digestibility coefficient of 0.39 when swine were fed a low-phytate barley diet, which was less than the value of 0.45 obtained in the present study. The low-phytate barley had a greater apparent P digestibility coefficient than the other dietary treatments, which was expected because this diet contained little phytate and thus would have a greater available P concentration.

Because the diets in the present study contained different amounts of nonphytate P and phytase, it was not possible to separate the effects of differences in nonphytate P content and endogenous phytase in the grains on the resultant P digestibility coefficients obtained. However, the fact that there were few differences in apparent total-tract P digestibility coefficients among the treatments, despite large differences in endogenous phytase activity, suggests that there was very little contribution to P digestibility from the phytase
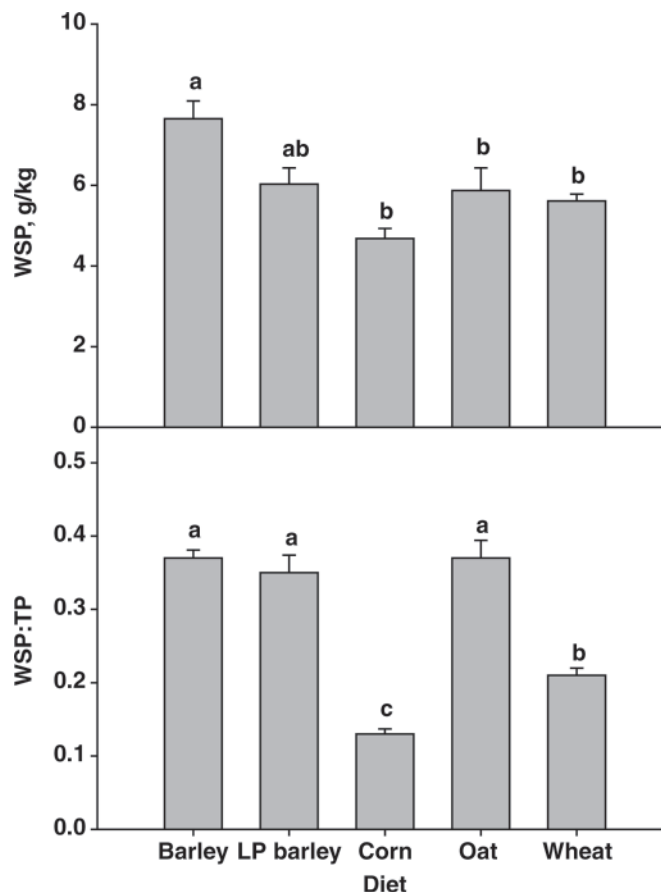

Figure 1. Fecal water-soluble P (WSP; top panel) and the ratio of fecal WSP to total P (WSP:TP; bottom panel) from swine fed a variety of cereal-based diets. Error bars represent the SD of the mean, and bars with the same letter (a–c) do not differ ($P < 0.05$). LP barley = low-phytate barley.

enzymes naturally associated with the grains. For instance, the high-fat–low-lignin oat diet had an apparent P digestibility coefficient of 0.39, which was not

Table 3. Phosphorus characterization of swine feces from diets composed of commercial-grade corn, Harrington barley, low-phytate barley, high-fat–low-lignin oats, or commercial-grade wheat fed to swine, determined by NaOH-EDTA extraction and solution ^{31}P nuclear magnetic resonance spectroscopy[1]

	NaOH-EDTA-extractable P				
Item	Total NaOH-EDTA P[2]	Phosphate	Phosphate monoesters[3]	Phytate	Pyrophosphate
Diet, g of P/kg of DM					
Commercial-grade corn	31.4[a]	14.9 (47)[b]	1.6 (5)[b]	14.3 (46)[a]	0.48 (2)
Harrington barley	18.5[bc]	13.8 (74)[a]	1.2 (6)[ab]	3.1 (17)[b]	0.39 (3)
Low-phytate barley	13.8[c]	10.3 (74)[a]	1.3 (10)[a]	1.9 (14)[b]	0.24 (2)
High-fat–low-lignin oats	14.9[c]	11.8 (79)[a]	0.7 (4)[b]	2.2 (15)[b]	0.22 (2)
Commercial-grade wheat	23.6[b]	15.8 (67)[ab]	1.8 (7)[ab]	5.7 (24)[b]	0.35 (2)
SEM	1.71	6.59	1.26	6.16	0.55
$P > F$	<0.0001	0.006	0.02	0.002	0.71

[a–c]Within a column, means without a common superscript differ ($P < 0.05$).

[1]All values are reported on a DM basis. Values in parentheses are the proportion (%) of the NaOH-EDTA-extracted P.

[2]Values are the means of 3 animals/feeding treatment.

[3]Values for phosphate monoesters include all monoesters other than phytate.

significantly different from wheat (0.35), even though we found a 7-fold greater phytase activity in the wheat vs. the oat diet.

The corn diet did not have a different apparent total-tract P digestibility coefficient from the other treatments (with the exception of the low-phytate barley). This suggests that the degradation of phytate in the diets occurred distal to the terminal ileum and that little P absorption took place. Although Rapp et al. (2001) did not calculate digestibility coefficients, they reported that 19 and 65% of total dietary phytate P was hydrolyzed at the terminal ileum in swine fed either corn- or wheat-based diets, respectively, which is similar to the trend observed in the present study. The corn diet had the least NDF concentration compared with the other diets, which had concentrations 1.4- to 2-fold greater. Increasing NDF, including nonstarch polysaccharides, can increase the population of microorganisms in the cecum and colon (Grieshop et al., 2001). This increase in microbial population could enhance the breakdown of phytate in the hindgut. This hypothesis is supported by the fact that as the NDF content of the diets increased, there was a linear increase in apparent total-tract phytate digestibility.

Few reports have been published on the P composition of feces obtained from swine fed different cereal grains. Furthermore, there is limited evidence of endogenous phytase secretion in the small intestine of swine (Jongbloed et al., 1992; Pointillart, 1994; Yi and Kornegay, 1996), which results in only small quantities of phytate hydrolyzed in the small intestine and poor availability of phytate-bound P. Therefore, it could be expected that because the utilization of phytate P by swine is poor, there should be significant excretion of intact dietary phytate in the feces. However, our data showed that the majority of P in feces was in the form of phosphate (47 to 79% of TP). The corn diet was the only diet that had a large amount of phytate in the feces (46% of TP), whereas the other treatments had less than 24%.

This finding is not consistent with our previous work (Leytem and Thacker, 2008), in which we found only small amounts of phytate (<6% of TP) in feces from swine fed a variety of cereal grains. The present study is also in contrast to the report by Leytem et al. (2004) that only trace amounts of phytate were found in the feces from swine fed diets containing either low- or high-phytate barley. The decreased phytate hydrolysis in the present study is likely due to the inclusion of inorganic P sources in the diet, which could inhibit the utilization of phytate in the diet. The negative effects of increased available P in the diet on phytate degradation have been demonstrated in several studies. Seynaeve et al. (2000) reported that increases in dietary phosphate decreased the phytase activity in ileal digesta of swine, which would decrease phytate hydrolysis in the upper gastrointestinal tract. Leytem et al. (2007) reported that when poultry were fed diets containing increasing supplemental P (40% increase in dietary available
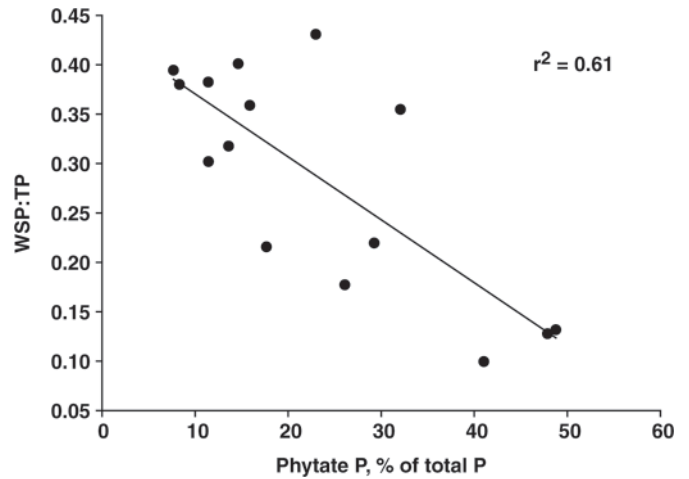

Figure 2. The relationship between the ratio of fecal water-soluble P (WSP) to total P (WSP:TP) and the percentage of total fecal P in the form of phytate.

P), the phytate content of the litter increased by 43%. Ballam et al. (1985) demonstrated that an increase in inorganic P in broiler diets significantly decreased intestinal phytate P hydrolysis. van der Klis and Versteegh (1996) demonstrated a decrease in phytate P hydrolysis of 26 to 48% in broilers fed diets with greater available P compared with diets containing less available P. Although much of this work was performed in poultry, it would be expected to be applicable to other nonruminant species such as swine.

Because swine do not produce appreciable amounts of phytase (Jongbloed et al., 1992; Pointillart, 1994; Yi and Kornegay, 1996), it would appear that other sources of phytase are functioning within the gastrointestinal tract of the pig. The negligible effect of endogenous cereal phytase on apparent P digestibility, combined with the fact that phytate hydrolysis exceeded 66% for all the cereal grains, with the exception of the corn diet, suggests that microbial activity in the hindgut was the most likely mechanism responsible for the high degree of phytate degradation in the present study. This hypothesis is supported by previous observations that bacteria in the hindgut hydrolyzed large amounts of the phytate contained in the digesta (Seynaeve et al., 2000).

The finding that relatively small concentrations of phytate were found in most of the swine feces does not necessarily mean that the P liberated from phytate is available to the pig. In nonruminant animals, there is little evidence of P absorption occurring past the distal ileum (Crenshaw, 2001). Therefore, any P liberated as a result of phytate hydrolysis in the hindgut will most likely not be absorbed and will be voided in the feces. This is supported by our results, which demonstrated that in spite of elevated total-tract phytate hydrolysis, apparent total-tract P digestibility coefficients were small. Even though the corn diet had a much smaller apparent phytate digestibility coefficient, the apparent P digestibility in this diet was similar to the other diets. Therefore, the majority of phytate P hydrolysis

occurred in the lower digestive tract, and the P made available from phytate was not absorbed by the animals and was simply excreted in the feces.

Sharpley and Moyer (2000) found a correlation of 98% between WSP in manure and the amount of P leached from a soil, suggesting that WSP is a good indicator of the potential of manure to contribute to P runoff or leaching after surface application. In the present study, there were few differences in fecal WSP concentrations. The Harrington barley diet resulted in significantly greater fecal WSP concentrations than the corn, oat, and wheat diets (which did not differ), but was not significantly different from the low-phytate barley diet. The fecal WSP reported in the present study (4.6 to 7.7 g/kg) are slightly greater than those found by Leytem and Thacker (2008), who reported concentrations ranging from 4.0 to 6.3 g/kg from swine fed cereal-based diets. The greater concentrations of fecal WSP found in the present study are likely a result of the greater dietary P concentrations. Maguire et al. (2005) reported that reducing dietary P concentrations resulted in a decrease in manure WSP ranging from 21 to 52%.

The WSP:TP was highly correlated with the percentage of TP in the feces that was in the form of phytate. Sequential extraction of manures has demonstrated that P compounds extracted in deionized water are predominantly inorganic P and that the majority of phytate P is extracted only with stronger extractions such as HCl or NaOH (Turner and Leytem, 2004). Therefore, manures that have a greater proportion of phytate P will have decreased WSP concentrations and WSP:TP. Examination of the available literature reveals a similar trend in swine feces (Baxter et al., 2003; Leytem and Thacker, 2008), broiler litter (Maguire et al., 2004; Toor et al., 2005), and manure from laying hens (Leytem et al., 2006).

The finding that the majority of the P contained in the feces of pigs fed cereal grains was present in the form of phosphate may have important implications for the fate of the P in swine feces when this is applied to land as fertilizer. Phosphate is relatively soluble in soils, whereas phytate is retained and is less likely to be lost in runoff (Anderson et al., 1974; Leytem et al., 2002). Our findings indicated that the majority of the P in swine feces was present as the more soluble phosphate form rather than as phytate. Therefore, care should be taken when applying swine manure to land to ensure that surface and groundwater resources are not contaminated.

We examined only the effect of diet on P composition in fresh feces and not a mixture of feces and urine after prolonged storage. Typically, when swine diets are not in excess of the dietary P requirement, very little P is excreted in the urine because of reabsorption of P in the kidneys. It was reported previously that in swine, urine P was less than 0.5% of TP excreted (Baxter et al., 2003; Beaulieu et al., 2007); thus, the contribu-

tion from urine would be very small. Typically, manure (mixture of feces and urine) is stored for a period of time before land application. Baxter et al. (2003) reported that phytate in stored swine manure decreased with increasing storage time. Therefore, one would expect that storage of the manures in the present study would result in a further decrease in phytate content, possibly resulting in an increase in manure WSP. Additionally, Leytem et al. (2006) reported that once manures are incorporated into soils, the manure phytate will break down over time, releasing soluble P, thereby ameliorating differences in manure WSP attributable to phytate content. Although the feces from the swine fed the corn diet had greater concentrations of phytate, and therefore less soluble P, the breakdown of phytate in storage and soils will release soluble P over time and will therefore ameliorate any benefits of this reduction in WSP.

In summary, our results indicate that the majority of P in the feces of pigs fed cereal grains is present in the form of phosphate and only small amounts of phytate are contained in the excreta. The exception to this was the corn diet, in which 45% of the total fecal P was in the form of phytate. As the NDF content of the diets increased, there was a linear increase in apparent total-tract phytate digestibility, but no increase in apparent P digestibility.

LITERATURE CITED

Anderson, G. E., E. G. Williams, and J. O. Moir. 1974. A comparison of the sorption of inorganic orthophosphate and inositol hexaphosphate by six acid solids. J. Soil Sci. 25:51–62.

AOAC. 1990. Official Methods of Analysis. 15th ed. Assoc. Off. Anal. Chem., Washington, DC.

Ballam, G. C., T. S. Nelson, and L. K. Kirby. 1985. Effect of different dietary levels of calcium and phosphorus. Nutr. Rep. Int. 32:909–913.

Baxter, C. A., B. C. Joern, D. Ragland, J. S. Sands, and O. Adeola. 2003. Phytase, high-available-phosphorus corn, and storage effects on phosphorus levels in pig excreta. J. Environ. Qual. 32:1481–1489.

Beaulieu, A. D., M. R. Bedford, and J. F. Patience. 2007. Supplementing corn or corn-barley diets with an *E. coli* derived phytase decreases total and soluble P output by weanling and growing pigs. Can. J. Anim. Sci. 87:353–364.

Bruce, J. A. M., and F. Sundstøl. 1995. The effect of microbial phytase in diets for pigs on apparent ileal and faecal digestibility, pH, and flow of digesta measurements in growing pigs fed a high-fibre diet. Can. J. Anim. Sci. 75:121–127.

Canadian Council on Animal Care. 1993. Guide to the Care and Use of Experimental Animals. Vol. 1. 2nd ed. Can. Counc. Anim. Care, Ottawa, Ontario, Canada.

Chen, P. S., T. Y. Toribara, and H. Warner. 1956. Microdetermination of phosphorus. Anal. Chem. 28:1756–1758.

Crenshaw, T. D. 2001. Calcium, phosphorus, vitamin D, and vitamin K in swine nutrition. Pages 49–66 in Swine Nutrition. A. J. Lewis and L. L. Southern, ed. CRC Press, Boca Raton, FL.

Cromwell, G. L. 1992. The biological availability of phosphorus in feedstuffs for pigs. Pig News Inform. 13:75–78.

Dorsch, J. A., A. Cook, K. A. Young, J. M. Anderson, A. T. Bauman, C. J. Volkmann, P. P. N. Murthy, and V. Raboy. 2003. Seed phosphorus and inositol phosphate phenotype of barley *low phytic acid* genotypes. Phytochemistry 62:691–706.

Fenton, T. W., and M. Fenton. 1979. An improved procedure for the determination of chromic oxide in feed and faeces. Can. J. Anim. Sci. 59:631–634.

Grieshop, C. M., D. E. Reese, and G. C. Fahey Jr 2001. Nonstarch polysaccharides and oligosaccharides in swine nutrition. Pages 107–130 in Swine Nutrition. A. J. Lewis and L. L. Southern, ed. CRC Press, Boca Raton, FL.

Harvey, B. L., and B. G. Rossnagel. 1984. Harrington barley. Can. J. Plant Sci. 64:193–194.

Jongbloed, A. W., Z. Mroz, and P. A. Kemme. 1992. The effect of supplementary *Aspergillus niger* phytase in diets for pigs on concentration and apparent digestibility of DM, total phosphorus, and phytic acid in different sections of the alimentary tract. J. Anim. Sci. 70:1159–1168.

Kornegay, E. T., and A. F. Harper. 1997. Environmental nutrition: Nutritional management strategies to reduce nutrient excretion of swine. Prof. Anim. Sci. 13:99–111.

Leytem, A. B., R. L. Mikkelsen, and J. W. Gilliam. 2002. Sorption of organic phosphorus compounds in Atlantic Costal Plain soils. Soil Sci. 167:652–658.

Leytem, A. B., P. W. Plumstead, R. O. Maguire, P. Kwanyuen, and J. Brake. 2007. What aspect of dietary modification in broilers controls litter water soluble phosphorus: Dietary phosphorus, phytase, or calcium? J. Environ. Qual. 36:453–463.

Leytem, A. B., D. R. Smith, T. J. Applegate, and P. A. Thacker. 2006. The influence of manure phytic acid on phosphorus solubility in calcareous soils. Soil Sci. Soc. Am. J. 70:1629–1638.

Leytem, A. B., and P. A. Thacker. 2008. Fecal phosphorus excretion and characterization from swine fed diets containing a variety of cereal grains. J. Anim. Vet. Adv. 7:113–120.

Leytem, A. B., B. L. Turner, and P. A. Thacker. 2004. Phosphorus composition of manure from swine fed low-phytate grains: Evidence for hydrolysis in the animal. J. Environ. Qual. 33:2380–2383.

Maguire, R. O., Z. Dou, J. T. Sims, J. Brake, and B. C. Joern. 2005. Dietary strategies for reduced phosphorus excretion and improved water quality. J. Environ. Qual. 34:2093–2103.

Maguire, R. O., J. T. Sims, W. W. Saylor, B. L. Turner, R. Angel, and T. J. Applegate. 2004. Influence of phytase addition to poultry diets on phosphorus forms and solubility in litters and amended soils. J. Environ. Qual. 33:2306–2316.

NRC. 1998. Nutrient Requirements of Swine. 10th ed. Natl. Acad. Press, Washington, DC.

Pointillart, A. 1994. The importance of cereal phytases. Feed Mix 2:12–15.

Powers, W. J., E. R. Fritz, W. Fehr, and R. Angel. 2006. Total and water-soluble phosphorus excretion from swine fed low-phytate soybeans. J. Anim. Sci. 84:1907–1915.

Raboy, V., D. B. Dickinson, and M. G. Neuffer. 1990. A survey of maize kernel mutants for variation in phytic acid. Maydica 35:383–390.

Rapp, C., H. J. Lantzsch, and W. Drochner. 2001. Hydrolysis of phytic acid by intrinsic plant and supplemental microbial phytase (*Aspergillus niger*) in the stomach and small intestine of minipigs fitted with re-entrant cannulas 3. Hydrolysis of phytic acid (IP_6) and occurrence of hydrolysis products (IP_5, IP_4, IP_3 and IP_2). J. Anim. Physiol. A Anim. Nutr. 85:420–430.

Ravindran, V., G. Ravindran, and S. Sivalogan. 1994. Total and phytate phosphorus contents of various foods and feedstuffs of plant origin. Food Chem. 50:133–136.

Schneider, B. H., and W. P. Flatt. 1975. The Evaluation of Feeds Through Digestibility Experiments. Univ. Georgia Press, Athens.

Seynaeve, M., G. Janssens, M. Hesta, C. van Nevel, and R. O. de Wilde. 2000. Effects of dietary Ca/P ratio, P level and microbial phytase supplementation on nutrient digestibilities in growing pigs: Breakdown of phytic acid, partition of P and phytase activity along the intestinal tract. J. Anim. Physiol. Anim. Nutr. (Berl.) 83:193–204.

Sharpley, A., and B. Moyer. 2000. Phosphorus forms in manure and compost and their release during simulated rainfall. J. Environ. Qual. 29:1462–1469.

Thacker, P. A., and B. G. Rossnagel. 2005. Performance of growing-finishing pigs fed diets containing normal or low lignin-high fat oat supplemented or unsupplemented with enzyme. J. Anim. Vet. Adv. 4:681–687.

Thacker, P. A., B. G. Rossnagel, and V. Raboy. 2003. Phosphorus digestibility in low phytate barley fed to finishing pigs. Can. J. Anim. Sci. 83:101–104.

Toor, G. S., J. D. Peak, and J. T. Sims. 2005. Phosphorus speciation in broiler litter and turkey manure produced from modified diets. J. Environ. Qual. 34:687–697.

Turner, B. L. 2004. Optimizing phosphorus characterization in animal manures by phosphorus-31 nuclear magnetic resonance spectroscopy. J. Environ. Qual. 33:757–766.

Turner, B. L., and A. B. Leytem. 2004. Phosphorus compounds in sequential extracts of animal manures: Chemical speciation and a novel fractionation procedure. Environ. Sci. Technol. 38:6101–6108.

Turner, B. L., N. Mahieu, and L. M. Condron. 2003. Phosphorus-31 nuclear magnetic resonance spectral assignments of phosphorus compounds in soil NaOH-EDTA extracts. Soil Sci. Soc. Am. J. 67:497–510.

van der Klis, J. D., and H. A. J. Versteegh. 1996. Phosphorus nutrition of poultry. Pages 71–83 in Recent Developments in Poultry Nutrition. P. C. Garnsworthy, J. Wiseman, and W. Haresign, ed. Nottingham Univ. Press, Nottingham, UK.

Van Soest, P. J., J. B. Robertson, and B. A. Lewis. 1991. Methods for dietary fiber, neutral detergent fiber, and non-starch polysaccharides in relation to animal nutrition. J. Dairy Sci. 74:3583–3597.

Yi, Z., and E. T. Kornegay. 1996. Sites of phytase activity in the gastrointestinal tract of young pigs. Anim. Feed Sci. Technol. 61:361–368.

Zasoski, R. J., and R. G. Burau. 1977. A rapid nitric-perchloric acid digestion method for multi-element tissue analysis. Commun. Soil Sci. Plant Anal. 8:425–436.

References

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