RESEARCHREPORT

ANIMAL SCIENCE — Nutritional Efficiency Consortium

TITLE: Effects of exogenous phytase on the apparent total tract digestibility of energy and phosphorus, and on the standardized total tract digestibility of P in corn, DDGS, HP DDG, and corn germ – **NPB #10-073**

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I. INDUSTRY SUMMARY

The objective of this research was to measure the effects on energy and P digestibility of adding 4 levels of exogenous enzyme to corn, DDGS, HP DDG, and corn germ when fed to growing pigs. The 4 levels of phytase were 0, 500, 1000, and 1500 FTU per kg of diet. It was a further objective to develop regression equations for the inclusion of phytase in each ingredient, which will allow users to calculate the digestibility of P in the 4 ingredients regardless of the level of phytase in the diet. A broken line analysis was also conducted to identify the level of phytase in each ingredient that maximizes P digestibility.

Results of this research have considerable value to US swine producers because it allows producers to optimize the use of distillers co-products and take advantage of the high digestibility of P in some of the co-products from the ethanol industry. Our previous research has demonstrated that the digestibility of P in co-products that have been fermented is much greater than in non-fermented co-products and in corn. It is also established that the addition of exogenous phytase to diets improve P digestibility. We have also demonstrated that the combination of phytase and DDGS in diets fed to pigs results in a very high digestibility of P. However,

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there is no information on the optimum levels of phytase in diets containing fermented or non-fermented corn co-products and it is not known how much the inclusion of phytase and corn co-products can reduce the excretion of P in the manure from pigs. There is also no information on the effects of phytase on the digestibility of P in corn co-products other than DDGS.

The present research addressed these questions and for the first time provided data to the US swine industry that allow feed companies and producers to optimize the use of phytase in diets containing corn and corn co-products. By optimizing the utilization of P, diet costs will be reduced, which will lower production costs of pigs, and thus, improve the competitiveness of US pork. Results of this research also showed reduction in the excretion of P from the animals, which will allow producers to apply more manure per acre and thus reduce the costs associated with manure applications.

Results of this research can be implemented immediately so the savings to pork producers can be obtained without delay and without requiring producers to invest in additional technology on their operations.

SCIENTIFIC ABSTRACT:

An experiment was conducted to measure the effects of graded levels of microbial phytase on the digestibility of energy and on the standardized total tract digestibility (STTD) of P in corn, distillers dried grains with solubles (DDGS), high protein distillers dried grains (HP-DDG), and corn germ. A second objective was to develop regression equations to predict the response of adding phytase to each of these ingredients. Four corn based diets, 4 DDGS based diets, 4 HP-DDG based diets, and 4 corn germ based diets were formulated. The 4 diets with each ingredient contained 0, 500, 1,000, or 1,500 phytase units (FTU) per kg (Optiphos 2000, Enzyvia, Sheridan, IN). A P-free diet was also formulated to measure the basal endogenous losses of P. A total of 102 pigs (initial BW: 18.2 ± 2.1 kg) were individually housed in metabolism cages equipped with a feeder and a nipple drinker and a screen floor that allowed for total collection of feces. Pigs were allotted to the 17 diets in a randomized complete block design with 6 replicates per diet. Phytase did not have an effect on the digestibility of energy. Supplementation of microbial phytase increased (linear, $P < 0.01$; quadratic, $P < 0.05$) the STTD of P in corn from 40.9 to 67.5, 64.5, and 74.9%, tended to increase (linear, $P = 0.07$) the STTD of P in

DDGS from 76.9 to 82.9, 82.5, and 83.0%, increased (linear, $P < 0.01$; quadratic, $P < 0.05$) the STTD of P in HP-DDG from 77.1 to 88.0, 84.1, and 86.9% , and increased (linear and quadratic, $P < 0.01$) the STTD of P in corn germ from 40.7 to 59.0, 64.4, and 63.2% in diets supplemented with 0, 500, 1,000, or 1,500 FTU/kg of phytase, respectively. Regression equations were developed to allow the calculation of the STTD of P with any level of phytase (Optiphos 2000, Enzyvia, Sheridan, IN) for each of the test ingredients. Therefore, results of this experiment allow the prediction of the amount of digestible P in corn and corn germ containing any level of phytase between 0 and 1,500 FTU.

Key words: digestibility, energy, endogenous losses, energy, phosphorus, phytase, pig

INTRODUCTION:

Corn contains approximately 0.26% P (NRC, 1998), but the digestibility is low because most of the P in corn is bound to phytate, which is poorly digested by pigs (Selle and Ravindran, 2008). Phytate has also been shown to bind other nutrients such as proteins, AA, and starch (Selle et al., 2000). Supplementation of exogenous phytase to corn and soybean meal (**SBM**) results in improved P utilization (Almeida and Stein, 2010), but research showing the effects of exogenous phytase on the digestibility of other nutrients have been inconsistent (Adeola and Sands, 2003). The low digestibility of P in corn is compensated by supplementation of diets with inorganic P. This practice has become expensive because of increasing prices of inorganic P. In an attempt to mitigate this problem, Almeida and Stein (2010) measured standardized total tract digestibility (**STTD**) of P in corn, SBM, and distillers dried grains with solubles (**DDGS**) with 0 or 500 phytase units (**FTU**) per kg. It was concluded from this work that the STTD of P is increased in corn and SBM, but not in DDGS if 500 FTU of microbial phytase is added to the diets. There are, however, no data on the effects of adding microbial phytase to other corn co-products such as high protein distillers dried grains (**HP-DDG**) and corn germ. The effects of adding graded levels of exogenous phytase to corn, DDGS, HP-DDG, and corn germ have also not been reported and the inclusion rate of phytase that is needed to maximize the STTD of P in these ingredients is not known. The objectives of this experiment were to measure the effects of graded levels of phytase on the ATTD of energy and P, and on the STTD of P in corn, DDGS, HP-DDG, and corn germ. A second objective was to develop regression equations to predict the response of adding different levels of phytase to each ingredient on the STTD of P.

STATED OBJECTIVES FROM ORIGINAL PROPOSAL:

The objectives of this experiment were to measure the effects of graded levels of phytase on the ATTD of energy and P, and on the STTD of P in corn, DDGS, HP-DDG, and corn germ. A second objective was to develop regression equations to predict the response of adding different levels of phytase to each ingredient on the STTD of P.

MATERIALS AND METHODS:

The protocol for the experiment was reviewed and approved by the Institutional Animal Care and Use Committee at the University of Illinois. Pigs used in the experiment were the offspring of Landrace boars that were mated to Large White sows (Genetiporc, Alexandria, MN).

Diets, Animals, and Experimental Design

Distillers dried grains with solubles and corn germ were obtained from Poet Nutrition, Coon Rapids, IA, and HP-DDG was obtained from Poet Nutrition, Corning, IA (Table 1). A commercial hybrid of corn was obtained locally. Seventeen diets were formulated (Tables 2 and 3). There were 4 corn based diets, 4 DDGS based diets, 4 HP-DDG based diets, and 4 corn germ based diets. Phytase (Optiphos 2000, Enzyvia, Sheridan, IN) was added at levels of 0, 500, 1,000, or 1,500 phytase units (**FTU**) per kg to the 4 diets with each ingredient. A P-free diet was used to measure the basal endogenous P loss (**EPL**) from the pigs. A total of 102 growing pigs (initial BW: 18.2 ± 2.1 kg) were housed in metabolism cages equipped with a feeder and a nipple drinker that allowed for total collection of feces. Pigs were allotted to the 17 diets in a randomized complete block design. The Experimental Animal Allotment Program (Kim and Lindemann, 2007) was used to allot pigs to the 17 diets based on BW with 6 replicate pigs per diet.

Feeding and Sample Collection

The daily amount of feed provided to the pigs was calculated as 3 times the estimated requirement for maintenance energy (i.e., 106 kcal ME per kg $^{0.75}$; NRC, 1998) and fed in 2 equal meals. Pigs were allowed ad libitum access to water throughout the experiment. There was a 5 d adaptation period to the diets, which was followed by total collection of feces from d 6 to 11 according to the marker to marker approach (Adeola, 2001). In the morning meal of d 6, chromic oxide was added to the diets to determine the beginning of collections, and on d 11, ferric oxide was added to the diets to determine the end of collections. Fecal samples were collected twice daily and stored at -20˚C immediately after collection.

Sample Analysis and Data Processing

Prior to analysis, fecal samples were dried in a forced air oven and finely ground through a 2 mm screen using a Thomas-Wiley mill (Model 4, Swedesboro, NJ). After wet ash sample preparation (method 975.03; AOAC Int., 2007), fecal samples, ingredients, and diets were analyzed for Ca and P by inductively coupled plasma (ICP) spectroscopy (method 985.01; AOAC Int., 2007), for DM by oven drying at 135˚C for 2 h (method 930.15; AOAC Int., 2007) and for GE using bomb calorimetry (model 6300, Parr Instruments, Moline, IL). Ingredients and diets were also analyzed for CP (method 990.03; AOAC Int., 2007) using an Elementar Rapid N-cube protein/nitrogen apparatus (Elementar Americas Inc., Mt. Laurel, NJ), ADF (method 973.18; AOAC Int., 2007), NDF (Holst, 1973), and for phytase activity (Phytex Method, version 1; Eurofins, Des Moines, IA). Ingredients were also analyzed for AA (method 982.30 E (a, b, c); AOAC Int., 2007) and for phytate (Eurofins, Des Moines, IA) using the method of Ellis et al. (1977).

The ATTD (%) of GE and P in each diet was calculated according to the following equation:

$$ATTD = [(Ni – Nf)/Ni] \times 100\%,$$

where Ni is the total energy (kcal) or P intake (g) from d 6 to 11 and Nf is the total fecal energy (kcal) or P output (g) originating from the feed that was provided from d 6 to 11 (Pedersen et al., 2007).

The basal EPL (mg/kg DMI) was measured from pigs fed the P-free diet according to the following equation:

$$\text{EPL (mg/kg DMI)} = ([\text{Pf/Fi}] \times 1{,}000 \times 1{,}000),$$

where EPL is the endogenous P loss and Fi is the total feed (g) intake from d 6 to 11 (Petersen and Stein, 2006).

Values for the daily EPL of pigs fed the P containing diets were calculated by multiplying the basal EPL by the average daily DMI of each pig during the 5 d collection period.

The STTD of P was calculated using the following equation:

$$\text{STTD (\%)} = ([\text{Pi} - \{\text{Pf} - \text{EPL}\}/\text{Pi}]) \times 100,$$

where STTD (%) is the standardized total tract digestibility of P.

Data were analyzed as a randomized complete block design using the Proc Mixed Procedure in SAS. The UNIVARIATE procedure in SAS was used to confirm that variances were homogenous and also to analyze for outliers, but no outliers were identified. The model included diet as the fixed effect and replicate as a random effect. No effects of replicate were observed, and therefore, replicate was removed from the final model. The effects of adding graded levels of phytase to each ingredient were analyzed by orthogonal polynomial contrasts. Appropriate coefficients for unequally spaced concentrations of supplemental phytase were obtained using the Proc IML of SAS. Estimates for the regression equations between STTD of P and analyzed phytase inclusion level were determined by submitting each treatment observations to Proc GLM of SAS. The pig was the experimental unit and an alpha value of 0.05 was used to assess significance among means.

RESULTS:

Throughout the adaptation period pigs remained healthy and readily consumed their diets. During the 5 d collection period, however, 1 pig that was fed the corn germ diet without phytase was diagnosed with pneumonia, and therefore, removed from the experiment. All of the remaining pigs were healthy until the end of the experiment.

The analyzed values for phytase activity of all diets were slightly less than the calculated values (Table 3). Because of this discrepancy, analyzed values for phytase activity were used in all statistical analysis. The basal EPL was measured at 206 mg/kg DMI from pigs fed the P-free diet.

Digestibility of Energy in Corn and Corn Co-Products

No differences in GE intake were observed among diets, however, exogenous phytase supplementation to the corn diet increased (P < 0.01) linearly the GE intake (Table 4). The concentration of GE in feces was not different among treatments. No linear or quadratic effects of phytase supplementation on the ATTD of GE were detected in any of the ingredients.

Digestibility of P in Corn

No differences in fecal output were detected among treatments (Table 5). Phytase increased (linear, P < 0.05) ADFI, P intake and EPL. The concentration of P in feces was reduced (linear and quadratic, P < 0.01) from 1.94 to 1.25, 1.15, and 0.94% for pigs that were fed diets that were supplemented with 0, 420, 720, or 1,100 FTU of phytase, respectively. Likewise, daily P output was linearly (P < 0.01) and quadratically (P < 0.05) reduced from 1.0 to 0.6, 0.7, and 0.6 g/d for pigs that were fed diets that were supplemented with 0, 420, 720, or 1,100 FTU of phytase, respectively. The ATTD and STTD of P were linearly (P < 0.01) and quadratically (P < 0.05) increased from 33.5 to 60.1, 57.0, and 67.4%, and from 40.9 to 67.5, 64.5, and 74.9% for pigs that were fed diets that were supplemented with 0, 420, 720, or 1,100 FTU of phytase, respectively.

Digestibility of P in DDGS

No differences in ADFI, P intake, daily fecal output, or EPL were observed among treatments. Concentration of P in feces decreased (linear, P < 0.01) as graded levels of phytase were added to the diets. Likewise, the daily P output was reduced (P < 0.05) by the addition of phytase to the diets. Addition of phytase to the diets tended (linear, P = 0.08) to increase the ATTD and STTD of P from 72.6 to 78.6, 78.2, and 78.6%, and from 76.9 to 82.9, 82.5, and 83.0% in pigs that were fed diets containing 130, 430, 770, or 1,100 FTU of phytase, respectively.

Digestibility of P in HP-DDG

There were no differences in ADFI, P intake, daily fecal output, or EPL among diets. Addition of phytase to the diets reduced (linear, $P < 0.01$; quadratic, $P < 0.05$) the concentration of P in feces. Phytase also reduced (linear, $P < 0.01$; quadratic, $P < 0.05$) P excretion from 0.4 to 0.3, 0.3, and 0.3 g/d in pigs that were fed diets containing 0, 500, 770, or 1,100 FTU of phytase, respectively. Addition of phytase to the diets increased (linear, $P < 0.01$; quadratic, $P < 0.05$) the ATTD of P from 68.6 to 79.5, 75.6, and 78.4%, and the STTD of P from 77.1 to 88.0, 84.1, and 86.9% in pigs that were fed diets containing 0, 500, 770, or 1,100 FTU of phytase, respectively.

Digestibility of P in Corn Germ

There were no differences in ADFI, P intake, daily fecal output, and EPL among treatments. Addition of phytase to the diets reduced (linear and quadratic, $P < 0.01$) the concentration of P in the feces. Likewise, addition of phytase to the diets reduced (linear and quadratic, $P < 0.01$) the daily P output from 2.2 to 1.6, 1.3, and 1.4 g/d for pigs that were fed diets containing 110, 390, 910, and 1,400 FTU of phytase, respectively. Addition of phytase to the diets increased (linear and quadratic, $P < 0.01$) the ATTD of P from 37.3 to 55.7, 63.0, and 59.8%, and the STTD of P from 40.7 to 59.0, 64.4, and 63.2% in pigs that were fed diets containing 110, 390, 910, and 1,400 FTU of phytase, respectively.

Regression Equations

Regression equations for the STTD of P as affected by microbial phytase in corn, HP-DDG, and corn germ are presented in Table 6. Addition of phytase to DDGS did not affect the STTD of P, and therefore, a regression equation for this ingredient is not presented.

DISCUSSION:

Effects of Phytase on the ATTD of GE

The concentration of GE in corn that was measured in the present experiment is in agreement with previously published value by Pedersen et al., (2007). The fact that phytase did not improve the ATTD of GE in the present experiment is in agreement with Liao et al., (2005) who showed no effects of phytase on the concentration of DE in corn-SBM, wheat-SBM, or barley-peas-canola meal diets. In their study, however, supplementation of phytase to wheat-SBM-canola meal diet tended ($P = 0.067$) to increase the concentration of DE. Phytate present in feed ingredients may form complexes with starch, and therefore, decrease the digestibility of this nutrient (Liao et al., 2002). This hypothesis, however, was not confirmed in the present experiment.

Phosphorus and Phytate P in Ingredients

The concentration of P that was measured in ingredients used in this experiment is in agreement with the values reported by Widmer et al. (2007) and NRC (1998). The phytic acid data demonstrates that on a percentage basis, corn and corn germ contain similar amounts of phytic acid (72 and 76% of total P, respectively), and that DDGS and HP-DDG also have similar concentrations of phytic acid, but these amounts are much less (30 and 28% of total P, respectively) than in corn and corn germ. The reason for these differences is that during the production of DDGS and HP-DDG, some of the phytic acid is hydrolyzed, which results in a reduced concentration of phytate-bound P.

ATTD and STTD Values of P in Ingredients without Phytase

The value for the ATTD of P in corn measured in the present experiment is in agreement with values reported by Bünzen et al. (2008) and Stein et al. (2009). The ATTD of P in corn germ was greater than the value reported by Widmer et al. (2007). This difference may be due to different processing of corn to obtain corn germ, or differences between corn hybrids that were used to obtain the corn germ. The ATTD of P in DDGS has been reported to vary from 50 to 69% (Pedersen et al., 2007; Almeida and Stein, 2009; Stein et al., 2009). In the present experiment, the value for the ATTD of P in DDGS is close to the greatest value in this range. This observation confirms that the ATTD of P in DDGS is much greater than in corn, which is likely a consequence of the reduced concentration of phytate in DDGS compared with corn. The ATTD of P in HP-DDG measured in this experiment is similar to the ATTD of P in DDGS, and slightly greater than the value reported by Widmer et al. (2007). This observation indicates that the ATTD of P in HP-DDG and DDGS are similar as would be expected from the similar content of phytate in these 2 ingredients. We are not aware of any other experiments in which the ATTD of P in these 2 ingredients has been measured.

The value for the basal EPL that was measured in this experiment is very close to previous values (Stein et al., 2006; Widmer et al., 2007; Almeida and Stein, 2009). Values for the STTD of P are expected to be greater than the ATTD values because they are corrected for basal EPL, and results from the present experiment confirm this hypothesis. If values for the ATTD of P are corrected for total EPL, true total tract digestibility values are obtained and these values are expected to be greater than STTD values. The value calculated for the STTD of P in corn in the present experiment is in agreement with this theory because it is within the range for the ATTD (31.9%) and true total tract digestibility (49.2%) of P in corn reported by Stein et al. (2009) and Wu et al. (2008), respectively. The value for the STTD of P in DDGS is in agreement with the value reported by Almeida and Stein, (2009). Likewise, the STTD of P in HP-DDG and in corn germ measured in the present experiment is in agreement with the STTD of P that was measured in HP-DDG and in corn germ by Widmer et al. (2007).

Effects of Phytase

Supplementation of diets with phytase resulted in increased STTD of P, which was expected because hydrolysis of phytate by microbial phytase liberates P in the intestines of pigs, and therefore, improves the digestibility of P (Selle and Ravindran, 2008). Phytase increases the STTD of P in corn and soybean meal (Almeida and Stein, 2009), but to our knowledge, there are no data on the effects of graded levels of phytase on the STTD of P in corn, DDGS, HP-DDG, and corn germ. Although the objective of this experiment was not to compare the STTD of P among ingredients, our data suggest that the effect of phytase on the STTD of P in corn and corn germ seems to be greater than the effect of phytase on the STTD of P in DDGS and HP-DDG. Corn and corn germ contain greater amounts of phytate than DDGS and HP-DDG. During the processing of corn to obtain ethanol, it is possible that some of the phytate present in corn is hydrolyzed. Therefore, the lack of substrate in DDGS and HP-DDG may inhibit the effectiveness of phytase in improving the STTD of P in these ingredients, which is not the case in corn and corn germ.

Regression Equations

The regression equations determined in this experiment allow calculation of the STTD of P with any level of phytase between 0 and 1,500 FTU (Optiphos 2000, Enzyvia, Sheridan, IN). The r^2 value for the regression equation of HP-DDG indicates that the model accounts for only 36% of the variation of the STTD of P, and that the relationship between graded levels of phytase and the STTD of P in HP-DDG is unlikely to be linear or quadratic. As a result, it is possible to predict the amount of digestible P in corn or corn germ containing different levels of phytase, but the regression equation for HP-DDG may not be adequate to predict the effect of phytase on the STTD of P. This suggests that the effect of phytase on the STTD of P in HP-DDG needs further investigation.

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TABLES:

Table 1. Analyzed nutrient composition of ingredients

	Ingredient			
Item	Corn	DDGS[1]	HP-DDG[2]	Corn germ
Ca, %	0.01	0.02	0.01	0.02
Total P, %	0.25	0.85	0.39	1.41
Phytate P, %	0.18	0.26	0.11	1.07
Non-phytate P[3], %	0.07	0.59	0.28	0.33
DM, %	86.67	91.18	91.35	91.45
CP, %	7.27	26.41	38.09	15.36
ADF	3.16	7.30	14.37	8.00
NDF	17.40	28.93	27.68	28.87
Phytase, FTU/kg[4]	< 70	< 70	< 70	180
Indispensable AA, %				
Arg	0.34	1.16	1.37	1.07
His	0.19	0.71	1.00	0.41
Ile	0.24	1.00	1.61	0.45
Leu	0.74	2.84	5.16	1.02
Lys	0.24	0.87	1.05	0.80
Met	0.14	0.51	0.80	0.25
Phe	0.32	1.18	2.08	0.58
Thr	0.23	0.98	1.34	0.51
Trp	0.05	0.20	0.22	0.10

Val	0.33	1.32	1.95	0.73
Dispensable AA, %				
Ala	0.47	1.78	2.82	0.88
Asp	0.44	1.67	2.35	1.10
Cys	0.16	0.57	0.76	0.30
Glu	1.09	3.51	6.10	1.79
Gly	0.28	1.05	1.26	0.77
Pro	0.53	1.87	3.23	0.90
Ser	0.29	1.13	1.60	0.58
Tyr	0.21	0.95	1.51	0.40

[1]DDGS = distillers dried grains with solubles.

[2] HP-DDG = high protein distillers dried grains.

[3]Calculated as the difference between phytate-P and total P.

[4]FTU = phytase units per kg.

Table 2. Ingredient composition of basal diets (as-fed basis)[1]

Ingredient, %	Corn	DDGS[2]	HP-DDG[2]	Corn germ	P-free
Ground corn	97.00	-	-	-	-
DDGS	-	50.00	-	-	-
HP-DDG	-	-	50.00	-	-
Corn germ	-	-	-	40.00	-
Sugar	-	20.00	20.00	20.00	-
Soybean oil	1.00	-	1.00	-	4.00
Ground limestone	1.20	1.20	1.20	1.20	0.80
Salt	0.40	0.40	0.40	0.40	0.40
Vitamin mineral premix[3]	0.30	0.30	0.30	0.30	0.30
Cornstarch	0.10	28.10	27.10	38.10	49.22
Potassium carbonate	-	-	-	-	0.40
Magnesium oxide	-	-	-	-	0.10
Solka floc[4]	-	-	-	-	4.00
Gelatin[5]	-	-	-	-	20.00
AA mixture[6]	-	-	-	-	0.78

[1]For each ingredient, 3 additional diets were formulated by adding 0.025, 0.050, and 0.075% of phytase (Optiphos 2000, Enzyvia, Sheridan, IN) at the expense of cornstarch. These levels of Optiphos were expected to create diets containing 500, 1,000, or 1,500 phytase units per kg.

[2] DDGS = distillers dried grains with solubles; HP-DDG = high protein distillers dried grains

[3] The vitamin-micromineral premix provided the following quantities of vitamins and micro minerals per kilogram of complete diet: Vitamin A, 11,128 IU; vitamin D_3, 2,204 IU; vitamin E, 66 IU; vitamin K, 1.42 mg; thiamin, 0.24 mg; riboflavin, 6.58 mg; pyridoxine, 0.24 mg; vitamin B_{12}, 0.03 mg; D-pantothenic acid, 23.5 mg; niacin, 44 mg; folic acid, 1.58 mg; biotin, 0.44 mg; Cu, 10 mg as copper sulfate; Fe, 125 mg as iron sulfate; I, 1.26 mg as potassium iodate; Mn, 60 mg as manganese sulfate; Se, 0.3 mg as sodium selenite; and Zn, 100 mg as zinc oxide.

[4] Fiber Sales and Development Corp., Urbana, OH.

[5] Pork gelatin obtained from Gelita Gelatine USA Inc., Sioux City, IA.

[6] Provided the following quantities (%) of AA per kg of complete diet: DL-methionine, 0.27; L-threonine, 0.08; L-tryptophan, 0.14; L-histidine, 0.08; L-isoleucine, 0.16; and L-valine, 0.05.

Table 3. Analyzed nutrient composition of diets (as-fed basis)

	Corn				DDGS[1]				HP-DDG[2]				Corn Germ				P-free
Phytase[3]	0	500	1000	1500	0	500	1000	1500	0	500	1000	1500	0	500	1000	1500	
Item																	
GE, kcal/kg	3,907	3,916	3,985	3,968	4,357	4,281	4,373	4,369	4,419	4,452	4,473	4,432	4,111	4,091	4,100	3,981	4,065
Ca, %	0.50	0.50	0.54	0.53	0.37	0.46	0.63	0.67	0.45	0.58	0.71	0.49	0.47	0.50	0.56	0.60	-
P, %	0.24	0.24	0.24	0.24	0.45	0.45	0.45	0.45	0.23	0.23	0.23	0.23	0.56	0.56	0.56	0.56	-
DM, %	86.70	86.85	86.91	86.93	92.57	92.81	93.27	94.08	93.32	93.07	93.43	93.39	93.07	93.21	92.01	92.31	91.59
CP, %	6.89	7.05	6.86	7.20	14.26	14.08	14.50	13.89	20.10	19.89	20.88	20.82	5.85	7.20	6.71	7.21	22.12
ADF	2.78	2.61	2.77	2.77	3.39	3.53	3.69	4.09	6.37	6.17	7.43	6.33	2.89	3.96	3.34	3.51	-
NDF	15.26	17.30	20.22	17.50	13.88	14.92	15.25	15.96	16.49	13.96	14.82	15.02	12.74	16.88	15.27	11.77	-
Phytase[3]	< 70	420	720	1,100	130	430	770	1,100	< 70	500	770	1,100	110	390	910	1,400	-

[1] DDGS = distillers dried grains with solubles.
[2] HP-DDG = high protein distillers dried grains.
[3] Expressed as phytase units per kg.

Table 4. Effects of phytase on the apparent total tract digestibility (ATTD) of GE in corn, distillers dried grains with solubles (DDGS), high protein distillers dried grains (HP-DDG), and corn germ[1]

Item	GE intake, kcal	GE in feces, kcal	ATTD of GE,%
Corn			
Corn + 0 FTU/kg	12,661	1,375	89.18
Corn + 500 FTU/kg	12,730	1,345	89.53
Corn + 1000 FTU/kg	12,938	1,560	87.98
Corn + 1500 FTU/kg	13,970	1,584	88.66
SEM	546.9	120.2	0.76
P, Linear	< 0.01	0.15	0.40
P, Quadratic	0.13	0.78	0.87
DDGS			
DDGS + 0 FTU/kg	15,668	2,250	85.65
DDGS + 500 FTU/kg	14,850	2,163	85.26
DDGS + 1000 FTU/kg	15,805	2,383	85.09
DDGS + 1500 FTU/kg	15,805	2,350	85.02
SEM	906.0	123.7	1.06
P, Linear	0.48	0.35	0.56
P, Quadratic	0.58	0.87	0.82
HP-DDG			
HP-DDG + 0 FTU/kg	12,688	1,539	87.84

HP-DDG + 500 FTU/kg	13,079	1,477	88.70
HP-DDG + 1000 FTU/kg	12,567	1,467	88.33
HP-DDG + 1500 FTU/kg	13,353	1,583	88.12
SEM	277.1	74.8	0.59
P, Linear	0.23	0.81	0.76
P, Quadratic	0.52	0.25	0.36
Corn germ			
Corn germ + 0 FTU/kg[2]	12,823	1,744	86.43
Corn germ + 500 FTU/kg	12,858	1,633	87.33
Corn germ + 1000 FTU/kg	12,928	1,621	87.46
Corn germ + 1500 FTU/kg	12,520	1,646	86.85
SEM	358.9	115.3	0.67
P, Linear	0.37	0.48	0.72
P, Quadratic	0.32	0.41	0.22

[1] Data are means of 6 observations per treatment.

[2] Data are means of 5 observations.

Table 5. Effects of phytase on P-balance, apparent total tract digestibility (ATTD), and standardized total tract digestibility (STTD) of P in corn, distillers dried grains with solubles (DDGS), high protein distillers dried grains (HP-DDG), and corn germ[1]

Item	Feed intake, g/d	P intake, g/d	Fecal output, g/d	P in feces, %	P output, g/d	ATTD of P, %	Endogenous P, mg/d[2]	STTD of P, %
Corn								
Corn + 0 FTU/kg	562	1.56	53.48	1.94	1.0	33.5	115.52	40.9
Corn + 500 FTU/kg	565	1.56	50.26	1.25	0.6	60.1	116.32	67.5
Corn + 1000 FTU/kg	564	1.56	58.94	1.15	0.7	57.0	116.24	64.5
Corn + 1500 FTU/kg	612	1.69	58.59	0.94	0.6	67.4	126.07	74.9
SEM	24.13	0.07	4.14	0.08	0.08	3.68	4.97	3.68
P, Linear	0.03	0.03	0.21	< 0.01	< 0.01	< 0.01	0.03	< 0.01
P, Quadratic	0.12	0.12	0.73	< 0.01	0.03	0.04	0.12	0.04
DDGS								
DDGS + 0 FTU/kg	666	3.20	92.28	0.95	0.88	72.6	137.14	76.9
DDGS + 500 FTU/kg	644	3.09	88.72	0.74	0.66	78.6	132.63	82.9
DDGS + 1000 FTU/kg	682	3.25	95.08	0.75	0.72	78.2	140.56	82.5

DDGS + 1500 FTU/kg	680	3.22	94.03	0.71	0.68	78.6	140.21	83.0
SEM	39.02	0.19	4.88	0.06	0.07	2.09	8.03	2.09
P, Linear	0.40	0.64	0.60	< 0.01	0.11	0.08	0.40	0.07
P, Quadratic	0.64	0.72	0.80	0.13	0.21	0.20	0.65	0.20
HP-DDG								
HP-DDG + 0 FTU/kg	536	1.29	53.91	0.75	0.4	68.6	110.39	77.1
HP-DDG + 500 FTU/kg	547	1.32	52.29	0.52	0.3	79.5	112.64	88.0
HP-DDG + 1000 FTU/kg	525	1.27	52.00	0.59	0.3	75.6	108.14	84.1
HP-DDG + 1500 FTU/kg	563	1.36	54.77	0.53	0.3	78.4	115.91	86.9
SEM	11.60	0.03	2.59	0.06	0.03	2.57	2.39	2.57
P, Linear	0.27	0.31	0.84	< 0.01	< 0.01	< 0.01	0.27	< 0.01
P, Quadratic	0.26	0.27	0.41	0.03	0.02	0.04	0.26	0.04
Corn germ								
Corn germ + 0 FTU/kg[3]	581	3.48	70.14	3.14	2.2	37.3	119.61	40.7
Corn germ + 500 FTU/kg	586	3.50	61.47	2.57	1.6	55.7	120.68	59.0
Corn germ + 1000	580	3.52	61.93	2.11	1.3	63.0	119.54	64.4

FTU/kg								
Corn germ + 1500 FTU/kg	581	3.51	60.26	2.35	1.4	59.8	119.62	63.2
SEM	15.80	0.09	3.67	0.16	0.11	2.37	3.25	2.21
P, Linear	0.91	0.69	0.43	< 0.01	< 0.01	< 0.01	0.91	< 0.01
P, Quadratic	0.81	0.76	0.25	0.02	< 0.01	< 0.01	0.81	< 0.01

[1] Data are means of 6 observations per treatment.

[2] Calculated by multiplying the basal EPL (206 mg/kg DMI) measured from pigs fed P-free diet by the DMI of each individual pig fed P containing diets.

[3] Data are means of 5 observations.

Table 6. Regression equations for the STTD of P in corn, HP-DDG, and corn germ against phytase units (FTU) per kilogram of ingredient.

Item	Regression equation	r^2	*P*-value
Corn	$42.34 + 0.059FTU – 0.000028FTU^2$	0.63	< 0.01
HP-DDG	$77.55 + 0.023FTU – 0.000014FTU^2$	0.36	< 0.01
Corn germ	$34.50 + 0.067FTU – 0.000034FTU^2$	0.79	< 0.01