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# Effects of graded levels of microbial phytase on the standardized total tract digestibility of phosphorus in corn and corn coproducts fed to pigs<sup>1</sup>

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**ABSTRACT:** An experiment was conducted to determine the influence of adding graded levels of microbial phytase to corn, distillers dried grains with solubles (DDGS), high-protein distillers dried grains (HP-DDG), and corn germ on the standardized total tract digestibility (STTD) of P. 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 were formulated to contain 0, 500, 1,000, or 1,500 phytase units (FTU)/kg. A P-free diet was also formulated to determine basal endogenous losses of P. A total of 102 pigs (initial BW:  $18.2 \pm 2.1$  kg) were individually housed in metabolism cages equipped with a feeder, 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 replicate pigs per diet. Pigs were fed their respective diets for 12 d, and feces were collected quantitatively from d 6 to 11. Supplementation with 500, 1,000, or 1,500 FTU of microbial phytase/kg 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%, respectively, tended to increase (linear,  $P = 0.07$ ) the STTD of P in DDGS from 76.9 to 82.9, 82.5, and 83.0%, respectively, 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%, respectively, 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%, respectively. Regression equations were developed to calculate the STTD of P in corn and corn germ, and  $R^2$  values were 0.63 and 0.79, respectively. However, for DDGS and HP-DDG, the  $R^2$  values were only 0.20 and 0.36, respectively, and these equations were, therefore, not considered adequate to predict the STTD of P. In conclusion, the increase in the STTD of P in corn and corn germ that is a result of microbial phytase can be predicted by regression equations, but microbial phytase has much less of an effect on the STTD of P in DDGS and HP-DDG and responses to addition of graded levels of phytase to these ingredients can, therefore, not be accurately predicted by regression equations.

**Key words:** digestibility, endogenous loss, phosphorus, phytase, pig, standardized total tract digestibility

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## INTRODUCTION

Corn contains approximately 0.26% P (NRC, 1998), but the digestibility is poor because most of the P in corn is bound to phytate, which is poorly digested by pigs (Selle and Ravindran, 2008). The poor digestibility of P in corn is compensated by supplementation of diets fed to pigs with inorganic P, but this has become

expensive because of increasing costs of inorganic P. In an attempt to ameliorate this problem, Almeida and Stein (2010) determined standardized total tract digestibility (STTD) of P in corn, soybean meal (SBM), and distillers dried grains with solubles (DDGS) with 0 or 500 phytase units (FTU) per kilogram. 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 effects of adding microbial phytase to other corn coproducts such as high-protein distillers dried grains (HP-DDG) and corn germ, and it is not known if addition of more than 500 FTU/kg of phytase to DDGS improves the STTD of P. The effects of adding graded levels of exogenous phytase to corn, DDGS, HP-DDG, and corn germ have also not been re-

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**Table 1.** Analyzed nutrient composition of corn, distillers dried grains with solubles (DDGS), high-protein distillers dried grain (HP-DDG), and corn germ

Item	Ingredient <sup>1</sup>			
	Corn	DDGS	HP-DDG	Corn germ
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
Ca, %	0.01	0.02	0.01	0.02
Total P, %	0.25	0.85	0.39	1.41
Phytate, %	0.64	0.91	0.41	3.80
Phytate P, <sup>2</sup> %	0.18	0.26	0.11	1.07
Nonphytate P, <sup>3</sup> %	0.07	0.59	0.28	0.33
Phytase, units/kg	<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

<sup>1</sup>Distillers dried grains with solubles was obtained from Poet Nutrition, Corning, IA, and HP-DDG and corn germ were obtained from Poet Nutrition, Coon Rapids, IA.

<sup>2</sup>Calculated as 28.2% of phytate (Tran and Sauvant, 2004).

<sup>3</sup>Calculated as the difference between phytate P and total P.

ported, 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, therefore, were to determine the influence of adding graded levels of microbial phytase to corn, DDGS, HP-DDG, and corn germ on the STTD of P and to develop regression equations to predict the response of adding different amounts of phytase to each ingredient.

## MATERIALS AND METHODS

The protocol for the experiment was reviewed and approved by the Institutional Animal Care and Use Committee of 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*

A commercial hybrid of corn was obtained locally. High-protein distillers dried grains and corn germ were obtained from Poet Nutrition, Coon Rapids, IA, and

DDGS was obtained from Poet Nutrition, Corning, IA (Table 1). Corn germ and HP-DGG are produced by some ethanol plants that remove the hulls and the germ from the corn before fermentation. The endosperm is then used for ethanol production, but because of the removal of hulls and germ before fermentation, the distilled grains that are produced from this process contain more CP and less fat and fiber than traditional DDGS and are, therefore, called HP-DDG. The hulls that are produced are usually fed to ruminant animals, but the corn germ is available for feeding pigs.

Four corn-based diets, 4 DDGS-based diets, 4 HP-DDG-based diets, and 4 corn germ-based diets were formulated (Tables 2 and 3). Phytase (Optiphos 2000, Enzyvia, Sheridan, IN) was added to supply 0, 500, 1,000, or 1,500 FTU per kilogram to the 4 diets with each ingredient. A P-free diet was used to measure the basal endogenous P loss from the pigs. A total of 102 growing pigs (initial BW = 18.2 ± 2.1 kg) were housed in metabolism cages that were equipped with a feeder, a nipple drinker, and a screen floor that allowed for total collection of feces. The Experimental Animal Allotment Program (Kim and Lindemann, 2007) was used

**Table 2.** Ingredient composition of basal diets (as-fed basis)<sup>1</sup>

Ingredient, %	Corn	DDGS <sup>2</sup>	HP-DDG <sup>2</sup>	Corn germ <sup>2</sup>	P free
Ground corn	97.00	—	—	—	—
Coproducts	—	50.00	50.00	40.00	—
Sucrose	—	20.00	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 <sup>3</sup>	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 <sup>4</sup>	—	—	—	—	4.00
Gelatin <sup>5</sup>	—	—	—	—	20.00
AA mixture <sup>6</sup>	—	—	—	—	0.78

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

<sup>2</sup>DDGS = distillers dried grains with solubles; HP-DDG = high-protein distillers dried grains; DDGS was obtained from Poet Nutrition, Corn- ing, IA, and HP-DDG and corn germ were obtained from Poet Nutrition, Coon Rapids, IA.

<sup>3</sup>Provided the following quantities of vitamins and microminerals per kilogram of complete diet: vitamin A as retinyl acetate, 11,128 IU; vitamin D<sub>3</sub> as cholecalciferol, 2,204 IU; vitamin E as DL- $\alpha$  tocopheryl acetate, 66 IU; vitamin K as menadione nicotinamide bisulfite, 1.42 mg; thiamine as thiamine mononitrate, 0.24 mg; riboflavin, 6.58 mg; pyridoxine as pyridoxine hydrochloride, 0.24 mg; vitamin B<sub>12</sub>, 0.03 mg; D-pantothenic acid as D-calcium pantothenate, 23.5 mg; niacin as nicotinamide and nicotinic acid, 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.

<sup>4</sup>Fiber Sales and Development Corp., Urbana, OH.

<sup>5</sup>Pork gelatin obtained from Gelita Gelatine USA Inc., Sioux City, IA.

<sup>6</sup>Provided the following quantities (%) of AA per kilogram 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.

to allot pigs to the 17 diets in a randomized complete block design based on BW with 6 replicate pigs per diet.

### Feeding and Sample Collection

Pigs were fed their assigned diets for 12 d. The daily amount of feed provided to the pigs was calculated as 3 times the estimated requirement for maintenance energy (i.e., 106 kcal of ME/kg<sup>0.75</sup>; NRC, 1998) and provided in 2 equal meals at 0700 and 1700 h. 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 at 0730 and 1730 h and stored at -20°C immediately after collection.

### Sample Analyses and Data Processing

Before analysis, fecal samples were dried in a forced-air oven and finely ground through a 2-mm screen (model 4, Thomas-Wiley, Swedesboro, NJ). After wet ash sample preparation (method 975.03; AOAC International, 2007), fecal samples, ingredients, and diets were analyzed for Ca and P by inductively coupled plasma spectroscopy (method 985.01; AOAC Interna-

tional, 2007) and for DM by oven drying at 135°C for 2 h (method 930.15; AOAC International, 2007). Ingredients and diets were also analyzed for CP (method 990.03; AOAC International, 2007) using a rapid N-cube protein/nitrogen apparatus (Elementar Americas Inc., Mt. Laurel, NJ), ADF (method 973.18; AOAC International, 2007), NDF (Holst, 1973), and phytase activity (Phytex Method, version 1, Eurofins, Des Moines, IA). Ingredients were also analyzed for AA [method 982.30 E (a, b, c); AOAC International, 2007] and for phytate (Eurofins) using the method of Ellis et al. (1977).

The apparent total tract digestibility (ATTD) and STTD of P and the endogenous P loss in each diet were calculated as described previously (Almeida and Stein, 2010). The concentration of phytate bound P in each ingredient was calculated as 28.2% of phytate (Tran and Sauvant, 2004), and nonphytate P was calculated as the difference between the concentration of total P and phytate-bound P.

Data were analyzed using the MIXED procedure (SAS Inst. Inc., Cary, NC). 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. Mean values of all treatments were calculated using the LSMEANS procedure. Effects of adding graded levels of phytase to each ingredient were analyzed by orthogonal polynomial contrasts. Analyzed concentrations of phytase in the diets were used in these calculations, and appropriate coefficients for unequally spaced concentrations of

Table 3. Analyzed nutrient composition of diets (as-fed basis)<sup>1</sup>

Item	Corn					DDGS <sup>2</sup>					HP-DDG <sup>2</sup>					Corn germ <sup>2</sup>				
	<70	420	720	1,100	130	430	770	1,100	<70	500	770	1,100	110	390	910	1,400	P			
	FTU/kg	FTU/kg	FTU/kg	FTU/kg	FTU/kg	FTU/kg	FTU/kg	FTU/kg	FTU/kg	FTU/kg	FTU/kg	FTU/kg	FTU/kg	FTU/kg	FTU/kg	FTU/kg	free			
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	—			
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	—			

<sup>1</sup>Phytase expressed as phytase units (FTU) per kilogram.

<sup>2</sup>DDGS = distillers dried grains with solubles; HP-DDG = high-protein distillers dried grains; DDGS was obtained from Poet Nutrition, Corning, IA, and HP-DDG and corn germ were obtained from Poet Nutrition, Coon Rapids, IA.

supplemental phytase were obtained using the interactive matrix language procedure in SAS. Regression equations to estimate the relationship between STTD of P and microbial phytase inclusion were generated by the REG procedure in SAS. The pig was the experimental unit, and an  $\alpha$ -value of 0.05 was used to assess significance among means. If the  $P$ -value was between 0.05 and 0.10, the difference was considered a trend.

## 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 failed to consume its allotted ration, and therefore, was removed from the experiment. All other pigs remained healthy until the conclusion of the experiment and consumed their diets.

The analyzed values for phytase activity of most diets were slightly less than the calculated values (Table 3). Because of this discrepancy, analyzed values for phytase activity were used in all statistical analyses. The basal endogenous P loss was determined at 206 mg/kg of DMI from pigs fed the P-free diet (Table 4).

### Digestibility of P in Corn

The analyzed levels of phytase in corn diets were <70, 420, 720, and 1,100 FTU/kg. No differences in fecal output were detected among treatments (Table 4). Pigs fed the diet that analyzed 1,100 FTU/kg of phytase had slightly greater (linear,  $P < 0.05$ ) ADFI, and therefore, also slightly greater (linear,  $P < 0.05$ ) P intake and endogenous P loss than pigs fed the other diets. 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 supplemented with <70, 420, 720, or 1,100 FTU of phytase/kg, 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 fed diets supplemented with <70, 420, 720, or 1,100 FTU of phytase/kg, respectively. The ATTD and STTD of P increased linearly ( $P < 0.01$ ) and quadratically ( $P < 0.05$ ) 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 fed diets supplemented with <70, 420, 720, or 1,100 FTU of phytase/kg, respectively.

### Digestibility of P in DDGS

The analyzed levels of phytase in DDGS-containing diets were 130, 430, 770, and 1,100 FTU/kg. No differences in ADFI, P intake, daily fecal output, P output, or daily endogenous P loss were observed among treatments. Concentration of P in feces decreased (linear,  $P < 0.01$ ) as graded levels of phytase were added to the diets. Addition of phytase to the diets tended (linear,  $P < 0.10$ ) 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,

**Table 4.** Effects of microbial 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<sup>1</sup>

Item <sup>2</sup>	Feed intake, g/d	P intake, g/d	Fecal output, g/d	P in feces, %	P output, g/d	ATTD of P, %	EPL, <sup>3</sup> mg/d	STTD of P, %
<b>Corn</b>								
Corn + 0 FTU/kg	562	1.56	53.48	1.94	1.0	33.5	115.52	40.9
Corn + 420 FTU/kg	565	1.56	50.26	1.25	0.6	60.1	116.32	67.5
Corn + 720 FTU/kg	564	1.56	58.94	1.15	0.7	57.0	116.24	64.5
Corn + 1,100 FTU/kg	612	1.69	58.59	0.94	0.6	67.4	126.07	74.9
SEM	24	0.07	4.14	0.08	0.1	3.7	4.97	3.7
<i>P</i> -value								
Linear	0.03	0.03	0.21	<0.01	<0.01	<0.01	0.03	<0.01
Quadratic	0.12	0.12	0.73	<0.01	0.03	0.04	0.12	0.04
<b>DDGS</b>								
DDGS + 130 FTU/kg	666	3.20	92.28	0.95	0.9	72.6	137.14	76.9
DDGS + 430 FTU/kg	644	3.09	88.72	0.74	0.7	78.6	132.63	82.9
DDGS + 770 FTU/kg	682	3.25	95.08	0.75	0.7	78.2	140.56	82.5
DDGS + 1,100 FTU/kg	680	3.22	94.03	0.71	0.7	78.6	140.21	83.0
SEM	39	0.19	4.88	0.06	0.1	2.1	8.03	2.1
<i>P</i> -value								
Linear	0.40	0.64	0.60	<0.01	0.11	0.08	0.40	0.07
Quadratic	0.64	0.72	0.80	0.13	0.21	0.20	0.65	0.20
<b>HP-DDG</b>								
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 + 770 FTU/kg	525	1.27	52.00	0.59	0.3	75.6	108.14	84.1
HP-DDG + 1,100 FTU/kg	563	1.36	54.77	0.53	0.3	78.4	115.91	86.9
SEM	12	0.03	2.59	0.06	0.0	2.6	2.39	2.6
<i>P</i> -value								
Linear	0.27	0.31	0.84	<0.01	<0.01	<0.01	0.27	<0.01
Quadratic	0.26	0.27	0.41	0.03	0.02	0.04	0.26	0.04
<b>Corn germ</b>								
Corn germ + 110 FTU/kg	581	3.48	70.14	3.14	2.2	37.3	119.61	40.7
Corn germ + 390 FTU/kg	586	3.50	61.47	2.57	1.6	55.7	120.68	59.0
Corn germ + 910 FTU/kg	580	3.52	61.93	2.11	1.3	63.0	119.54	64.4
Corn germ + 1,400 FTU/kg	581	3.51	60.26	2.35	1.4	59.8	119.62	63.2
SEM	16	0.09	3.67	0.16	0.1	2.4	3.25	2.4
<i>P</i> -value								
Linear	0.91	0.69	0.43	<0.01	<0.01	<0.01	0.91	<0.01
Quadratic	0.81	0.76	0.25	0.02	<0.01	<0.01	0.81	<0.01

<sup>1</sup>Data are means of 6 observations per treatment, except for corn germ (5 observations).

<sup>2</sup>FTU = phytase units. Values refer to analyzed concentrations.

<sup>3</sup>EPL = basal endogenous P loss. The daily EPL was calculated by multiplying the EPL by the DMI of each pig fed P-containing diets. The EPL (206 mg/kg of DMI) was determined from pigs fed the P-free diet.

82.5, and 83.0% in pigs fed diets containing 130, 430, 770, or 1,100 FTU of phytase/kg, respectively.

78.4%, and the STTD of P from 77.1 to 88.0, 84.1, and 86.9% in pigs fed diets containing <70, 500, 770, or 1,100 FTU of phytase/kg, respectively.

### Digestibility of P in HP-DDG

The analyzed levels of phytase in the diets that contained HP-DDG were <70, 500, 770, and 1,100 FTU/kg. There were no differences in ADFI, P intake, daily fecal output, or daily endogenous P loss 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 fed diets containing <70, 500, 770, or 1,100 FTU of phytase/kg, 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

### Digestibility of P in Corn Germ

The analyzed levels of phytase in corn germ-containing diets were 110, 390, 910, and 1,400 FTU/kg. There were no differences in ADFI, P intake, daily fecal output, or daily endogenous P loss among treatments. Addition of phytase to the diets reduced (linear,  $P < 0.01$ ; quadratic,  $P < 0.05$ ) the concentration of P in the feces, and the daily P output was reduced (linear and quadratic,  $P < 0.01$ ) from 2.2 to 1.6, 1.3, and 1.4 g/d for pigs fed diets containing 110, 390, 910, and 1,400 FTU of phytase/kg, respectively. However, addition of phytase to the diets increased (linear and quadratic,

**Table 5.** Regression equations for the effects of microbial phytase on the standardized total tract digestibility of P in corn, distillers dried grains with solubles (DDGS), high-protein distillers dried grains (HP-DDG), and corn germ<sup>1</sup>

Item	Regression equation	R <sup>2</sup>	SE			P-value			
			Intercept	Linear term	Quadratic term	Model	Intercept	Linear term	Quadratic term
Corn	42.34 + 0.059 FTU – 0.000028 FTU <sup>2</sup>	0.63	3.95	0.02	0.000014	<0.01	<0.01	<0.01	0.05
DDGS	74.73 + 0.021 FTU – 0.000013 FTU <sup>2</sup>	0.20	3.21	0.01	0.000010	0.10	<0.01	0.06	0.17
HP-DDG	77.55 + 0.023 FTU – 0.000014 FTU <sup>2</sup>	0.36	2.15	0.01	0.000008	<0.01	<0.01	<0.01	0.06
Corn germ	35.50 + 0.067 FTU – 0.000034 FTU <sup>2</sup>	0.79	2.86	0.01	0.000006	<0.01	<0.01	<0.01	<0.01

<sup>1</sup>FTU = phytase units.

$P < 0.01$ ) 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 fed diets containing 110, 390, 910, or 1,400 FTU of phytase/kg, respectively.

### Regression Equations

The quadratic response for the STTD of P to the addition of increasing levels of phytase to corn is presented by the following equation (Table 5):

$$\text{STTD of P, \%} = 42.34 + 0.059 \text{ FTU} - 0.000028 \text{ FTU}^2 \quad (R^2 = 0.63).$$

The following equation was generated for DDGS:

$$\text{STTD of P, \%} = 74.73 + 0.021 \text{ FTU} - 0.000013 \text{ FTU}^2 \quad (R^2 = 0.20).$$

For HP-DDG, the following equation was generated:

$$\text{STTD of P, \%} = 77.55 + 0.023 \text{ FTU} - 0.000014 \text{ FTU}^2 \quad (R^2 = 0.36).$$

For corn germ, the response to microbial phytase could be described by the following equation:

$$\text{STTD of P, \%} = 34.50 + 0.067 \text{ FTU} - 0.000034 \text{ FTU}^2 \quad (R^2 = 0.79).$$

## DISCUSSION

### P and Phytate P in Ingredients

The concentration of P that was determined in corn, DDGS, HP-DDG, and corn germ is in agreement with values reported by Widmer et al. (2007) and NRC (1998). Of the total molecular weight of phytate (660.04 g·mol<sup>-1</sup>; Selle et al., 2009), P accounts for 28.2% because phytate binds 6 molecules of P (molecular weight

= 30.974 g·mol<sup>-1</sup>; Ham, 2008). On a percentage basis, corn and corn germ contain similar amounts of phytate-bound P (72 and 76% of total P, respectively). The concentration of phytate-bound P in corn on a percentage basis is within the range (61 to 77%) that has been reported, but the concentration of phytate-bound P in corn germ is slightly greater (76 vs. 65%) than previous data (Eeckhout and DePaepe, 1994). Distillers dried grains with solubles and HP-DDG also have similar concentrations of phytate-bound P, but these concentrations are much less (30 and 28% of total P, respectively) than in corn and corn germ. Lopez et al. (1983) and Mahgoub and Elhag (1998) observed that fermentation of corn and sorghum caused a reduction in the concentration of phytate in the grains. Therefore, it is possible that during fermentation of corn in the production of DDGS and HP-DDG, some of the phytate may be hydrolyzed, which results in a reduced concentration of phytate-bound P in these ingredients compared with corn. The concentration of phytate-bound P in DDGS has been reported to be 35% (Yáñez et al., 2011), and data from the present experiment are in agreement with this value, but we are not aware of other data for the concentration of phytate-bound P in HP-DDG.

### ATTD and STTD Values of P in Ingredients Without Phytase

The value for the ATTD of P in corn without phytase that was determined in this experiment is in agreement with values reported by Bünzen et al. (2008) and Stein et al. (2009), but the ATTD of P in corn germ without phytase was slightly greater than the value reported by Widmer et al. (2007). This difference may be due to different processing of corn to obtain corn germ, differences between corn hybrids that were used to obtain the corn germ, or differences in the concentration of P in corn from one year to another. The ATTD of P in DDGS without phytase varies between 50 and 69% (Pedersen et al., 2007; Stein et al., 2009; Almeida and Stein, 2010). 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-bound P in DDGS compared with corn. The ATTD of P in HP-DDG without phytase determined in this experiment concurs with the ATTD of P in DDGS, and is 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 proportion of P that is bound in phytate in these 2 ingredients. We are not aware of other experiments in which the ATTD of P in DDGS and HP-DDG has been compared.

The value for the basal endogenous P loss that was determined in this experiment is very close to previous values (Stein et al., 2006; Widmer et al., 2007; Almeida and Stein, 2010). This value was used to correct ATTD values for the basal endogenous P loss to calculate values for STTD of P. Endogenous losses of P have sometimes been expressed in milligrams per kilogram of BW (Jongbloed, 1987; Rodehutschord et al., 1998). However, to be able to use values for endogenous losses to correct values for ATTD of P to calculate values for STTD, it is necessary that endogenous losses are expressed relative to DMI or in grams per day (Petersen and Stein, 2006; Bünzen et al., 2008).

Values for the STTD of P are expected to be greater than the ATTD values but less than values for true total tract digestibility of P. The value calculated for the STTD of P in corn without phytase in the present experiment is in agreement with this theory because it is between the ATTD (31.9%) and the true total tract digestibility (49.2%) of P in corn reported by Stein et al. (2009) and by Wu et al. (2008), respectively. The value for the STTD of P in DDGS without phytase is in agreement with the value reported by Almeida and Stein, (2010), but values for the STTD of P in HP-DDG and in corn germ determined in the present experiment are slightly greater than values reported by Widmer et al. (2007).

### *Effects of Phytase*

Supplementation of diets with phytase resulted in increased ATTD and STTD of P, which was expected because hydrolysis of phytate by microbial phytase liberates P in the gastrointestinal tract 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, 2010), but to our knowledge, there are no data on the effects of graded amounts of phytase on the STTD of P in corn, DDGS, HP-DDG, and corn germ. The responses on the STTD of P of adding the least quantities of phytase to corn or DDGS that were obtained in this experiment are in agreement with the values reported by Almeida and Stein (2010). Although it was not the objective of this experiment to compare the STTD of P among

ingredients, our data indicate 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. It is likely that the relatively low concentration of phytate-bound P in DDGS and HP-DDG may reduce the effectiveness of phytase in improving the STTD of P in these ingredients, which is the reason for the reduced effect of phytase in DDGS and HP-DDG compared with corn and corn germ. Nevertheless, the values for STTD of P that were determined for DDGS and HP-DDG indicate that the P in these ingredients is readily digestible by pigs.

### *Regression Equations*

The regression equations determined in this experiment allow calculation of the STTD of P with any amount of phytase between <70 and 1,100, or between 110 and 1,400 FTU/kg for corn and corn germ, respectively. If pigs that are fed diets containing corn or corn germ do not need a maximum release of P from these ingredients to meet their requirement for P, a reduced concentration of phytase may be used. Thus, the equations that are presented in Table 5 may be used to predict the STTD of P in corn and corn germ with inclusion of any amount of phytase that is less than 1,100 or 1,400 FTU/kg, respectively. As a consequence, this approach may result in a more economical use of phytase.

The fact that there were no effects of graded amounts of phytase on the STTD of P in DDGS indicates that for this ingredient, the STTD of P determined for DDGS without phytase may be used regardless of the inclusion of phytase. However, the digestibility of P in the source of DDGS that was used in this experiment is in the upper end of the range of values reported for DDGS (Pedersen et al., 2007; Stein et al., 2009; Almeida and Stein, 2010), and it is possible that if a source of DDGS with decreased digestibility is used, phytase may improve the STTD of P. The quadratic effect of graded amounts of phytase on the STTD of P in HP-DDG indicates that phytase increased the STTD of P in HP-DDG, but only to 500 FTU/kg. Although a regression equation could be developed, this equation had an  $R^2$  value of only 0.36, which means that the model accounts for only 36% of the variation of the STTD of P. This equation, therefore, may not accurately predict the response to graded amounts of microbial phytase in HP-DDG.

In conclusion, the STTD of P in corn and corn germ can be increased by the addition of microbial phytase to the diet, and the response of increasing dietary levels of phytase on the STTD of P in corn and corn germ can be predicted by regression equations. Phosphorus in DDGS and HP-DDG is well digested by pigs even if no microbial phytase is used. This is probably the result of a relatively low concentration of phytate-bound P in DDGS and HP-DDG, which limited the effect of microbial phytase on the STTD of P in these ingredients.

## LITERATURE CITED

- Adeola, L. 2001. Digestion and balance techniques in pigs. Pages 903–916 in *Swine Nutrition*. A. J. Lewis and L. L. Southern, ed. CRC Press, Washington, DC.
- Almeida, F. N., and H. H. Stein. 2010. Performance and phosphorus balance of pigs fed diets formulated on the basis of values for standardized total tract digestibility of phosphorus. *J. Anim. Sci.* 88:2968–2977.
- AOAC International. 2007. *Official Methods of Analysis*. 18th ed. Rev. 2. W. Howitz and G. W. Latimer Jr., ed. AOAC Int., Gaithersburg, MD.
- Bünzen, S., H. S. Rostagno, D. C. Lopes, F. A. M. Hashimoto, P. C. Gomes, and L. R. Apolônio. 2008. Digestibilidade do fosforo de alimentos de origem vegetal determinada em suínos em crescimento e terminação. *R. Bras. Zootec.* 37:1236–1242.
- Eeckhout, W., and M. De Paepe. 1994. Total phosphorus, phytate-phosphorus and phytase activity in plant feedstuffs. *Anim. Feed Sci. Technol.* 47:19–29.
- Ellis, R., E. R. Morris, and C. Philpot. 1977. Quantitative determination of phytate in the presence of high inorganic phosphate. *Anal. Biochem.* 77:536–539.
- Ham, B. 2008. Periodic table of elements. Pages 92–94 in *The Periodic Table*. Chelsea House, New York, NY.
- Holst, D. O. 1973. Holst filtration apparatus for Van Soest detergent fiber analysis. *J. AOAC* 56:1352–1356.
- Jongbloed, A. W. 1987. Phosphorus in the feeding of pigs: Effect of diet on the absorption and retention of phosphorus by growing pigs. PhD Diss. Wageningen Agricultural Univ., Wageningen, the Netherlands.
- Kim, B. G., and M. D. Lindemann. 2007. A new spreadsheet method for the experimental animal allotment. *J. Anim. Sci.* 85(Suppl. 2):218. (Abstr.)
- Lopez, Y., D. T. Gordon, and M. L. Fields. 1983. Release of phosphorus from phytate by natural lactic acid fermentation. *J. Food Sci.* 48:953–955.
- Mahgoub, S. E. O., and S. A. Elhag. 1998. Effect of milling, soaking, malting, heat-treatment and fermentation on phytate level of four Sudanese sorghum cultivars. *Food Chem.* 61:77–80.
- NRC. 1998. *Nutrient Requirements of Swine*. 10th rev. ed. Natl. Acad. Press, Washington, DC.
- Pedersen, C., M. G. Boersma, and H. H. Stein. 2007. Digestibility of energy and phosphorus in ten samples of distillers dried grains with solubles fed to growing pigs. *J. Anim. Sci.* 85:1168–1176.
- Petersen, G. I., and H. H. Stein. 2006. Novel procedure for estimating endogenous losses and measurement of apparent and true digestibility of phosphorus by growing pigs. *J. Anim. Sci.* 84:2126–2132.
- Rodehutscord, M., R. Haverkamp, and E. Pfeffer. 1998. Inevitable losses of phosphorus in pigs estimated from balance data using diets deficient in phosphorus. *Arch. Anim. Nutr.* 51:27–38.
- Selle, P. H., A. J. Cowieson, and V. Ravindran. 2009. Consequences of calcium interactions with phytate and phytase for poultry and pigs. *Livest. Sci.* 124:126–141.
- Selle, P. H., and V. Ravindran. 2008. Phytate-degrading enzymes in pig nutrition. *Livest. Sci.* 113:99–122.
- Stein, H. H., M. G. Boersma, and C. Pedersen. 2006. Apparent and true total tract digestibility of phosphorus in field peas (*Pisum sativum* L.) by growing pigs. *Can. J. Anim. Sci.* 86:523–525.
- Stein, H. H., S. P. Connot, and C. Pedersen. 2009. Energy and nutrient digestibility in four sources of distillers dried grains with solubles produced from corn grown within a narrow geographical area and fed to growing pigs. *Asian-australas. J. Anim. Sci.* 22:1016–1025.
- Tran, G., and D. Sauvant. 2004. Chemical data and nutritional value. Pages 17–24 in *Tables of Composition and Nutritional Value of Feed Materials: Pigs, Poultry, Cattle, Sheep, Goats, Rabbits, Horses, Fish*. D. Sauvant, J. M. Pérez, and G. Tran, ed. Institut National de la Recherche Agronomique, Association Française de Zootechnie, Paris, France.
- Widmer, M. R., L. M. McGinnis, and H. H. Stein. 2007. Energy, phosphorus, and amino acid digestibility of high-protein distillers dried grains and corn germ fed to growing pigs. *J. Anim. Sci.* 85:2994–3003.
- Wu, Z., Z. Ruan, Y. G. Zhang, Y. Q. Hou, Y. L. Yin, T. J. Li, R. L. Huang, W. Y. Chu, X. F. Kong, B. Gao, and L. X. Chen. 2008. True digestibility of phosphorus in different resources of feed ingredients in growing pigs. *Asian-australas. J. Anim. Sci.* 21:107–119.
- Yáñez, J. L., E. Beltranena, M. Cervantes, and R. T. Zijlstra. 2011. Effect of phytase and xylanase supplementation or particle size on nutrient digestibility of diets containing distillers dried grains with solubles cofermented from wheat and corn in ileal-cannulated grower pigs. *J. Anim. Sci.* 89:113–123.

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