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Productive performance of commercial growing and finishing pigs supplemented with a *Buttiauxella* phytase as a total replacement of inorganic phosphate

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ABSTRACT

The objective of this study was to test if a novel phytase from *Buttiauxella* sp. can replace all added inorganic phosphate in a diet with reduced Ca and metabolizable energy (ME) fed to commercial pigs from 12 kg body weight (BW) until slaughter, whilst maintaining performance and carcass quality parameters. Four dietary treatments were tested in a completely randomized design with 9 replicate pens, each containing 31 mixed sex Newsham Choice pigs. Diets included a positive control (PC) based on corn, soybean meal, wheat middling and bakery meal, meeting all nutrient requirement of pigs; a negative control (NC) excluded inorganic phosphate and with reduced Ca (−0.13%) and ME (−0.15 MJ/kg); and NC supplemented with *Buttiauxella* phytase at 500 or 1,000 FTU/kg feed. Diets were fed *ad libitum* in mash form in 5 phases: starter (12 to 25 kg BW), grower 1 (25 to 50 kg BW) and 2 (50 to 75 kg BW), and finisher 1 (75 to 100 kg BW) and 2 (100 kg BW to slaughter). The NC group showed lower ($P < 0.05$) average daily feed intake (ADFI) and average daily gain (ADG) in starter and grower phases, lower gain to feed ratio (G:F) in starter and grower 1 compared with PC. Pigs receiving the high dose of phytase of 1,000 FTU/kg had improved performance vs. the 500 FTU/kg phytase treatment in starter and grower 1 phase compared with the PC in grower 1 phase. Increasing phytase dose resulted in a linear increase in ADG (12 to 120 kg BW) and G:F (50 to 75 kg BW). A comparison of treatment groups over the full production period from 12 kg BW until slaughter showed that both 500 and 1,000 FTU/kg phytase treatments were able to maintain growth performance and carcass characteristics compared with PC. The application of *Buttiauxella* phytase could therefore be used as an effective strategy to replace all inorganic phosphate in diets of pigs fed corn, soybean meal, wheat middling and bakery meal based diets from 12 kg BW. An economic analysis showed greater return from both phytase treatments vs. the PC and favored the higher phytase dose at 1,000 FTU/kg vs. the traditional dose of 500 FTU/kg. The latter was mainly related to the improved performance of the higher dose in younger pigs to 75 kg BW.

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1. Introduction

Phytase enzymes are routinely added to animal feeds in order to increase the utilization of phytate-bound phosphorus (P) from plant based ingredients, reducing the need to add inorganic sources of this mineral to diet formulations (Selle and Ravindran, 2008). The need to improve the utilization of dietary P and reduce inorganic P added to diets was first raised in the 1990s when high levels of P excretion in manure from intensively raised animals resulted in environmental concerns that resulted in legislation to limit the application of P in manure onto farmland in many countries. From

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the producer's point of view, improving the dietary utilization of P from feed materials and reducing P excretion represents a saving in diet formulation costs while reducing the environmental impact when manure is used as fertilizer. Phytase has traditionally been used at a dose of 500 FTU/kg feed that resulted in phytate degradation in a range of 45% to 60% by the end of the small intestine in pigs and broilers (Dersjant-Li et al., 2015). Recently, new phytase enzymes have been produced, including a *Buttiauxella* sp. phytase that are more efficacious in improving total tract P digestion and conferred additional benefits at dose rates above 500 FTU/kg (Bento et al., 2012; Zeng et al., 2015). Trials with broiler chickens noted a higher efficacy of *Buttiauxella* phytase when compared with *Escherichia coli* phytases (Kumar et al., 2012). In other study *Buttiauxella* phytase at 1,000 FTU/kg was able to breakdown up to 91.5% of the dietary phytate (based on IP6 disappearance rate at the ileum) in broilers (Li et al., 2016). In growing pigs, Zeng et al. (2016) observed that supplementation of *Buttiauxella* phytase at 1,000 FTU/kg increased ileal phytate P digestibility from 11% in negative control (NC) to 71%. In weaned piglets, *Buttiauxella* phytase at 1,000 and 2,000 FTU/kg improved phytate P digestibility from 39% in NC to 76% and 83%, respectively at the end of the small intestine (Dersjant-Li and Dusel, 2016). Similarly, increasing *Buttiauxella* phytase dose to 2,000 FTU/kg increased apparent total tract digestibility of P from 57.3% in the NC to 86.5% in weaned piglets fed corn and soybean meal (SBM) based diets (Dersjant-Li et al., 2017a). In that study, 2,000 FTU/kg phytase replaced 0.24% inorganic P from dicalcium phosphate (DCP, based on analyzed P levels) while maintaining body weight gain and feed efficiency compared with positive control (PC) in piglets. In a recent study (Dersjant-Li et al., 2017b), it was reported that *Buttiauxella* phytase at 1,000 FTU/kg could totally replace inorganic phosphate in grower pigs from 30 to 85 kg BW, fed European type wheat/barley based diets. However, limited information is available on using phytase to total replace inorganic P in commercial corn and SBM based diets in pigs raised under commercial production settings.

The objective of this study was to test if a *Buttiauxella* phytase at 500 or 1,000 FTU/kg could completely replace inorganic phosphate in diets with reduced Ca and metabolizable energy (ME) fed to commercial pigs from 12 kg BW until slaughter (around 130 kg BW), whilst maintaining performance and carcass quality parameters.

2. Materials and methods

The experiment was conducted in a commercial research facility in the USA, conducted under Good Laboratory Practice and Good Clinical Practice guidelines and according to all the restrictions provided under Animal and Human Welfare Codes/Laboratory Practice Codes.

2.1. Animals

Newsham Choice piglets were weaned between 18 and 21 days of age and fed a common corn and SBM based commercial diets for the first 3 weeks after weaning. At the start of the trial, a total of 1,116 piglets (average BW 11.9 ± 0.69 kg) were allocated to 4 treatment diets, arranged as 9 replicate pens, in a completely randomized design, each pen contained 31 pigs. Pen size was 305 cm \times 720 cm and no bedding materials were used. Piglets were allocated to pens based similar average body weight and equal sexes per pen at the start of the trial. Animals were vaccinated with Enterisol ileitis vaccine, Ingelvac CircoFLEX (Boehringer Ingelheim and Vetmedica Inc., St. Joseph, USA) and Porcilis M Hyo (Merck Animal Health, Madison, USA). Mecadox 10 was administered via

feed at 0.05% during the initial feeding program, followed by Tylan 40 (at 0.05% dose) through the grower and finisher phases.

2.2. Dietary treatments

Mash diets were fed to pigs *ad libitum* in 5 phases of 28 d each to slaughter. For each of the phases, a PC diet was formulated using corn and soybean meal, wheat middling and bakery meal with adequate Ca, digestible P and meeting or exceeding all nutrient requirement based on NRC (2012), Table 1. A NC diet was formulated without the addition of any inorganic phosphate and with reduced Ca (0.13% lower than PC) and ME (0.15 MJ/kg lower than PC). In addition to the reduction in P following removal of inorganic P, the Ca and ME in NC diet were reduced to account for the potential of improved utilization of these nutrients by phytase. In PC diets, the formulated Ca to total P ratio is in a range of 1.3 (phase 1) to 0.94 (phase 5), which is close to the NRC recommendation of 1.25 to 1 (NRC, 2012). The ME reduction was obtained by increasing the use of by-products in the feed at the expense of added fat. Treatments 3 and 4 were identical in composition to the NC but contained a new generation 6-phytase from *Buttiauxella* sp. (EC 3.1.3.26), expressed in *Trichoderma reesei* (Axta PHY, Danisco Animal Nutrition/DuPont IB, Marlborough, United Kingdom) at inclusion levels of 500 or 1,000 FTU/kg feed. One FTU is defined as the quantity of enzyme that releases 1 μ mol of inorganic P per minute from 5.0 mmol/L sodium phytate at pH 5.5 at 37 °C (AOAC, 2000). Feed and water were supplied *ad libitum*. Feed samples were taken after feed processing and analyzed for phytase (Yu et al., 2012), Ca and P (ICP in Feed Samples – AOAC 965.17/985.01) and phytate content (Analytical Biochemistry Vol. 77: 536 to 539 [1977] by Eurofins, Des Moines, USA).

2.3. Measurements

All animals were monitored for health status, which was recorded along with mortality and cause of death. Pigs were weighed on the first day of the trial (start at 12 kg BW) and then at 28 day intervals thereafter, until slaughter weight at around 130 kg. All performance parameters including average daily feed intake (ADFI), average daily gain (ADG) and gain to feed ratio (G:F) were corrected for mortality (calculated based on pig days, e.g., how many days each animal in the pen was alive). Due to the leg weakness and animal welfare concerns, pigs in NC group were switched to the PC diet after finisher 1 (on attaining 100 kg body weight in the PC fed group). Only 3 pens in the NC treatment reached similar market weight at the same time that pigs from other treatments were slaughtered. All pigs were slaughtered at a commercial facility (Cargill Ltd, Beardstown, IL, USA) and carcass components were measured with certified standard commercial processing procedures.

2.4. Statistics

Pen was the experimental unit. Performance and carcass data were analyzed by one-way ANOVA in the Fit Model platform of JMP 11 (SAS Institute, Cary, North Carolina, USA) and means separation was conducted according to Tukey's HSD test. Significance was determined at $P < 0.05$; $P < 0.10$ was taken to indicate a trend. Linear response was analyzed with increasing phytase dose from 0 (NC) to 1,000 FTU/kg.

3. Results

The performance results on ADFI, ADG and G:F were all corrected for mortality and are presented in Table 2 for each feeding

Table 1Basal diet ingredients (as fed, % of the diet) and nutrients composition in a 5 phase feeding program from 12 kg body weight till slaughter.¹

Item	Starter		Grower 1		Grower 2		Finisher 1		Finisher 2	
	12 to 25 kg		25 to 50 kg		50 to 75 kg		75 to 100 kg		100 to slaughter	
	PC	NC	PC	NC	PC	NC	PC	NC	PC	NC
Ingredient, %										
Corn	50.96	56.79	58.39	60.58	57.22	61.19	59.58	64.07	64.62	66.78
SBM 47.8%	33.55	33.64	22.18	21.79	18.83	18.8	11.7	11.79	5.01	4.76
Wheat middling	5	5	15	15	20	17.87	25	22.21	27	26.62
Bakery meal	5	0.61	0	0	0	0	0	0	0	0
Limestone	1.46	1.43	1.45	1.34	1.31	1.14	1.17	0.96	1.08	0.8
Fat (A/V) ²	2.29	1.34	1.66	0.47	1.62	0.33	1.56	0.25	1.31	0.22
DCP	0.72	0	0.57	0	0.39	0	0.29	0	0.17	0
Salt	0.5	0.62	0.18	0.18	0.17	0.18	0.17	0.2	0.17	0.17
Vitamin premix ³	0.15	0.15	0.15	0.15	0.15	0.15	0.15	0.15	0.15	0.15
Mineral premix ⁴	0.13	0.13	0.13	0.13	0.13	0.13	0.13	0.13	0.13	0.13
Mecadox 10	0.05	0.05	0	0	0	0	0	0	0	0
Tylan 40	0	0	0.05	0.05	0.05	0.05	0.05	0.05	0.05	0.05
L-Lys HCl	0.14	0.14	0.21	0.21	0.12	0.13	0.18	0.19	0.24	0.24
DL-Met	0.06	0.05	0.05	0.05	0.01	0.04	0.05	0.01	0.01	0.01
L-Thr	0	0	0.01	0.01	0	0	0	0	0.04	0.03
Calculated nutrients, %										
DE, MJ/kg	14.8	14.6	14.4	14.2	14.3	14.2	14.2	14.0	14.0	13.9
ME, MJ/kg	13.8	13.7	13.6	13.5	13.6	13.5	13.6	13.5	13.6	13.5
NE, MJ/kg	10.1	10.0	10.1	10.0	10.1	10.1	10.2	10.2	10.3	10.3
CP	22	22	18	18	17	17	14.5	14.5	12	12
AID Lys	1.15	1.15	0.94	0.94	0.81	0.81	0.69	0.69	0.57	0.57
AID Met	0.37	0.37	0.30	0.29	0.24	0.24	0.21	0.21	0.17	0.17
AID SAA	0.65	0.65	0.53	0.53	0.46	0.46	0.40	0.40	0.33	0.33
AID Thr	0.67	0.68	0.54	0.54	0.50	0.50	0.41	0.41	0.35	0.35
AID Trp	0.23	0.23	0.18	0.18	0.17	0.17	0.14	0.14	0.11	0.11
Ca	0.76	0.63	0.72	0.59	0.64	0.51	0.56	0.43	0.50	0.37
Total P	0.57	0.42	0.58	0.46	0.57	0.48	0.57	0.49	0.53	0.50
Available P	0.29	0.14	0.26	0.14	0.23	0.15	0.21	0.15	0.18	0.15
Digestible P	0.24	0.15	0.22	0.15	0.21	0.15	0.19	0.15	0.17	0.14
Analyzed nutrients ⁵ , %										
Ca	1.07	1.01	1.09	0.96	0.75	0.61	0.72	0.54	0.46	NA ⁶
P	0.59	0.41	0.53	0.46	0.51	0.49	0.48	0.45	0.5	NA
Phytate P	0.28	0.27	0.30	0.33	0.36	0.34	0.37	0.35	0.38	NA
Phytase, FTU ⁷	196	80	526	417	463	416	644	633	NA	NA

DCP = dicalcium phosphate; AID = apparent ileal digestible lysine; SAA = sulphur amino acids.

¹ Phytase (Axta PHY, Danisco Animal Nutrition/DuPont IB) was top dressed to the NC diet. PC: positive control. NC: negative control.² Fat (A/V) refers to a mixing of animal and vegetable fat.³ Supplied the following per kg of finished feed: 9,921 IU vitamin A; 1,544 IU vitamin D₃; 53 IU vitamin E; 4 mg vitamin K; 27.5 mg pantothenic acid; 8 mg riboflavin; 40 mg niacin; 0.03 mg vitamin B₁₂.⁴ Supplied the following per kg of finished feed: 13.5 mg Cu from copper sulphate; 120 mg Zn from zinc sulphate; 94 mg Fe from ferrous sulphate; 36 mg Mn manganese sulphate (50%) and manganese oxide (50%); 0.3 mg I from calcium iodate; 0.3 mg Se from sodium selenite.⁵ Analyzed Ca, P and phytate P levels in phytase treatments are similar to NC, for each phase.⁶ NA: not analyzed, NC finisher 2 diets were not fed. The NC + phytase treatments had an analyzed Ca level of 0.52%.⁷ Phytase activity in the diets expressed as FTU/kg and analyzed by Eurofins Scientific, Inc. (Des Moines, USA). The analyzed average phytase activity across all phases was 591 FTU/kg for NC + 500 FTU treatment and 1,516 FTU/kg for NC + 1,000 FTU treatment, after deduction of the phytase activity in NC diets.

phase, with the cumulative data over all phases shown in Table 3. The NC diet resulted in lower ($P < 0.05$) ADFI, ADG in starter, grower 1 and 2 phases, and decreased G:F ($P < 0.05$) in starter and grower 1 phases compared with PC group. During the starter phase of 12 to 25 kg BW, phytase diets containing 500 FTU/kg improved ADG ($P < 0.05$) compared with NC, while phytase at 1,000 FTU/kg improved ($P < 0.05$) ADG, ADFI and G:F compared with NC, but did not differ from pigs fed the PC diet. During grower phase 1 (25 to 50 kg BW), phytase inclusion at 500 and 1,000 FTU improved ($P < 0.05$) ADFI, ADG and G:F compared with the NC diet. Supplementation with phytase at 1,000 FTU/kg improved ($P < 0.05$) ADFI and ADG compared with the 500 FTU/kg diets, and resulted in greater ($P < 0.05$) ADG and G:F compared with pigs fed the PC diet. During grower phase 2 (50 to 75 kg BW), both 500 and 1,000 FTU/kg phytase improved ($P < 0.05$) ADFI and ADG compared with NC and were not different from the PC. In the finisher 1 feeding phase (75 to 100 kg BW), no significant treatment effects were observed for ADFI, ADG or G:F. Body weight at the end of the finisher phase 1 was lower ($P < 0.05$) in the NC fed groups, which

was improved ($P < 0.05$) with phytase supplementation, whereby phytase at 1,000 FTU/kg resulted in greater ($P < 0.05$) body weights compared with pigs fed phytase at 500 FTU/kg. The body weight for pigs fed the PC diet was intermediate between the 2 phytase inclusion levels. No significant differences were found on mortality rate in each phase. However, at the end of finisher 1 phase, pigs fed the NC diet were switched to the PC diet, hence the performance data are not shown for the finisher 2 phase. For the whole finisher phase (75 kg till slaughter), NC excluded, the best ADG and G:F were observed with 500 FTU/kg phytase, which were greater than that recorded for pigs fed the PC ($P < 0.05$). Phytase at 1,000 FTU/kg did not differ from PC. A linear increase in ADFI and ADG with increasing phytase dose from 0 (NC) to 1,000 FTU/kg was observed in all phases up to finisher 1, while similar response was seen for G:F in starter, grower 1 and whole grower phase from 25 to 75 kg BW.

Overall, from the starter phase until the end of the grower period (Table 3) for pigs weighing from 12 to 75 kg, those fed the NC diet had reduced ($P < 0.05$) ADFI, ADG and G:F compared with PC.

Table 2
The impact of adding different levels of *Buttiauxella* phytase to a negative control diet on performance of pigs during each feeding phase.

Item	PC ¹	NC ¹	NC + 500 FTU Phytase	NC + 1,000 FTU Phytase	SEM	P-value	P-value Linear ²
Starter phase (12 to 25 kg)							
BW at start, kg	11.9	11.9	11.9	11.9	0.24	1	
ADG, kg	0.57 ^a	0.43 ^c	0.47 ^b	0.54 ^a	0.009	<0.0001	<0.0001
ADFI, kg	0.99 ^a	0.85 ^c	0.88 ^{bc}	0.95 ^{ab}	0.020	<0.0001	0.0007
G:F	0.574 ^a	0.511 ^b	0.533 ^b	0.564 ^a	0.0069	<0.0001	<0.0001
Mortality, %	0.35	0.00	0.69	1.39	0.54	0.33	0.1
Grower 1 phase (25 to 50 kg)							
BW start, kg	27.3 ^a	23.6 ^b	24.6 ^b	26.4 ^a	0.44	<0.0001	<0.0001
ADG, kg	0.78 ^b	0.62 ^d	0.72 ^c	0.83 ^a	0.013	<0.0001	<0.0001
ADFI, kg	1.75 ^a	1.44 ^c	1.59 ^b	1.78 ^a	0.036	<0.0001	<0.0001
G:F	0.444 ^{bc}	0.431 ^c	0.453 ^{ab}	0.467 ^a	0.0045	<0.0001	<0.0001
Mortality, %	0.69	1.39	0.35	0.35	0.51	0.43	0.17
Grower 2 phase (50 to 75 kg)							
BW start, kg	49.2 ^a	41.1 ^c	44.8 ^b	49.7 ^a	0.75	<0.0001	<0.0001
ADG, kg	0.90 ^a	0.78 ^b	0.88 ^a	0.92 ^a	0.016	<0.0001	<0.0001
ADFI, kg	2.34 ^a	2.06 ^b	2.23 ^a	2.39 ^a	0.044	<0.0001	<0.0001
G:F	0.383	0.375	0.395	0.385	0.006	0.25	0.37
Mortality, %	1.39	1.79	0.35	0.70	0.51	0.35	0.18
BW end of grower phase, kg	74.7 ^b	66.9 ^a	69.8 ^a	75.8 ^b	0.95	<0.0001	<0.0001
Finisher 1 phase (75 to 100 kg)							
BW end of finisher 1, kg	98.4 ^{ab}	87.2 ^c	94.8 ^b	100.0 ^a	1.15	<0.0001	<0.0001
ADG, kg	0.80	0.78	0.83	0.84	0.020	0.18	0.046
ADFI, kg	2.69 ^{xy}	2.57 ^y	2.67 ^{xy}	2.79 ^x	0.057	0.085	0.01
G:F	0.298	0.305	0.310	0.301	0.006	0.50	0.7
Mortality, %	1.40	2.21	2.45	0.36	0.73	0.19	0.21
Whole finisher 75 kg to slaughter ³							
BW at slaughter, kg	130.5		130.9	132.7	1.236	0.44	
ADG, kg	0.78 ^b		0.83 ^a	0.79 ^{ab}	0.013	0.0297	
ADFI, kg	2.97		2.95	3.03	0.053	0.58	
G:F	0.264 ^b		0.282 ^a	0.262 ^b	0.037	<0.0001	

^{a, b, c} Values in the same row not sharing the same superscript differ at $P < 0.05$.

^{xy} Values in the same row not sharing the same superscript tend to differ at $P < 0.10$.

¹ Negative control (NC) diet was formulated without added inorganic phosphate, 0.15 MJ/kg less ME and 0.13% less Ca vs. the positive control (PC).

² Linear regression was analyzed with increasing phytase dose from 0 (NC) to 1,000 FTU/kg.

³ Pigs fed the NC diet switched to the PC diet at the end of finisher 1 and not used in the calculation for PC group. Only 3 replicates in NC reached market weight and therefore NC treatment was not included for calculating overall performance for whole finisher phase.

Table 3
The impact of adding different levels of *Buttiauxella* phytase to a negative control diet on accumulated performance of pigs.

Item	PC ¹	NC ¹	NC + 500 FTU Phytase	NC + 1,000 FTU Phytase	SEM	P-value	P-value linear ²
12 to 75 kg BW							
ADG, kg	0.74 ^a	0.60 ^c	0.69 ^b	0.76 ^a	0.011	<0.0001	<0.0001
ADFI, kg	1.70 ^a	1.46 ^b	1.58 ^b	1.72 ^a	0.032	<0.0001	<0.0001
G:F	0.441 ^b	0.421 ^a	0.440 ^b	0.447 ^b	0.005	0.0021	0.0006
Mortality, %	2.43	3.13	1.39	2.43	0.93	0.62	0.63
12 to 100 kg BW							
ADG, kg	0.77 ^{ab}	0.67 ^c	0.74 ^b	0.78 ^a	0.009	<0.0001	<0.0001
ADFI, kg	1.95 ^a	1.73 ^b	1.85 ^{ab}	1.99 ^a	0.036	<0.0001	<0.0001
G:F	0.393	0.387	0.397	0.394	0.004	0.268	0.16
Mortality, %	3.82	5.21	3.82	2.78	1.22	0.58	0.22
12 to 120 kg BW							
ADG, kg	0.78 ^{ab}	0.69 ^c	0.75 ^b	0.79 ^a	0.009	<0.0001	<0.0001
ADFI, kg	2.17 ^a	1.98 ^b	2.09 ^{ab}	2.20 ^a	0.037	0.0007	0.0001
G:F	0.357 ^{xy}	0.350 ^y	0.361 ^x	0.358 ^{xy}	0.003	0.064	0.06
Mortality, %	4.51	5.56	5.21	4.51	1.47	0.94	0.65
12 kg BW to slaughter ³							
ADG, kg	0.76		0.75	0.78	0.008	0.106	
ADFI, kg	2.25		2.18	2.29	0.03	0.106	
G:F	0.339		0.346	0.340	0.0025	0.20	
Mortality, %	5.21		6.25	5.21	1.38	0.83	

^{a, b, c} Values in the same row not sharing the same superscript differ at $P < 0.05$.

^{xy} Values in the same row not sharing the same superscript tend to differ at $P < 0.10$.

¹ Negative control (NC) diet was formulated without added inorganic phosphate, 0.15 MJ/kg less ME and 0.13% less Ca vs. the positive control (PC).

² Linear regression was analyzed with increasing phytase dose from 0 (NC) to 1,000 FTU/kg.

³ Pigs fed the NC diet switched to the PC diet at the end of finisher 1. Only 3 replicates in NC reached market weight and therefore NC treatment was not included for calculating overall performance from 12 kg BW till slaughter.

Table 4The impact of adding different levels of *Buttiauxella* phytase to a negative control diet on carcass quality.¹

Item	PC	NC + 500 FTU phytase	NC + 1,000 FTU phytase	SEM	P-value
Carcass weight, kg	96.59	96.45	98.57	0.919	0.21
Lean carcass, %	53.3	53.2	53.0	0.126	0.20
Yield, %	74.0 ^{x,y}	73.7 ^y	74.3 ^x	0.184	0.075
Muscle depth, mm	63.78 ^{ab}	63.91 ^a	62.63 ^b	0.334	0.022
10th rib back fat, mm	18.94 ^y	19.72 ^x	18.99 ^y	0.232	0.052

PC = positive control.

^{a, b} Values in the same row not sharing the same superscript differ at $P < 0.05$.^{x, y} Values in the same row not sharing the same superscript tend to differ at $P < 0.10$.¹ Only 3 replicates in negative control (NC) reached market weight and therefore NC treatment was not included.

Pigs fed diets supplemented with phytase at 1,000 FTU/kg attained the same levels of ADG, ADFI and G:F as the PC. The high phytase dose of 1,000 FTU/kg improved ($P < 0.05$) ADFI and ADG compared with 500 FTU/kg phytase, however, no significant effect was seen for G:F cumulatively, from the starter to finisher 1 (pigs weighing from 12 to 100 kg), both 500 and 1,000 FTU/kg phytase addition improved ($P < 0.05$) ADG vs. NC and maintained ADG at the same level as seen for pigs fed the PC diet. Supplementation with 1,000 FTU/kg resulted in greater ($P < 0.05$) ADG compared with 500 FTU/kg. Differences in G:F between 500 and 1,000 FTU/kg were not significant. For pigs weighing from 12 to 120 kg, phytase at 1,000 FTU/kg had greater ($P < 0.05$) ADG compared with 500 FTU/kg, while phytase at 500 FTU/kg had greater ($P < 0.05$) ADG and tended to show a greater G:F than that pigs fed the NC ($P < 0.10$). For the entire growth period to slaughter (12 to 130 kg BW), the NC group was not included in the analysis because only three pens in this treatment reached market weight. There were no significant differences between both phytase inclusion levels and the PC fed group.

Because only 3 pens reached market size from the NC fed group, this treatment was excluded from the carcass analysis (Table 4). The pigs receiving phytase at 1,000 FTU/kg feed tended to have greater percentage yield ($P < 0.10$) compared with 500 FTU/kg. Muscle depth was the greatest ($P < 0.05$) for the pigs fed the 500 FTU/kg diets. There was a tendency ($P < 0.10$) for greater 10th rib backfat in pigs fed the 500 FTU/kg diets compared with PC and 1,000 FTU/kg. No significant differences were found for body weight at slaughter, carcass weight and percentage of lean carcass between the PC diet and 2 phytase treatments.

4. Discussion

The analyzed Ca levels, except the finisher 2 phase, were about 20% to 60% higher than calculated levels while analyzed P levels were similar to calculated P levels. The higher Ca could be attributed to additional Ca in limestone form being used as carrier in some feed additives or as a flow agent in SBM and that was not considered in feed formulation. The higher Ca levels occurred to both PC and NC diets and it is not known to what extent this could have affected the response. Previous research has shown a high Ca level or high Ca to P ratio may have a negative impact on performance of pigs (Qian et al., 1996) while excess Ca could also be expected to reduce P digestibility (Létourneau-Montminy et al., 2012). This negative impact applies to all treatments, therefore it is expected that this will not have a major impact on the comparison of the results between PC and phytase treatments. Dicalcium phosphate was totally removed from NC. Based on the DCP inclusion level in PC diets, it can be estimated that the inorganic P removal was 0.14%, 0.11%, 0.07%, 0.05% and 0.03% in the 5 phases respectively. The analyzed Ca reduction in NC diets (in the first 4 phases) was 0.06%, 0.13%, 0.14% and 0.18% compared with PC, confirming an average reduction level of 0.13%.

As expected, pigs fed the NC diet with reduced P, Ca and ME had lower ADFI, ADG and G:F compared with those receiving the PC, which is in agreement with many previous studies (Kies et al., 2006; Zeng et al., 2015).

In the current study, cumulative effects of poor performance in NC diets were observed for the whole trial period, from 12 kg BW until the end of finisher phase 1 when pigs had to be removed from the trial (switched to PC diets). Similarly, in earlier research (Harper et al., 1997) NC diets with reduced available P and Ca caused poorer productive performance in grower and finisher pigs, while phytase supplementation linearly increased ADG in a dose-dependent manner. In piglets (Zeng et al., 2015, 2016, initial BW of 9.5 kg) fed a corn and SBM based diet for 4 weeks, it was observed that a reduction of 0.18% non-phytate P and 0.16% Ca in NC diet significantly reduced ADG and increased feed to gain ratio (F:G) compared with the PC that was formulated to be adequate in available P and Ca. In that study, supplementation of 500, 1,000 and 20,000 FTU/kg *Buttiauxella* phytase linearly improved ADG and reduced F:G ratio, which was closely related to increased phytate degradation rate with increasing phytase dose. Furthermore, in a study in piglets (Jones et al., 2010), a linear response on ADG and F:G ratio was observed with increasing doses (from 200 to 1,000 FTU/kg) of an *E. coli* phytase. Pigs fed the low digestible P, corn and soybean meal based NC diet had a reduction in performance during the 21-day trial period. Brana et al. (2006) observed a linear response in ADG and F:G ratio with increasing doses of *E. coli* phytase from 250 to 1,000 FTU/kg in piglets, grower and finisher phases. In the current study, reductions in ADG were 24%, 20% and 13% in piglets, grower 1 and grower 2 phases, respectively, in NC treatment compared with PC. This is clearly related to the greater degree of digestible P deficiency in the early phases. In the finisher 1 phase, no significant differences were found between the NC and PC dietary groups, which could have been related to: 1) lower level of reduction of the digestible P in the finisher NC diet compared with the PC, due to a lower digestible P requirement; or 2) the pigs in NC group had lower starting BW so the pigs might had compensatory growth. The P deficiency experienced in the NC group after continuous feeding of low digestible P diet from 12 kg BW until finisher 1 (e.g., leg weakness), resulted in these pigs having to be switched to PC diets after finisher 1 for animal welfare reasons. In contrast, all pigs receiving the NC diet with supplemental phytase maintained good health and performance with no significant differences in mortality between phytase treatments and the PC control groups observed.

The cumulative performance data from 12 kg BW to the end of grower phase showed that the addition of phytase at 1,000 FTU/kg improved ADFI and ADG above that achieved with 500 FTU/kg. A notable result from this study is that the phytase at 1,000 FTU/kg showed improved ADG and G:F compared with PC during grower 1 phase. Similar responses were seen in grower pigs from 30 to 85 kg BW, when *Buttiauxella* phytase dosed at 1,000 FTU/kg

improved ADG by 5.3% vs. a positive control (Dersjant-Li et al., 2017b). Since the PC diet was formulated to meet the animals P requirements, the improved performance above that on the PC supports observations that the higher dose phytase could impact the utilization of other nutrients such as energy or amino acids (Woyengo and Nyachoti, 2013). Phytase can not only breakdown phytate and increase the availability of phytate P, but also reduce the negative impacts of phytate on digestion of other nutrients such as amino acids and energy (Selle et al., 2012). In weaned piglets, it has previously been reported that supplementation of an *E. coli* phytase increased ADG, apparent ileal digestibility of CP and amino acids compared with pigs fed a NC diet (Yáñez et al., 2013). In other studies, using *Buttiauxella* phytase, it has been reported that supplementation with this enzyme improved energy and amino acids digestibility in piglets (Zeng et al., 2015), grower pigs (Adedokun et al., 2015; Lizardo et al., 2015) and in broilers (Amerah et al., 2014). Amerah et al. (2014) observed that inclusion of the same phytase at 1,000 FTU/kg improved mean amino acid digestibility by 10 percentage-points compared with the control. In the current study, phytase at 1,000 FTU/kg, added to a NC diet that is lower in ME, maintained or improved ADG and G:F compared with PC, further supporting a potential extra-phosphoric response.

Overall, from 12 kg BW until slaughter, there were no significant differences between pigs fed either the phytase treatments or the PC diet, indicating that phytase at 500 or 1,000 FTU/kg can replace all DCP and replace 0.13% Ca and 0.15 MJ/kg ME in the diet, while maintaining growth performance. When comparing phytase dose rates, the 1,000 FTU/kg dose resulted in greater ADG in the piglets and grower pig phases and better G:F in piglets phase than 500 FTU/kg. Increasing phytase dose from 0 (NC) to 1,000 FTU/kg resulted in incremental increases in ADFI and ADG in all phases up to finisher 1 and increased G:F up to 75 kg. This would support that it is beneficial to use high doses of phytase in piglets and grower phases, and that phytase can replace all inorganic phosphate in pigs fed corn, soybean meal, wheat middling and bakery meal based diets.

The complete replacement of DCP by phytase in the NC diet, together with a reduction of Ca and ME, did not affect carcass weight, lean carcass and yield percentage. Although loin muscle depth was statistically different between the 2 phytase dose levels, they were not significantly different from those fed the PC diets. Pigs fed diets with phytase maintained the same carcass parameters compared with those receiving the PC diets. Harper et al. (1997) reported that carcass quality was not affected by phytase supplementation or available P level. In growing and finishing pigs, the reduction in Ca and available P in the NC diet reduced hot carcass weight, dressing percent and kilograms of carcass lean, while addition of phytase maintained carcass characteristics similar to the PC diet (Shelton et al., 2004).

Replacing DCP and lower dietary energy level based on phytase contribution can be expected to reduce the feed cost. Using data obtained from this study, the cost benefit was calculated for 1,000 pigs. This calculation was based on the actual feed cost, the housing cost and the income from slaughtered pigs with adjustment for mortality. At the time of the study, it was estimated that the net improvement in profit per pig in 500 and 1,000 FTU/kg phytase treatments of \$3.58 and \$4.21, respectively, compared with pigs on the PC diet. This indicated a substantial financial advantage of the higher phytase doses, mainly due to the improved performance in piglets and grower phases.

5. Conclusion

In conclusion, 500 to 1,000 FTU/kg *Buttiauxella* sp. phytase was able to totally replace inorganic phosphate and maintain performance and carcass characteristics in commercial pigs fed corn, soybean meal, wheat middling and bakery meal based diets from 12 kg BW till slaughter. In this study, phytase at 1,000 FTU/kg gave the best responses in younger pigs, whereas, in the finishing period up to slaughter, feeding phytase at 500 FTU/kg diets resulted in the best G:F.

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