# **Degradation of dietary fiber in the stomach, small intestine, and large intestine of growing pigs fed corn- or wheat-based diets without or with microbial xylanas[e1](#page-0-0)**

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**ABSTRACT:** An experiment was conducted to test the hypothesis that microbial xylanases may contribute to the degradation of fiber in wheat and wheat-based diets and in corn and corn-based diets along the intestinal tract of pigs. Twenty-four growing barrows (initial BW:  $28.51 \pm 1.86$  kg) were prepared with a T-cannula in the proximal duodenum and another T-cannula in the distal ileum and allotted to a replicated  $12 \times 4$  Youden square design with 12 diets and four 18-d periods. Two diets based on corn and soybean meal (**SBM**) or corn, SBM, and 30% distillers dried grains with solubles (**DDGS**) were formulated and two diets based on wheat and SBM or wheat, SBM, and 30% wheat middlings were also formulated. The four diets were formulated without microbial xylanase, or with one of two microbial xylanases (xylanase A or xylanase B) for a total of 12 diets. Feces and urine were collected on days 8 to 13, ileal digesta were collected on days 15 and 16, and duodenal digesta were collected on days 17 and 18 of each period. The apparent duodenal digestibility (**ADD**), apparent ileal digestibility (**AID**), and apparent total tract digestibility (**ATTD**) of GE, nutrients, and dietary fiber were calculated. Results

indicated that the AID of GE in corn-SBM or wheat-SBM diets was greater  $(P < 0.05)$  than in the corn-SBM-DDGS and wheat-SBM-wheat middlings diets, but no difference was observed for the AID of dietary fiber between wheat-SBM and wheat-SBM-wheat middlings diets. The ATTD of dietary fiber was also greater  $(P < 0.05)$ in corn-SBM and wheat-SBM diets compared with corn-SBM-DDGS and wheat-SBM-wheat middlings diets, which indicates that the concentration of dietary fiber may influence the degree of fermentation of fiber. Inclusion of xylanase A or B improved  $(P < 0.05)$  the ADD and the ATTD of dietary fiber in wheat-based diets, indicating activity of xylanase in the gastro-intestinal tract of pigs. Inclusion of xylanase A improved  $(P < 0.05)$  the concentration of DE and ME in wheat-SBM-wheat middlings diets and xylanase B improved  $(P < 0.05)$  the concentration of DE in wheat-based diets and improved  $(P < 0.05)$  the concentration of the ME in wheat-SBM diet. In conclusion, the xylanases used in this experiment improved the digestibility of dietary fiber in the stomach and hindgut and improved the energy status of pigs fed wheat-based diets, but not of pigs fed corn-based diets.

**Key words:** dietary fiber, digestibility, energy, pigs, stomach, xylanase

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## **INTRODUCTION**

Corn and wheat and co-products from these grains contain considerable quantities of arabinoxylans [\(Jaworski et al., 2015](#page-13-0)), but the response to microbial xylanases is often greater in wheatbased diets than in corn-based diets although the

reason for this observation has not been elucidated. Degradation of non-starch polysaccharides in the intestinal tract varies depending on the structure and physicochemical characteristics of the non-starch polysaccharides [\(Bach Knudsen, 2001](#page-13-1)), which indicates that the rate of degradation of non-starch polysaccharides, and consequently the effect of microbial xylanase, may vary among ingredients. Results of a recent experiment indicated that most of the soluble dietary fiber (**SDF**) was fermented prior to the colon, whereas most fermentation of insoluble dietary fiber (**IDF**) takes place in the colon [\(Jaworski and Stein,](#page-13-2)  [2017\)](#page-13-2). Fiber in DDGS or soybean hulls is less fermentable compared with fiber in wheat middlings further indicating that differences among different types of fiber exist ([Jaworski and Stein, 2017\)](#page-13-2). It is also possible that the type of fiber influences the site in the intestinal tract where fiber will be fermented and if that is the case, it is likely that microbial xylanase will have different activities in different sections of the intestinal tract. However, degradation of individual dietary fiber fractions in corn and wheatbased diets in different sections of the gastrointestinal tract of pigs has not been reported. Therefore, an experiment was conducted to test the hypothesis that microbial xylanases contribute to the degradation of fiber in wheat and wheat-based diets and in corn and corn-based diets at different sites of the intestinal tract. The objectives of the experiment were to quantify the degradation of dietary fiber fractions in the stomach, small intestine, and large intestine of pigs and to determine the effect of xylanase on degradation of dietary fiber fractions in corn and wheat and their co-products.

## **MATERIALS AND METHODS**

The protocol for this experiment was approved by the Institutional Animal Care and Use Committee at the University of Illinois. Pigs that were the offspring of PIC L359 boars mated to Camborough females (Pig Improvement Company, Hendersonville, TN) were used.

### *Animals, Housing, Diets, and Experimental Design*

Twenty-four growing barrows (initial BW:  $28.51 \pm 1.86$  kg) were prepared with a T-cannula in the proximal duodenum and another T-cannula in the distal ileum [\(Stein et al., 1998](#page-14-0)). Pigs were housed individually in metabolism crates with a fully slatted floor, a fecal collection screen, a urine tray, a feeder, and a nipple drinker. Feeding of experimental diets was initiated 7 d after surgery. Water was available at all times.

Two diets based on corn and SBM or corn, SBM, and 30% distillers dried grains with solubles (**DDGS**) were formulated to meet nutrient requirements for 25 to 50 kg growing pigs ([NRC,](#page-14-1)  [2012](#page-14-1); [Table 1\)](#page-2-0). Two additional diets based on wheat and SBM or wheat, SBM, and 30% wheat middlings were also formulated. The four diets were formulated without microbial xylanase, or with one of two microbial xylanases (16,000 units per kg of xylanase A or xylanase B; Danisco Animal Nutrition-DuPont Industrial Biosciences, Marlborough, UK) for a total of 12 diets. All diets contained microbial phytase (1,000 phytase units per kg; Axtra PHY; Danisco Animal Nutrition-DuPont Industrial Biosciences, Waukesha, WI). Titanium dioxide was included at 0.40% in all diets as an indigestible marker.

The 24 pigs were allotted to a replicated  $12 \times 4$ Youden square design with 12 diets and four periods using the Balanced Latin Square Designer ([Kim and Stein, 2009](#page-13-3)). Within each period, two pigs received each diet for a total of eight replicate pigs per diet for the four periods. The daily feed allowance was calculated to provide 3.2 times the estimated requirement for maintenance energy (i.e., 197 kcal ME/kg $0.6$ ; [NRC, 2012](#page-14-1)) and was divided into two equal meals that were fed at 0800 and 1600 hours, respectively. All diets were fed in a meal form. The BW of each pig was recorded at the beginning of the experiment and at the end of each period.

Each period lasted 18 d. The initial 7 d was an adaptation period to the diets. Feces and urine were collected from the feed provided from days 8 to 13 following the marker to marker approach [\(Adeola,](#page-13-4) [2001\)](#page-13-4). Ileal digesta were collected on days 15 and 16, and duodenal digesta were collected on days 17 and 18 ([González-Vega et al., 2014\)](#page-13-5). Digesta were collected by attaching a 225-mL plastic bag to the cannula barrel, which allowed digesta to flow into the bag. Bags were replaced every 30 min or whenever full. Immediately after collection, digesta were stored at  $-20$  °C.

The total volume of urine was measured when collected, and  $20\%$  of the volume was stored at  $-$ 20 °C. At the end of each collection period, urine was thawed, filtered through cheesecloth, subsampled, and freeze-dried for analysis. At the conclusion of the experiment, the duodenal digesta and ileal digesta were thawed, sub-sampled, lyophilized, and then ground. Fecal samples were thawed, mixed, dried for 120 h in a 65 °C drying oven, and ground through a 1-mm screen in a Wiley Mill (Model 4; Thomas Scientific, Swedesboro, NJ), and then subsampled.

<span id="page-2-0"></span>**Table 1.** Ingredient composition and calculated chemical composition of experimental diets

Item	Corn-SBM <sup>1</sup>	Corn-SBM- DDGS <sup>1</sup>	Wheat-SBM	Wheat-SBM-wheat middlings
Ingredient, %				
Corn	71.40	47.55		
<b>DDGS</b>		30.00		
Wheat			73.75	44.88
Wheat middlings			$\overline{\phantom{a}}$	30.00
SBM, 48% CP	24.00	18.00	22.00	21.00
Soybean oil	1.00	1.00	1.00	1.00
Limestone	1.17	1.44	1.40	1.45
Dicalcium phosphate	0.56	0.18	0.14	
L-Lys HCl, 78% Lys	0.28	0.33	0.21	0.17
DL-Met	0.03			
L-Thr	0.06			
Vitamin-mineral premix <sup>2</sup>	0.30	0.30	0.30	0.30
Sodium chloride	0.30	0.30	0.30	0.30
Titanium dioxide	0.40	0.40	0.40	0.40
Phytase premix <sup>3, 4</sup>	0.50	0.50	0.50	0.50
Calculated values <sup>5</sup>				
NE, kcal/kg	2,476	2,416	2,348	2,247
CP, %	17.38	20.76	21.24	21.31
Ca, %	0.66	0.66	0.66	0.66
Standardized total tract digestible P, %	0.31	0.31	0.31	0.40
Amino acids <sup>6</sup> , %				
Arg	1.01	1.04	1.12	1.23
His	0.42	0.48	0.47	0.49
Ile	0.62	0.69	0.73	0.71
Leu	1.36	2.08	1.30	1.28
Lys	0.98	0.98	0.98	0.98
Met	0.28	0.32	0.27	0.27
$Met + Cys$	0.56	0.61	0.62	0.61
Phe	0.74	0.87	0.89	0.86
Thr	0.59	0.60	0.60	0.60
Trp	0.18	0.17	0.24	0.24
Val	0.69	0.81	0.80	0.81

 ${}^{1}$ SBM = soybean meal; DDGS = distillers dried grains with solubles.

2 Provided the following quantities of vitamins and micro-minerals per kilogram of complete diet: vitamin A as retinyl acetate, 11,136 IU; vitamin D<sub>3</sub> as cholecalciferol, 2,208 IU; vitamin E as  $PL-a$  tocopheryl acetate, 66 IU; vitamin K as menadione dimethylprimidinol bisulfite, 1.42 mg; thiamin as thiamine mononitrate, 0.24 mg; riboflavin, 6.59 mg; pyridoxine as pyridoxine hydrochloride, 0.24 mg; vitamin  $B_{12}$ , 0.03 mg; p-pantothenic acid as p-calcium pantothenate, 23.5 mg; niacin, 44.1 mg; folic acid, 1.59 mg; biotin, 0.44 mg; Cu, 20 mg as copper sulfate and copper chloride; Fe, 126 mg as ferrous sulfate; I, 1.26 mg as ethylenediamine dihydriodide; Mn, 60.2 mg as manganese sulfate; Se, 0.3 mg as sodium selenite and selenium yeast; and Zn, 125.1 mg as zinc sulfate.

3 The phytase premix contained 200,000 units of microbial phytase (Axtra PHY; Danisco Animal Nutrition-DuPont Industrial Biosciences, Waukesha, WI) per kilogram, which resulted in addition of 1,000 units per kilogram of microbial phytase in the complete diet.

4 Four additional diets that were identical to the diets above were formulated by including xylanase A in the phytase premix and another four diets were formulated by including xylanase B in the phytase premix. Each of the two xylanases was included in the premixes in quantities that provided 16,000 units of xylanase in the final diet. Both xylanases A and B were experimental xylanases produced by Danisco Animal Nutrition-DuPont Industrial Biosciences (Marlborough, UK).

<sup>5</sup>Calculated from [NRC \(2012\)](#page-14-1).

6 Amino acids are indicated as standardized ileal digestible AA.

#### *Chemical Analyses*

All ingredients, diets, duodenal digesta, ileal digesta, and fecal samples were analyzed for DM (Method 930.15; [AOAC Int., 2007\)](#page-13-6) and ash (Method 942.05; [AOAC Int., 2007](#page-13-6)). These samples were also analyzed for ADF and NDF using Ankom Technology method 12 and 13, respectively (Ankom2000 Fiber Analyzer, Ankom Technology, Macedon, NY) and ADL using Ankom Technology method 9 (Ankom Daisy<sup>II</sup> Incubator, Ankom

Technology, Macedon, NY). Samples were also analyzed for SDF and IDF according to Method 991.43 [\(AOAC Int., 2007](#page-13-6)) using the Ankom<sup>TDF</sup> Dietary Fiber Analyzer (Ankom Technology, Macedon, NY).

Samples were analyzed for CP using the combustion procedure (Method 990.03; [AOAC Int.,](#page-13-6)  [2007](#page-13-6)) on an Elementar Rapid N-cube protein/ nitrogen apparatus (Elementar Americas Inc., Mt. Laurel, NJ); aspartic acid was used as a calibration standard, and CP was calculated as  $N \times 6.25$ . Diets and ingredients were analyzed for AA on a Hitachi AA Analyzer, Model No. L8800 (Hitachi High Technologies America, Inc., Pleasanton, CA) using ninhydrin for postcolumn derivatization and norleucine as the internal standard (Method 982.30 E (a, b, c); [AOAC Int., 2007\)](#page-13-6). All diets and ingredients were analyzed for acid hydrolyzed ether extract using 3*N* HCl on the ANKOM<sup>HCl</sup> Hydrolysis System (ANKOM Feed Technology, Macedon, NY) followed by crude fat extraction using petroleum ether on an ANKOM<sup>XT15</sup> Extractor (Method: AOCS Am 5-04; ANKOM Feed Technology, Macedon, NY).

Titanium concentration in diets, duodenal digesta, and ileal digesta samples was analyzed following the procedure of [Myers et al. \(2004\)](#page-13-7). All ingredients, diets, duodenal digesta, ileal digesta, freeze-dried urine, and fecal samples were analyzed in duplicate for GE using bomb calorimetry (Model 6300; Parr Instruments, Moline, IL), with benzoic acid used as a calibration standard. Diets and ingredients were also analyzed for Ca and P by inductively coupled plasma-optical emission spectrometry (Method 985.01 A, B, and C; [AOAC Int.,](#page-13-6)  [2007](#page-13-6)) after wet ash sample preparation [Method 975.03 B(b); [AOAC Int., 2007\]](#page-13-6).

## *Calculations*

Values for cellulose, insoluble hemicellulose, total dietary fiber (**TDF**), non-starch polysaccharides, insoluble non-starch polysaccharides, and non-cellulosic non-starch polysaccharides were calculated in ingredients, diets, duodenal digesta, ileal digesta, and fecal samples [\(Table 2\)](#page-3-0). The apparent

<span id="page-3-0"></span>**Table 2.** Calculation of dietary fiber components

duodenal digestibility (**ADD**), the apparent ileal digestibility (**AID**), and the apparent total tract digestibility (**ATTD**) of GE in each diet were calculated [\(Stein et al., 2007;](#page-14-2) [NRC, 2012](#page-14-1)), and the GE in feces and urine samples was subtracted from the GE in diets to calculate DE and ME of each diet [\(Adeola, 2001\)](#page-13-4). The ADD, AID, and ATTD of DM, ash, OM, CP, ADL, ADF, NDF, cellulose, insoluble hemicellulose, IDF, SDF, TDF, non-starch polysaccharides, insoluble non-starch polysaccharides, and non-cellulosic non-starch polysaccharides were also calculated.

### *Statistical Analyses*

All data were analyzed following a  $2 \times 2 \times$ 3 design with two types of diets (corn based or wheat based), two levels of fiber (low or high), and three microbial xylanase treatments (none, xylanase A, or xylanase B) using the MIXED procedure of SAS (SAS Inst. Inc., Cary, NC) with pig as the experimental unit. The model included diet, fiber, xylanase, diet  $\times$  fiber, diet  $\times$  xylanase, fiber  $\times$  xylanase, and diet  $\times$  fiber  $\times$  xylanase as fixed effects, and pig and period as random effects. Least square means were calculated for each independent variable, and means were separated using the PDIFF option. The significance among dietary treatments was determined at  $P \leq 0.05$  for all analyses.

#### **RESULTS**

## *Ingredients and Diets*

The analyzed nutrient composition of ingredients and diets was close to expected values ([Tables 3](#page-4-0) and [4\)](#page-5-0). The analyzed values for xylanase in all diets containing xylanase A or xylanase B were more than 16,000 units of xylanase per kg. The analyzed phytase values were between 636 and 828 phytase units for all corn-based diets, close to 1,000 phytase units per kilogram for the wheat-SBM diets, and wheat-SBM-wheat middlings diets contained between 1,376 and 1,549 phytase units per kg.



<span id="page-4-0"></span>



1 DDGS = distillers dried grains and solubles; SBM = soybean meal.

2 AEE = acid hydrolyzed ether extract.

3 Calculated values: OM = DM—ash; cellulose = ADF—ADL; insoluble hemicellulose = NDF—ADF; total dietary fiber = insoluble dietary fiber + soluble dietary fiber; nonstarch polysaccharides = total dietary fiber—ADL; insoluble nonstarch polysaccharides = nonstarch polysaccharides—soluble dietary fiber; noncellulosic nonstarch polysaccharide = nonstarch polysaccharide—cellulose.

## *Apparent Duodenal, Ileal, and Total Tract Digestibility*

Addition of xylanase B to the corn-SBM diet reduced ( $P < 0.05$ ) the ADD of GE, DM, and OM, but that was not the case if added to the corn-SBM-DDGS diets or wheat-based diets (grain source  $\times$  fiber concentration  $\times$  xylanase interaction,

*P* < 0.05; [Table 5\)](#page-7-0). Inclusion of xylanase A improved  $(P < 0.05)$  the ADD of GE, DM, and OM in wheat-SBM-wheat middlings diets, but no difference was observed if added to the wheat-SBM or corn-based diets (grain source  $\times$  fiber concentration  $\times$  xylanase interaction,  $P < 0.05$ ). There was an interaction  $(P = 0.01)$  between fiber concentration and xylanase

### <span id="page-5-0"></span>**Table 4.** Analyzed composition of diets



 ${}^{1}SBM =$  soybean meal.

2 Xylanases A and B were experimental xylanases supplied by Danisco Animal Nutrition-DuPont Industrial Biosciences (Marlborough, UK).

3 AEE = acid hydrolyzed ether extract.

4 Calculated values: OM = DM—ash; cellulose = ADF—ADL; insoluble hemicellulose = NDF—ADF; total dietary fiber = insoluble dietary fiber + soluble dietary fiber; nonstarch polysaccharides = total dietary fiber—ADL; insoluble non-starch polysaccharides = nonstarch polysaccharides—soluble dietary fiber; non-cellulosic nonstarch polysaccharide = nonstarch polysaccharide—cellulose.

for the ADD of ash with xylanase A reducing the ADD of ash in the corn-SBM diet, but xylanase B increased the ADD of ash in the wheat-SBM diet.

The ADD of SDF was reduced if xylanase B was added to the corn-SBM diet, but not to the corn-SBM-DDGS diet, but for the wheat-based diets, no differences among treatments were observed for the ADD of SDF (fiber concentration  $\times$  xylanase interaction,  $P \leq 0.05$ ). Addition of xylanase B to wheat-based diets improved  $(P < 0.05)$  the ADD of ADF and non-cellulosic non-starch polysaccharides, but no difference was observed if added to the corn-based diets (grain source  $\times$  xylanase interaction,  $P < 0.05$ ). Inclusion of xylanase A or B to the wheat-based diets improved  $(P < 0.05)$  the ADD of NDF, cellulose, insoluble hemicellulose, IDF, TDF, non-starch polysaccharides, and insoluble non-starch polysaccharides, but no difference was observed if xylanase was added to the cornbased diets (grain source × xylanase interaction, *P*  $< 0.05$ ).

The ADD of CP in wheat-SBM-wheat middlings diets was greater ( $P < 0.05$ ) than in wheat-SBM diets, but no difference was observed between corn-SBM and corn-SBM-DDGS diets (grain source  $\times$  fiber concentration interaction,  $P \le 0.05$ ). The ADD of ADF in corn-SBM diets was less  $(P < 0.05)$  than in the corn-SBM-DDGS diets, but the ADD of ADF in wheat-SBM diets was greater  $(P < 0.05)$  than in the wheat-SBM-wheat middlings diets (grain source  $\times$  fiber concentration interaction, *P* < 0.05). The ADD of NDF in wheat-SBM diets was greater  $(P < 0.05)$  than in wheat-SBM-wheat middlings diets, but no difference was observed between corn-SBM and corn-SBM-DDGS diets (grain source  $\times$  fiber concentration interaction, *P* < 0.05). The ADD of ADL and insoluble hemicellulose in corn-SBM-DDGS diets was greater  $(P < 0.05)$  than in corn-SBM diets, but no difference was observed between wheat-SBM and wheat-SBM-wheat middlings diets (grain source  $\times$  fiber concentration interaction,  $P < 0.05$ ).

The AID of NDF, cellulose, insoluble hemicellulose, and SDF in corn-SBM-DDGS diets was greater ( $P < 0.05$ ) than in corn-SBM diets, but no difference was observed between wheat-SBM and wheat-SBM-wheat middlings diets (grain source  $\times$ fiber concentration interaction,  $P \le 0.05$ ; [Table 6](#page-8-0)). The improvement in the AID of ADL and ADF was greater  $(P < 0.05)$  if DDGS was added to the corn-based diets than if wheat middlings was added to the wheat-based diets (grain source  $\times$  fiber concentration interaction,  $P \le 0.05$ ). The AID of ash in wheat-SBM diets was greater than  $(P < 0.05)$ in wheat-SBM-wheat middlings diets, but no difference was observed between corn-SBM and corn-SBM-DDGS diets (grain source × fiber concentration interaction,  $P \leq 0.05$ ). The AID of GE in corn-based diets was greater  $(P < 0.05)$  than in wheat-based diets and the AID of CP, IDF, TDF, non-starch polysaccharides, insoluble non-starch polysaccharides, and non-cellulosic non-starch polysaccharides in wheat-based diets was greater  $(P < 0.05)$  than in corn-based diets. The AID of GE, DM, OM, and CP in diets without DDGS or wheat middlings was greater  $(P < 0.05)$  than in diets containing DDGS or wheat middlings.

Inclusion of xylanase A or B improved  $(P < 0.05)$  the ATTD of GE, DM, OM, NDF, insoluble hemicellulose, IDF, TDF, and insoluble non-starch polysaccharides in wheat-based diets, but that was not the case for corn-based diets (grain source  $\times$  xylanase interaction,  $P \le 0.05$ ; [Table 7](#page-9-0)). The ATTD of ash, ADF, NDF, cellulose, and insoluble hemicellulose in wheat-SBM diets was greater  $(P < 0.05)$  than in wheat-SBM-wheat middlings diets, but no difference was observed between corn-SBM and corn-SBM-DDGS diets (grain source  $\times$  fiber concentration interaction,  $P < 0.05$ ). The reduction in ATTD of insoluble non-starch polysaccharides was greater  $(P < 0.05)$  if DDGS was added to corn-based diets than if wheat middlings was added to wheat-based diets, and the improvement in ATTD of ADL was greater  $(P < 0.05)$  if DDGS was added to corn-based diets than if wheat middlings was added to wheat-based diets (grain source  $\times$  fiber concentration interaction,  $P \le 0.05$ ).

The ATTD of SDF in corn-SBM-DDGS diets was greater  $(P < 0.05)$  than in corn-SBM diets, but no difference was observed between wheat-SBM and wheat-SBM-wheat middlings diets (grain source  $\times$  fiber concentration interaction,  $P \le 0.05$ ). The ATTD of non-cellulosic non-starch polysaccharides in corn-SBM diets was greater  $(P < 0.05)$ than in corn-SBM-DDGS diets, but less  $(P < 0.05)$ in wheat-SBM diets than in wheat-SBM-wheat middlings diets (grain source  $\times$  fiber concentration interaction,  $P \le 0.05$ ). The ATTD of CP and non-starch polysaccharides in wheat-based diets was greater  $(P < 0.05)$  than in corn-based diets and the ATTD of CP and non-starch polysaccharides in diets without DDGS and wheat middlings was greater  $(P < 0.05)$  than in diets with DDGS and wheat middlings.

## *Concentration of DE and ME in the Diets*

The reduction in the concentration of DE and ME was greater  $(P < 0.05)$  if wheat middlings was added to the wheat-based diets than if DDGS was added to the corn-based diets (grain source  $\times$  fiber concentration interaction,  $P < 0.05$ ; [Table 8](#page-10-0)). The concentration of DE in wheat-based diets was improved ( $P < 0.05$ ) if xylanase B was used, but no



Data are means of eight observations per treatment. 1Data are means of eight observations per treatment.

 $PDF$  = insoluble dietary fiber; SDF = soluble dietary fiber; TDF = total dietary fiber; NSP = nonstarch polysaccharides.  $21DF =$  insoluble dietary fiber; SDF = soluble dietary fiber; TDF = total dietary fiber; NSP = nonstarch polysaccharides.

<span id="page-7-0"></span> ${}^{3}\mathrm{SBM}$  = soybean meal. 3SBM = soybean meal.

Xylanases A and B were experimental xylanases supplied by Danisco Animal Nutrition-DuPont Industrial Biosciences (Marlborough, UK). 4Xylanases A and B were experimental xylanases supplied by Danisco Animal Nutrition-DuPont Industrial Biosciences (Marlborough, UK).

5DDGS = distillers dried grains with solubles. 5DDGS = distillers dried grains with solubles.





1Data are means of eight observations per treatment.

'Data are means of eight observations per treatment.<br>2DF = insoluble dietary fiber; SDF = soluble dietary fiber; TDF = total dietary fiber; NSP = nonstarch polysaccharides. 2IDF = insoluble dietary fiber; SDF = soluble dietary fiber; TDF = total dietary fiber; NSP = nonstarch polysaccharides.

<span id="page-8-0"></span> ${}^{3}SBM =$  soybean meal. 3SBM = soybean meal.

"Xylanases A and B were experimental xylanases supplied by Danisco Animal Nutrition-DuPont Industrial Biosciences (Marlborough, UK). 4Xylanases A and B were experimental xylanases supplied by Danisco Animal Nutrition-DuPont Industrial Biosciences (Marlborough, UK).

5DDGS = distillers dried grains with solubles. 5DDGS = distillers dried grains with solubles.



**Table 7.** Apparent total tract digestibility of energy, nutrients, and dietary fiber of pigs fed corn- and wheat-based diets, %1

1Data are means of eight observations per treatment. 1Data are means of eight observations per treatment.

 ${}^{2}IDF$  = insoluble dietary fiber; SDF = soluble dietary fiber; TDF = total dietary fiber; NSP = nonstarch polysaccharides. 2IDF = insoluble dietary fiber; SDF = soluble dietary fiber; TDF = total dietary fiber; NSP = nonstarch polysaccharides.

<span id="page-9-0"></span> ${}^3{\rm SBM}$  = soybean meal. 3SBM = soybean meal.

"Xylanases A and B were experimental xylanases supplied by Danisco Animal Nutrition-DuPont Industrial Biosoiences (Marlborough, UK). 4Xylanases A and B were experimental xylanases supplied by Danisco Animal Nutrition-DuPont Industrial Biosciences (Marlborough, UK). 5DDGS = distillers dried grains with solubles. 5DDGS = distillers dried grains with solubles.

<span id="page-10-0"></span>

difference was observed if xylanase B was added to the corn-based diets (grain source  $\times$  xylanase interaction,  $P < 0.05$ ). Inclusion of xylanase B improved  $(P < 0.05)$  the concentration of ME in wheat-SBM diets and xylanase A improved ( $P < 0.05$ ) the concentration of DE and ME in wheat-SBM-wheat middlings diets.

## **DISCUSSION**

All diets were fortified with the same concentrations of microbial phytase, but it is likely that wheat and wheat middlings contained endogenous phytase, which resulted in the greater analyzed concentrations of phytase in these diets compared with the corn-based diets. Because diets were not heat treated, the endogenous phytase was likely intact in the feed and although the bio-efficacy of the endogenous wheat phytase is less than that of the microbial phytase, the analyzed values reflect the total concentration of microbial and endogenous phytase. The microbial phytase and the xylanase that were added to the diets were prepared as one premix and the fact that phytase did not appear to be included above the intended level indicates that premix inclusion in the diets was as intended. The much greater analyzed values for xylanase B compared with intended levels, therefore, appears to be

a result of an unintended overage of xylanase B in the premix.

Commercial xylanases are usually included in diets at concentrations of ~4,000 units. The reason the intended inclusion in this experiment was 16,000 units was that we hypothesized that by including superdosing levels of xylanase we might have a greater chance of increasing the digestibility of energy in the corn-based diets, where a xylanase response is often elusive. Thus, because the hypothesis was that xylanase increases degradation of fiber, we ensured that there was sufficient amount of the enzyme even in the high-fiber diets.

The Youden square design that was used in this experiment resulted in pigs being fed four different diets during the experiment. This design was used to minimize the number of animals that needed to have intestinal cannulas installed, but the design assumes that there is no carry-over effect from one diet to another. The 7-d adaptation period that was used for each diet is believed to be sufficient to obtain a steady fermentation of fiber because it has been demonstrated that fiber fermentation is maximized after 5 d of feeding a diet [\(Jaworski et al.,](#page-13-8) [2016](#page-13-8)). In other similar studies, a 5-d adaptation period has been used ([Kiarie et al., 2016\)](#page-13-9), further indicating that the 7-d adaptation period used in this experiment likely was adequate.

The low ADD of GE, DM, OM, CP, and all dietary fiber components that was observed for all diets was expected because there is limited absorption of these components in the stomach [\(Wilfart](#page-14-3) [et al., 2007](#page-14-3); [Cadogan and Choct, 2015\)](#page-13-10). The low ADD of most fiber components indicates that insoluble fiber is not fermented in the stomach. However, it appears that, particularly for wheatbased diets, some solubilization of non-cellulosic non-starch polysaccharides takes place in the stomach or the first part of the duodenum. The reason some of the SDF disappeared in the stomach likely is that some parts of fiber solubilize in the liquid environment in the stomach, as previously reported in sows ([Planas, 1999](#page-14-4)). It appears that SDF from wheat and wheat middlings are more soluble in the early part of the digestive tract than SDF from corn or corn-DDGS, but microbial xylanases do not appear to influence solubility of dietary fiber in the stomach of pigs fed corn-based diets.

The AID of GE in the corn-SBM diets that was calculated in this experiment is in agreement with values from previous experiments ([Urriola](#page-14-5)  [and Stein, 2012;](#page-14-5) [Gutierrez et al., 2016](#page-13-11)), although lower AID of DM and OM also has been reported ([Passos et al., 2015](#page-14-6)). Likewise, the AID of GE in the corn-SBM-DDGS diets was within the range of previous estimates ([Urriola and Stein, 2010](#page-14-7); [Ndou et al., 2015;](#page-13-12) [Gutierrez et al., 2016;](#page-13-11) [Moran](#page-13-13)  [et al., 2016](#page-13-13); [Jaworski and Stein, 2017\)](#page-13-2). However, the observation that there was no effect of xylanase supplementation on the AID of GE in the corn-SBM or DDGS diets is in contrast with previous reports, where supplementation of xylanase improved ([Ndou et al., 2015](#page-13-12)) or reduced [\(Moran et al., 2016](#page-13-13)) the AID of GE in corn-SBM-DDGS diets.

The reason the AID of GE, DM, OM, and CP in the corn-SBM-DDGS diets was less than in corn-SBM diets is most likely that DDGS increased the dietary fiber concentration, thereby reducing digestibility and increasing endogenous nutrient losses ([Grieshop et al., 2001](#page-13-14); [Souffrant, 2001](#page-14-8); [Urriola](#page-14-7)  [and Stein, 2010](#page-14-7)). These results are in agreement with previous data ([Gutierrez et al., 2016;](#page-13-11) [Jaworski](#page-13-2)  [and Stein, 2017\)](#page-13-2). The observation that the AID of NDF and ADF was greater in the corn-SBM-DDGS diets than in the corn-SBM diets is likely a result of the fact that there was more substrate in the corn-SBM-DDGS diets than in the corn-SBM diets, and this observation is also in agreement with previous data [\(Urriola and Stein, 2010](#page-14-7)). However, a lack of a difference in the AID of NDF and ADF between a corn-SBM-DDGS diets and corn-SBM diets has also been reported [\(Urriola and Stein,](#page-14-7)  [2010](#page-14-7); [Gutierrez et al., 2016](#page-13-11); [Jaworski and Stein,](#page-13-2) [2017](#page-13-2)). The negative AID of ADF, cellulose, and SDF that was observed in this experiment may have been a result of the fact that some compounds that are secreted by the animal into the intestinal tract are analyzed as fiber (Cervantes‐[Pahm et al., 2014](#page-13-15); [Montoya et al., 2015,](#page-13-16) [2016,](#page-13-17) [2017\)](#page-13-18). Likewise, the negative AID of ADL that was observed may be a result of cutin and other non-lignin carbohydrates that are analyzed as lignin in the ADL procedure ([Van Soest and Wine, 1968;](#page-14-9) [Cherney, 2000\)](#page-13-19).

The AID of GE in the wheat-SBM diets is in agreement with previous data [\(Cadogan and](#page-13-10) [Choct, 2015](#page-13-10)) and the AID of GE in the wheat-SBM-wheat middlings diets is also in agreement with data by [Jaworski and Stein \(2017\)](#page-13-2), but a lower AID of GE has also been reported [\(Moran et al.,](#page-13-13)  [2016](#page-13-13)). There is, however, a considerable difference in the composition of wheat middlings among suppliers, which may be the reason different results for the AID of GE have been observed. The greater AID of GE, DM, OM, and CP that was observed for the wheat-SBM diets than for the wheat-SBMwheat middlings diets is likely a result of greater concentration of dietary fiber in wheat middlings than in wheat and SBM, which may have resulted in reduced digestibility of other nutrients as was also observed for the corn-SBM-DDGS diets. The observation that there was no difference in the AID of fiber fractions between the wheat-SBM and the wheat-SBM-wheat middlings diets, which has also been reported previously ([Jaworski and Stein,](#page-13-2) [2017](#page-13-2)), indicates that the fiber fractions from wheat are fermented at the same rate regardless of the concentration in the diet. The low, but highly variable, AID of dietary fiber fractions that was observed in this experiment is in agreement with previous data ([Bach Knudsen et al., 2013](#page-13-20); [Jaworski and Stein,](#page-13-2) [2017](#page-13-2)), and reflects the fact that there is limited fermentation of dietary fiber in the small intestine of pigs. The lack of responses to the xylanases on AID of GE or nutrients is in agreement with some previous data [\(Yáñez et al., 2011;](#page-14-10) [Moran et al., 2016](#page-13-13)), although a positive response to xylanase has also been reported ([Diebold et al., 2004;](#page-13-21) [Nortey et al.,](#page-14-11) [2007](#page-14-11)).

The observed values for ATTD of DM, GE, CP, ADF, NDF, cellulose, and insoluble hemicellulose in all diets are within the range of reported data ([Yin et al., 2000;](#page-14-12) [Urriola and Stein, 2010](#page-14-7); [Gutierrez et al., 2016](#page-13-11); [Moran et al., 2016;](#page-13-13) [Jaworski](#page-13-2) [and Stein, 2017](#page-13-2); [Tsai et al., 2017\)](#page-14-13) and the observed values for the ATTD of insoluble hemicellulose, IDF, TDF, non-starch polysaccharides, insoluble non-starch polysaccharides, and non-cellulosic non-starch polysaccharides are in agreement with previous data ([Jaworski and Stein, 2017](#page-13-2)). The observation that values for the ATTD of cellulose is greater than the ATTD of hemicellulose in corn-SBM and corn-SBM-DDGS diets, but not in diets based on wheat-SBM or wheat-SBM-wheat middlings indicates that the insoluble hemicellulose in corn is less fermentable than in wheat, whereas the cellulose in corn may be more fermentable than in wheat. Fermentability of cellulose is related to the proportion of amorphous cellulose, and the present results indicate that cellulose from corn may be more amorphous and less crystalline than cellulose from wheat. This difference in the fermentability of cellulose between corn- and wheat-based diets is the reason the ATTD of ADF is similar to the ATTD of NDF in corn-based diets, whereas the ATTD of ADF is much less than of NDF in wheat-based diets as was observed in this experiment.

The greater ATTD of GE, DM, CP, and OM in the corn-SBM and wheat-SBM diets than in corn-SBM-DDGS and wheat-SBM-wheat middlings diets is likely a result of the greater concentration of dietary fiber in DDGS and wheat middlings than in corn, wheat, and SBM, which may have reduced nutrient digestibility in diets containing DDGS or wheat middlings. Dietary fiber may serve as a structural barrier for digestion because of hindering the access of digestive enzymes to starch, CP, and possibly other nutrients ([Jørgensen, 1996;](#page-13-22) [Le Gall,](#page-13-23) [2009](#page-13-23); [de Vries, 2014](#page-14-14)). The observation that the ATTD of IDF, TDF, non-starch polysaccharides, insoluble non-starch polysaccharides, and non-cellulosic non-starch polysaccharides in corn-SBM diets is greater than in the corn-SBM-DDGS diets indicates that these dietary fiber components in corn are more fermentable if present in the diet in reduced concentrations. This observation also indicates that the fermentation process during ethanol production does not solubilize or de-lignify dietary fiber in corn, which is in agreement with data, indicating that acid extrusion of DDGS did not affect the degradation of non-starch polysaccharides in corn DDGS ([de Vries et al., 2014\)](#page-14-14). The observation that the ATTD of NDF, ADF, and cellulose in corn-based diets is not influenced by the presence of DDGS in the diets indicates that dietary fiber components in corn and DDGS are fermented to the same degree regardless of the concentration in the diet, which is likely a result of the fact that the percentage of arabinoxylans and cellulose in the non-starch polysaccharides of DDGS is not different from that of corn [\(Jaworski et al., 2015\)](#page-13-0).

The observed greater ATTD of NDF, ADF, cellulose, insoluble hemicellulose, IDF, TDF, non-starch polysaccharides, insoluble non-starch polysaccharides, and non-cellulosic non-starch polysaccharides in the wheat-SBM diets compared with the wheat-SBM-wheat middlings diet indicates that the fermentability of dietary fiber in wheat may be reduced with increased fiber concentration in the diet. The digestibility of these dietary fiber fractions is likely influenced by their structural arrangement in the cell wall, which makes them less susceptible to digestive enzymes ([Bach Knudsen, 1993](#page-13-24); [Jørgensen,](#page-13-22) [1996](#page-13-22); [Le Gall, 2009;](#page-13-23) [de Vries, 2014\)](#page-14-14).

The observation that the DE and ME in corn diets are greater than in wheat diets was expected because corn contains more starch and less NDF compared with wheat [\(NRC, 2012\)](#page-14-1). The reduction in DE and ME with increased fiber level in the diets, as observed in the wheat-SBM-wheat middlings diets, is in agreement with previous data ([Stewart](#page-14-15) [et al., 2013](#page-14-15); [Jaworski and Stein, 2017\)](#page-13-2).

The lack of a response to xylanase in the cornbased diets indicates that the microbial xylanases used in this experiment are not effective in hydrolyzing the glycosidic and ester bonds in the arabinoxylans in corn and DDGS even when included at very high concentrations. A positive response to xylanase to both corn- and wheat-based diets was reported (Kiarie et al., 2016), but it is possible that the lack of a response in this experiment is because different xylanases were used. The reason the response to xylanase addition to cornbased diets was less than to wheat-based diets may be that the arabinoxylans in corn and DDGS are lignified, highly branched, and linked to structural proteins, which make it difficult for microbial and exogenous enzymes to ferment arabinoxylans and other dietary fiber components in corn or DDGS compared with wheat or wheat middlings [\(Saulnier](#page-14-16) [et al., 1995](#page-14-16); [Saha and Bothast, 1999\)](#page-14-17). It is also possible that the three-dimensional structure of the arabinoxylans in corn fiber is different from that in wheat fiber and that this hinders xylanase activity in corn fiber. However, because we did not measure the three-dimensional structure of arabinoxylans in this experiment we cannot confirm this hypothesis. The improvement in the ADD and ATTD of some nutrients in the wheat-SBM-wheat middlings diet in response to both xylanases is likely a result of increased fermentability of arabinoxylans because the calculated ATTD of hemicellulose increased. The observed responses to xylanase B in both wheat-SBM and the wheat-SBM-wheat middlings diets indicate that xylanase B may be included in wheat-based diets to improve digestibility of nutrients and fermentability of dietary fiber fractions. The positive effects of xylanase on DE and ME in the wheat-based diets are in agreement with previous data ([Nortey et al., 2007;](#page-14-11) [Olukosi et al., 2007](#page-14-18)), although a lack of a positive response to xylanase has also been reported [\(Yáñez et al., 2011](#page-14-10)). It is possible that this effect is not only a result of increased utilization of carbohydrates, but may also be related to increased digestibility of fat [\(Adeola](#page-13-25) [and Cowieson, 2011\)](#page-13-25). The observed improvement in both DE and ME in wheat-SBM diets and wheat-SBM-wheat middlings diets with the inclusion of xylanase B and xylanase A, respectively, indicates that xylanase B may be included in wheat-based diets with less concentration of dietary fiber and xylanase A may be more effective in wheat-based diets with greater concentration of dietary fiber.

### **CONCLUSION**

Digestion of energy and fermentation of dietary fiber occur mainly in the small intestine and hindgut of the pigs, respectively. The ATTD of dietary fiber is greater in corn-SBM and wheat-SBM diets compared with diets containing DDGS or wheat middlings, which indicates that the concentration of dietary fiber may influence the degree of fermentation of dietary fiber. Microbial xylanase improved the ATTD of energy and dietary fiber and the concentration of DE and ME in wheat-based diets. The microbial xylanases used in this experiment improved the dietary fiber digestibility in the stomach and hindgut of the pigs and improved energy status of pigs fed wheat-based diets, but not corn-based diets.

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