



## Effects of Inclusion Levels of Wheat Bran and Body Weight on Ileal and Fecal Digestibility in Growing Pigs

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**ABSTRACT:** The objective of this study was to determine the effects of graded inclusions of wheat bran (0%, 9.65%, 48.25% wheat bran) and two growth stages (from 32.5 to 47.2 kg and 59.4 to 78.7 kg, respectively) on the apparent ileal digestibility (AID), apparent total tract digestibility (ATTD) and hindgut fermentation of nutrients and energy in growing pigs. Six light pigs (initial body weight [BW] 32.5±2.1 kg) and six heavy pigs (initial BW 59.4±3.2 kg) were surgically prepared with a T-cannula in the distal ileum. A difference method was used to calculate the nutrient and energy digestibility of wheat bran by means of comparison with a basal diet consisting of corn-soybean meal (0% wheat bran). Two additional diets were formulated by replacing 9.65% and 48.25% wheat bran by the basal diet, respectively. Each group of pigs was allotted to a 6×3 Youden square design, and pigs were fed to three experimental diets during three 11-d periods. Hindgut fermentation values were calculated as the differences between ATTD and AID values. For the wheat bran diets, the AID and ATTD of dry matter (DM), ash, organic matter (OM), carbohydrates (CHO), gross energy (GE), and digestible energy (DE) decreased with increasing inclusion levels of wheat bran ( $p<0.05$ ). While only AID of CHO and ATTD of DM, ash, OM, CHO, GE, and DE content differed ( $p<0.05$ ) when considering the BW effect. For the wheat bran ingredient, there was a wider variation effect ( $p<0.01$ ) on the nutrient and energy digestibility of wheat bran in 9.65% inclusion level due to the coefficient of variation (CV) of the nutrient and energy digestibility being higher at 9.65% compared to 48.25% inclusion level of wheat bran. Digestible energy content of wheat bran at 48.25% inclusion level (4.8 and 6.7 MJ/kg of DM, respectively) fermented by hindgut was significantly higher ( $p<0.05$ ) than that in 9.65% wheat bran inclusion level (2.56 and 2.12 MJ/kg of DM, respectively), which was also affected ( $p<0.05$ ) by two growth stages. This increase in hindgut fermentation caused the difference in ileal DE ( $p<0.05$ ) to disappear at total tract level. All in all, increasing wheat bran levels in diets negatively influences the digestibility of some nutrients in pigs, while it positively affects the DE fermentation in the hindgut. (**Key Words:** Digestive Sites, Hindgut Fermentation, Dietary Fiber, Digestibility, Wheat Bran, Growth Stage)

### INTRODUCTION

It is known that the efficiency of nutrient and energy digestibility of feed ingredients in pig diets is usually affected by physicochemical characteristics (Le Goff and Noblet, 2001), dietary supplements, processing methods (De Vries et al., 2012), animal factors, feeding levels (Noblet and Shi, 1994) and other factors. In addition, different sites of digestion in the small intestine and hindgut

fermentation (Varel, 1987; Bastianelli et al., 1996) will also affect the digestibility. A few studies comparing ileal and total tract digestion in diets have been developed for pigs (Usry et al., 1991; Shi and Noblet, 1993; Wilfart et al., 2007; Chen et al., 2013). It is usually believed that the nutritional significance of different digestive sites of digestion is quite different because the efficiency of energy utilization of hindgut utilization is lower. However, most studies did not aim at the digestibility of feed ingredients at different digestive sites.

Wheat bran, a common type of fibrous feed ingredient, is widely used in swine diets. Previous data (Huang et al., 2013) in our lab indicated that 10% wheat middlings in the

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corn soybean meal diets provided significant lower digestible energy (DE) content against the higher inclusion levels of wheat middlings. In addition, no differences were observed for the DE content provided by wheat bran (Zhang, 2012; Huang et al., 2014) fed to pigs in two different growing stages (body weight [BW] around 31 to 50 kg and 61 to 75 kg, respectively). It can be also assumed that ileal digestibility remains relatively constant over a BW range. In addition, higher DE content was supposed to be supplied by hindgut fermentation when heavier pigs were fed to increasing inclusion level of fiber in pig diets (Shi and Noblet, 1994). However, there are no studies conducted to compare nutrient and energy digestibility of wheat bran at different digestive sites, considering graded inclusion levels of wheat bran fed to growing pigs at two growth stages. This will improve understanding the fibrous feed ingredients use in growing pigs.

Therefore, the objectives were to quantify the contributions of the small intestine and large intestine to apparent total tract digestibility (ATTD) of nutrients and energy of diets formulated by graded inclusions of wheat bran, and the effects of graded inclusions of wheat bran on the nutrient digestibility and DE content at two growth stages, calculated by the difference method.

## MATERIALS AND METHODS

The Institutional Animal Care and Use Committee at China Agricultural University (Beijing, China) reviewed and approved the protocols used in the study.

### Diets and feeding

All the analyzed chemical composition of feed ingredients was presented in Table 1. A corn-soybean meal

**Table 1.** Analyzed composition of feed ingredients (% , as fed basis)

Items	Corn	Soybean meal	Wheat bran
Dry matter	86.31	88.05	88.29
Crude protein	7.62	43.62	17.50
Ether extract	3.42	1.10	2.83
NDF	10.93	13.29	37.88
ADF	2.63	7.88	11.13
Hemi-cellulose <sup>1</sup>	8.30	5.40	26.75
Ash	0.92	5.28	5.12
Organic matter	85.38	82.76	83.17
Carbohydrates	88.03	50.00	74.55
Total dietary fiber	12.92	19.25	42.44
Soluble dietary fiber	1.72	2.31	4.11
Insoluble dietary fiber	11.2	16.94	38.33
Gross energy (MJ/kg)	16.12	17.15	16.89

NDF, neutral detergent fiber; ADF, Acid detergent fiber.

<sup>1</sup> Hemi-cellulose = NDF-ADF; Organic matter = dry matter-ash; Carbohydrates = 100-(ash+crude protein+ether extract).

diet with 0% wheat bran was formulated as the basal diet. Two other experimental diets contained 9.65% and 48.25% wheat bran, which was added at the expense of corn and soybean meal to provide the graded levels of dietary fiber (Table 2). Vitamins and minerals were supplemented in all diets to meet or exceed the estimated nutrient requirements for growing pigs recommended by NRC (1998). Chromic oxide (0.3%) was included in all diets as an indigestible maker. The daily allowance was adjusted to 2.6 times the maintenance requirement for energy (i.e., 106 kcal of ME/kg of BW<sup>0.75</sup>; NRC, 1998). Diets were provided to pigs in mash form.

### Animals, experimental design and samples collection

Six light crossbred (Duroc×Landrace×Yorkshire) growing pigs (initial BW: 32.5±2.1 kg) and six heavy crossbred (Duroc×Landrace×Yorkshire) growing pigs (initial BW: 59.4±3.2 kg) were surgically prepared with a T-cannula in the distal ileum according to the producer of Ren et al. (2011). All pigs were housed in stainless steel metabolism cages (1.4×0.45×0.6 m<sup>3</sup>). Water was freely

**Table 2.** Ingredients and composition of the experimental diets

Items (%)	Level of wheat bran (%)		
	0	9.65	48.25
Ingredients			
Corn	73.88	66.49	36.94
Soybean meal	22.34	20.11	11.17
Wheat bran	0	9.65	48.25
L-Lysine·HCl <sup>1</sup>	0.28	0.25	0.14
Dicalcium phosphate	1.4	1.4	1.4
Calcium carbonate	0.9	0.9	0.9
Chromic oxide	0.3	0.3	0.3
Sodium chloride	0.4	0.4	0.4
Vitamin-mineral premix <sup>2</sup>	0.5	0.5	0.5
Analyzed chemical composition (% , as fed basis)			
Dry matter	87.50	88.29	88.08
Crude protein	15.82	16.47	16.84
Neutral detergent fiber	12.16	14.12	24.17
Acid detergent fiber	3.98	4.51	7.23
Ether extract	2.57	2.68	2.63
Ash	5.14	5.02	6.35
Total dietary fiber	14.82	18.4	31.86
Insoluble dietary fiber	9.66	11.83	20.93
Soluble dietary fiber	5.16	6.57	10.93
Gross energy, MJ/kg	15.89	16.07	16.12

<sup>1</sup> L-lysine·HCl was provided by DaCheng Group, ChangChun, China.

<sup>2</sup> Provided the following quantities of vitamins and minerals per kg of complete diet: Mn, 50 mg (MnO); Fe, 125 mg (FeSO<sub>4</sub>·H<sub>2</sub>O); Zn, 125 mg (ZnO); Cu, 150 mg (CuSO<sub>4</sub>·5H<sub>2</sub>O); I, 50 mg (CaI<sub>2</sub>); Se, 0.48 mg (Na<sub>2</sub>SeO<sub>3</sub>), retinyl acetate, 4,500 IU; cholecalciferol, 1,350 IU; DL- $\alpha$ -tocopheryl acetate, 13.5 mg; menadione sodium bisulfite complex, 2.7 mg; niacin, 18 mg; Vitamin B<sub>12</sub>, 27.6  $\mu$ g; thiamine, 0.6 mg; pyridoxine, 0.9 mg; riboflavin, 1.8 mg; D-Ca-pantothenate, 10.8 mg; nicotinic acid, 30.3 mg; choline chloride, 210 mg.

available from a low-pressure drinking nipple. The room temperature was kept at  $22\pm 2^{\circ}\text{C}$  for the duration of the experiment.

Each group of six pigs was used in a  $6\times 3$  Youden square design, and pigs were fed to three experimental diets during three 11-d period. Fecal samples were collected via grab sampling from d 8 to 9 of each period after a 7-d adaptation period, and ileal digesta samples were collected in each period from 08:00 to 18:00 h on d 10 and 11. The digesta collection method was similar to the procedure provided by Huang et al. (2012).

Digesta and feces samples were collected in plastic bags and stored at a  $-20^{\circ}\text{C}$  immediately after each collection. All samples were thawed, mixed within animal and diet, and the sub-sample then after were lyophilized in a vacuum-freeze dryer (Tofflon Freezing Drying Systems, Minhang District, Shanghai, China) and ground through a 1-mm screen for further chemical analysis.

### Chemical analyses

The methods used to analyze the digesta, feces, ingredient and diets were similar with the descriptions provided by Huang et al. (2014). All samples were analyzed for dry matter (DM, procedure 4.1.06; AOAC 2000), ether extract (EE) (Thiex et al., 2003), crude protein (CP) (Thiex et al., 2002), neutral detergent fiber (NDF), acid detergent fiber (ADF) (procedure 4.6.03; AOAC 2000), and Ash (procedure 4.1.10; AOAC, 2000). Neutral detergent fiber and ADF were determined using fiber bags and fiber analyzer equipment (Fiber Analyzer, Ankom Technology, Macedon, NY, USA). The concentration of NDF was analyzed using heat stable  $\alpha$ -amylase and sodium sulfite without correction for insoluble ash. The gross energy (GE) of all samples was measured using an Automatic Adiabatic Oxygen Bomb Calorimeter (Parr 6300 Calorimeter, Moline, IL, USA). Total dietary fiber (TDF) and insoluble dietary fiber (IDF) of ingredients and diets were determined according to the method provided by Prosky et al. (1992). The concentration of soluble dietary fiber (SDF) was calculated as the difference between TDF and IDF values.

### Calculations

Apparent ileal digestibility (AID) and ATTD of DM, CP, EE, NDF, ADF, ash, organic matter (OM), carbohydrates (CHO), and DE content of experimental diets were calculated in all diets according to the equation provided by Chen et al. (2013).

$$\text{AD}_{\text{nutrient}} = [1 - (\text{Nutrient}_{(\text{ileum/feces})} / \text{Nutrient}_{(\text{diet})}) \times (\text{Cr}_2\text{O}_3_{(\text{diet})} / \text{Cr}_2\text{O}_3_{(\text{ileum/feces})})] \times 100 \quad (1)$$

Where  $\text{AD}_{\text{nutrient}}$  is the AID or ATTD of a nutrient or energy in the diet (%),  $\text{nutrient}_{(\text{diet})}$  and  $\text{nutrient}_{(\text{ileum/feces})}$  is a

nutrient (g) or energy (MJ/kg DM) content in the diet and the ileal or fecal samples, respectively, and  $\text{Cr}_2\text{O}_3_{(\text{diet})}$  and  $\text{Cr}_2\text{O}_3_{(\text{ileum/feces})}$  are the  $\text{Cr}_2\text{O}_3$  concentrations (g/kg) in the diet and the ileal or fecal samples, respectively.

The AID and ATTD of DM, CP, EE, NDF, ADF, ash, OM, CHO, and DE content in wheat bran were calculated by the difference method (Fan and Sauer, 1995) using the equation below:

$$\text{AD}_{\text{nutrient}} = [(\text{AD}_{\text{assay}} - \text{AD}_{\text{control}}) \times \text{Nutrient}_{\text{control}}] / (1 - \text{Nutrient}_{\text{control}}) \quad (2)$$

Where  $\text{AD}_{\text{nutrient}}$  is the AID or ATTD of a nutrient in the ingredient and diets (%),  $\text{AD}_{\text{assay}}$  is the AID or ATTD of nutrient in the assay diet (%),  $\text{AD}_{\text{control}}$  is the AID and ATTD of nutrient in the control diet, and  $\text{Nutrient}_{\text{control}}$  is the contribution of the nutrient from the control diet to the assay diet. The hindgut fermentation of nutrients and energy was calculated according to the differences between ATTD and AID values (Urriola and Stein, 2012).

### Statistical analysis

Normal distribution and equal variances of the data were determined using the UNIVARIATE procedure of SAS (SAS Inst. Inc., Cary, NC, USA). Data were analyzed by general linear model using the MIXED procedure of SAS (Littell et al., 1998). Pig was considered the experimental unit. The diets, inclusion level, growth stages, and inclusion level  $\times$  growth stage were considered as fixed effects, where pigs and periods were treated as random effects. No interactions ( $p > 0.05$ ) were observed for level  $\times$  growth stage effect in the study. If significant differences were found, the Student Newman Keul's test was used to test the significance of differences between means. The contrast option was also used to compare the effects of growth stages on the digestibility data. In all analyses, the differences were considered significant if  $p < 0.05$ .

## RESULTS

### The apparent ileal digestibility, apparent total tract digestibility and hindgut fermentation of diets

The effects of three inclusion levels of wheat bran, two growth stages of growing pigs and their interactions on the AID, ATTD, and hindgut fermentation of nutrients and energy content in diets are presented in Table 3. For all digestibility values, there were no interactions between the growth stages and inclusion levels ( $p > 0.05$ ). As for the influence of inclusions of wheat bran on dietary digestibility, the AID and ATTD values of DM, ADF, ash, OM, CHO, GE, and DE content in 48.25% wheat bran diets were lower ( $p < 0.05$ ) than other diets. In contrast, the AID and ATTD of

**Table 3.** Effects of dietary wheat bran levels and growth stages on apparent ileal digestibility, apparent total tract of digestibility and hindgut fermentation of nutrients and energy in growing pigs<sup>1</sup> (% , unless otherwise specified )

Items	Growth stage 1			Growth stage 2			SEM	p-value		
	Basal diet	9.65% wheat bran diet	48.25% wheat bran diet	Basal diet	9.65% wheat bran diet	48.25% wheat bran diet		Levels	Levels× BW	BW
Apparent ileal digestibility (%)										
Dry matter	64.52 <sup>ab</sup>	62.49 <sup>ab</sup>	50.86 <sup>c</sup>	67.94 <sup>a</sup>	64.33 <sup>ab</sup>	50.31 <sup>c</sup>	0.48	<0.01	0.28	0.11
Crude protein	71.73	68.24	62.44	66.43	66.09	64.29	1.12	0.13	0.46	0.39
Ether extract	44.58	46.92	42.49	53.22	48.13	43.74	2.08	0.56	0.71	0.38
NDF	36.05	29.63	39.77	41.26	37.96	33.90	2.17	0.72	0.41	0.53
ADF	7.99	8.94	21.99	10.04	13.11	23.11	2.52	0.33	0.21	0.59
Ash	24.07 <sup>ab</sup>	17.67 <sup>ab</sup>	3.86 <sup>ab</sup>	31.36 <sup>a</sup>	14.27 <sup>ab</sup>	-0.42 <sup>b</sup>	2.70	<0.01	0.63	0.99
Organic matter	68.74 <sup>ab</sup>	66.77 <sup>b</sup>	56.56 <sup>c</sup>	71.71 <sup>a</sup>	68.72 <sup>ab</sup>	56.13 <sup>c</sup>	0.43	<0.01	0.29	0.09
Carbohydrates	70.50 <sup>b</sup>	68.60 <sup>b</sup>	57.57 <sup>c</sup>	75.03 <sup>a</sup>	71.16 <sup>b</sup>	56.32 <sup>c</sup>	0.45	<0.01	0.05	<0.05
Gross energy	66.63 <sup>a</sup>	64.76 <sup>a</sup>	54.72 <sup>b</sup>	69.51 <sup>a</sup>	66.53 <sup>a</sup>	55.39 <sup>b</sup>	0.45	<0.01	0.63	0.06
DE, MJ/kg of DM	11.91 <sup>ab</sup>	11.53 <sup>b</sup>	9.61 <sup>c</sup>	12.54 <sup>a</sup>	11.79 <sup>ab</sup>	9.43 <sup>c</sup>	0.08	<0.01	0.19	0.15
Apparent total tract digestibility (%)										
Dry matter	83.35 <sup>a</sup>	80.96 <sup>a</sup>	73.49 <sup>c</sup>	84.86 <sup>a</sup>	83.82 <sup>a</sup>	77.65 <sup>b</sup>	0.50	<0.01	0.56	<0.05
Crude protein	79.61	78.17	78.46	82.26	78.20	79.65	0.77	0.37	0.79	0.38
Ether extract	37.41	31.64	19.86	26.69	30.22	31.72	2.51	0.56	0.23	0.92
NDF	66.71	53.57	61.45	68.62	58.07	60.68	1.81	<0.05	0.94	0.48
ADF	60.42	43.97	42.73	59.02	53.92	49.21	2.2	<0.05	0.56	0.26
Ash	49.99 <sup>ab</sup>	44.46 <sup>b</sup>	35.23 <sup>c</sup>	59.68 <sup>a</sup>	53.77 <sup>ab</sup>	49.62 <sup>a</sup>	1.23	<0.01	0.73	<0.01
Organic matter	86.53 <sup>a</sup>	84.06 <sup>a</sup>	77.87 <sup>b</sup>	87.51 <sup>a</sup>	86.37 <sup>a</sup>	80.76 <sup>b</sup>	0.44	<0.01	0.64	<0.05
Carbohydrates	90.52 <sup>a</sup>	87.92 <sup>a</sup>	80.98 <sup>b</sup>	91.61 <sup>a</sup>	90.59 <sup>a</sup>	83.49 <sup>b</sup>	0.39	<0.01	0.65	<0.05
Gross energy	83.57 <sup>a</sup>	80.96 <sup>ab</sup>	74.12 <sup>c</sup>	85.08 <sup>a</sup>	83.72 <sup>a</sup>	78.09 <sup>b</sup>	0.54	<0.01	0.66	<0.05
DE (MJ/kg of DM)	15.87 <sup>a</sup>	15.39 <sup>a</sup>	13.94 <sup>b</sup>	16.28 <sup>a</sup>	16.02 <sup>a</sup>	14.52 <sup>b</sup>	0.12	<0.01	0.92	<0.05
Hindgut fermentation (%)										
Dry matter	18.84	18.46	22.63	16.92	19.48	27.35	0.70	0.35	0.15	0.51
Crude protein	7.88	9.93	16.02	15.86	12.11	15.36	1.74	0.85	0.78	0.48
Ether extract	-7.17	-15.28	-22.63	-12.03	-17.92	-26.52	2.39	0.26	0.09	0.96
NDF	30.65	23.94	21.68	27.36	20.12	26.79	2.40	0.65	0.19	0.89
ADF	52.41	35.03	20.74	48.99	30.81	36.11	2.69	0.09	0.30	0.60
Ash	25.92	26.79	31.37	28.32	39.50	50.03	3.39	0.40	0.40	0.17
Organic matter	17.79	17.29	21.31	15.81	17.65	24.63	0.64	0.48	0.25	0.89
Carbohydrates	20.02	19.31	23.41	16.58	19.43	27.18	0.60	0.20	0.21	0.53
Gross energy	16.94	16.21	19.40	15.57	17.22	22.70	0.78	0.85	0.69	0.76
DE (MJ/kg of DM)	3.95 <sup>c</sup>	3.86 <sup>cd</sup>	4.33 <sup>ab</sup>	3.74 <sup>d</sup>	4.23 <sup>b</sup>	5.09 <sup>a</sup>	0.09	<0.01	0.99	0.99

SEM, standard error of the mean; BW, body weight; NDF, neutral detergent fiber; ADF, acid detergent fiber; DE, digestible energy.

<sup>1</sup> Mean values from six replications.<sup>a-d</sup> Values within a row lacking a common superscript letter are different (p<0.05).

CP and EE showed no differences.

As for the effects of two different growth stages, the AID and hindgut fermentation of most nutrients and energy content were unaffected, with only an observed effect of the AID of CHO (p<0.05). On the other hand, the ATTD values of DM, ash, OM, CHO, GE, and DE content were significantly influenced by growth stages (p<0.05).

As for the hindgut fermentation, DE content of 48.25% wheat bran diets provided higher DE (p<0.01) than other inclusion levels of wheat bran diets, but it was not affected by growth stages. Ether extract digestibility values were all

negative in the hindgut for diets.

### The apparent ileal digestibility, apparent total tract digestibility, and hindgut fermentation of ingredients

The effects of two inclusion levels, two growth stages of growing pigs and their interaction on AID, ATTD and hindgut fermentation of nutrients and energy content in wheat bran calculated by the difference method are presented in Table 4. As for the influence of inclusion levels on digestibility of wheat bran ingredient, it was indicated that there was more variation in AID, ATTD and hindgut

**Table 4.** Effects of inclusion levels and growth stages on apparent ileal digestibility, apparent total tract digestibility and hindgut fermentation of nutrients and energy of wheat bran in growing pigs calculated by difference method<sup>1</sup> (% , unless otherwise specified )

Items	Growth stage 1		Growth stage 2		SEM	p-value		
	9.65% wheat bran	48.25% wheat bran	9.65% wheat bran	48.25% wheat bran		Levels	Levels ×BW	BW
Apparent ileal digestibility (%)								
Dry matter	63.84 <sup>a</sup>	39.11 <sup>b</sup>	67.51 <sup>a</sup>	37.90 <sup>b</sup>	1.82	<0.01	0.54	0.83
Crude protein	61.36	60.41	65.73	66.38	5.88	0.08	0.06	<0.05
Ether extract	41.48	60.46	21.76	52.59	18.02	0.12	0.20	0.21
NDF	-17.81 <sup>a</sup>	44.43 <sup>b</sup>	-19.29 <sup>a</sup>	43.51 <sup>b</sup>	2.35	<0.01	0.41	0.45
ADF	-18.67	30.38	-2.30	32.06	6.78	0.76	0.84	0.66
Ash	-14.37	-17.41	-37.79	-38.66	11.92	0.48	0.10	0.18
Organic matter	52.38 <sup>b</sup>	44.52 <sup>a</sup>	59.12 <sup>b</sup>	40.55 <sup>a</sup>	1.60	<0.05	0.70	0.96
Carbohydrates	54.79 <sup>b</sup>	43.61 <sup>ab</sup>	38.02 <sup>b</sup>	36.12 <sup>b</sup>	2.65	<0.05	0.52	0.19
DE (MJ/kg of DM)	9.01 <sup>b</sup>	7.60 <sup>ab</sup>	8.07 <sup>ab</sup>	6.59 <sup>a</sup>	0.24	<0.05	0.40	0.76
Apparent total tract digestibility (%)								
Dry matter	82.98 <sup>a</sup>	66.51 <sup>b</sup>	89.26 <sup>a</sup>	77.30 <sup>a</sup>	3.04	<0.01	0.10	<0.05
Crude protein	86.72	85.77	85.27	86.69	6.54	0.81	0.95	0.73
Ether extract	-9.26	16.71	-3.27	34.17	26.56	0.56	0.67	0.41
NDF	-9.51 <sup>a</sup>	51.42 <sup>b</sup>	-6.24 <sup>a</sup>	63.10 <sup>b</sup>	8.33	<0.01	0.62	0.86
ADF	21.32	39.24	40.55	51.49	9.95	0.69	0.97	0.60
Ash	23.29 <sup>a</sup>	59.13 <sup>b</sup>	5.80 <sup>a</sup>	77.19 <sup>b</sup>	13.11	<0.05	0.85	0.43
Organic matter	66.15 <sup>a</sup>	69.38 <sup>a</sup>	76.49 <sup>b</sup>	78.28 <sup>b</sup>	3.13	<0.05	0.32	<0.05
Carbohydrates	65.88 <sup>a</sup>	70.28 <sup>ab</sup>	66.27 <sup>a</sup>	78.66 <sup>b</sup>	3.56	<0.05	0.05	<0.05
DE (MJ/kg of DM)	11.57	12.40	10.19	13.29	0.92	0.45	0.40	0.80
Hindgut fermentation (%)								
Dry matter	19.15	27.41	21.75	39.39	5.87	0.45	0.37	0.20
Crude protein	25.36	25.36	19.54	20.31	6.55	0.51	0.52	0.34
Ether extract	-50.74	-43.75	-25.03	-18.43	23.58	0.65	0.90	0.96
NDF	8.30 <sup>a</sup>	6.96 <sup>a</sup>	18.35 <sup>b</sup>	19.60 <sup>b</sup>	1.80	<0.05	0.18	0.46
ADF	-2.65	8.86	42.85	49.42	15.71	0.25	0.47	0.77
Ash	31.08 <sup>a</sup>	76.54 <sup>ab</sup>	43.59 <sup>a</sup>	85.85 <sup>b</sup>	9.82	<0.05	0.85	0.89
Organic matter	13.77 <sup>a</sup>	24.87 <sup>b</sup>	17.36 <sup>a</sup>	37.73 <sup>c</sup>	3.14	<0.05	0.30	<0.05
Carbohydrates	11.09 <sup>a</sup>	26.67 <sup>ab</sup>	28.25 <sup>ab</sup>	42.54 <sup>c</sup>	4.31	<0.05	0.33	<0.05
DE (MJ/kg of DM)	2.56 <sup>a</sup>	4.80 <sup>b</sup>	2.12 <sup>a</sup>	6.70 <sup>c</sup>	0.45	<0.05	0.46	<0.05

SEM, standard error of the mean; BW, body weight; NDF, neutral detergent fiber; ADF, acid detergent fiber; DE, digestible energy.

<sup>1</sup> Mean values from six replications.<sup>a-d</sup> Values within a row lacking a common superscript letter are different (p<0.05).

fermentation values in inclusion level of 9.65% than inclusion level of 48.25% wheat bran calculated by difference method. The AID of DM, OM, and CHO, and ATTD of DM at inclusion level of 9.65% wheat bran were higher (p<0.05) than at inclusion level of 48.25% wheat bran. And the other AID and ATTD of nutrients and energy were higher at higher inclusion level. The hindgut fermentation of NDF, ash, OM, CHO, and DE content were affected by the effect of inclusion level of wheat bran (p<0.05). Inclusion level of 48.25% wheat bran showed higher digestibility values for these nutrients and energy contents.

As for the BW effects, differences (p<0.05) were observed in AID of CP, ATTD of DM, OM, CHO and hindgut fermentation of OM, CHO, and DE content. In

addition, the hindgut fermentation of OM, CHO, and DE content were significantly higher in heavier pigs (p<0.05). For example, for the inclusion level 48.25% wheat bran, heavier pigs provided 6.7 MJ/kg of DM of DE content, which was significantly higher than DE content fermented by light pigs (2.12 MJ/kg of DM).

## DISCUSSION

### Digestive utilization of diets

In the present study, most AID of nutrients and energy were not affected by BW except for CHO digestibility. It may be that the small intestine is relatively fully developed at 20 kg, while the large intestine keeps developing until 150 kg in pigs (Fernandez, 1986). Therefore, Noblet and

Shi (1994) recommended that 60 kg pigs used in digestibility trials will be more representative for the whole growing-finishing period, especially for higher dietary fiber diets. Wheat bran is one of the most effective fiber sources for increasing the rate of passage in the digestive tract (Jørgensen et al., 1996). The concentration of fiber is by far the most important factor for accelerating the amount of CHO that passes from the small to the large intestine (Erik et al., 2013). More than 30% of the CHO in the current study that arrived into the large intestine were fermented as they passed along the large intestine with substantial differences among nutrients. In contrast, the ATTD of most nutrients (DM, ash, OM, CHO, and GE) and DE content showed significant differences between two growth stages, which was mainly caused by the effects of hindgut fermentation. The amount of fiber digestion in 48.25% wheat bran diets had significantly higher NDF or ADF utilizations because of higher dietary fiber content in the diets, although no differences were observed for hindgut fermentation. However, the ATTD of ash was lower for diets with high wheat bran inclusion levels compared to the low ones. This could be explained by increased endogenous secretions in high fiber diets, or decreased hydrolysis and absorption of nutrients with these diets (Wilfart et al., 2007).

Digestion in the hindgut is affected by the time that digesta is subjected to fermentation, and a rapid passage of digesta may diminish the effectiveness of this process (Morel et al., 2006). Although this research showed that there were no differences between the diets for hindgut fermentation of all the nutrients, except for the DE content. This might be due to the higher digestion amount of fiber in higher level of wheat bran supplemented diet in comparison to its counterparts. The hindgut fermentation supplied DE content in the current study, which is in agreement with the results of Shi and Noblet (1993), who found a value of 15% to 20% for growing pigs. However, the variation in this contribution to DE is high and it can be easily be influenced by the high fiber inclusion levels of fiber diets and growth stages from the present study. The negative values of hindgut fermentation of EE with for all diets could be due to the synthesis of VFA's by microbial bacteria. But it was not affected by fiber levels in the current study, which is not consistent with the result by Kil et al. (2010), reporting that fiber can increase the fat synthesis. All in all, the small intestine and large intestine do not have the same implications in the digestive process, and their contributions differed depending on the different chemical composition of diets and pigs used in the study.

#### **Digestive utilization of ingredients**

In the current study, it was indicated that there was a wider variation in AID and ATTD of most nutrients and DE content of wheat bran at 9.65% inclusion level (data not

shown). These results are similar to those of Huang et al. (2013). The reason was that lower inclusion level of wheat bran would cause the higher calculation error (Adeola, 2001) by difference method because of their lower nutrient contributions. In addition, nutrient interaction is an important factor causing variation in digestibility values. Further, some chemical components are not easily digestible and results can also be influenced by endogenous synthesis, contamination in the digestive sites and analysis methods (Wilfart et al., 2007). Therefore, when the inclusion level of wheat bran was 48.25%, values showed that ATTD of nutrients and energy were more accurate and comparable with the previous researches (Graham et al., 1986; Oeckel et al., 2005; Huang et al., 2013). Semi-purified diets were also used to formulate and determine the nutrient digestibility of ingredients (Urriola et al., 2010, 2012). However, a comparison between the two digestibility determination methods had not been done yet, except for amino acid digestibility (Fan and Sauer, 1995). Therefore, it is recommended to use increasing inclusion levels of ingredients to determine the digestibility of feed ingredients, for this will reduce the risk of variations and interactions.

Unexpectedly, ileal DE content of wheat bran at low inclusion level was higher than at high inclusion level of wheat bran. This might be explained by higher inclusion levels of wheat bran accelerating the passage rate of digesta and reducing the retention time to incompletely digest the nutrients in the small intestine. However, because of the effects of hindgut fermentation of fiber and utilization of other by-pass nutrients, this study showed that higher inclusion levels of wheat bran had higher DE contents, especially for heavier pigs. This increase in DE content is also shown in the fermentation coefficient of wheat bran, which were up to 38.7% and 50.4% in the two growth stages, respectively, at high wheat bran inclusion levels, while at low inclusion levels these values were which were significantly higher than two lower inclusion level of wheat bran (22.1% and 20.8%, respectively). There are potentially two processes involved in increasing hindgut fermentation after an adaptation period to fibrous ingredients: adaptation of the microflora (population, species and enzyme) in the digestive tract to the new substrate (Edwards, 1993) and adaptation of the digestive tract itself (proliferation, digestive juices) to the high fiber by-products (Johnson, 1988). In addition, one of our studies showed that feeding 20% wheat middings in the nursery diets improved the DE content of wheat middlings after 3 wk adaptation (unpublished data). So it might be that a short adaptation period causes to underestimation of the potential DE of fibrous ingredients. In short, the nutritional and biochemical mechanism of fiber digestion in the hindgut are important for understanding the fiber utilization. Fibrous feed ingredients at high inclusion levels especially in high fiber

diets provided much more DE content in the hindgut by fermentation than at lower inclusion levels.

## CONCLUSION

In summary, increasing the inclusion level of ingredients can reduce the risk of calculation errors when determining the nutrient and energy digestibility of fibrous ingredients. This research also showed a higher nutrient digestibility of wheat bran ingredients by pigs when increasing inclusion levels are used. In addition, DE content of wheat bran could be better fermented in the large intestine at higher inclusion due to the increasing ability of hindgut fermentation.

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## CONFLICT OF INTEREST

We certify that there is no conflict of interest with any financial organization regarding the material discussed in the manuscript.

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