NON RUMINANT NUTRITION

Net Energy of high-protein sunflower meal fed to growing pigs and effect of dietary phosphorus on measured values of NE

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Abstract

An experiment was carried out to determine energy values of high-protein sunflower meal (HP-SFM) and to compare the energy values of HP-SFM determined using either a phosphorus (P)-deficient basal diet or a P-adequate basal diet. Twenty-four growing barrows were randomly assigned to 1 of 4 dietary treatments with 6 replicates per treatment. Four experimental diets including 2 basal diets containing 2 levels of standardized total tract digestible P (i.e., P-deficient and P-adequate) and the other 2 diets containing 30% HP-SFM with each basal diet (i.e., HP-SFM 1 diet and HP-SFM 2 diet) were formulated to determine the energy values of HP-SFM and to compare energy values of HP-SFM determined by the difference method using 2 basal diets. Pigs were fed diets for 15 d including 10 d for adaptation and 5 d for total collections. Pigs were then moved to indirect calorimetry chambers to determine total heat production (THP) and fasting heat production (FHP). A reduced ($P < 0.01$) amount of nitrogen was retained in pigs fed the P-deficient basal diet compared with those fed the other diets. The THP of pigs fed the HP-SFM 1 and 2 diets was greater ($P < 0.01$) than those fed the P-deficient basal diet with the intermediate value for pigs fed the P-adequate basal diet. The retained energy (RE) as protein of pigs fed the P-deficient basal diet was less ($P < 0.01$) but RE as lipid was greater ($P < 0.01$) than those fed the P-adequate basal, or HP-SFM 1 and 2 diets. However, there was no difference in FHP of pigs among the dietary treatments. The NE of HP-SFM determined using the P-deficient basal diet was 2,062 kcal/kg, as-fed basis, whereas the value determined using the P-adequate basal diet was 2,151 kcal/kg. Although no differences were observed in energy values, the amount of P in basal diet might affect energy balance by modifying N utilization, thus, a diet containing adequate amount of P is a more suitable basal diet when the difference method is used for calculation of NE in a feed ingredient.

Key words: indirect calorimetry, net energy, phosphorus, pig, sunflower meal

Introduction

Cost-effective coproducts have been widely used in the swine industry to reduce total production cost due to a drastic increase in feed cost over the last decade (Woyengo et al., 2014). Sunflower meal (SFM), which is obtained after oil extraction from sunflower seeds, is sometimes used as a protein source in livestock diets (Waititu et al., 2018a, 2018b). Furthermore, an

advanced dehulling technology has been developed for further processing of the dehulled SFM to produce a high-protein SFM (HP-SFM), now available for use in livestock diets.

Of the available energy systems, the NE system provides a more accurate estimate of the dietary energy available to pigs because DE and ME systems often overestimate the energy contents of diets and ingredients high in fiber or protein

(Noblet et al., 1994). Therefore, the NE value should be used to express the energy content of HP-SFM to accurately estimate available dietary energy in HP-SFM when fed to pigs. However, no information on NE of HP-SFM fed to growing pigs has been reported.

The difference method has often been used to determine energy contents of feed ingredients (Velayudhan et al., 2015; Kim et al., 2017, 2018) because some ingredients cannot be fed alone due to low palatability and anti-nutritional factors (Kong and Adeola, 2014). This method requires a basal diet and a test diet in which a portion of the basal diet is replaced with the test ingredient to calculate energy contents of the test ingredient. Depending on the nutrient composition of the basal diet and the test ingredient, diets can be nutritionally unbalanced or deficient in specific nutrients. This may significantly affect the absolute NE value of the test ingredient because this diet cannot fully support optimal growth and lean tissue gain of pigs (Noblet et al., 1993).

In a previous study, we found that a basal diet that is nutritionally unbalanced with respect to AAs may be inappropriate to determine NE value of feed ingredients (Kim et al., 2018). Thus, it is prudent to suppose that content of other essential nutrients, specifically phosphorus (P), in a basal diet may also affect the absolute NE value of feed ingredients determined by the difference method because P also plays important roles in the body, thereby affecting growth and development of pigs. Indeed, insufficient P supply led to a reduction in feed intake and BW gain (Baker et al., 2013), and an increase in urinary nitrogen (N) excretion due to reduced protein deposition (Varley et al., 2011). However, dietary P contents had no effects on digestibility of DM, GE, AA, and fat (Johnston et al., 2004).

Based on these observations, it can be speculated that DE of the ingredient determined using P-deficient and P-adequate basal diets would not be different because digestibility of energy and nutrients are not affected by dietary P content, but ME would be affected because of increased urinary N excretion, therefore increasing urinary energy loss. The NE of ingredient would also be influenced due to an increase in lipid deposition rather than protein deposition (Noblet and van Milgen, 2013). Therefore, the objectives of the current study were (i) to determine DE, ME, and NE of HP-SFM and (ii) to compare the energy values of HP-SFM determined using either the P-deficient basal diet or the P-adequate basal diet.

Materials and Methods

The experimental protocol used in this experiment was reviewed and approved by the University of Manitoba Animal Care Committee, and pigs were handled according to the guidelines described by the Canadian Council on Animal Care (2009).

Animals, housing, and experimental diets

Twenty-four growing barrows [initial BW of 19.3 ± 1.8 kg] that were the offspring of Tempo boars mated to TN70 females (Topigs Norsvin, Winnipeg, MB, Canada) were used in the current study. All pigs were individually housed in adjustable metabolism crates (1.8×0.6 m) located in a temperature-controlled room (24 ± 1 °C). The metabolism crates were equipped with smooth transparent plastic sides and plastic-covered expanded metal sheet flooring with a screen underneath for fecal collection and a stainless-steel urine tray underneath the fecal screen, which allowed for the total, but separate, collection of feces and urine from each pig.

Corn and soybean meal (SBM) used in the current study were obtained from the Glenlea Research Station, University of Manitoba, and HP-SFM was acquired from Bunge Global Innovation, Spain (Table 1). The sunflower was grown in Bulgaria. Four experimental diets included two corn-SBM-based basal diets containing 2 levels of standardized total tract digestible (STTD) P [i.e., P-deficient basal diet (formulated without inorganic P inclusion), and P-adequate basal diet (formulated with inorganic P inclusion)]. Two additional diets were formulated by adding 30% HP-SBM to each basal diet (Table 2). To formulate the diets containing 30% of HP-SFM, 30% of each basal diet was replaced with the same amount of HP-SFM. Therefore, the DE, ME, and NE of HP-SFM were determined by the difference method (Adeola, 2001) using the P-deficient or P-adequate diet as a basal diet. Amino acids, vitamins, and minerals except P were included in the experimental diets to meet or exceed the requirements for growing pigs (NRC, 2012).

Experimental design and procedure

The current study was carried out in 2 consecutive periods (12 pigs per period) using similar experimental conditions and procedures and same facility because only 3 indirect calorimetry chambers were available for the measurement of heat production (HP) at the same time. Pigs were randomly allotted to 1 of the 4 dietary treatments with 3 replicates per treatment (per period).

Pigs were fed their experimental diets at 550 kcal ME/kg BW^{0.60} per day on the basis of BW on days 1, 5, and 10 (Noblet et al., 1994). During the study, experimental diets were provided to pigs at 0900 h once daily and pigs were trained to consume their daily feed allowance within 1 h after feeding. Pigs had free access to water via a low-pressure nipple drinker throughout the study. In each experimental period, pigs were fed experimental diets for 15 d, including 10 d of adaptation period to the experimental condition and diet. Total collection of feces and urine were performed separately from days 11 to 15 to determine DE and ME as previously described by Kim and Nyachoti, (2017). Feces were collected once daily in the morning, weighed, and immediately stored at -20 °C. Urine collection was initiated on day 11 at 0900 h and terminated on day 16 at 0900 h. Urine excretion was also collected once daily in a plastic container containing 20 mL of 3 N HCl to minimize N losses. The collected urine was weighed and a 5% aliquot was stored at -20 °C. At the end of each collection period, urine samples were thawed, and thoroughly mixed, and a sub-sample was obtained by filtering with glass wool to remove non-urinary components.

Table 1. Analyzed nutrient composition of corn, soybean meal, and high-protein sunflower meal (HP-SFM), as-fed basis

Item ¹	Ingredient		
	Corn	Soybean meal	HP-SFM
DM, %	89.3	91.0	92.7
GE, kcal/kg	3,908	4,219	4,205
CP, %	8.3	48.0	48.5
Ether extract, %	3.1	1.0	0.9
NDF, %	9.8	6.7	13.1
ADF, %	2.8	5.3	9.0
Ash, %	0.9	6.2	9.4
Calcium, %	0.01	0.43	0.43
Phosphorus, %	0.21	0.72	1.87

¹DM, dry matter; GE, gross energy; CP, crude protein; NDF, neutral detergent fiber; ADF, acid detergent fiber.

Table 2. Ingredient and nutrient composition of experimental diets, as-fed basis

Item, %	Diets ¹			
	P-deficient basal	P-adequate basal	HP-SFM 1	HP-SFM 2
Ingredients				
Corn	68.460	68.460	47.922	47.922
Soybean meal	28.200	28.200	19.740	19.740
HP-SFM ²	—	—	30.000	30.000
Vegetable oil	1.000	1.000	0.700	0.700
Corn starch	0.700	—	0.490	—
Lysine-HCl	0.100	0.100	0.070	0.070
Limestone	1.000	1.000	0.700	0.700
Monocalcium phosphate	—	0.700	—	0.490
Salt	0.400	0.400	0.280	0.280
Vitamin–mineral premix ³	0.140	0.140	0.098	0.098
Calculated composition, %				
ME, kcal/kg	3,372	3,343	3,131	3,111
SID ⁴ lysine	0.98	0.98	1.09	1.09
SID methionine	0.28	0.28	0.40	0.40
STTD ⁵ phosphorus	0.16	0.30	0.22	0.32
Analyzed composition, %				
DM	88.6	88.9	90.1	90.1
GE, kcal/kg	3,949	3,914	4,027	4,004
CP	19.2	19.2	27.4	26.9
Ether extract	2.7	2.8	2.7	2.6
NDF	8.7	8.2	9.6	9.9
ADF	3.3	3.2	4.8	4.9
Ash	3.2	4.0	5.1	5.7
Calcium	0.41	0.67	0.42	0.59
Phosphorus	0.33	0.48	0.81	0.90

¹P-deficient basal, a corn–soybean meal-based basal diet without inorganic phosphorus inclusion; P-adequate basal, a corn–soybean meal-based basal diet with inorganic phosphorus inclusion; HP-SFM 1, a diet containing 70% of P-deficient basal and 30% of high-protein sunflower meal; HP-SFM 2, a diet containing a 70% of P-adequate basal and 30% of high-protein sunflower meal.

²HP-SFM, high-protein sunflower meal.

³Supplied the following per kilogram of finished feed: vitamin A, 2,000 IU; vitamin D, 200 IU; vitamin E, 40 IU; vitamin K, 2 mg; choline, 350 mg; pantothenic acid, 14 mg; riboflavin, 7 mg; folic acid, 1 mg; niacin, 21 mg; thiamin, 1.5 mg; vitamin B₆, 2.5 mg; biotin, 70 µg; vitamin B₁₂, 20 mg; Cu, 10 mg; Zn, 110 mg; Fe, 120 mg; Mn, 10 mg; I, 0.4 mg; Se, 0.3 mg.

⁴SID, standardized ileal digestible.

⁵STTD, standardized total tract digestible.

Before and after each experimental period, the accuracy of indirect calorimetry chambers was tested by burning ethanol in an indirect calorimetry system, based on the stoichiometric equation of ethanol burning. Theoretically, the ratio of CO₂ produced to O₂ consumed as a result of ethanol burning results in a respiratory quotient (RQ) of 0.667 (Benedict and Tompkins, 1916). An RQ value between 0.640 and 0.690 was considered acceptable and no accuracy problem was observed during the entire experiments. On day 16, 3 pigs were transferred to the indirect calorimetry chambers (1.22 × 0.61 × 0.91 m; Columbus Instruments, Columbus, OH) from the metabolism crates within 1 h of consuming their daily feed allowance. Then, the HP by pigs was calculated continuously for 24 h (fed state) followed by

12 h (fasting state) of fasting heat production (FHP) based on O₂ consumption, CO₂ production, and urinary N excretion. The next sets of 3 pigs were moved to the indirect calorimetric chambers every 2 days (i.e., on days 18, 20, and 22) for measuring HP and FHP. Pigs were assigned to 1 of 3 indirect calorimetry chambers in a randomized complete block design using the chamber as a blocking factor to avoid possible confounding effects. Pigs had unlimited access to water via a nipple drinker during the HP and FHP measurement, and urine voided during the measurements of HP (24 h) and FHP (12 h) was collected separately, weighed, sub-sampled, and stored at –20 °C until required for N analysis. The temperature inside the chamber was maintained at 22 °C ± 1 °C using the air conditioner. Personnel movement in the chamber room was limited, except for the regular check and urine collection, to avoid stressing pigs during the measurements of HP and FHP.

Sample preparation and chemical analyses

Within pig, collected fecal samples were pooled and dried in a forced-air drying oven at 60 °C for 5 d and finely ground before chemical analysis. Urine samples from metabolism crates and indirect calorimetry chambers were thawed, filtered through glass wool, and transferred separately into a plastic vial for each pig. Diet and ingredient samples were ground through a 1-mm screen in a Thomas Wiley mill model 4 (Labwrench, Midland, ON, Canada) and thoroughly mixed before chemical analysis. Samples of diets and ingredients were analyzed in duplicate for DM, GE, CP (N × 6.25), ether extract (EE), NDF, ADF, ash, calcium (Ca), and P, whereas fecal and urine samples were analyzed in duplicate for DM, GE, and CP. Dry matter (method 934.01), EE (method 920.39A), and ash (942.05) contents were determined according to the Association of Official Analytical Chemists (AOAC, 2006). GE content was measured using an isoperibol bomb calorimeter (model 6400; Parr Instrument, Moline, IL), which had been calibrated using benzoic acid as a standard. Nitrogen concentration was determined by the combustion method (984.13A-D; AOAC, 2006) using the LECO N analyzer (model CNS-2000; LECO Corp., St. Joseph, MI), and the N content was used to calculate the CP content (N × 6.25). The ADF and NDF contents were determined according to the method of Goering and van Soest (1970) using heat stable α-amylase (Sigma number A3306; Sigma-Aldrich, St. Louis, MO) and sodium sulfite. The Ca (method 968.08) and P (method 946.06) concentrations were determined according to the AOAC (2006) and read on a Varian inductively coupled plasma mass spectrometer (Varian Inc., Palo Alto, CA).

The GE content of urine was determined as previously described by Kim and Nyachoti (2017). Briefly, 0.5 g of cellulose was dried in a drying oven at 103 °C for 24 h, and 2 mL of urine sample were added to the dried cellulose, and the final weight of the resulting mixture was recorded. The urine–cellulose mixture and a sample of pure cellulose were dried at 50 °C for 24 h to determine dried urine content. The GE of the dried urine–cellulose mixture and cellulose itself were determined using an isoperibol bomb calorimeter as described above, from which the GE of urine samples were calculated by the difference method.

Calculations

The apparent total tract digestibility (ATTD) of DM, GE, and CP, and N retention rate of pigs were calculated by the total collection method using the following equations:

$$\text{ATTD (\%)} = 100 \times [(\text{CI} - \text{CO}_{\text{feces}}) / \text{CI}] ,$$

where CI is the component intake (g) and CO_{feces} is the component output in feces (g).

$$\text{Nitrogen retention (\%)} = 100 \times [(NI - NO_{feces} - NO_{urine}) / NI],$$

where NI is the N intake, and NO_{feces} and NO_{urine} are the N output in feces and urine (g), respectively.

The HP and FHP (Brouwer, 1965), retained energy (RE; Noblet et al., 1994), and NE content (Noblet et al., 1994) were calculated using the following equations:

$$HP = 3.866 \times O_2 + 1.200 \times CO_2 - 1.431 \times \text{urinary N excretion},$$

where HP is in kilocalories, O_2 is oxygen consumption in liters, CO_2 is carbon dioxide production in liters, and urinary N excretion is total urinary N excretion in grams. The FHP was also calculated using the same equation for HP.

$$RE = ME - HP,$$

where RE, ME, and HP are in kilocalories per day.

Retained energy as protein (RE_p) was calculated from N retention by pigs ($N \times 6.25 \times 5.68$, kcal/g) according to Ewan (2001). Retained energy as lipid (RE_l) was calculated as the difference between RE and RE_p (Labussière et al., 2009).

$$DE = (GE_i - GE_f) / DMI$$

$$ME = (GE_i - GE_f - GE_u) / DMI,$$

$$NE = (RE + FHP) / DMI,$$

where DE, ME, and NE are the DE, ME, and NE contents of a diet in kcal/kg of DM, respectively. The GE_i , GE_f , and GE_u are the total GE intake, total fecal GE output, and total urinary GE output in kilocalories, respectively. The RE and FHP are in kilocalories per day, and DMI is DM intake in kilograms.

The RQ was calculated with the ratio of CO_2 production to O_2 consumption (Noblet et al., 2001). The DE, ME, and NE of HP-SFM were calculated by subtracting the energy contribution of the

basal diet (i.e., P-deficient basal diet, P-adequate basal diet) from the energy content of the diets containing 30% of HP-SFM.

Statistical analysis

The UNIVARIATE procedure (SAS Inst. Inc., Cary, NC) was used to verify homogeneity of variances, but no outlier was identified for all measurements. All data were analyzed using the MIXED procedure of SAS, and the individual pig served as the experimental unit. The initial model included diet and period as the fixed effect, and pig as the random effect for statistical analysis. However, the effect of period was not significant in this study; therefore, it was excluded in the final model. The LSMEANS procedure was used to calculate mean values and the PDIF option with the Tukey's adjustment was used to separate means. Differences were considered significant at $P < 0.05$.

Results

All pigs adapted well to their experimental diets and conditions, remained healthy and readily consumed their daily feed allowance throughout the experimental period.

ATTD of DM, GE, and CP and nitrogen utilization of diets

The ATTD of DM and GE was greater ($P < 0.01$) in the P-deficient and P-adequate basal diets than in the HP-SFM 1 and 2 diets (Table 3). However, the ATTD of CP was not affected by the dietary treatment.

The N intake (g) and fecal N excretion (g) of pigs fed the P-deficient and P-adequate basal diets was lower ($P < 0.01$) compared with those fed the HP-SFM 1 and 2 diets. The urinary N excretion (g) was lower ($P < 0.01$) in pigs fed the P-adequate basal diet than those fed the P-deficient basal diet and the HP-SFM 1 and 2 diets. However, a lower ($P < 0.01$) amount of N (g) was retained in pigs fed the P-deficient basal diet compared with those fed the other diets. Pigs fed the P-adequate basal diet had higher ($P < 0.01$) N retention (%) than those fed the other diets.

Energy utilization and content of experimental diets

The energy utilization of experimental diets fed to growing pigs and energy values of experimental diets are presented in

Table 3. Apparent total tract digestibility (ATTD) of DM, GE, and CP, and nitrogen (N) balance of experimental diets fed to growing pigs

Item	Dietary treatment ¹				SEM	P-value
	P-deficient basal	P-adequate basal	HP-SFM 1	HP-SFM 2		
ATTD, %						
DM	88.7 ^a	89.1 ^a	85.0 ^b	85.1 ^b	0.45	< 0.001
GE	87.4 ^a	87.8 ^a	85.1 ^b	85.3 ^b	0.54	0.003
CP	86.8	87.9	85.8	85.9	0.90	0.333
N balance, g/d						
N intake	33.0 ^b	32.8 ^b	47.1 ^a	46.9 ^a	0.95	< 0.001
Fecal excretion	4.3 ^b	4.0 ^b	6.7 ^a	6.7 ^a	0.46	< 0.001
Urinary excretion	14.2 ^a	6.9 ^b	17.0 ^a	16.2 ^a	1.11	< 0.001
N retained	14.5 ^b	21.9 ^a	23.4 ^a	24.1 ^a	1.50	0.001
N retained, %	43.9 ^b	66.8 ^a	49.6 ^b	51.2 ^b	2.88	0.005

¹P-deficient basal, a corn-soybean meal-based basal diet without inorganic phosphorus inclusion; P-adequate basal, a corn-soybean meal-based basal diet with inorganic phosphorus inclusion; HP-SFM 1, a diet containing 70% of P-deficient basal and 30% of high-protein sunflower meal; HP-SFM 2, a diet containing a 70% of P-adequate basal and 30% of high-protein sunflower meal.

^{a,b}Within a low, means without a common superscript differ ($P < 0.05$).

Table 4. Energy utilization of experimental diets fed to growing pigs and energy values of experimental diets

Item	Dietary treatment ¹				SEM	P-value
	P-deficient basal	P-adequate basal	HP-SFM 1	HP-SFM 2		
Energy value, kcal/kg DM						
DE	3,897 ^a	3,864 ^{ab}	3,805 ^{ab}	3,794 ^b	24.2	0.021
ME	3,701 ^a	3,707 ^a	3,580 ^b	3,578 ^b	24.0	0.001
THP ²	1,791 ^b	1,937 ^{ab}	2,111 ^a	2,102 ^a	58.3	0.004
Total RE ³	1,900 ^a	1,773 ^a	1,475 ^b	1,473 ^b	65.5	<0.001
As protein	489 ^b	778 ^a	805 ^a	841 ^a	55.0	0.001
As lipid	1,411 ^a	995 ^b	670 ^b	632 ^b	93.6	<0.001
FHP ⁴	1,208	1,269	1,368	1,352	50.7	0.130
NE ⁵	3,108 ^a	3,042 ^a	2,843 ^b	2,825 ^b	46.5	0.001
Respiratory quotient						
Fed state	1.03	1.02	1.06	1.04	0.017	0.274
Fasting state	0.73	0.75	0.72	0.71	0.011	0.070
Energy utilization						
ME:DE	0.950 ^{ab}	0.959 ^a	0.941 ^b	0.943 ^b	0.0026	0.001
NE:DE	0.799 ^a	0.786 ^{ab}	0.746 ^b	0.745 ^b	0.0122	0.014
NE:ME	0.841 ^a	0.820 ^{ab}	0.793 ^{ab}	0.791 ^b	0.0123	0.028

¹P-deficient basal, a corn-soybean meal-based basal diet without inorganic phosphorus inclusion; P-adequate basal, a corn-soybean meal-based basal diet with inorganic phosphorus inclusion; HP-SFM 1, a diet containing 70% of P-deficient basal and 30% of high-protein sunflower meal; HP-SFM 2, a diet containing a 70% of P-adequate basal and 30% of high-protein sunflower meal.

²Total heat production = $(3.87 \times O_2 + 1.20 \times CO_2 - 1.43 \times \text{urinary N})/\text{DMI}$.

³Total retained energy = ME – HP; retained energy as protein was calculated according to Ewan (2001) as N retention (g) $\times 6.25 \times 5.68$ (kcal/g).

Whereas, retained energy as lipid was calculated as the difference between total retained energy and retained energy as protein.

⁴Fasting heat production = $(3.87 \times O_2 + 1.20 \times CO_2 - 1.43 \times \text{urinary N})/\text{DMI}$.

⁵NE = (RE + FHP)/DMI.

^{a,b}Within a low, means without a common superscript differ ($P < 0.05$).

Table 4. The DE of the P-deficient basal diet was higher ($P < 0.05$) than that of the HP-SFM 2 diet, however, that of P-adequate basal and HP-SFM 1 diet were not different. A higher ($P < 0.01$) ME value was observed in the P-deficient and P-adequate basal diets than in the HP-SFM 1 and 2 diets. The total heat production (THP) of pigs fed the HP-SFM 1 and 2 diets was higher ($P < 0.01$) than those fed the P-deficient basal diet with the intermediate value for pigs fed the P-adequate diet. The total RE was higher ($P < 0.01$) in pigs fed the P-deficient and P-adequate basal diets than in those fed the HP-SFM 1 and 2 diets. The RE_p of pigs fed the P-deficient basal diet was lower ($P < 0.01$) but RE_L was higher ($P < 0.01$) than those fed the P-adequate basal, and HP-SFM 1 and 2 diets. However, there was no difference in the FHP of pigs among the dietary treatments. The NE of the P-deficient and P-adequate diets was higher ($P < 0.01$) than that of the HP-SFM 1 and 2 diets.

The P-adequate basal diet had a higher ($P < 0.01$) ME:DE ratio compared with the HP-SFM 1 and 2 diets, with the P-deficient basal diet being intermediate. In addition, the NE:DE ratio of the P-deficient basal diet was higher ($P < 0.01$) compared with that of the HP-SFM 1 and 2 diets. Similarly, a higher ($P < 0.01$) NE:ME ratio was observed in the P-deficient basal diet than in the HP-SFM 2 diet.

Energy values of high-protein sunflower meal

The DE, ME, and NE of HP-SFM determined using the P-deficient basal diet were 3,328, 3,057, and 2,062 kcal/kg, as-fed basis, respectively, whereas these values determined using the P-adequate basal diet were 3,366, 3,037, and 2,151 kcal/kg, respectively (Table 5). However, there was no differences in energy values between values determined using either the P-deficient basal diet or the P-adequate basal diet.

Table 5. DE, ME, and NE of high-protein sunflower meal (HP-SFM) fed to growing pigs determined by the difference method using phosphorus (P)-deficient basal and P-adequate basal diets

	Energy content of HP-SFM			
Item	P-deficient basal	P-adequate basal	SEM	P-value
As-fed basis, kcal/kg				
DE	3,328	3,366	98.8	0.793
ME	3,057	3,037	90.3	0.881
NE	2,062	2,151	82.6	0.469
DM basis, kcal/kg				
DE	3,591	3,631	106.6	0.795
ME	3,298	3,276	97.4	0.879
NE	2,224	2,320	89.2	0.470

Discussion

Chemical composition of ingredients and diets

Sunflower meal, a coproduct of sunflower seed crushing industry, is one of the most widely used protein sources in swine diets (Dadalt et al., 2016). The CP content of HP-SFM (48.5%) used in the current study was greater than that of dehulled SFM reported by NRC (2012; 39.9%). In addition, the NDF and ADF contents were less than the NRC (2012) values, which is mainly due to the differences in the method and degree of oil extraction processing. A new processing technique has been developed to improve the quality of dehulling and to allow further reductions in fiber components of SFM, by which the CP content of HP-SFM can reach approximately 46%, and fiber contents can be reduced

significantly (Waititu et al., 2018a, 2018b). Crude protein was inversely correlated with crude fiber, NDF, ADF, indicating that the greater CP of HP-SFM indicating that the greater CP content of HP-SFM was mainly because of greater extent of dehulling (Villamide and San Juan, 1998). The nutritive values of HP-SFM fed to broiler chickens (Waititu et al., 2018a) and standardized ileal digestibility of AA in pigs (Dadalt et al., 2016) have been reported. However, no information on energy values of HP-SFM fed to pigs has been reported although DE, ME, and NE contents in regular SFM has been reported recently (Li et al., 2018). The DE, ME, and NE values of dehulled SFM were 2,584, 2,307, and 1,550 kcal/kg DM reported by Li et al. (2018) and 2,840, 2,569, and 1,482 kcal/kg according to the NRC (2012), respectively, which were lower than the values in the current study. Energy contents of a diet and an ingredient high in fiber and protein tend to be overestimated when determined with the DE and ME systems (Noblet et al., 1994), which necessitates the determination of the NE value for HP-SFM to facilitate its effective utilization.

Nutrient compositions of the corn and SBM used in the current study are in agreement with reported values (NRC, 2012; Kim et al., 2017, 2018). Also, the analyzed nutrient and energy contents of experimental diets were in agreement with values calculated from the inclusion level and analyzed composition of the ingredients. As expected, GE, CP, NDF, and ADF contents increased as a result of HP-SFM inclusion in each basal diet.

ATTD of DM, GE, and CP and nitrogen utilization of diets

In the current study, a lower ATTD of DM and GE was observed in the diets containing HP-SFM compared with the two basal diets, which is most likely attributable to the addition of HP-SFM resulting in increased dietary fiber as reported in previous studies (Kim and Nyachoti, 2017; Kim et al., 2018; Li et al., 2018). However, the extent of reduction in the ATTD of DM as a result of SFM inclusion to the basal diet (percentage change; 4.3%) was much less than the value (percentage change; 11.6%) reported in the study of Li et al. (2018) although similar level of SFM (29.5%) was included in the corn-SBM basal diet. The fiber contents in SFM likely led to this observation because the SFM used in the current study was extensively dehulled SFM, and therefore contained less NDF and ADF compared with the SFM used in their study (NDF, 13.1% vs. 43.5%; ADF, 9.0% vs. 29.0%).

Phosphorus and protein deposition are closely correlated and it has been estimated that ~35 g of P is required to retain each kilogram of body protein (NRC, 2012). Standardized total tract digestible P in the P-deficient basal diet was 0.16% which was about half of the recommended P requirement for this BW class of pigs (i.e., 0.31%); therefore, an increase in urinary N excretion and decreases in daily N retention (g) and N retention rate (%) by pigs fed the P-deficient diet were expected although other essential nutrients (e.g., AA, Ca) were included to meet or exceed the requirements. Indeed, pigs fed the P-deficient basal diet retained less N compared with those fed the other diets in the current study. This result is in agreement with data by Varley et al. (2011) that indicate that pigs fed a diet containing 0.45% of total P had less daily N retention (g) and N retention rate (%) compared with those fed a diet containing 0.64% total P. The lack of a difference in daily N retention (g) among pigs fed the P-adequate basal diet and HP-SFM 1 and 2 diets was likely due to the high P content in HP-SFM (1.87%). To formulate the SFM 1 and 2 diets, 30% of each basal diet was replaced with the same amount of HP-SFM, which elevated dietary concentration of STTD P. As a consequence, when HP-SFM was added to the

P-deficient basal diet, the dietary concentration of P was at the requirement.

Energy utilization and content of experimental diets

The ME:DE ratio was highest for the P-adequate basal diet and lowest for the HP-SFM 1 and 2 diets with the value for the P-deficient basal diet being intermediate in the current study. The higher CP content in the diets containing HP-SFM (on average 27.1%) compared with the requirement of pigs and greater N excretion by pigs fed the P-deficient basal diet compared with those fed the P-adequate basal diet may explain this observation. It has been well demonstrated that excessive AA intake more than the requirement for protein deposition is catabolized during metabolism and excreted through urine, which results in increased urinary energy loss and therefore decreased ME:DE ratio (Noblet and Perez, 1993; Le Bellego et al., 2001).

In the current study, pigs fed the diets containing HP-SFM had greater THP compared with those fed the P-deficient basal diet, which is likely attributable to an increase in dietary CP content as a consequence of the addition of HP-SFM (Noblet et al., 1994; Le Bellego et al., 2001; Noblet and van Milgen, 2013). Replacing 30% of basal diet with 30% of HP-SFM resulted in a decrease in dietary starch but an increase in dietary CP. Dietary protein has greater heat increment per unit of energy associated with metabolic utilization of energy than starch because dietary protein ingestion increases protein turnover and energy cost related to urea synthesis for urinary excretion, resulting in less energetic efficiency (Noblet et al., 1994, 2001; Le Bellego et al., 2001). The observation that pigs fed the diets containing HP-SFM retained less energy than those fed the two basal diets is also likely due to an increase in THP associated with high dietary protein.

The average FHP estimate of 1,299 kcal/kg DM (154 kcal/BW^{0.6}) observed in the current study was within the range (from 143 to 252 kcal/BW^{0.6}) observed in similar studies conducted in the same facility and with similar experimental procedures (Ayoade et al., 2012; Heo et al., 2014; Velayudhan et al., 2015; Kim and Nyachoti, 2017).

Energy values of high-protein sunflower meal determined using a phosphorus-deficient basal diet and a phosphorus-adequate basal diet

The difference method has been increasingly used to determine NE value of feed ingredients, and several studies have reported no difference between determined NE using indirect calorimetry and predicted NE from prediction equations (Heo et al., 2014; Kim and Nyachoti, 2017; Li et al., 2018). However, calculation of the NE value of a test ingredient determined by the difference method can be biased by nutrient composition of the basal diet if nutrients composition of the basal diet is insufficient to support optimal growth of pigs (Kim et al., 2018). This is because the efficiency of dietary ME utilization depends on final utilization of dietary energy (i.e., deposition as protein vs. deposition as lipid). According to Noblet and van Milgen (2013), the energetic efficiency of dietary ME utilization is greater for lipid deposition than for protein deposition (80% vs. 60%). Therefore, dietary NE for pigs should be measured using balanced diets that meet nutrient requirements to support optimal growth and lean tissue gain (Noblet et al., 1993; Noblet and van Milgen, 2013). Indeed, Li et al. (2018) reported different NE values for corn fed to growing pigs when it was supplemented with dietary crystalline AA. Also, in our previous study (Kim et al., 2018), it was concluded that

the corn-SBM basal diet that meets the nutrient requirement of pigs is a more suitable basal diet than the corn basal diet that contains insufficient AA to support growth and lean tissue gain.

In the current study, effects of dietary P of a basal diet on energy values of HP-SFM determined by the difference method were evaluated. As hypothesized, the DE of HP-SFM determined using the P-deficient and P-adequate basal diets was not different (3,328 vs. 3,366 kcal/kg, as-fed basis), which is supported by no differences in the ATTD of DM, GE, and CP between the two basal diets. Contrary to our hypothesis, however, no differences in the ME (3,057 vs. 3,037 kcal/kg, as-fed basis) and NE (2,062 vs. 2,151 kcal/kg, as-fed basis) of HP-SFM determined using the P-deficient basal diet and the P-adequate basal diet were observed. Theoretically, significantly greater urinary N excretion observed in pigs fed the P-deficient basal diet should be extended to a decrease in ME value of an ingredient because greater urinary N excretion, therefore greater urinary energy loss would lead to underestimation of dietary ME (Noblet and van Milgen, 2013). The lower RE_p and the greater RE_l that were observed in pigs fed the P-deficient basal diet than in pigs fed the P-adequate basal diet did not result in different NE values in HP-SFM despite the fact that the energetic efficiency of dietary ME utilization for protein deposition is less than for lipid deposition (Noblet and van Milgen, 2013). Although there were reasonable supporting data to have different ME and NE values of HP-SFM determined by the P-deficient and P-adequate basal diets in the current study, we failed to observe differences, which may be explained by one of the inherent problems associated with using the difference method (Kil et al., 2011). The energy values calculated with this method are generally associated with large SEM values. In this study, the SEM values for the ME and NE of HP-SFM were much greater than the SEM values for the ME and NE of the diets (ME; 24 vs. 90.3, NE; 46.5 vs. 82.6). Although P content did not affect energy values in HP-SFM in the present study, given N balance seemed more maintainable with P-adequate diet compared with P-deficient diet, thus dietary P content might affect energy balance by modifying N utilization. To generate more reliable values, thus, dietary P should be provided adequate amounts when dietary energy contents are measured.

In conclusion, the DE, ME, and NE of HP-SFM determined in the current study were, on average, 3,347, 3,047, and 2,107 kcal/kg, respectively, as-fed basis. The energy values determined using P-deficient and P-adequate diets were not different. However, because P might affect energy balance by modifying N utilization, a diet containing adequate amount of P is more suitable basal diet when the difference method is used for the calculation of NE value in a feed ingredient.

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Conflict of interest statement

None declared.

Literature Cited

Adeola, O. A. 2001. Digestion and balance techniques in pigs. In: A. J. Lewis and L. L. Southern, editors, *Swine nutrition*. 2nd ed. Boca Raton, FL: CRC Press; p. 903–916.

- AOAC. 2006. *Official methods of analysis*. 18th ed., Washington, DC: Association of Official Analytical Chemists.
- Ayoade, D. I., E. Kiarie, M. A. Trinidad Neto, and C. M. Nyachoti. 2012. Net energy of diets containing wheat-corn distillers dried grains with solubles as determined by indirect calorimetry, comparative slaughter, and chemical composition methods. *J. Anim. Sci.* **90**:4373–4379. doi:10.2527/jas.2011-4858.
- Baker, S. R., B. G. Kim, and H. H. Stein. 2013. Comparison of values for standardized total tract digestibility and relative bioavailability of phosphorus in dicalcium phosphate and distillers dried grains with solubles fed to growing pigs. *J. Anim. Sci.* **91**:203–210. doi:10.2527/jas.2010-3776.
- Benedict, F. G., and E. H. Tompkins. 1916. Respiratory exchange, with a description of a respiration apparatus for clinical use. *N. Engl. J. Med.* **174**:898–909. doi:10.1056/NEJM191606221742502
- Brouwer, E. 1965. Report of subcommittee on constants and factors. In: *Proc. 3rd EAAP Symp. Energy Metab.* Troon Publication No. 11. London: Academic; p. 441–443.
- Canadian Council on Animal Care. 2009. *CCAC guidelines on: the care and use of farm animals in research, teaching and testing*. Ottawa, ON, Canada: Canadian Council on Animal Care.
- Dadalt, J. C., D. E. Velayudhan, M. T. Neto, B. A. Slominski, and C. M. Nyachoti. 2016. Ileal amino acid digestibility in high protein sunflower meal and pea protein isolate fed to growing pigs with or without multi-carbohydrase supplementation. *Anim. Feed Sci. Technol.* **221**:62–69. doi:10.1016/j.anifeedsci.2016.08.015
- Ewan, R. C. 2001. Energy utilization in swine nutrition. In: A. J. Lewis and L. L. Southern, editors, *Swine nutrition*. 2nd ed. Washington, DC: CRC Press; p. 85–94.
- Goering, H. K., and P. J. van Soest. 1970. *Forage fiber analyses (apparatus, reagents, procedures and some applications)*. Agric. Handbook No. 379. Washington, DC: ARS-USDA.
- Heo, J. M., D. Adewole, and M. Nyachoti. 2014. Determination of the net energy content of canola meal from *Brassica napus* yellow and *Brassica juncea* yellow fed to growing pigs using indirect calorimetry. *Anim. Sci. J.* **85**:751–756. doi:10.1111/asj.12196
- Johnston, S. L., S. B. Williams, L. L. Southern, T. D. Bidner, L. D. Bunting, J. O. Matthews, and B. M. Olcott. 2004. Effect of phytase addition and dietary calcium and phosphorus levels on plasma metabolites and ileal and total-tract nutrient digestibility in pigs. *J. Anim. Sci.* **82**:705–714. doi:10.2527/2004.823705x
- Kil, D. Y., F. Ji, L. L. Stewart, R. B. Hinson, A. D. Beaulieu, G. L. Allee, J. F. Patience, J. E. Pettigrew, and H. H. Stein. 2011. Net energy of soybean oil and choice white grease in diets fed to growing and finishing pigs. *J. Anim. Sci.* **89**:448–459. doi:10.2527/jas.2010-3233
- Kim, J. W., B. Koo, and C. M. Nyachoti. 2017. Digestible, metabolizable, and net energy of camelina cake fed to growing pigs and additivity of energy in mixed diets. *J. Anim. Sci.* **95**:4037–4044. doi:10.2527/jas2017.1759
- Kim, J. W., B. Koo, and C. M. Nyachoti. 2018. Net energy content of canola meal fed to growing pigs and effect of experimental methodology on energy values. *J. Anim. Sci.* **96**:1441–1452. doi:10.1093/jas/sky039
- Kim, J. W., and C. M. Nyachoti. 2017. Net energy of hemp hulls and processed hemp hull products fed to growing pigs and the comparison of net energy determined via indirect calorimetry and calculated from prediction equations. *J. Anim. Sci.* **95**:2649–2657. doi:10.2527/jas.2016.1255
- Kong, C., and O. Adeola. 2014. Evaluation of amino Acid and energy utilization in feedstuff for Swine and poultry diets. *Asian-Australas. J. Anim. Sci.* **27**:917–925. doi:10.5713/ajas.2014.r.02
- Labussiere, E., G. Maxin, S. Dubois, J. van Milgen, G. Bertrand, and J. Noblet. 2009. Effect of feed intake on heat production and protein and fat deposition in milk-fed veal calves. *Animal* **3**:557–567. doi:10.1017/S1751731108003777

- Le Bellego, L., J. van Milgen, S. Dubois, and J. Noblet. 2001. Energy utilization of low-protein diets in growing pigs. *J. Anim. Sci.* 79:1259–1271. doi:10.2527/2001.7951259x
- Li, Y, Z. Li, H. Liu, J. Noblet, L. Liu, D. Li, F. Wang, and C. Lai. 2018. Net energy content of rice bran, corn germ meal, corn gluten feed, peanut meal, and sunflower meal in growing pigs. *Asian-Australas. J. Anim. Sci.* 9:1481–1490. doi:10.5713/ajas.17.0829
- Noblet, J., H. Fortune, C. Dupire, and S. Dubois. 1993. Digestible, metabolizable and net energy values of 13 feedstuffs for growing pigs: effect of energy system. *Anim. Feed Sci. Technol.* 42:131–149. doi:10.1016/0377-8401(93)90029-J
- Noblet, J., H. Fortune, X. S. Shi, and S. Dubois. 1994. Prediction of net energy value of feeds for growing pigs. *J. Anim. Sci.* 72:344–354. doi:10.2527/1994.722344x
- Noblet, J., L. Le Bellego, J. van Milgen, and S. Dubois. 2001. Effects of reduced dietary protein level and fat addition on heat production and nitrogen and energy balance in growing pigs. *Anim. Res.* 50:227–238. doi:10.1051/animres:2001129
- Noblet, J., and J. M. Perez. 1993. Prediction of digestibility of nutrients and energy values of pig diets from chemical analysis. *J. Anim. Sci.* 71:3389–3398. doi:10.2527/1993.71123389x
- Noblet, J., and J. van Milgen. 2013. Energy and energy metabolism in swine. In: L. I. Chiba, editor, *Sustainable Swine Nutrition*. 1st ed. Ames, IA: John Wiley & Sons; p. 23–57.
- NRC. 2012. *Nutrient requirements of swine*. 11th rev. ed. Washington, DC: National Academic Press.
- Varley, P. F., J. J. Callan, and J. V. O'Doherty. 2011. Effect of dietary phosphorus and calcium level and phytase addition on performance, bone parameters, apparent nutrient digestibility, mineral and nitrogen utilization of weaner pigs and the subsequent effect on finisher pig bone parameters. *Anim. Feed Sci. Technol.* 165:201–209. doi:10.1016/j.anifeedsci.2011.02.017
- Velayudhan, D. E., J. M. Heo, and C. M. Nyachoti. 2015. Net energy content of dry extruded-expelled soybean meal fed with or without enzyme supplementation to growing pigs as determined by indirect calorimetry. *J. Anim. Sci.* 93:3402–3409. doi:10.2527/jas.2014-8514
- Villamide, M. J., and L. D. San Juan. 1998. Effect of chemical composition of sunflower seed meal on its true metabolizable energy and amino acid digestibility. *Poult. Sci.* 77:1884–1892. doi:10.1093/ps/77.12.1884
- Waititu, S. M., J. W. Kim, N. Sanjayan, P. Leterme, and C. M. Nyachoti. 2018a. Metabolizable energy and standardized ileal digestible amino acid contents of a high-protein sunflower meal fed to broiler chicks. *Can. J. Anim. Sci.* 98:517–524. doi:10.1139/cjas-2017-0158
- Waititu, S. M., N. Sanjayan, M. M. Hossain, P. Leterme, and C. M. Nyachoti. 2018b. Improvement of the nutritional value of high-protein sunflower meal for broiler chickens using multi-enzyme mixtures. *Poult. Sci.* 97:1245–1252. doi:10.3382/ps/pex418
- Woyengo, T. A., E. Beltranena, and R. T. Zijlstra. 2014. Nonruminant nutrition symposium: controlling feed cost by including alternative ingredients into pig diets: a review. *J. Anim. Sci.* 92:1293–1305. doi:10.2527/jas.2013-7169