Evaluation of collection method and diet effects on apparent digestibility and energy values of swine diets[1]

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ABSTRACT: Two experiments were conducted to investigate the effects of collection method and diet type on digestibility coefficients. In Exp. 1, 24 barrows were fed either a corn–soybean meal (CSBM) diet or CSBM with 20% dried distillers' grains with solubles (CSBM-DDGS). In Exp. 2, the effects of basal diet and collection method on determination of dried distillers' grains with solubles (DDGS) digestibility were studied using 24 barrows. The 4 diets used in Exp. 2 were: a CSBM (basal 1) , a barley–canola meal (BCM; basal 2), 80% basal 1 with 20% DDGS (CSBM-DDGS), and 80% basal 2 with 20% DDGS (BCM-DDGS). In both experiments, feces were collected using a time-based collection method (DY) or a "marker-to-marker" collection method (MM). Diets contained 0.5% of titanium dioxide (TiO_2) for estimating digestibility using the index marker approach (IM). The apparent total tract digestibility (ATTD) of DM and GE were lower ($P < 0.05$) in the CSBM-DDGS diet than in the CSBM diet in Exp. 1 but were not different in Exp. 2. All the estimates of BCM-based diets were consistently lower ($P < 0.05$) than those of CSBM-based diets. In Exp. 1, digestibility coefficients determined by the DY and MM were not different from each other, whereas those estimates were lower ($P < 0.05$) using the IM than those using the total collection approach (TC; DY and MM). In Exp. 2, interactions ($P < 0.05$) were observed between diet type and method for dietary digestibility coefficients. Digestibility and energy values estimated by the DY and MM were not different in pigs fed CSBM-based diets and the BCM-DDGS diet, whereas those estimates were greater ($P < 0.05$) using the DY than those using the MM in pigs fed the BCM. There were no interactions between basal diet and method for estimating DDGS digestibility. The ATTD of DM and GE of DDGS using the MM were greater ($P < 0.05$) than those using the IM, and ATTD of N tended to be greater ($P < 0.10$) using the MM than that using the IM. All estimates using the DY were not different from those using the MM or the IM, except that DE of DDGS was greater ($P < 0.05$) using the DY than when using the IM. Digestibility estimates of DDGS were not affected by basal diets. The mean DE and ME (as-fed basis) of DDGS were 3,994 and 3,688 kcal/kg, respectively, when estimated using the basal 1 diet and were 3,919 and 3,547 kcal/kg, respectively, when estimated using the basal 2 diet. In conclusion, both collection methods can be used to estimate energy and nutrient digestibility of diets and DDGS when using CSBM-based diets.

Key words: collection method, diet, digestibility, pig

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INTRODUCTION

In vivo techniques for estimating digestibility of diets include the total collection approach (**TC**) and the index marker approach (**IM**). The assumption of the TC is that with timed feeding and constant daily feed intake over a sufficiently long adaptation period, daily fecal output remains constant (Adeola, 2001). The IM, however, avoids the quantitative collection of feces by

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mixing an indigestible marker in the diets (Kavanagh et al., 2001). For the TC, feces can be collected using the time-based collection method (**DY**; Liu et al., 2012) or the "marker-to-marker" collection method (**MM**; Pedersen et al., 2007a). To date, no experiments have directly compared the energy and nutrient digestibility estimated using the 2 collection methods.

To estimate the digestibility of a certain ingredient, the difference method is commonly used and assumes that there are no interactions between the digestibility values of a component in the test feedstuff and the basal diet (Adeola, 2001). However, this assumption may not be true. The digestibility estimates of a test feedstuff may be changed if different basal diets are used or contain dramatically diverse nutrient compositions. Various basal diets have been used to investigate the energy and nutrient digestibility of ingredients, but few studies have compared the effects of using different basal diets on the estimation of digestibility coefficients (May and Bell, 1971; Widyaratne and Zijlstra, 2007; Stein et al., 2009).

The objective of this study was to compare the DY and MM relative to estimating the digestibility values of diets or dried distillers' grains with solubles (**DDGS**) when corn–soybean meal (**CSBM**) or barley–canola meal (**BCM**) basal diets were used. Additionally, the IM was used to compare nutrient and energy digestibility of pigs with values determined using the TC. Results from this study will provide a scientific basis for using different methodologies when conducting digestibility trials.

Table 1. Ingredient and chemical composition of the experimental diets used in Exp. 1 (as-fed basis)

Item	Diet[1]			
	CSBM		CSBM -DDGS	
Ingredients, %				
DDGS	–		20	
Corn	77.90		61.62	
Soybean meal, 47.5% CP	18.20		15.48	
Corn oil	1.10		0.35	
Limestone, ground	0.80		1.25	
Dicalcium phosphate, 18.5% P	0.80		0.10	
L-Lys HCL, 78%	0.10		0.10	
Salt	0.25		0.25	
Vitamin premix[2]	0.20		0.20	
Trace mineral premix[3]	0.15		0.15	
TiO_2	0.50		0.50	
Calculated composition				
ME, kcal/kg	3,343		3,343	
CP, %	12.69		14.53	
Fermentable fiber, %	9.95		13.84	
SID[4] Lys, %	0.70		0.69	
Ca, %	0.56		0.56	
STTD[5] P, %	0.25		0.22	
Analyzed composition	Replicate 1	Replicate 2	Replicate 1	Replicate 2
DM, %	89.00	88.85	89.67	88.93
CP, %	14.31	14.63	17.69	18.69
NDF, %	10.72	11.23	14.22	17.01
TiO_2, %	0.49	0.44	0.45	0.43
GE, kcal/kg	3,944	3,945	4,106	4,080

[1]CSBM = corn–soybean meal; DDGS = dried distillers' grains with solubles.

[2]Vitamin premix supplied, per kilogram of diet, 5,500 IU vitamin A (as retinyl acetate), 550 IU vitamin D (as cholecalciferol), 30 IU vitamin E (as tocopheryl acetate), 4.4 mg vitamin K (as menadione dimethylpyrimidinol bisulfate), 11.0 mg riboflavin, 22.05 mg D-pantothenic acid, 33.0 mg niacin, and 33.0 mg vitamin B_{12} (as cyanocobalamin).

[3]Trace mineral premix contained 10 mg/kg copper (as $CuSO_4 \cdot 5H_2O$), 0.25 mg/kg iodine (as $Ca(IO_3) \cdot H_2O$), 125 mg/kg iron (as $FeSO_4FeSO_4 \cdot 2H_2O$), 15 mg/kg manganese (MnO), 0.3 mg/kg selenium (Na_2SeO_3), and 125 mg/kg zinc ($ZnSO_4 \cdot H_2O$).

[4]SID = standardized ileal digestible.

[5]STTD = standardized total tract digestible.

MATERIALS AND METHODS

The experimental protocol was reviewed and approved by the Institutional Animal Care and Use Committee of the University of Nebraska, Lincoln.

For both experiments, diets were formulated to meet or exceed the nutrient requirements of pigs at 75 to 100 kg BW according to the NRC (2012). All diets contained 0.5% TiO_2 as an index marker for the determination of nutrient digestibility using the IM.

Experiment 1

Twenty-four terminal crossbred barrows in 2 replicates (87.9 ± 2.2 and 88.5 ± 2.6 kg initial BW, respectively) were randomly allotted to 4 treatments (diet × collection method factorial arrangements; 6 pigs per treatment). Pigs were individually penned in a temperature-controlled room containing 12 metabolism crates. Pigs were allowed 10 d to adapt to the diets and the crates, after which the specific collection methods were initiated. Feed intake was determined based on the lowest ad libitum intake during the early adaptation period and remained approximately 2.8 kg/d (3.3 ×ME for maintenance) through out the experiment. Pigs were given 2 equal meals at 0700 and 1700 h and ad libitum access to water for the entire duration of the experiment.

Diets consisted of CSBM and CSBM with 20% DDGS were formulated to be isocaloric (ME basis; Table 1). Within each dietary treatment, fecal collection of pigs was conducted using either the DY or the MM. For pigs assigned the DY, feces were collected for 96 h starting at 0700 h on d 11 and ceased at 0700 h on d 15 of the experimental period. Feed intake recording began at the morning meal of d 11 and ceased immediately before the morning meal on d 15. For pigs assigned the MM, the initiation and termination of fecal collections were marked by the addition of 0.5% carmen indigo into the morning meal on d 11 and 15. During the collection period of each method, feces were collected at 0700 and 1700 h daily and the total quantities of feces were stored at −20°C. The same fecal sample for individual pigs was also used to represent a sample for the IM. Urine was continuously collected for 96 h, and collection was initiated on d 11 at 0700 h and ceased on d 15 at 0700 h for all pigs. Urine was collected and weighed in plastic buckets containing 65 mL of 6 *N* HCl. Ten percent of the collected urine was subsampled daily in the morning and stored at −20°C immediately after collections. At the end of the experiment, urine samples were thawed and mixed within individual pig and subsampled for analysis.

Experiment 2

Twenty-four barrows in 2 replicates (90.3 ± 2.1 and 90.9 ± 2.4 kg initial BW, respectively) were individually penned in metabolism crates and fed 1 of 4 experimental diets (6 pigs per treatment). Pigs were adapted to diets and crates for 9 d, after which the collection period (4 d) was initiated. The feed intake of all pigs were adjusted to 2.4 kg/d (>2.5 ×ME for maintenance) during the early adaptation period according to the lowest daily ad libitum feed intake of the pigs and remained constant during the remainder of the experimental period.

The CSBM (basal 1)and BCM (basal 2) diets were formulated to contain similar standardized ileal digestible Lys and Ca and standardized total tract digestible P to meet the NRC (2012) requirements of a 90-kg pig but were not isocaloric. The CSBM with 20% DDGS (**CSBM-DDGS**) diet consisted of 80% CSBM (basal 1) and 20% DDGS, and the (BCM-DDGS) diet consisted of 80% basal 2 with 20% DDGS. Fat was not used in this experiment; therefore, the BCM-DDGS and BCM diets were 5 to 7% lower in ME compared with NRC (2012).

The DY and MM were used for each pig to obtain separate fecal collections. The DY was initiated at 0700 on d 10 of the experimental period; meanwhile, the respective meals mixed with 0.5% marker (carmen indigo) were fed to all the pigs. Feces excreted from the beginning of DY to the appearance of the first marker were collected in Bag 1. At 0700 h on d 14, DY was ceased and the second marked meal was fed. Feces excreted from the appearance of the first marker to the end of DY period were collected into Bag 2. Feces excreted after that and until the appearance of the second marker were collected in Bag 3. The sum of the amount of feces in Bag 1 and Bag 2 represented fecal collection using the DY, and the addition of Bag 2 and Bag 3 represented fecal collection using the MM. A pooled fecal sample from the 3 bags for individual pigs was used for nutrient, energy, and Ti analysis using the IM. Fecal and urine collection protocols were the same as described for Exp. 1 except that the amount of urine that some pigs excreted decreased relative to expectations in this study; therefore, a constant amount (10 to 50%) of the daily collected urine was stored for individual pigs.

Chemical Analysis

Diet samples and oven-dried (100°C) fecal samples were ground through a 1.0-mm screen before analysis. Feed and fecal samples were analyzed for DM (procedure 930.15; AOAC, 1995), N (TruSpec N Determinator; Leco Corporation, St. Joseph, MI; procedure 984.13; AOAC, 1995), and Ti (Leone, 1973). Feed samples were also analyzed for NDF (Van Soest et al., 1991). The energy concentration in feed, fecal, and urine samples were determined by bomb calorimetry (Parr 1241 Calorimeter; Parr Instrument Co., Moline, IL). Ether extract content of DDGS and diets used in Exp. 2 was determined as described in AOAC (2000) method 920.39. In Exp. 2, energy and chemical analysis of fecal subsamples from the 3 bags for individual pigs were measured separately. Data of Bag 1 and Bag 2 were pooled for determining digestibility values using the DY, and data of Bag 2 and Bag 3 were pooled for estimates using the MM. In proportion to the fecal collection of each bag relative to the total amount, subsamples of the 3 bags were pooled to represent a 50-g sample. Subsequently, DM, N, GE, and Ti of these fecal samples were measured for the determination of digestibility estimates using the IM.

Calculations and Statistical Analysis

In Exp. 1 and 2, nutrient and energy digestibility and balance of complete diets using the TC and IM were calculated as described by Adeola (2001). Titanium dioxide recovered in the feces relative to feed intake was calculated as described by Jagger et al. (1992). In Exp. 2, the digestibility and energy values of DDGS were calculated using the difference method by subtracting the contribution of basal diet from the respective total diet (Adeola, 2001; Baker and Stein, 2009). Average estimates of basal diet in each replicate ($n = 3$) were used to calculate the digestibility and energy values of DDGS in the total diet for pigs in the same replicate (Fan and Sauer, 1995). The energy values of diets and DDGS were calculated on a DM basis and subsequently converted to an as-fed basis (based on the analyzed DM).

All data were analyzed by GLIMMIX procedure (SAS Inst. Inc., Cary, NC). Both experiments were completely randomized designs, in which the individual pig was considered the experimental unit and a random effect. The 2 replicates were also considered a random effect.

Table 2. Ingredient and chemical composition of the experimental diets used in Exp. 2 (as-fed basis)

Item		Diet[1]			
		CSBM	CSBM-DDGS	BCM	BCM-DDGS
Ingredient, %					
Corn		76.83	61.46	–	–
Soybean meal, 47.5% CP		20.50	16.40	–	–
Barley		–	–	68.35	54.68
Canola meal		–	–	29.50	23.60
DDGS		–	20.00	–	20.00
L-Lys HCl, 78%		0.15	0.12	0.15	0.12
L-Trp		–	–	0.05	0.04
Dicalcium phosphate, 18.5% P		0.65	0.52	0.30	0.24
Limestone		0.77	0.62	0.55	0.44
Salt		0.25	0.20	0.25	0.20
Trace mineral premix[2]		0.15	0.12	0.15	0.12
Vitamin premix[3]		0.20	0.16	0.20	0.16
TiO_2		0.50	0.40	0.50	0.40
Calculated composition					
ME, kcal/kg		3,290	3,319	3,071	3,144
Fermentable fiber, %		10.45	14.06	11.71	15.07
CP, %		16.26	18.47	19.97	21.45
SID[4] Lys, %		0.80	0.73	0.80	0.73
Ca, %		0.52	0.44	0.52	0.44
STTD[5] P, %		0.24	0.28	0.24	0.28
Analyzed nutrients[6]	DDGS				
DM, %	91.3	89.5	89.7	92.4	92.2
CP, %	27.3	16.0	18.5	22.4	23.8
NDF, %	37.7	15.5	18.1	35.1	34.1
Ether extract, %	11.95	2.64	4.30	4.93	6.67
TiO_2, %	–	0.46	0.36	0.47	0.39
GE, kcal/kg	4,808	3,953	4,140	4,246	4,358

[1]CSBM = corn–soybean meal; CSBM-DDGS = CSBM with 20% dried distillers' grains with solubles (DDGS); BCM = barley–canola meal; BCM-DDGS = 80% basal 2 with 20% DDGS (BCM-DDGS).

[2]Trace mineral premix contained 10 mg/kg copper (as $CuSO_4{\cdot}5H_2O$), 0.25 mg/kg iodine (as $Ca(IO_3){\cdot}H_2O$), 125 mg/kg iron (as $FeSO_4FeSO_4{\cdot}2H_2O$), 15 mg/kg manganese (MnO), 0.3 mg/kg selenium (Na_2SeO_3), and 125 mg/kg zinc ($ZnSO_4{\cdot}H_2O$).

[3]Vitamin premix supplied, per kilogram of diet, 5,500 IU vitamin A (as retinyl acetate), 550 IU vitamin D (as cholecalciferol), 30 IU vitamin E (as tocopheryl acetate), 4.4 mg vitamin K (as menadione dimethylpyrimidinol bisulfate), 11.0 mg riboflavin, 22.05 mg D-pantothenic acid, 33.0 mg niacin, and 33.0 mg vitamin B_{12} (as cyanocobalamin).

[4]SID = standard ileal digestible.

[5]STTD = standardized total tract digestible.

[6]Data represents means of dietary nutrient concentrations from 2 replicates.

In Exp. 1, data from the 2 replicates were combined and analyzed as 2 × 2 factorial for the comparison of collection method and diet. The statistic model was

$$Y_{ijk} = \mu + \alpha_i + \beta_j + (\alpha\beta)_{ij} + \gamma_k + \omega_{ik} + s_{ijk},$$

in which Y_{ijk} is the individual observation of the *k*th pig, μ is the overall mean, αi is the effect of the ith diet, β_j is the effect of the jth collection method, $(\alpha\beta)_{ij}$ is the interaction of diet and collection method, γ_k is the variance due to replicates, ω_{ik} is the variance among animals, and s_{ijk} is the residual error. To compare the TC with the IM, data were analyzed as a 2 × 2 factorial arrangement in a split-plot, completely randomized design. Collection methods (DY vs. MM) were considered the whole-plot units and the approaches (TC vs. IM) were the split-plot units. Diet, collection method (DY vs. MM), and approach (TC vs. IM) were considered fixed effects.

In Exp. 2, data were analyzed as a 4 × 3 factorial in a split plot for comparisons of dietary digestibility and as a 2 × 3 factorial in a split plot for comparisons of DDGS digestibility. Pigs were the whole-plot units and methods (DY, MM, or IM) were the split-plot units. Observations from 1 pig fed the basal 2 diet were not used because of low feed intake during the early experimental period. The statistic model is as follows:

$$Y_{ijk} = \mu + \alpha_i + \beta_j + (\alpha\beta)_{ij} + \gamma_k + \omega_{ik} + s_{ijk},$$

in which Y_{ijk} is the individual observation of the *k*th pig, μ is the average digestibility estimates of diet or DDGS, α_i is the effect of the ith diet or basal diet, β_j is the effect of the jth collection method, $(\alpha\beta)_{ij}$ is the interaction of diet and collection method, γ_k is the variance due to replicates, ω_{ik} is the variance among animals, and s_{ijk} is the residual error. All means are presented as least squares means (±SEM). A *P*-value less than 0.05 was considered significant, and $0.05 < P < 0.10$ was denoted as a trend.

RESULTS

Diets used in each replicate were independently analyzed for both experiments. Nitrogen and GE contents were similar between the 2 replicates within each diet (Tables 1 and 2). On a cumulative basis, more than 94% of the TiO_2 was recovered in the feces. Dietary treatment and collection method did not affect the recovery rate of TiO_2 in Exp. 1(Table 3).

In Exp. 1, there were no interactions between diet and collection method for any of the analyzed variables (Table 3). The apparent total tract digestibility (**ATTD**) of DM and GE were 2.88 and 2.77% greater, respectively ($P < 0.05$), in pigs fed the CSBM diet compared to pigs fed the CSBM-DDGS diet. However, no differences in N digestibility or apparent DE and ME were observed between pigs fed the CSBM diet and pigs fed the CSBM-DDGS diet. Dietary digestibility estimates were not affected by using different collection methods (DY vs. MM), but ME values tended to be lower ($P < 0.10$) when estimated using the MM than when estimated using the DY. Comparing digestibility variables calculated using the IM vs. those calculated using the TC (Table 4), there were no 2-way or 3-way interactions observed in any analyzed values

Table 3. The effects of diet and collection method on the digestibilities of nutrients and energy values in pigs (Exp. 1; data were analyzed only using the total collection method)

	CSBM[2]		CSBM-DDGS[3]		Main effects[4]		
	Collection method		Collection method			Diet	Method
Analysis[1]	DY	MM	DY	MM	SED	*P*-value	*P*-value
Digestibility							
DM, %	89.07	88.61	86.59	85.32	0.518	0.001	0.110
N, %	87.87	87.71	88.13	86.93	0.610	0.672	0.282
GE, %	88.66	88.34	86.45	85.00	0.555	0.001	0.126
DE, kcal/kg[5]	3,497	3,484	3,538	3,479	22.55	0.428	0.127
ME, kcal/kg[5]	3,389	3,372	3,397	3,340	21.05	0.565	0.097
TiO_2 recovery, %	94.06	94.18	95.26	100.70	2.582	0.152	0.296

[1]*n* = 6 per treatment.

[2]CSBM = corn–soybean meal; DY = time-based collection method; MM = "marker-to-marker" collection method.

[3]CSBM-DDGS = CSBM with 20% dried distillers' grains with solubles.

[4]There were no interactions between diet and collection method ($P > 0.10$).

[5]As-fed basis.

among diet, collection method (DY and MM), and approach (TC and IM; data not shown). The ATTD of DM, N, and GE were approximately 0.5 percentage units lower ($P < 0.05$) when estimated using the IM than when estimated using the TC. Apparent DE (as-fed basis) was also lower ($P < 0.05$) when measured using the IM than when measured using the TC (3,479 vs. 3,500 kcal/kg, respectively) as was ME (3,354 vs. 3,374 kcal/kg, respectively).

In Exp. 2, there was an interaction ($P < 0.05$) between diet type and method for apparent dietary nutrient and energy digestibility (Table 5). The ATTD of DM, N, and GE and apparent DE and ME values for pigs fed the CSBM-based diets were greater ($P < 0.05$) than those of pigs fed the BCM-based diets. Digestibility coefficients were not different for pigs fed the CSBM diet vs. pigs fed the CSBM-DDGS diet, regardless of the methodology used. Comparing the BCM diet with the BCM-DDGS diet, the ATTD of DM and GE were not different, regardless of the method, whereas N digestibility along with DE and ME content for pigs fed the BCM-DDGS diet were greater ($P < 0.05$) than those for pigs fed the BCM diet when the MM was used.

The apparent dietary digestibility and energy values were not affected by methodology for pigs fed the CSBM-based diets. However, for pigs fed the BCM diet, all estimates were greater ($P < 0.05$) when calculated using the DY than when calculated using the MM and IM. Using the MM, the ATTD of DM and N were greater ($P < 0.05$) than when using the IM, and the ATTD of GE, DE, and ME tended to be greater ($P < 0.10$) using the MM than when using the IM. For pigs fed the BCM-DDGS diet, estimates calculated using the DY and MM were not different and were greater ($P < 0.05$) than those calculated using the IM. The TiO_2 recovered in feces tended to be affected ($P < 0.10$) by diets but not by collection method (Table 5).

Digestibility estimates and energy values of DDGS were not affected by basal diet type (Table 6). The ATTD of DM and GE, DE, and ME of DDGS using the MM were greater ($P < 0.05$) than when using the IM, and the ATTD of N tended to be greater ($P < 0.10$) using the MM than when using the IM. All the estimates of DDGS using the DY were not different from those using the MM or the IM, except that DE of DDGS was greater ($P < 0.05$) when calculated using the DY than when calculated using IM. The mean DE and ME (as-fed basis) of DDGS were 3,994 and 3,688, respectively, when estimated using the basal 1 diet and were 3,919 and 3,547 kcal/kg, respectively, when estimated using the basal 2 diet.

DISCUSSION

Diet Effects

Urriola and Stein (2012) reported that the ATTD of DM and GE was decreased by inclusion of 30% DDGS in a CSBM diet. This is supported by Wilfart

Table 4. The effects of approaches (total collection vs. index method) on digestibilities of nutrients and energy values in pigs (Exp. 1)

	Approaches[1]			Main effect
Analysis	Total collection	Index marker approach	SED	of approach *P*-value
Digestibility				
DM, %	87.40	86.91	0.157	0.006
N, %	87.66	87.15	0.160	0.005
GE, %	87.11	86.60	0.164	0.006
DE, kcal/kg[2]	3,500	3,479	6.56	0.006
ME, kcal/kg[2]	3,374	3,354	6.56	0.006

[1]*n* = 24 per approach, where digestibility estimates for individual pigs were compared using the total collection approach vs. the index marker approach.

[2]As-fed basis.

Table 5. The effects of diet and collection method on the determination of digestibilities of nutrients and energy values in pigs (Exp. 2)

		Diet[2]				SED		*P*-values		
Responses[1]	Method	CSBM	CSBM-DDGS	BCM	BCM-DDGS	Diet	Method	Diet[3]	Method	Diet × method
Digestibility										
DM, %	DY	88.49	87.12	75.38[a]	75.27[a]	1.299	0.296	0.003	0.001	0.001
	MM	88.50	87.31	73.61[b]	75.44[a]					
	IM	88.82	86.52	72.25[c]	72.64[b]					
N, %	DY	89.29	89.39	70.18[a]	72.89[a]	1.881	0.373	0.003	0.001	0.002
	MM	88.98	89.34	68.30[b]	73.04[a]					
	IM	89.16	88.56	65.70[c]	69.66[b]					
GE, %	DY	88.70	87.54	73.11[a]	73.95[a]	1.509	0.320	0.003	0.001	0.002
	MM	88.70	87.72	71.29[b]	74.16[a]					
	IM	89.07	87.02	69.93[bc]	71.37[b]					
DE, kcal/kg[4]	DY	3,507	3,624	3,088[a]	3,223[a]	74.0	13.6	0.011	0.001	0.002
	MM	3,506	3,631	3,012[b]	3,232[a]					
	IM	3,521	3,602	2,969[bc]	3,110[b]					
ME, kcal/kg[4]	DY	3,397	3,481	2,953[a]	3,048[a]	68.5	13.6	0.008	0.001	0.002
	MM	3,397	3,488	2,876[b]	3,057[a]					
	IM	3,412	3,459	2,818[bc]	2,936[b]					
TiO_2 recovery, %	DY	110.83	96.13	93.97	95.76	4.395	1.881	0.086	0.503	0.451
	MM	110.65	94.95	100.87	95.35					

[a–c]For the same response criteria, means within the column not sharing a common superscript letter are different ($P < 0.05$).

[1]$n = 5$ for pigs fed the barley–canola meal (BCM) diet calculated using the time-based collection method (DY) or the "marker-to-marker" collection method (MM); $n = 6$ for the 2 groups of pigs fed the corn–soybean meal (CSBM)–based meal and those fed the 80% basal 2 with 20% DDGS (BCM-DDGS). diet calculated using the DY or MM; $n = 6$ for all the means calculated using the index marker approach (IM).

[2]CSBM-DDGS = CSBM with 20% dried distillers' grains with solubles.

[3]All the digestibility coefficients for pigs fed CSBM-based diets were greater ($P < 0.05$) than for BCM-based diets regardless of method, and there were no differences between the CSBM diet and the CSBM-DDGS diet regardless of method.

[4]As-fed basis.

et al. (2007), who reported that increasing dietary fiber decreases the ATTD of DM and GE. The addition of fiber to swine diets decreases the DE concentration of the diet (Galassi et al., 2010). The concentrations of ADF, NDF, and total dietary fiber in DDGS (9.9, 25.3, and 42.1%, respectively) are about 3 times greater than in corn (Stein and Shurson, 2009). Urriola et al. (2010) reported that the ATTD of total dietary fiber in DDGS was only 46%, which may contribute to the decreased ATTD of DM and GE in corn-based coproducts. Furthermore, dietary fiber increased the ileal flow rate of most nutrients, which subsequently decreased the apparent digestibility of carbohydrates and energy (Serena et al., 2008). In Exp. 1, the CSBM-DDGS diet had 4.6% greater NDF concentration than the CSBM diet, which resulted in decreased ATTD of DM and GE in the CSBM-DDGS diet compared with CSBM diet. However, differences of digestibility estimates between the CSBM and CSBM-DDGS diets were not significant in Exp. 2. This may be partially attributed to lower daily feed intake in Exp. 2 than in Exp. 1 (2.4 vs. 2.8 kg). Haydon et al. (1984) reported an increased disappearance of absorbable nutrients in the large intestine with decreasing feed intake, indicating an increased nutrient digestibility. At lower feed intake, the longer retention time (Seerley et al., 1962) and less nonprotein substrate for microbes' requirement may result in increased deamination of AA by microbes and absorption in the large intestine (Fuller and Reeds, 1998). Contrasting results have been published on the effects of feed intake on apparent energy digestibility of pigs. No difference was observed for the ATTD of GE at different feed intake levels (Moter and Stein, 2004). In contrast, Haydon et al. (1984) reported increased energy digestibility with decreased feed intake. Harris et al. (2012) selected pigs based on their residual feed intake (**RFI**) and concluded that the digestibility values for DM, CP, and GE were greater in the low-RFI pigs than in the high-RFI pigs, whereas P digestibility did not differ between the pig lines.

In addition, the consistently lower digestibility values of BCM-based diets compared with CSBM-based diets may be due to greater NDF concentrations in the BCM diets. The ATTD of GE in canola meal was 60.3 to 68.6%, depending on different NDF contents of varieties (Le et al., 2012). Results of the current study are in agreement with studies that showed the inclusion of canola meal decreased the apparent digestibility of diets (Sanjayan, 2013). Barley contains greater

Table 6. The effects of basal diet and collection method on the digestibility of nutrients and energy values of dried distillers' grains with solubles (DDGS) in pigs (Exp. 2)

		DDGS[3]		SED		*P*-values		
Responses[1]	Method[2]	Basal 1	Basal 2	Basal diet	Method	Basal diet	Method	Basal diet × method
Digestibility								
DM, %	DY[ab]	81.89	77.81	10.73	2.24	0.863	0.007	0.298
	MM[a]	82.70	85.18					
	IM[b]	77.54	74.23					
N, %	DY	89.24	84.63	14.06	2.61	0.867	0.155	0.518
	MM	89.82	90.65					
	IM	87.14	82.90					
GE, %	DY[ab]	83.52	79.36	10.69	2.17	0.863	0.018	0.328
	MM[a]	84.32	86.07					
	IM[b]	80.28	76.40					
DE, kcal/kg[4]	DY[a]	4,016	4,024	560	126	0.905	0.033	0.256
	MM[a]	4,054	4,138					
	IM[b]	3,913	3,594					
ME, kcal/kg[4]	DY[ab]	3,685	3,490	501	110	0.805	0.067	0.192
	MM[a]	3,723	3,812					
	IM[b]	3,656	3,338					

[a,b]Means for the same response not sharing a common superscript letter are different ($P < 0.05$).

[1]$n = 6$ per basal diet.

[2]DY = time-based collection method; MM = "marker-to-marker" collection method; IM = index marker approach.

[3]Basal 1 = 80% corn–soybean meal; Basal 2 = 80% barley–canola meal.

[4]As-fed basis.

CP but also greater fiber content (ADF and NDF) than corn (Pedersen et al., 2007b), which may be one of the factors for the decreased digestibility estimates in pigs fed BCM-based diets.

The different fiber concentrations of the CSBM and CSBM-DDGS diets, in both experiments, did not affect estimates of N digestibility. This result was inconsistent with some other digestibility studies where a decreased N digestibility as dietary fiber concentration increased was observed (Wilfart et al., 2007; Urriola and Stein, 2012). Comparatively, no differences were observed for the average ATTD of N in DDGS vs. corn diets (Pedersen et al., 2007a; Liu et al., 2012). A lower ATTD of N can be explained by increased endogenous N losses, decreased hydrolysis and absorption of nutrients, or both (Wilfart et al., 2007). The ileal endogenous losses (g/kg DM intake) increased with the increased concentration of barley bran but was not affected by the different concentrations of barley hulls (Leterme et al., 2000), indicating that endogenous N loss is affected by the dietary fiber sources. Some studies have shown that when fiber source does not contribute significant amounts of protein to the diet, an increase in the concentration of fiber does not affect protein digestibility (NRC, 1998). The 20% DDGS contributed 5.3% protein, representing 30% in the diet, which may help explain the observation that N digestibility was not different between the 2 CSBM-based diets in the present study. Similarly, the canola meal contributed about 50% of the total CP content to the BCM diet; additionally, the NDF content of BCM-based diets was much greater than that of the CSBM-based diets. Therefore, N digestibility of BCM-based diets was decreased compared with the CSBM-based diets.

In Exp.1, the analyzed ME values of the CSBM and DDGS diet, averaging 3,381 and 3,368 kcal/kg, respectively, were similar to the calculated values (3,343 kcal/kg). In Exp. 2, the analyzed ME values of CSBM-based diets (3,402 and 3,476 kcal/kg, respectively) were greater than the predicted NRC (2012) values (3,290 and 3,319 kcal/kg, respectively), whereas ME values for BCM-based diets (2,882 and 3,014 kcal/kg, respectively) were lower than the NRC values (3,071 and 3,144 kcal/kg, respectively). This may due to the different nutrient composition (fat, protein, NDF, etc.) of various ingredient sources. The ME of most practical swine diets used in North America is 94 to 97% of DE (NRC, 2012). In both experiments, ME percentages in DE for the CSBM and the CSBM-DDGS diet were 97 and 96%, respectively, whereas ME:DE ratios were 95.4 and 94.5% for the BCM and BCM-DDGS diets, respectively. Pedersen et al. (2007a) reported that DE and ME values of DDGS and corn were not different and indicated that with greater CP concentration in DDGS, the increased urine N excretion may cause the decreased ME:DE ratio compared with corn.

Basal Diet Effect on Dried Distillers' Grains with Solubles Digestibility

Using the basal 1 diet in Exp. 2, the ATTD of DM, N, GE, DE, and ME of DDGS were slightly greater than the upper ranges of DDGS estimates reported by others (Pedersen et al., 2007a; Stein et al., 2009). This is because the GE of DDGS used in the current study (4,808 kcal/kg) was also greater than the greatest value (4,324 kcal/kg) among the 14 DDGS sources previously cited. When the basal 2 diet was used, the observations were more variable, which was likely due to relatively greater NDF concentrations that increased the variability of hindgut digestibility among animals (Wilfart et al., 2007). This may partially contribute to the lack of differences in apparent nutrient and energy digestibility of DDGS between basal diets. Another reason may be the relatively lower dietary inclusion level of DDGS compared with levels used in other studies (Pedersen et al., 2007a; Stein et al., 2009). Fan and Sauer (1995) suggested that increasing the dietary inclusion level of the test ingredient in the total diet decreased the SE of the estimates. However, Le et al. (2012) also showed that the DE content of canola meal was not affected by dietary inclusion levels.

Using corn and wheat as the basal diet, respectively, energy digestibility of DDGS in growing pigs was estimated to be 75.1 and 78.7%, respectively, from 2 different studies (Widyaratne and Zijlstra, 2007; Stein et. al., 2009). The difference of CP contributions to the total diet for the basal diet and the test feedstuff may affect the digestibility estimates of the ingredient (May and Bell, 1971). Moreover, the greater NDF concentration in BCM-based diets may affect nutrient composition in the excreta because dietary fiber increases endogenous and exogenous losses (Schulze et al., 1994; Leterme et al., 2000). It was possible that the endogenous and exogenous losses due to the substitution of DDGS were underestimated and subsequently affect the determination of DDGS digestibilities using the basal 2 diet.

Day vs. the Marker-to-Marker Collection Method

To date, few studies have directly compared the digestibility estimates for pigs using the DY vs. the MM. By indirectly comparing the DY and MM, Lammers et al. (2008) reported that the apparent DE and ME concentrations of crude glycerol were not affected by collection methods. The digestibility values estimated using the DY and MM were similar except for the basal 2 diet. This difference was likely due to the lesser amount of fecal collection by the DY (1,977 g) than by the MM (2,146 g) for pigs fed the basal 2 diet (data not shown). For the MM, the marker appearances were recorded only at feeding times. The approximate transit durations between the first and second marker were 96, 91, 98, and 96 h, respectively, for the CSBM, CSBM-DDGS, BCM, and BCM-DDGS diets. The collection duration for the DY method, however, was fixed at 96 h. Therefore, the longer collection duration caused less feces to be collected in the DY than in the MM for the basal 2 diet. The assumption of the constant daily fecal output in the TC methodology may not always be true. A longer collection period may reduce the mean difference estimates of the collection durations between the DY and MM.

There were essentially no differences between the DY and MM for estimating nutrient and energy digestibility of DDGS. Using the basal 2 diet, numerically lower nutrient and energy digestibility coefficients of DDGS estimated using the DY than those estimated using the MM were observed. This was likely due to greater digestibility estimates of the basal 2 diet calculated using the DY than those calculated using the MM, resulting in underestimation by the DY or overestimation by the MM for the determination of DDGS digestibility. The similar DE and ME values of DDGS estimated using the DY and MM indicated that either method may be used for estimating DDGS digestibility.

The Total Collection Approach vs. the Index Marker Approach

In Exp. 1, the differences between mean digestibility estimates of the TC (DY and MM) and the IM were only about 0.5% percentage units for the digestibility estimates and 20 kcal/kg for dietary energy values. Significant differences were still observed because of the relatively decreased SE compared with the total collection model (DY vs. MM). The concentrations of DM, N, and GE used in the IM are the same values that used in the TC. Data were analyzed as a split-plot structure so that estimates calculated using the IM and TC were compared within the individual pigs (whole-plot units). The variance among animals was eliminated. Therefore, the reduced digestibility coefficients estimated by the IM compared with those estimated by the TC were primarily contributed by the differences of TiO_2 recovery relative to 100%. Other studies also showed that the recovery of index marker in the feces was correlated with the digestibility coefficients (Kavanagh et al., 2001).

In Exp. 2, fecal samples in 3 bags for individual pigs were pooled and separately measured for data using the IM. Similar digestibility estimates and dietary energy values were observed for the CSBM-based diets when using the IM, DY, and MM, indicating that the IM is suitable for estimating CSBM-based diets. Nevertheless, the IM seemed to underestimate digestibility of BCM-based diets and DDGS compared with the TC. The TiO2 recovery rates were similar between the DY and MM when CSBM-based diets and the BCM-DDGS diet were fed; however, the recovery rates related to the 2 collection methods were not consistent when the basal 2 diet was fed, which were mainly due to the different amounts of fecal collections between the DY and MM previously discussed.

There were essentially no differences between the DY and MM for estimation of DDGS digestibility. It appears that either collection method can be used to estimate digestibility values of diets and DDGS when using CSBM-based diets. No significant differences were detected for digestibility of DDGS using different basal diets. Although digestibility estimates were lower using the IM than when using the TC, the differences were not substantial for the CSBM diets and the relative ranking of diets/ingredients was similar between methods.

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