Determination and prediction of digestible and metabolizable energy concentrations in byproduct feed ingredients fed to growing pigs

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Objective: An experiment was conducted to determine digestible energy (DE) and metabolizable energy (ME) of different byproduct feed ingredients fed to growing pigs, and to generate prediction equations for the DE and ME in feed ingredients.

Methods: Twelve barrows with an initial mean body weight of 31.8 kg were individually housed in metabolism crates that were equipped with a feeder and a nipple drinker. A 12×10 incomplete Latin square design was employed with 12 dietary treatments, 10 periods, and 12 animals. A basal diet was prepared to mainly contain the corn and soybean meal (SBM). Eleven additional diets were formulated to contain 30% of each test ingredient. All diets contained the same proportion of corn:SBM ratio at 4.14:1. The difference procedure was used to calculate the DE and ME in experimental ingredients. The in vitro dry matter disappearance for each test ingredient was determined.

Results: The DE and ME values in the SBM sources were greater (p<0.05) than those in other ingredients except high-protein distillers dried grains. However, DE and ME values in tapioca distillers dried grains (TDDG) were the lowest (p<0.05). The most suitable regression equations for the DE and ME concentrations (kcal/kg on the dry matter [DM] basis) in the test ingredients were: DE = 5,528–(156×ash)–(32.4×neutral detergent fiber [NDF]) with root mean square error = 232, R² = 0.958, and p<0.001; ME = 5,243–(153 ash)–(30.7×NDF) with root mean square error = 277, R² = 0.936, and p<0.001. All independent variables are in % on the DM basis.

Conclusion: The energy concentrations were greater in the SBM sources and were the least in the TDDG. The ash and NDF concentrations can be used to estimate the energy concentrations in the byproducts from oil-extraction and distillation processes.

Keywords: Feedstuff; Prediction Models; Protein Supplements; Swine

INTRODUCTION

Oilseed meals are used primarily as a protein source [1], but play a role as an energy source in swine diets. Soybean meal (SBM) is one of the most commonly used oilseed meals in the swine diet. However, alternative feed ingredients, which can replace the SBM in the swine diet, are needed as the price of SBM has been continuously increasing. An accurate determination of energy concentrations of the ingredients is important to use relatively cheaper feed ingredients in the swine diet. However, studies about energy concentrations in various protein sources for pigs are limited.

The digestible energy (DE) and metabolizable energy (ME) concentrations of the feed ingredients are ideally determined via animal experiment, which is the most accurate method. However, because animal experiments are time-consuming and costly, equations for predicting the energy concentrations of feed ingredients can be used as an alternative method [2]. Addi-
tionally, the in vitro dry matter disappearance (IVDMD) of ingredients can also be useful for predicting energy concentration in ingredients for swine diets [3]. However, the use of equations can be limited to the range of nutrient compositions in the ingredients that were used to generate the equations [4,5]. We hypothesized that energy concentrations in the feed ingredients with large range of chemical composition can be estimated using prediction equations with the IVDMD as an independent variable. The objectives were to determine the DE and ME of 9 byproducts from the oil-extraction process and 2 byproducts from distillation process fed to growing pigs and to generate equations that predict the DE and ME of byproduct feed ingredients.

MATERIALS AND METHODS

Animal care
The experimental procedure was approved by the Institutional Animal Care and Use Committee at Konkuk University (KU 12062).

Diet and feeding
Twelve barrows with a mean initial body weight of 31.8 kg (standard deviation = 2.7) were used to determine the DE and ME concentrations of sesame meal produced in Korea, two sources of dehulled SBM produced in Korea (SBM-KD1 and SBM-KD2), SBM produced in India (SBM-I), high-protein distillers dried grains (HPDDG) produced from corn in the USA, perilla meal (PM) produced in Korea, canola meal produced in Indonesia, copra meal produced in the Philippines, corn germ meal produced in Korea, palm kernel expellers produced in Malaysia, and tapioca distillers dried grains (TDDG) produced in China (Table 1). The palm kernel product was classified as the expellers and tapioca distillers dried grains (TDDG) produced in China, perilla meal (PM) produced in Korea, canola meal produced in Indonesia, copra meal produced in the Philippines, corn germ meal produced in Korea, palm kernel expellers produced in Malaysia, and tapioca distillers dried grains (TDDG) produced in China (Table 1). The palm kernel product was classified as the expellers because the concentration of ether extract in the feed ingredient was 6.97% [6].

The pigs were placed in metabolic cages equipped with a feeder and a nipple drinker. A 12×10 incomplete Latin square design was employed with 12 dietary treatments, 10 periods, and 12 animals. Potential carryover effects were balanced using a spreadsheet-based program [7]. The quantity of feed provided daily per pig was calculated as approximately 2.7 times the estimated energy requirement for maintenance (i.e., 106 kcal of ME per kg body weight0.75) adjusted in the NRC [8] based on the calculated ME concentration in the diets. The feed was divided into two equal meals and fed to pigs at 0730 and 1630. Water was available at all times. Body weight was measured at the end of each period to determine feed allowance.

A basal diet contained corn and SBM as the sole energy sources. Eleven additional diets were formulated to contain 30% of each test ingredient (Table 2). All diets contained the same proportion of corn:SBM ratio at 4.14:1. Vitamins and minerals were adequate to meet requirement estimates in the literature [8].

Sample collection
An experimental period consisted of a 4-d adaptation period and a 4-d collection period. Feed refusals were collected and dried in a forced-air drying oven at 55°C until constant weight, and then weighed after cooling at room temperature. Feces were quantitatively collected according to the marker-to-marker procedure [9]. Chromic oxide was used as an indigestible marker and was included at 0.5% in morning meals on d 5 and 9. Fecal collection was started when the green color of marker begin to appear in the feces, and ended when the green color appeared again. Urine was collected from 1400 on d 5 to 1400 on d 9 using plastic containers including a 200 mL of 2 N HCl. A 200 mL aliquot of urine from each animal was placed in a plastic bottle. All feces and the urine were stored at −20°C immediately after collection.

Chemical analysis

Table 1. Energy and nutrient composition of experimental ingredients1 (as-is basis)

| Item                | Ingredient              | Ingredient                          | Ingredient                          | Ingredient                          |
|---------------------|-------------------------|-------------------------------------|-------------------------------------|-------------------------------------|
| Dry matter (%)      | Sesame meal             | Soybean meal-dehulled-Korea 1       | Soybean meal-dehulled-Korea 2       | Soybean meal-India                  |
|                     | 97.0                    | 90.2                                | 90.2                                | 90.1                                |
| Gross energy (kcal/kg) | 4,688                   | 4,299                               | 4,332                               | 4,221                               |
| Crude protein (%)   | 50.0                    | 47.1                                | 47.4                                | 39.6                                |
| Ether extract (%)   | 6.05                    | 2.46                                | 0.74                                | 0.84                                |
| Crude fiber (%)     | 9.3                     | 4.6                                 | 5.7                                 | 5.1                                 |
| Ash (%)             | 11.0                    | 6.2                                 | 6.3                                 | 6.3                                 |
| Neutral detergent fiber (%) | 28.1                    | 7.4                                 | 8.7                                 | 9.6                                 |
| Acid detergent fiber (%) | 17.5                    | 7.2                                 | 9.1                                 | 8.2                                 |
| Calcium (%)         | 2.15                    | 0.64                                | 0.67                                | 0.70                                |
| Phosphorus (%)      | 1.32                    | 0.64                                | 0.62                                | 0.53                                |

1 Data are the mean of duplicate analyses of each ingredient.
Table 2. Ingredient composition and analyzed composition of experimental diets (as-fed basis)

| Item                                      | Basal          | Sesame meal | Soybean meal-dehulled-Korea 1 | Soybean meal-dehulled-Korea 2 | Soybean meal-India | High-protein distillers dried grains | Perilla meal | Canola meal | Copra meal | Corn germ meal | Palm kernel expellers | Tapioca distillers dried grains |
|-------------------------------------------|----------------|-------------|--------------------------------|--------------------------------|-------------------|--------------------------------------|--------------|-------------|------------|---------------|------------------------|-------------------------------|
| Ingredient (%)                            | 78.60          | 54.44       | 54.44                          | 54.44                          | 54.44             | 54.44                                | 54.44        | 54.44       | 54.44      | 54.44          | 54.44                   | 54.44                          |
| Crude protein (%)                         | 19.00          | 13.16       | 13.16                          | 13.16                          | 13.16             | 13.16                                | 13.16        | 13.16       | 13.16      | 13.16          | 13.16                   | 13.16                          |
| Gross energy (kcal/kg)                    | 4,003          | 4,087       | 4,047                          | 4,007                          | 4,087             | 4,137                                | 4,032        | 4,087       | 4,137      | 4,087          | 4,087                  | 4,087                          |
| Dry matter (%)                            | 548            | 543         | 548                            | 548                            | 548               | 548                                  | 548          | 548         | 548        | 548            | 548                    | 548                            |
| Ash (%)                                   | 9.7            | 9.7         | 9.7                            | 9.7                            | 9.7               | 9.7                                  | 9.7          | 9.7         | 9.7         | 9.7            | 9.7                    | 9.7                            |
| 1) Provided the following quantities per kg of complete diet: vitamin A, 25,000 IU; vitamin D₃, 4,000 IU; vitamin E, 50 IU; vitamin K, 5.0 mg; thiamin, 4.9 mg; riboflavin, 10.0 mg; pyridoxine, 4.9 mg; vitamin B₆, 0.06 mg; pantothenic acid, 37.5 mg; folic acid, 1.10 mg; niacin, 62 mg; biotin, 0.06 mg; Cu, 25 mg as copper sulfate; Fe, 268 mg as iron sulfate; I, 5.0 mg as potassium iodate; Mn, 125 mg as manganese sulfate; Se, 0.38 mg as sodium selenite; Zn, 313 mg as zinc oxide; and butylated hydroxytoluene, 50 mg.

The fecal samples were dried in a forced-air drying oven at 55°C and ground before analysis. All diet and fecal samples were dried in a forced-air drying oven at 135°C for 2 h to analyze dry matter [10]. The urine samples were dried according to a method described previously [11]. Approximately 3 mL of the urine sample was added to a cotton ball (0.3 to 0.4 g) placed in a stainless steel crucible. The weight of crucible, cotton ball, and urine was recorded, and then the samples were dried in a freeze dryer for 24 h. Samples of the diets, ingredients, feces, and urine were analyzed for gross energy (GE) concentration using a bomb calorimeter (C 2000; IKA, Staufen, Germany). Ingredient samples were analyzed for crude protein (CP; method 990.03), ether extract (method 920.39), crude fiber (method 978.10) and ash (method 942.05) [12]. Diet and ingredient samples were also analyzed for neutral detergent fiber (NDF; method 2002.04), acid detergent fiber (ADF; method 973.18), calcium (method 978.02), and phosphorus (method 946.06) [12]. The diet samples were also analyzed for the CP and ash according to the aforementioned procedures. Duplicate analyses were performed for all the samples, but the GE concentration was analyzed in triplicate.

Calculation

After the chemical analyses, energy digestibility and metabolizability were calculated using the amount of energy intake and excreted feces and urine. The DE and ME concentrations in the sum of corn and SBM in the basal diet were calculated by dividing energy concentration in the basal diet by the sum of corn and SBM concentrations. The DE and ME concentrations of the test ingredients were calculated using a difference procedure [9].

In vitro dry matter disappearance

The IVDMD of 11 ingredients was determined using procedures reported in previous studies [13-15] with minor modification. The procedure consisted of three steps, and each step simulated digestion in the stomach, small intestine, and large intestine of pigs. In the first step, 0.5 g of ingredient sample was placed in a 100-mL flask with 25 mL of phosphate buffer solution (0.1 M, pH 6.0) and 10 mL of 0.2 M HCl. Then the pH was adjusted to 2.0 using a 1 M HCl or 1 M NaOH solution, and 1 mL of pepsin solution (25 mg/mL; ≥250 units/mg solid, P7000, Pepsin from porcine gastric mucosa, Sigma-Aldrich, St. Louis, MO,
USA) was added. The test flasks were incubated in a shaking incubator at 39°C for 2 h.

In the second step, 10 mL of phosphate buffer solution (0.2 M, pH 6.8) and 5 mL of 0.6 M NaOH solution were added in the test flasks. Then the pH was adjusted to 6.8, and 1 mL of pancreatin solution (100 mg/mL; 4xUSB, P1750, Pancreatin from porcine pancreas, Sigma-Aldrich, USA) was added. Then the test flasks were incubated in a shaking incubator at 39°C for 4 h.

In the third step, 10 mL of 0.2 M ethylenediaminetetraacetic acid solution was added in the test flasks. The pH was adjusted to 4.8. As a substitution of microbial enzyme, 0.5 mL of Viscozyme (VZ10, Viscozyme L, Sigma-Aldrich, USA) was added. Then the test flasks were incubated in a shaking incubator for 18 h at 39°C.

Following the incubation, undigested residues were filtered in glass filter crucibles containing 500 mg of celite as filter aid using the Fibertec System (Fibertec System 1021 Cold Extractor, Tecator, Höganäs, Sweden). Undigested residues in glass filter crucibles were rinsed twice with 10 mL of 95% ethanol and 99.5% acetone. Then, glass filter crucibles with undigested samples were dried at 130°C for 6 h. After 1 h cooling in a desiccator, glass filter crucibles were weighed. The IVDMD for each ingredient was measured in triplicate.

### Statistical analysis
Data were analyzed using the MIXED procedure of SAS (SAS Inst. Inc., Cary, NC, USA). Outliers (difference from median >2×interquartile range) were removed from the dataset for the final statistical analysis. The model included dietary treatment as a fixed variable and animal and period as random variables. Least squares means of each treatment were calculated, and the difference in means was tested using the PDIVF option with the Tukey’s adjustment. The experimental unit was a pig, and the statistical significance was set at p-value <0.05.

### RESULTS

#### Nutrient composition
Values for the GE of the ingredients ranged from 3,875 to 4,924 kcal/kg on an as-is basis (Table 1). The CP concentration of the ingredients ranged from 15.3% to 50.0%, and the NDF concentration ranged from 7.35% to 61.4% on an as-is basis.

#### Digestible and metabolizable energy
Feed intake during the collection period was greater (p<0.05) for the basal, palm kernel expellers, and TDDG diets than that for the HPDDG and canola meal diets (Table 3). Energy digestibility of the basal and SBM-containing diets was greater (p<0.05) than that of the other diets. The DE concentration in the SBM-KD1 diet was greater (p<0.05) than that in the other experimental diets except the SBM-KD2 diet. The ME concentration in the SBM-KD1 diet was also greater (p<0.05) than that in the other diets except the SBM-KD2 and SBM-I diets. The DE and ME in the TDDG diet were the lowest (p<0.05) among the experimental diets. The DE and ME (kcal/kg on an as-fed basis) in the three sources of SBM ingredients were greater (p<0.05) than those in the other experimental ingredients except the HPDDG (Table 4). The DE and ME in the TDDG were also the lowest

### Table 3. Energy utilization of basal and experimental diets containing test ingredients fed to growing pigs

| Item                        | Diet                         | Observation (n) | SEM | p-value |
|-----------------------------|-----------------------------|----------------|-----|---------|
|                             | Basal                        | 10             | 0.15| <0.001 |
|                             | Sesame meal                  | 10             |     |         |
|                             | Soybean meal                 | 9              |     |         |
|                             | Soybean meal-dehulled-Korea 1| 9              |     |         |
|                             | Soybean meal-dehulled-Korea 2| 8              |     |         |
|                             | Soybean meal-India           | 10             |     |         |
|                             | High-protein distillers dried grains | 8     |     |         |
|                             | Perilla meal                 | 9              |     |         |
|                             | Canola meal                  | 5              |     |         |
|                             | Copra meal                   | 8              |     |         |
|                             | Corn germ meal               | 10             |     |         |
|                             | Palm kernel expellers        | 8              |     |         |
|                             | Tapioca distillers dried grains | 8     |     |         |

SEMs, standard error of the mean; GE, gross energy; DE, digestible energy; ME, metabolizable energy.

* Means within a row without a common superscript letter differ (p<0.05).
(p<0.05) among the experimental ingredients.

**Prediction equations for energy concentrations and energy digestibility**

The DE and ME in the ingredient samples were correlated (p<0.05) with the crude fiber, ash, NDF, ADF, IVDMD, and DE:GE ratio (Table 5). A high correlation (p<0.001) was observed between the DE and ME. The NDF and ADF were negatively correlated (p<0.01) with the DE in the byproduct feed ingredients. The R² and p-values of the equation and independent variables were used to evaluate the suitability of the prediction equations, and 3 prediction equations for each of DE and ME were chosen based on the suitability (Tables 6 and 7). The most suitable regression equation for the DE in the byproduct feed ingredients was equation 2: DE (kcal/kg on the dry matter basis) = 5,243−(153×ash)−(30.7×NDF) with root mean square error = 277, R² = 0.936, and p-value <0.001. All independent variables are presented in % on the dry matter basis. A linear relationship was observed between the energy digestibility and IVDMD (r² = 0.534 and p = 0.011; Figure 1).

**DISCUSSION**

Most nutrient compositions of ingredients were within range of previous studies [2,4]. In this study, the lowest DE and ME values in the TDDG diet can be explained mainly by the largest fecal energy output in the pigs fed the TDDG diet. Although GE intake by pigs fed the TDDG diet was not different from most of the other experimental diets, the dry feces output of pigs fed the TDDG diet was the greatest among the experimental diets. The large quantity of fecal output may be caused by the high fiber concentration in the TDDG, which increases passage rate of digesta and lowers time for digestion and absorption of nutrients [16,17]. Therefore, fecal GE output of pigs fed the

| Item          | Observation (n) | As-fed basis | Dry matter basis |
|---------------|----------------|--------------|------------------|
| GE (kcal/kg)  | 4,688          | 4,299        | 4,332            |
| DE (kcal/kg)  | 2,592          | 3,923        | 3,725            |
| ME (kcal/kg)  | 2,269          | 3,782        | 3,552            |
| DE:GE ratio   | 0.55           | 0.91         | 0.86             |
| ME:DE ratio   | 0.88           | 0.96         | 0.95             |
| ME:GE ratio   | 0.48           | 0.88         | 0.82             |
| Dry matter basis |              |              |                  |
| GE (kcal/kg)  | 4,832          | 4,767        | 4,802            |
| DE (kcal/kg)  | 2,630          | 4,381        | 4,063            |
| ME (kcal/kg)  | 2,279          | 4,222        | 3,872            |

SEM, standard error of the mean; GE, gross energy; DE, digestible energy; ME, metabolizable energy.

*p<0.05; ** p<0.01; *** p<0.001.
Table 6. Regression equations for digestible energy in byproduct feed ingredients for growing pigs (kcal/kg dry matter basis)

| Regression coefficient parameter (% dry matter basis) | Statistical parameter |
|------------------------------------------------------|-----------------------|
| Equation 1                                           |                       |
| Intercept                                            | 6,084                 |
| CP                                                   | -10.1                 |
| CF                                                   | -                     |
| Ash                                                  | -153                  |
| NDF                                                  | -37.7                 |
| SE                                                   | 542                   |
| p-value                                              | < 0.001               |
| Equation 2                                           | 5,528                 |
| CF                                                   | -                     |
| Ash                                                  | -156                  |
| NDF                                                  | -32.4                 |
| SE                                                   | 194                   |
| p-value                                              | < 0.001               |
| Equation 3                                           | 4,860                 |
| CP                                                   | -142                  |
| CF                                                   | -                     |
| Ash                                                  | -                      |
| NDF                                                  | -399                  |
| SE                                                   | 265                   |
| p-value                                              | < 0.001               |

RMSE, root mean square error; SE, standard error.

Table 7. Regression equations for metabolizable energy in byproduct feed ingredients for growing pigs (kcal/kg dry matter basis)

| Regression coefficient parameter (% dry matter basis) | Statistical parameter |
|------------------------------------------------------|-----------------------|
| Equation 1                                           |                       |
| Intercept                                            | 6,231                 |
| CP                                                   | -17.9                 |
| CF                                                   | -                     |
| Ash                                                  | -148                  |
| NDF                                                  | -40.1                 |
| SE                                                   | 576                   |
| p-value                                              | < 0.001               |
| Equation 2                                           | 5,243                 |
| CP                                                   | 0.109                 |
| CF                                                   | -                     |
| Ash                                                  | -153                  |
| NDF                                                  | -30.7                 |
| SE                                                   | 232                   |
| p-value                                              | < 0.001               |
| Equation 3                                           | 4,578                 |
| CP                                                   | -136                  |
| CF                                                   | -                     |
| Ash                                                  | -                      |
| NDF                                                  | -436                  |
| SE                                                   | 290                   |
| p-value                                              | < 0.001               |

RMSE, root mean square error; SE, standard error.

TDDG diet was greater than that of pigs fed the other experimental diets except the PM diet despite being the lowest GE in dry feces. For these reasons, the DE in the TDDG diet may be less than that in the other experimental diets. The TDDG diet had the lowest ME value, which may have occurred because the TDDG diet had the lowest DE and the urinary GE output of pigs fed the TDDG diet was not different from most of the other experimental diets.

The DE and ME in the sesame meal were less than values in the literature [2,4], which appear to be due to the greater NDF and ADF concentrations in the sesame meal used in this experiment than the fiber concentrations in the literature [2,4]. Dietary fiber negatively affects the energy utilization [16,18]. Thus, although the GE of sesame meal in this experiment was similar to values in the literature, the DE:GE ratio was less in this experiment than that reported in the literature [2,4].

The GE, DE, and ME in the two sources of SBM-KD were within the range of previous values [2,4,19,20]. The DE, ME, and DE:GE ratio in the SBM-I were similar to the previous values [2,4].

The DE and ME in the HPDDG were less than previous values [4,11,21,22], but were similar with a previous value [23]. The GE in the HPDDG used in this experiment was within the range of previous values, but the DE:GE ratio was less than that in previous studies, resulting in a lower DE and ME in the HPDDG used in this experiment. We cannot clearly explain why
energy digestibility was less compared with previous studies; however, it may be a result of unknown factors, such as region, variety, manufacturing process, or the presence of anti-nutritional factors.

The energy concentrations and nutrient composition in the canola meal determined were comparable with previous values [4,20,24,25]. In the present study, the average daily feed intake for the pigs fed the canola meal diet was the least among the pigs fed other diets. The glucosinolate which is an anti-nutritional factor in the canola meal may contribute to the low feed intake. It has been known that the dietary glucosinolates have an adverse effect on the feed intake for pigs [26]. The GE, DE, and ME in the copra meal used in this experiment were less than those reported in the literature [4,27]. In particular, the DE:GE ratio of copra meal was less in our study compared with the previous values. This reason may be that the NDF and ADF concentrations in the copra meal used in this experiment were greater than those used previously, and a relatively large proportion of non-starch polysaccharides, such as mannans, may have been present in copra meal [28], which can be an anti-nutritional factor. The concentrations and digestibility of energy in the corn germ meal were within the range of previous values [2,4,21,23,29]. The GE, DE, and ME in the palm kernel expellers were also within the range of previous studies [2,4,5,27].

The DE and ME in the PM and TDDG for pigs have not been reported. The DE:GE ratios of PM and TDDG were considerably less than those of other test ingredients. However, the CP concentration in the PM and TDDG was relatively greater than that in corn, and the CP concentration in the PM was fairly comparable to the CP in the SBM. Therefore, the PM and TDDG would be good alternative ingredients if studies are conducted to improve the energy efficiency of PM and TDDG. Further research is needed to determine the amino acid composition and digestibility of PM and TDDG.

In this study, there was a negative correlation between the fiber and DE concentration in the test ingredients, which agree with previous studies [30]. Although the most suitable equation for the ME was equation 1 considering the root mean square error, R², and p-value for the model, the CP as the independent variable was excluded because no significant correlation was found between the ME and CP. In a previous study [3], the IVDMD was highly correlated with energy digestibility in an in vivo experiment, and was a good predictor to estimate energy digestibility. A strong relationship between the energy digestibility and IVDMD was also observed in this experiment.

In conclusion, the three sources of SBM had greater energy concentrations than that in most of the byproduct feed ingredients and had greater energy digestibility than that in other byproduct feed ingredients fed to growing pigs. The ash and NDF were useful for estimating energy concentrations in the byproduct feed ingredients. The IVDMD was also useful to estimate energy digestibility.

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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