Animal Nutrition 2 (2016) 357–360

Contents lists available at ScienceDirect

Animal Nutrition

journal homepage: http://www.keaipublishing.com/en/journals/aninu/

Original Research Article

Nutrient profile and digestibility of tubers and agro-industrial coproducts determined using an *in vitro* model of swine

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ARTICLE INFO

Article history:
Received 13 May 2016
Received in revised form 25 June 2016
Accepted 20 July 2016
Available online 26 July 2016

Keywords:
Alternative feedstuffs
Coproducts
In vitro digestibility
Swine
Tubers

ABSTRACT

Exploring and evaluating alternative feed ingredients to be used in swine diet is essential due to highly variable cost and limited availability of conventional feed ingredients. Tubers and agro-industrial coproducts could provide the basis for producing affordable swine feed. However, information on the nutritional value of these potential alternative feedstuffs is necessary while considering their use in swine feeding program. Four tubers (purple sweet potato [PSP], okinawan sweet potato, taro and cassava) and 3 coproducts (okara, wheat millrun [WMR] and barley brewers grain [BBG]) were analyzed for their proximate nutrients, starch, fibers and gross energy (GE) content. Two independent *in vitro* studies were carried out for tubers and coproducts to determine their nutrients digestibility using a 3-step enzymatic assay (which mimics the digestion occurring in the gastrointestinal tract of swine) with 9 replicates of each sample digested in 3 batches equally. All replicate samples were used to determine *in vitro* dry matter digestibility (IVDDM) while 2 replicates from each batch were used to determine *in vitro* GE digestibility (IVDGE). Among tubers, CP content was the highest in taro (8.8%) and the lowest in cassava (3.7%), while CP content among coproducts was the highest in okara (22.7%) and the lowest in WMR (11.8%). Ether extract content among tubers ranged from 1.1% to 2.8%. The GE content among tubers, ranged from 4,134 to 4,334 kcal/kg whereas among coproducts it ranged from 4,270 to 4,794 kcal/kg. Among tubers, IVDDM for PSP was significantly higher (86.8%, $P < 0.001$) than taro (70.3%). Among coproducts, IVDDM of okara (74.1%) was significantly higher ($P < 0.05$) than BBG (61.3%). In conclusion, both tubers and coproducts can be used as a partial substitute of conventional energy feedstuffs in swine diets as these are rich in GE and other nutrients and are fairly digestible.

1. Introduction

Market availability of conventional feedstuffs like corn, wheat and soybean meal (SBM) are quite variable and their price fluctuates depending on production and their demand for human and livestock consumption and biofuel production. Including alternative feedstuffs in the swine diets has the potential not only to support the swine industry economically but also enhancing the environmental sustainability. This is particularly true for places where the conventional feedstuffs cannot be grown or destined cost-effectively to swine feeding. In such case, alternative feeding systems need to be developed and evaluated, which can partially or completely replace the inclusion of conventional feedstuffs in swine diets. Potential alternative feedstuffs such as distillers dried grains with soluble (Avelar et al., 2010; Jha et al., 2013; Agyekum et al., 2014), oil seed cakes (Yin et al., 1994; Seneviratne et al., 2010), and wheat mill run (WMR) (Nortey et al., 2008; Jha et al., 2012) have been studied and are widely used in swine feeding program.

Tubers such as purple sweet potato (PSP), okinawan sweet potato (OSP), taro and cassava along with agro-industrial coproducts such as okara (coproduct from tofu making process), wheat mill run, and barley brewers grain (BBG) could together provide basis

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Peer review under responsibility of Chinese Association of Animal Science and Veterinary Medicine.

Production and Hosting by Elsevier on behalf of KeAi

http://dx.doi.org/10.1016/j.aninu.2016.07.004

2405-6545/

for producing more affordable locally manufactured feed for swine. Tubers are rich in starch and can serve as a good source of energy whereas coproducts are intermediate products with fair amount of energy and protein with high fiber content. Some of these agricultural products and coproducts are evaluated and used elsewhere. However, agro-climatic conditions and farming systems largely differ from location to location, which influences the nutritional profile of tubers and their utilization in animals. Additionally, processing conditions affect the nutritional value of coproducts. Thus, nutritional value of these tubers and coproducts from particular origin needs to be evaluated before being used in swine feed.

2. Materials and methods

2.1. Feed ingredients and nutritional analysis

Four tubers (PSP, OSP, cassava, and taro) and 3 coproducts (WMR, BBG, and okara) were obtained from a local market of Honolulu, HI, USA. All the tuber samples were chopped and dried in a hot air oven (135°C for 2 h) to get the dry matter. The dried samples were ground to pass through 1 mm screen using a laboratory Wiley mill (Thomas Scientific, Swedesboro, NJ, USA). The sub samples were further ground using 0.5 mm screen size for some nutritional analysis. Nutrient profile and *in vitro* digestibility of nutrients were determined using 1 mm size sample, while starch was determined using 0.5 mm size sample. Proximate analysis of the samples was conducted according to the Association of Official Analytical Chemists (AOAC) standard procedures (AOAC, 2006) with specific methods as follows: DM (135°C for 2 h, method 930.15), ash (method 942.05), CP using a LECO analyzer (LECO CN-2000, Leco Corp., St. Joseph, MI, USA), (method 976.05, CP = N × 6.25). Acid detergent fiber (ADF, method 973.18) and neutral detergent fiber (NDF, method 2002.04) was determined using Ankom$_{200}$ Fiber Analyzer (Ankom Technology, Macedon, NY, USA). Heat stable α-amylase and sodium sulfite was used for NDF determination. Total starch content (method 996.11) was determined using a commercial test kits (Megazyme International Ltd., Ireland, UK). Gross energy content was determined using an oxygen bomb calorimeter (Parr Isoperibol Bomb Calorimeter 6200, Parr Instrument Co., Moline, IL, USA).

2.2. In vitro digestion

Three steps *in vitro* enzymatic digestion technique (Boisen and Fernández, 1997) was used to determine the apparent total tract digestibility of DM and GE in swine. Briefly, 1 g of sample was weighed in a 250 mL conical flask. Then, 50 mL of phosphate buffer solution 1 (0.1 mol/L, pH 6.0) was added to the flask followed by 20 mL of HCl solution (0.2 mol/L). The pH was adjusted to 2.0 by mixing with 1 mol/L HCl or 1 mol/L NaOH. One milliliter of chloramphenicol (Sigma C-0378, Sigma–Aldrich Corp., St. Louis, MO, USA) solution was added to prevent bacterial growth, which might take place during hydrolysis. Two milliliter of freshly prepared pepsin (Sigma P-0609, 800–2,500 units/mg, Sigma–Aldrich Corp., St. Louis, MO, USA) solution was added to the flask. The pepsin solution was made by mixing 0.75 g of pepsin to 30 mL of ultra-pure water. The flask was then closed with a rubber stopper and incubated in a water bath at 39°C under gentle agitation (50 rpm) for 2 h.

The 20 mL phosphate buffer solution 2 (0.2 mol/L, pH 6.8) and 10 mL of 0.6 mol/L NaOH was then added to the solution in the flask. The pH was adjusted to 6.8 with 1 mol/L HCl or 1 mol/L NaOH. Six milliliter of fresh pancreatin solution, made by mixing 3 g of pancreatin (Sigma P-1750, Sigma–Aldrich Corp., St. Louis, MO, USA) to 90 mL of ultrapure water, was added, and hydrolysis was continued under the same conditions for 4 h.

At the end of the of incubation, 20 mL of a 0.2 mol/L EDTA solution was added to the flask, and the pH was adjusted to 4.8 with a 30% acetic acid solution. Then 1 mL of Viscozyme (a multienzyme complex obtained from *Aspergillus aculeatus* containing cellulase, β-glucanase, arabinase, xylanase, mannanase, and pectinase; Novozymes, Bagsvaerd, Denmark) was added and the flask was incubated under same conditions for 18 h.

The undigested residue was then collected in a filtration unit using a porcelain filtration funnel lined with pre-weighed filter paper (Whatman no. 54; Whatman Inc., Florham Park, NJ, USA). All the material was transferred with double distilled water to the funnel. The residue, along with the filter paper, was dried overnight at 80°C and weighed the next day.

Two independent studies of tubers and coproducts were carried out. The experimental scheme for the *in vitro* digestion was as follows:

4 tubers × 3 bottles repeated over 3 batches;
3 agro-industrial coproducts × 3 bottles repeated over 3 batches.

2.3. Calculations and statistical analyses

All replicate samples were used to determine *in vitro* dry matter digestibility (IVDDM) while 2 replicates from each batch were used to determine *in vitro* GE digestibility (IVDGE). The IVDDM and IVDGE were calculated as follows:

$$\mathrm{IVDDM} = (\mathrm{DWH} - \mathrm{DWR})/\mathrm{DWH} \times 100,$$

where DWH = Dry weight of sample before hydrolysis; DWR = Dry weight of residue.

$$\mathrm{IVDGE} = [(\text{g sample} \times \mathrm{DMs} \times \mathrm{GEs}) - (\text{g residue} \times \mathrm{GEr})]/(\text{g sample} \times \mathrm{DMs} \times \mathrm{GEs}),$$

where DMs = % Dry matter of sample; GEs = Gross energy of sample; GEr = Gross energy of residue.

The digestible energy (DE) and metabolizable energy (ME) were calculated using equation number 24 and 45, respectively (Noblet and Perez, 1993) and net energy (NE) using equation number 11 (Noblet et al., 1994).

$$\mathrm{DE} = 4,162 - 9.4 \times \mathrm{ash} + 2 \times \mathrm{CP} + 3.9 \times \mathrm{EE} - 2.7 \times \mathrm{hemicellulose} - 4.5 \times \mathrm{ADF};$$

$$\mathrm{ME} = 0.997 \times \mathrm{DE} - 0.68 \times \mathrm{CP} + 0.23 \times \mathrm{EE};$$

$$\mathrm{NE} = 2,875 + 4.38 \times \mathrm{EE} + 0.67 \times \mathrm{starch} - 5.5 \times \mathrm{ash} - 2.01 \times (\mathrm{NDF} - \mathrm{ADF}) - 4.02 \times \mathrm{ADF}.$$

The IVDDM and IVDGE of feedstuffs were compared using the MIXED procedure of SAS (SAS v9.2, SAS Institute Inc., Cary, NC, USA), where feedstuffs were treated as fixed factor and batch as random factor. Means were separated using the Tukey method using pdmix macro of SAS. Differences were considered significant if $P < 0.05$.

3. Results and discussion

As shown in Table 1, CP content among tubers ranged from 3.7% to 8.8%. Taro contained the highest amount of CP (8.8%) and cassava had the lowest amount of CP (3.7%). Among coproducts, okara had the highest CP content (22.7%) and WMR had the lowest CP content

Table 1
Nutrient profile of tubers and agro-industrial coproducts (% basis).

Feedstuff	DM	Ash	CP	EE	ADF	NDF	Hemi-cellulose	Starch	GE[1]	DE[1,2]	ME[1,2]	NE[1,3]
Purple sweet potato	94.8	2.0	4.8	2.8	5.7	8.0	2.3	47.0	4,134	4,135	4,120	2,882
Okinawa sweet potato	94.5	2.8	5.3	2.0	8.1	9.7	1.5	51.7	4,154	4,116	4,100	2,869
Taro	97.7	2.4	8.8	1.9	10.4	11.5	1.1	48.4	4,333	4,117	4,099	2,860
Cassava	89.3	4.1	3.7	1.1	6.5	11.3	4.8	60.9	4,193	4,095	4,080	2,863
Wheat millrun	96.7	1.8	11.8	4.1	24.2	35.0	10.8	56.4	4,794	4,049	4,029	2,803
Barley brewers grain	97.1	8.7	15.9	1.8	34.1	42.1	8.0	46.7	4,270	3,947	3,924	2,715
Okara	92.7	5.2	22.7	13.7	19.7	31.0	11.3	61.2	4,707	4,095	4,071	2,847

DM = dry matter; CP = crude protein; EE = ether extract; ADF = acid detergent fiber; NDF = neutral detergent fiber; GE = gross energy; DE = digestible energy; ME = metabolizable energy; NE = net energy.
[1] Values presented as kcal/kg.
[2] Digestible energy and ME were calculated using equation number 24 and 45, respectively (Noblet and Perez, 1993).
[3] Net energy was calculated using equation number 11 (Noblet et al., 1994).

(11.8%). The PSP had the highest amount of EE (2.8%) and cassava the lowest amount of EE (1.1%). The ADF content of tubers ranged from 5.7% to 10.4%, taro had the highest and PSP had the lowest. The ADF content of the coproducts ranged from 19.7% to 34.1% with the highest value for BBG and the lowest for okara. The NDF content of all the tubers ranged from 8.0% to 11.5% with the highest value for taro and the lowest for PSP. There was a very small difference in the NDF content of taro and cassava. Both ADF and NDF content were higher in taro, whereas, NDF content in coproducts ranged from 31.0% to 42.1%. Crude protein content of both PSP and OSP was slightly lower than that reported (6.4%) by Dominguez (1992). Okara has the potential to be used in the animal feed, as the CP content was fairly high. Although okara has almost half of the CP content of SBM, it remains more advantageous over cereal grains such as corn, which contains almost half or even lower CP in general. Okara can also be viewed as a potential source of energy along with protein because of their high fat content. This was also reflected by their GE content (4,707 kcal/kg) and starch content (61.2%). The ADF content of PSP in this study was similar to observed value by Dominguez (1992) who reported ADF content of PSP to be 5.5%. However, ADF content of OSP observed in this study was slightly higher than those previous studies.

Starch content of tubers ranged from 47.0% to 60.9% with cassava containing the highest and taro containing the lowest. Starch content of coproducts ranged from 46.7% to 56.4%. The GE content of tubers ranged from 4,134 to 4,333 kcal/kg, taro had the highest amount of energy and PSP had the lowest. Among coproducts, BBG had the lowest (4,270 kcal/kg) and WMR (4,794 kcal/kg) had the highest GE value. Among tubers, IVDDM was significantly higher in PSP (86.8%, $P < 0.001$) than taro (70.3%), others were in between (Table 2). Among coproducts, IVDDM was significantly higher in okara (74.1%, $P < 0.05$) compared with BBG (61.3%) (Table 3). Among tubers, IVDGE was significantly higher in PSP (87.5%, $P < 0.001$) than taro (64.9%), while other tubers were ranged between these two. Among coproducts, IVDGE was significantly higher in okara (66.2%, $P < 0.05$) compared with BBG (43.0%).

The IVDDM of PSP (86.8%) was the highest among all tested feedstuffs. This can be attributed to low NDF and ADF content in PSP. The ADF is inversely related to digestibility, i.e., higher the ADF content in the feedstuff, lower the digestibility observed. Lower digestibility of taro can be due to presence of relatively higher amount of fiber than other tubers. Difference between the NDF and ADF content was also found to be the least in taro, indicating least amount of hemicelluloses; hence, taro was the least digestible feedstuff among the tubers as hemicelluloses are relatively better utilized than cellulose in the intestine of pig. Cassava had higher amount of hemicellulose (4.7%), hence, its IVDDM was higher than that of taro. Lower digestibility in taro might be due to inclusion of taro without peeling the outermost skin as higher amount of fiber is present on its outer skin.

Although energy content in taro was higher than other tubers studied, the amount of energy which is actually utilized in the body of swine is very less, as IVDGE was very low. Energy content in PSP is highly digestible and only low amount of PSP will be sufficient to meet a particular energy requirement of swine; however large amount of taro will be required to meet the same energy requirement as its energy digestibility is lower. Relative growth capacity of young piglets is high but their voluntary feed intake capacity is low, thereby, PSP can serve as potential energy dense diet for piglets. On the other hand, energy requirement of sows in the gestation period is low but they are still capable of eating more than they actually need. For such sows, taro having low energy digestibility can serve as potential source of energy as the actual amount of energy digested will be low. Starch content in cassava was high while taro had the least amount of starch. High amount of fiber and low amount of starch can be the limiting factor in inclusion of the taro,

Table 2
In vitro dry matter and gross energy digestibility of tubers.

Variable	Sample replicate	Purple sweet potato	Okinawan sweet potato	Taro	Cassava	SEM	*P*-value
Dry matter	9	86.8[a]	81.6[b]	70.3[c]	82.1[b]	0.010	<0.0001
Gross energy	6	87.5[a]	82.3[b]	64.9[c]	83.1[b]	0.011	<0.0001

[a,b,c] Means with different superscripts within the columns are significantly different ($P < 0.05$).

Table 3
In vitro dry matter and gross energy digestibility of coproducts.

Variable	Sample replicate	Wheat mill run	Barley brewers grain	Okara	SEM	*P*-value
Dry matter	9	69.9[b]	61.3[c]	74.1[a]	0.009	<0.0001
Gross energy	6	53.0[b]	43.0[c]	66.2[a]	0.015	<0.0001

[a,b,c] Means with different superscripts within the columns are significantly different ($P < 0.05$).

especially in young piglets. However, it can serve as an important source of energy feedstuff in places where it is largely produced and its digestibility can be increased by peeling off the outermost skin which is more fibrous.

The IVDDM of okara was fairly high (74.1%), which implies that 74% of nutrients in okara will be available to pigs upon its ingestion. The DE content of okara in this study was higher than SBM and expeller pressed canola meal (3,919 and 3,752 kcal/kg, respectively) (Seneviratne et al., 2010). Similarly, NE value of okara was higher than SBM (2,210 kcal/kg) and expeller pressed canola meal (2,550 kcal/kg; Seneviratne et al., 2010). This higher DE and NE value of okara may have resulted due to the higher EE content. Efficiency of energy utilization is decreased by presence of high amount of fiber. However, presence of higher amount of fat might have counteracted the negative effect of fiber. Each 1% increase in dietary fat causes an increase in the average daily gain by 2% in grower pigs and 1% in finisher pigs (DeRouchey, 2007).

4. Conclusions

Tubers and coproducts, both are rich in energy and other nutrients. Both IVDDM and IVDGE vary among tubers. The PSP had the highest *in vitro* digestibility of all nutrients, while taro had the lowest. Cassava and okinawan sweet potato had similar *in vitro* DM and GE digestibility. Coproducts are rich in fiber, protein as well as other nutrients. The IVDDM was higher in okara, whereas IVDDM of BBG and WMR were almost the same. Tubers can be used as partial substitute of common energy ingredients in pig diets, especially for subsistent farming system where these products are grown and are widely available whereas coproducts can also be used to replace traditional feed ingredients to some extent and can serve as potential source of protein as well as energy. All the tubers and coproducts studied showed a potential to be used in swine diet but these feedstuffs need to be subjected to animal trials to have a better idea about *in vivo* digestibility, inclusion percentage and palatability or the voluntary feed intake by animals.

Conflict of interest statement

We certify that there is no conflict of interests with any financial, professional or personal that might have influenced the performance or presentation of the work described in this manuscript.

Acknowledgements

This work was supported by the USDA, National Institute for Food and Agriculture, Hatch Project HAW02030-H, managed by the College of Tropical Agriculture and Human Resources, University of Hawaii at Manoa. Authors acknowledge Amit K. Singh for his help in lab works.

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