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Original Article

Effects of mangosteen wood vinegar as a potential additive on nutrient digestibility in growing pigs

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Abstract

The aim of the experiment was to determine the effects of mangosteen wood vinegar as a potential additive on nutrient digestibility in growing pigs. A 4x4 Latin square design was used in this study. Four crossbred (Duroc x Landrace x Large White) barrows averaging 19.41±0.81 kg in body weight were allotted 4 diets, mixed with mangosteen wood vinegar at levels of 0, 0.2, 0.4, and 0.8%, respectively. The pigs were raised in individual metabolism crates. Feces and urine samples were collected 4 times a day for 5 days for data collection. The results showed that the apparent nutrient digestibility of dry matter, ether extract, crude fiber, ash, nitrogen-free extract, apparent biological value, and digestible energy were not significantly different (P>0.05) in pigs fed with different diets. However, pigs fed with the diets mixed with mangosteen wood vinegar at levels of 0.2, 0.4, and 0.8%, respectively had significantly (P<0.05) higher digestible crude protein than pigs fed with the control diet and pigs fed with the diets mixed with mangosteen wood vinegar at levels 0.4 and 0.8% had significantly (P<0.05) higher metabolizable energy than pigs fed with the control diet. Moreover, Pearson's correlation coefficients with increasing levels of mangosteen wood vinegar showed a positive correlation at their apparent nutrient, energy digestibility, and protein quality evaluation. In conclusion, our results suggest that mangosteen wood vinegar could be used as a potential additive in increasing digestibility efficiency in growing pigs.

Keywords: mangosteen wood vinegar, potential additive, nutrient digestibility, growing pigs

1. Introduction

Nowadays there are many agricultural products for consumers and herdsmen who are interested in treating organic animals and seriously want to reduce the use of chemicals and antibiotics. Long term use of antibiotics can cause microorganisms to become resistant to antibiotics and chemicals can be contaminants in animal production. Acidic feed additives, in particular organic acids, are widely used in the animal industry in order to reduce microbial contamination of the animal feed. These additives impact the health of the animals (Izat *et al.,* 1990; Ruhnke *et al.,* 2015; Thompson & Hinton, 1997) and improve the food-producing animals by avoiding the use of antibiotics (Wang *et al.,* 2012). Organic acids are also known as effective preservatives which protect stored feeds against undesirable bacterial or fungal growth (Frank, 1994; Suiryanrayna & Ramana, 2015). Indirect effects of organic acids include reduction of gastric emptying rate, increased intestinal enzyme secretion and activity, additional energy source, improved protein digestion, increased mineral availability (Bühler, 2009; Kim *et al.,* 2005; Partanen 2001; Partanen & Mroz 1999; Ravindran & Kornegay 1993), and beneficial effects on swine performance (Mroz *et al.,* 2006; Upadhaya *et al.,* 2014; Wang *et al.,* 2009a).

Wood vinegar is the product obtained by distilling the smoke arising from burning wood and it is a complex mixture of 80-90% water and 10-20% organic compounds. Wood vinegar can be obtained from many different sources of raw materials such as wood and bamboo (Wititsiri, 2011).

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The major composition of wood vinegar is acetic acid (Rui *et al.,* 2014) and it also contains several phenolic compounds, such as guaiacol and cresol, and organic acids like acetic, formic, and propionic acids (Choi *et al.,* 2009). Acetic acid and phenolic compounds contained in wood vinegar have been reported to be anti-germination agents (Wang *et al.,* 2012; Yatagai *et al.,* 2002). Supplementation with bamboo liquids improved the growth performance of ducks and pigs (Kook *et al.,* 2002a, 2002b; Yan *et al.,* 2012). Choi *et al.* (2009) reported that wood vinegar improved the performance of weanling pigs by improving the nutrient digestibility and reducing harmful intestinal problems. Similarly, a study by Wang *et al.* (2012) reported that bamboo vinegar in the feed exerts an impact on the fecal bacterial community of piglets. In addition, Yan *et al.* (2012) reported that dietary bamboo vinegar supplementation increased growth performance and apparent total tract digestibility, along with its beneficial effect on the intestinal microbial population, meat quality, and fecal noxious gas concentrations. Although organic acid supplementation was initially targeted for weaned piglets, there is growing evidence that dietary acidification may also be beneficial for fattening pigs.

Previous studies showed that the burned woods could produce wood vinegar, but the chemical composition or quality of wood vinegar might have differences depending on the type of wood or the methods of obtaining the wood vinegar. On the other hand, sometimes the quality of wood vinegar from a commercial production might fluctuate due to factors in the process. In southern Thailand there are numerous types of wood, such as mangosteen tree wood with its large number of branches which is used to make wooden decorations. Mangosteen wood is also used to make charcoal. However, there has been no information on the potential effects of mangosteen wood vinegar on utilization of growing pig diets. Therefore, the object of this study was to evaluate the effects of mangosteen wood vinegar as a potential additive on nutrient digestibility of growing pigs.

2. Materials and Methods

The experimental protocols describing the management and care of animals were reviewed and approved by the Animal Care and Use Committee of the Institute of Animal for Scientific Purposes Development (National Research Council of Thailand, NRCT; Code Number. U1-01783-2558).

2.1 Preparation of mangosteen wood vinegar

Wood vinegar from the branch waste of mangosteen wood *(Garcinia mangostana* Linn*)* was produced through collaborations with farmers in the village of Cherewong, Ampher Lansaka, Nakhon Sri Thammarat Province, Thailand. The feedstock was introduced into a 200-liter fuel tank as a charcoal brazier equipped with a flame tunnel whose chimney was closed during the process of carbonization. The charcoal brazier was also equipped with a temperature gauge and sensors to indicate inside chamber and smoke outlet temperatures. The chamber was slowly heated up to 400 °C. The produced gas phase and smoke were condensed by a water-cooled condenser at the outlet temperature of 80-150 °C to give a brown liquid called bio-oil (Theapparat *et al.,* 2014). Bio-oil was kept in a closed container for protection from light and stored at an ambient temperature for six months. After phase separation was accomplished, a clear upper phase solution of wood vinegar was decanted. The sample was kept in a sealed vial for further studies.

2.2 Evaluation of the physicochemical characteristics of wood vinegar

The crude wood vinegars were centrifuged at 5,000 revolutions per minute for 20 min and then filtered through a 0.45 µm×13 mm nylon syringe filter to remove clear char particles. The acidity was measured using a pH meter (Mettler PEP 20, Greifensee, Switzerland) calibrated in standard buffer solutions (pH 4.0, 7.0, and 10.0). The total acid content (TAC) was indicated following Oasmaa *et al.* (2010). The amount of potassium hydroxide (KOH) in milligrams needed to neutrallize the acids in 1 g of wood vinegar using Autotitrator (Mettler T50, Greifensee, Switzerland) was evaluated and calculated using the following equation:

$$\text{TAN (mg of KOH g}^{-1}) = (\text{EP}n - \text{C31}) \times \text{C01} \times \text{C03/C00}$$

where EP*n* is the consumption of KOH at the final equivalent point (ml), C00 is the weight of the sample (g), C01 is 0.1 (concentration of the titrant, mol/l), C03 is 56.106 [M (KOH) in g/mol], and C31 is the consumption in zero titration (ml). A Karl Fischer titrator (Mettler DL18, Greifensee, Switzerland) was used to determine the amount of water in the wood vinegar samples. The titrator was calibrated with dry methanol. A wood vinegar sample was dropped in a container and titrated with the Karl Fischer reagent until the end point was reached. The results were the mean water content ± the relative standard deviation of three different measurements of each sample. Identification of the chemical compositions, a gas chromatograph (model 5890; Hewlett-Packard; Palo Alto, CA, USA) equipped with a mass spectrometer selective detector 5972 (GC/MS) was employed according to Theapparat *et al.* (2014). The conditions for gas chromategraphy (GC) analysis were as follows: HP-INNOWAX capillary column (60 m×0.5 mm internal diameter, 0.25 µm film thickness), carrier gas, helium at 2.0 ml min^{-1} flow rate, and splitless injection mode at 230 °C. The oven temperature profile was established as follows: initial 62 °C hold time of 6 min, 62-115 °C at 10 °C per min, 115-215 °C at 3 °C per min, with the final hold time of 15 min resulting in a total run time of 55.46 min. The volume of sample injected was approximately 1 µl. Chemstation software (Rev. A06.03[509]; Agilent Technology, Palo Alto, CA, USA) was used to control the operation of the GC system as well as data acquisition and the analysis of the chromatograms. Mass spectra were recorded at an ionization energy of 70 eV. The components were identified by comparing their mass spectra with those in the commercial mass spectral library database (Version 275; Wiley, NY, USA). The results were accepted when the constituents were identified with a match percentage >90%. For some components that had a very low response, co-elution by standards was used to confirm these components.

2.3 Experimental design and data collection

The four (Duroc x Landrauce x Large White) barrows, with an average weight of 19.41±0.81 kg, were randomly allotted into a 4x4 Latin square design. All pigs were individually housed in metabolic crates. Each metabolic crate was constructed in 50x120x75 cm sizes and had floors made with strong slatted plastic and had a trapezoid stainless steel sheet placed below each crate in which urine flowed through a stainless steel funnel to a plastic container covered with a clean filter cloth for contamination protection. The composition of the experimental diets was shown in Table 1. The 4 experimental diets were mixed with mangosteen wood vinegar at levels of 0, 0.2, 0.4, and 0.8%. The nutrient requirement for pigs weighing 20 to 60 kg was computed at a level sufficient to meet or exceed the nutrient requirements recommended by the National Research Council (1998 and 1988).

During the first 3 days, the individual pigs were housed in the metabolic crates to adapt to the metabolic crate and achieve a feed intake of approximately 4-5% of the pigs' body weight. After the initial adaptation period, each experimental period consisted of 7 days of experimental diet acclimation (*ad libitum*) which had ratios of prior and new diet (%) as follows: day 1 (80:20); day 2 (60:40); day 3 (40:60); day 4 (20:80); day 5 (100); day 6 (100); day 7 (100), and a 5–day collection period. Each experimental pig was weighed at the initial weight of each collection period. Feed allowance was 80-90% of total feed intake in the preliminary period. The first morning of the initiation of the 5-day collection period, 1.0% Cr_2O_3 was mixed with experimental diets as a marker to detect the beginning of fecal collection (Prawit *et al.,* 2017). Drinking water was available at 2.5 times that of feed intake.

All collected feces were weighed separately every day, in the morning (8:00 am) and in the evening (4:00 pm). Twenty percent of the sample was kept in plastic bags containing 10 ml of 10% formalin to prevent bacteria growth and stored at −20 °C. The collection buckets under the metabolic crates for the excreted urine contained 25 ml of 25% H_2SO_4 to prevent N losses through ammonia evaporation. The volume of excreted urine was weighed and 10% of the daily urine volume was kept and stored at −20 °C. One percent Cr_2O_3 was given again on the last morning of the end of the 5-day period to indicate when to terminate the fecal collection. At the end of the collection period, we returned to the 7-day experimental diet acclimation (*ad libitum*) and began the next collection period in which the experimental diets were not repeatedly circulated to each pig that would obtain the same 4 experimental diets. Prior to chemical analyses, the fecal samples were dried in a forced-draft oven at 55 °C and ground to pass through a 1-mm screen. Dry matter and nutrients digestibility (%) evaluations were conducted according to the methods described by Banerjee (1978). Apparent biological values were conducted according to the methods described by Pellet and Young (1980) and the digestible and metabolizable energy were determined using the following equations by Matterson *et al.* (1965); Silveira *et al.* (2015).

2.4 Chemical analysis

The dry matter, crude protein, ether extract, crude fiber, and ash of the experimental diets and feces samples were determined according to the methods of Association of Official Analytical Chemists (2000). Gross energy contents of

Table 1. Composition and chemical analysis of experimental diets for growing pigs (20-60 kg) (% as fed basis).

Ingredients (%)		Diet 1	Diet 2	Diet 3	Diet 4
Broken rice		43.24	46.58	49.91	56.60
Corn meal		30.14	26.52	22.90	15.65
Fish meal (55% crude protein)		8.87	8.87	8.87	8.87
Soybean meal (44% crude protein)		16.44	16.52	16.61	16.77
Dicalcium phosphate		0.31	0.31	0.31	0.31
L-lysine		0.20	0.20	0.20	0.20
Vit-Min-premix[1]		0.60	0.60	0.60	0.60
Salt		0.20	0.20	0.20	0.20
Mangosteen wood vinegar		-	0.20	0.40	0.80
Total		100.00	100.00	100.00	100.00
Calculated nutrients (%)	(NRC)				
Dry matter	-	88.39	88.23	88.08	87.76
Crude protein	18.00[3]	18.00	18.00	18.00	18.00
Ether extract	-	2.77	2.68	2.59	2.41
Crude fiber	-	2.98	2.93	2.88	2.77
Calcium	0.74[2]	0.74	0.74	0.74	0.74
Avail. Phosphorus	0.33[2]	0.37	0.37	0.37	0.37
Total Lysine	0.95[3]	1.13	1.13	1.13	0.14
Total Methionine + Cysteine	0.54[3]	0.62	0.62	0.62	0.62
Total Tryptophan	0.17[3]	0.19	0.19	0.19	0.19
Total Threonine	0.61[3]	0.68	0.68	0.68	0.68
Digestible energy (kcal/kg)	3,400.00[3]	-	-	-	-
Metabolizable energy (kcal/kg)	3,265.00[3]	3,265.00	3,265.00	3,265.00	3,265.00
Analyzed composition (%)					
Dry matter		87.85	87.68	87.55	87.38
Crude protein		17.11	17.29	17.61	17.47
Ether extract		2.70	2.55	2.47	2.36
Crude fiber		3.03	2.82	2.79	2.67
Ash		7.02	7.11	7.64	7.51
Nitrogen-free extract[4]		57.99	57.91	57.04	57.37
Gross energy (kcal/kg)		4,218.21	4,237.17	4,272.63	4,297.11

Note: [1] Each kg contains vit. A 800,000 IU, vit. D 80,000 IU , vit. E 3,000 IU, vit. K 700 mg, vit. B1100 mg, vit. B2 1,000 mg, pantothenic acid 5,000 mg, niacin 7,500 mg, choline chloride 27,000 mg, vit. B6100 mg, vit. B125 mg, biotin 16 mg, folic acid 33 mg, Fe 80 g, Zn 110 g, Cu 11 g, Mn 22 g, I 0.22 g, Se 0.18 g. and santoquin 0.5 g.
[2] NRC (1998).
[3] NRC (1988).
[4] Nitrogen-free extract = 100 – (%moisture + %ash + %ether extract + %crude fiber + %crude protein).

the experimental diets, feces, and urine samples were measured using an isoperibol bomb calorimeter (Leco AC-500).

2.5 Statistical analysis

The 4x4 Latin square experiments were statistically analyzed according to methods described by Steel and Torrie (1960). Duncan's multiple range test was used to compare the means of the treatments (Duncan, 1955). Variability in the data was expressed as the pooled standard error (SE) and a $P<0.05$ was considered to be statistically significant. Correlation between increasing levels of mangosteen wood vinegar and their apparent nutrient, energy digestibility, and protein quality evaluation were assessed by Pearson's correlation test using the CORR procedure and the level of significance was set at $P<0.05$. All data was computed using a computer program.

3. Results

3.1 Physicochemical property of mangosteen wood vinegar

The physicochemical properties of wood vinegar from the branch wastes of mangosteen *(Garcinia mangostana* Linn*)* were evaluated. The results showed that water was the main composition (86.90 %wt of wood vinegars) and the pH was 4.16. The TAC was 150.54±3.11 mg KOH g^{-1}. The 63 components of the crude mangosteen wood vinegars were identified by gas chromatography and mass spectrometry and expressed as a percentage of relative contents (Table 2). Analysis of the identified chemical constituents showed that organic acids (29.10%) and phenols (26.16%) were the major constituents. Alcohols (1.33%), furan derivatives (10.26 %), aldehydes and ketones (8.08%), heterocyclic aromatic compounds (2.64%), and other compounds (3.11%) were identified as minor constituents. Acetic acid was the largest content (23.22%) of the wood vinegar obtained from mangosteen branch biomass waste in this study. It has been suggested that acetic acid originated from the acetyl group in the hemicellulose under pyrolysis conditions (Kartal *et al.,* 2004). In addition, propionic acid and butanoic acid were also found in small amounts (4.11% and 1.62%, respectively). Methoxyphenols were the major proportion of the phenolic group in the mangosteen branch wood vinegar (17.70%). This was in accordance with another study which found that methoxyphenols were the major phenol group in wood vinegar from wood and bamboo (Theapparat *et al.,* 2014). Moreover, phenol derivatives were also found at a high concentration (8.46%). Phenolic constituents in wood vinegar are derived from the lignin component in plant biomass (Mohan *et al.,* 2006). Alcohol, aldehydes, ketones, and furans were also detected in small amounts from the pyrolysis reaction of cellulose and hemicellulose (Mohan *et al.,* 2006).

3.2 Nutrient, energy digestibility and protein quality evaluation

The results showed that the feed intake was between 805.00 to 812.50 g/day. Nutrient digestibility (%) of dry matter, ether extract, crude fiber, ash, nitrogen-free extract,

Table 2. Chemical composition of mangosteen wood vinegar.

Compound	Composition (%)
Organic acid	
Acetic acid	23.22
Propionic acid	4.11
Butanoic acid	1.62
2-butenoic acid	0.15
Total organic acids	29.10
Alcohols	
Methanol	1.33
Total alcohols	1.33
Furans	
2-methyl-furan	
Tetrahydro-2-furanmethanol	0.56
Dihydroxy-2(3H)-furanone	3.3
2,5-dihydro-3,5-dimethyl-2-furanone	0.41
2-furanmethanol	0.42
3-methyl-2(5H)-furanone	0.4
Tetrahydro-2H-pyran-2-one	0.48
3-hydroxy-2methyl-2H-pyran-4-one	0.83
Total furans	6.40
Phenol derivatives	
Phenol	5.88
3-methylphenol	1.39
4-methylphenol	1.19
Total phenol derivatives	8.46
Methoxyphenol derivatives	
2-methoxyphenol	2.79
2-methoxy-4-methylphenol	1.21
4-ethyl-2-methoxyphenol	0.82
2,6-dimethoxyphenol	9.69
4-methylsyringol	3.19
Total methoxy phenol derivatives	17.70
Total phenol derivatives	26.16
Aldehyde and ketones	
3-hydroxy-2-butanone	0.33
1-hydroxy-2-propanone	1.57
3-methyl-2-cyclopenten-1-one	1.13
2-ethylcyclohexanone	0.15
3,5-dimethyl cyclopentenolone	0.3
2-hydroxy-3methyl-2 cyclopenten-1-one	2.78
3,4-dimethyl-2-butenoic acid gamma lactone	0.41
2-ethyl-2-hydroxy-2cyclopenten-1-one	1.1
Acetic acid methyl ester	0.31
Total aldehydes and ketones	8.08
Heterocyclic aromatic compound	
Pyridine	0.37
2-methylpyridine	0.22
3-methoxypyridine	0.48
1-methylpiperidine	0.66
4-methyl-1H-imidazole	0.24
2-ethyl-1H-imidazole	0.21
2-methyl-3-pyridinol	0.15
3-pyridinol	0.31
Total heterocyclic aromatic compounds	2.64
Other compound	
Acetamide	0.65
N.N-dimethyl-methanamine	0.37
2,3,5-trimethoxytoluene	2.09
Total other compounds	3.11

apparent biological value, digestible energy, and metabolizable energy were not significantly different ($P>0.05$) in growing pigs fed with different diets. The results found that apparent digestibility (%) of dry matter, nitrogen-free extract, apparent biological value, and metabolizable energy tended to improve with increasing mangosteen wood vinegar levels (0, 0.2, 0.4, and 0.8, respectively). Apparent digestibility (%) of dry matter averaged 83.11, 86.97, 87.04, and 87.10% (0, 0.2, 0.4, and 0.8, respectively), nitrogen-free extract averaged 90.96, 93.11, 93.43, and 93.58% (0, 0.2, 0.4, and 0.8, respectively), apparent biological value averaged at 82.26, 84.89, 87.79, and 87.98% (0, 0.2, 0.4, and 0.8, respectively), and the metabolizable energy averages were 76.47, 78.57, 80.71, and 81.62% (0, 0.2, 0.4, and 0.8, respectively). Apparent digestibility (%) of ether extract, crude fiber, ash, and digestible energy was similar for all diets. Ether extract averaged 83.05, 84.17, 84.30, and 84.47% (0, 0.2, 0.4, and 0.8, respectively), crude fiber averaged 81.33, 81.48, 81.70, and 81.72% (0, 0.2, 0.4, and 0.8, respectively), ash averaged 83.00, 85.07, 86.60, and 86.81% (0, 0.2, 0.4, and 0.8, respectively), and digestible energy averaged 84.38, 84.11, 85.28, and 86.89% (0, 0.2, 0.4, and 0.8, respectively). However, the results of apparent digestibility (%) of crude protein found that growing pigs fed with mangosteen wood vinegar at every level had significantly (P<0.05) higher digestible crude protein than pigs fed with the control diet 89.67, 89.49, 89.01, and 84.05% (0.8, 0.4, 0.2, and 0, respectively). Also, the metabolizable energy of pigs fed with mangosteen wood vinegar at levels of 0.8 and 0.4% in the diets had significantly (P<0.05) higher metabolizable energy than pigs fed the control diet (3,507.35, 3,448.43, and 3225.71 kcal/kg) (Table 3). Increasing levels of mangosteen wood vinegar show that there was a very low positive correlation at dry matter, ether extract, crude fiber, ash, nitrogen-free extract digestibility, and digestible energy (r=0.35, 0.08, 0.05, 0.34, 0.28, and 0.19, respectively) but it appeared there was quite a moderate positive correlation at crude protein digestibility, apparent biological value, and metabolizable energy (r=0.50, 0.50, and 0.44, respectively). On the other hand a clear significant negative correlation was found between decreasing levels of fiber content. In particular, the apparent digestibility (%) of crude protein showed a statistically significant (r=-0.58; P<0.05) (Table 4).

Table 4. Pearson's correlation coefficients between increasing levels of mangosteen wood vinegar and decreasing levels of fiber content on their apparent nutrient and energy digestibility, and protein quality evaluation.

	Increasing levels of mangosteen wood vinegar	Decreasing levels of fiber content
Dry matter	0.35	- 0.45
Crude protein	0.50	- 0.58*
Ether extract	0.08	- 0.10
Crude fiber	0.05	- 0.05
Ash	0.34	- 0.37
Nitrogen-free extract	0.28	- 0.33
Apparent biological value	0.50	- 0.52
Digestible energy	0.19	- 0.15
Metabolizable energy	0.44	- 0.45

***** P<0.05.

4. Discussion

Mangosteen wood vinegar supplementation in growing pig diets could be anticipated to increase digestion of crude protein and metabolizable energy. Previous studies about wood vinegar utilization were mostly focused on antimicrobial activity. However, there were also studies showing that wood vinegar had positive achievements of supplementation in animal feed. Choi *et al.* (2009) reported that different levels of wood vinegar added to the diets as dietary treatments (0, 0.1, 0.2, and 0.3%) found that apparent fecal digestibility of dry matter, gross energy, and crude protein was significantly higher (P<0.05) in pigs fed the control diet and the performance of pigs fed wood vinegar was superior to those fed organic acid. Yan *et al.* (2012) found that the use 0.1 and 0.2% of bamboo vinegar led to a greater apparent total tract digestibility of dry matter and nitrogen (P<0.05) than pigs fed a control diet. This study observed that by increasing mangosteen wood vinegar at a level of 0.2, 0.4, and 0.8% (diet 2, diet 3, and diet 4, respectively), there would

Table 3. Effects of mangosteen wood vinegar supplementation on digestibility percentages of apparent nutrient, energy digestibility and protein quality evaluation in growing pigs (20 to 60 kg).

Items	Experimental diets				Mean	SEM	P-value
	Diet 1	Diet 2	Diet 3	Diet 4			
Feed intake (g/day)	805.00	810.00	812.50	812.50	810.00	-	-
Dry matter (%)	83.11	86.97	87.04	87.10	86.06	1.12	0.11
Crude protein (%)	84.05[b]	89.01[a]	89.49[a]	89.67[a]	88.06	1.19	0.04
Ether extract (%)	83.05	84.17	84.30	84.47	84.00	1.52	0.90
Crude fiber (%)	81.33	81.48	81.70	81.72	81.56	1.86	0.99
Ash (%)	83.00	85.07	86.60	86.81	85.37	2.44	0.68
Nitrogen-free extract (%)	90.96	93.11	93.43	93.58	93.77	1.01	0.28
Apparent biological value (%)	82.26	84.89	87.79	87.98	85.73	1.84	0.19
Digestible energy (%)	84.38	84.11	85.28	86.89	85.17	1.23	0.44
Metabolizable energy (%)	76.47	78.57	80.71	81.62	79.34	1.30	0.10
Digestible energy (kcal/kg)	3,559.28	3,563.91	3,643.60	3,733.64	3,625.11	52.88	0.16
Metabolizable energy (kcal/kg)	3,225.71[b]	3,329.53[ab]	3,448.43[a]	3,507.35[a]	3,377.76	55.78	0.04

[a, b] Means in the same row with different superscripts differ (P<0.05).

be high digestibility of crude protein due to the fact that chemical constituents of mangosteen wood vinegar in this study showed a high level of organic acids (29.10%), in particular, acetic acid (23.22%) and propionic acid (4.11%). Several studies reported previously that dietary acidification might be beneficial for the performance of pigs because organic acids consist of one proton and one anion. The effect of the proton of an organic acid is acidification of the feed while the anion inhibits the growth of microbes (Schutt, 2011). Another important effect is a low gastric pH that optimizes the pepsin activity and improves the digestibility of protein and decreases the rate of gastric emptying. Furthermore, it was reported that short-chain fatty acids have stimulatory effects on both endocrine and exocrine pancreatic secretions in pigs (Suiryanrayna & Ramana, 2015). Organic acids that stimulate exocrine pancreatic secretion of enzymes and bicarbonate improve protein and fat digestion (Lückstädt & Mellor, 2011). Also, organic acids enhance the apparent total tract digestibility and improve growth performance. It is concluded that organic acids and their salts increase the protein utilization especially in weaner pigs and improve the production indices (Suiryanrayna & Ramana, 2015). Moreover, Mosenthin *et al.* (1992), Mroz *et al.* (1997), and Suiryanrayna and Ramana (2015) reported that the apparent ileal digestibility of protein and amino acids improved in fattening pigs by the addition of organic acids, and acetic acid could influence nutrient digestibility by modulating the balance of intestinal microflora and pathogens in ovariectomized rats (Kishi *et al.,* 1999; Yan *et al.,* 2012). Likewise, this study observed that metabolizable energy, particularly at levels of 0.4 and 0.8% mangosteen wood vinegar, had significantly ($P<0.05$) higher metabolizable energy than pigs fed with a control diet. It might also be possible that increased crude protein digestibility and increased digestible energy could also increase metabolizable energy. Kirchgessner and Roth (1982) and Ravindran and Kornegay (1993) reported that metabolic reactions may be influenced by the addition of acidifiers and stimulatory effects of organic acids on intermediary metabolism resulting in improved energy or amino acid utilization or both.

5. Conclusions

The results of our study showed that mangosteen wood vinegar added to the diet of growing pigs improved the apparent nutrient digestibility. In particular, it increased the digestion of crude protein and metabolizable energy without impairing the digestibility of other nutrients. Therefore, our results suggest that mangosteen wood vinegar can be used as an alternative potential additive to increase the efficiency of digestibility in growing pigs.

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