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## Evaluation of agro-industrial by products as potential local feed for ruminant animals: volatile fatty acid and NH<sub>3</sub> concentration, gas production and methane emission

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# Evaluation of agro-industrial by products as potential local feed for ruminant animals: volatile fatty acid and NH<sub>3</sub> concentration, gas production and methane emission

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**Abstract.** Due to high cost of conventional feed, the use of agro-industrial by products has been widely known as alternative for animal feed. The aims of this experiment is to get information about the potency of agro-industrial by product in Aceh that can be utilized as feed for ruminant animals based on volatile fatty acid concentration, gas production and methane emission. Eight kinds of agro-industrial by product in Aceh from three different location products (sago residues, coconut meal, soybean-ketchup by product, coffee pulp, cacao pod, sago tree, corncob, and rice brand) were used in this study. Prior to analyses, all samples were dehydrated at the temperature of 60<sup>0</sup>C for 24 h, then crushed to pass a 1 mm sieve. Crushed samples were analysed to determine volatile fatty acid concentration, gas production and methane emission. Incubation was carried out in water bath with three replicates for 48 h with the temperature of 39<sup>0</sup>C. The production of gas was collected and periodically documented at 2, 4, 6, 8, 12, 24, 36 and 48 h after incubation by using a syringe. The VFA production from agro-industrial by products in this study was not significantly different ( $P>0.05$ ) from one by-products and others from  $83\pm 2.26$  mmol/l for cacao pod to  $123\pm 16.1$  mmol/l for coffee pulp. The production of NH<sub>3</sub> was with the range of 5.72 mmol/l to 15.71 mmol/l for for sago trees and coconut meal respectively. Total gas production from agro-industrial by products was 69.90 ml g/DM to 210.40 ml g/DM with methane gas production in the range of 33.88 – 35.18 ml/g DM after agro-industrial by products were incubated for 48 h. In conclusion, based on the fermentation parameters in this study, local agro-industrial by products in Aceh Province can be utilized as sources of alternative animal feed as replacement of conventional. Based on fermentation parameters measured in this study (VFA, NH<sub>3</sub>, gas production total) were still in optimal range for fermentation process in animal production

## 1. Introduction

Due to high cost of conventional feed, advances in industrial biotechnology can utilize agro-industrial by products as animal feed for ruminant animals. It has been well known that feedstuff's availability



for animal production in the worldwide was decreased due to altered function of land use and climate change [1]. Alternative feed resources, therefore, should be found out to fulfil the requirement of animals for production in the future.

Several studies have been published relating to utilization of agro-industrial products as alternatives animal feedstuffs for animal production [2, 3, 4]. Other advantages to use agro industrial by products as animal feed besides decrease cost of feed price are that to reduce pollution and to support zero waste production program. Ruminant animals were different from non-ruminant animals due to their ability to digest fibres containing agro by products from industries for their maintenance, growth, production and reproduction. Therefore, utilization of agro-industrial by products as potential feed for animals was promising.

The potency of Aceh Province to produce agro-industrial residues is well known with the rich of natural resources to be processed in agro industries. Several agro-industrial by product have been identified in Aceh Province such as sago residues, coconut meal, ketchup residues, coffee pulp, cacao pod, sago tree, corncob, and rice brand. The potency of agro-industrial residues as alternative feedstuffs have been reported [5, 6]. However, information relating the nutrient availabilities from agro-industrial products of local feedstuffs for animal production including digestibility was still limited. National Research Council (NRC) has been used as reference for feed formulation in Indonesia since long time ago. It should be consideration that chemical contents of feedstuffs were different from regions influencing by several factors such as environment, climate, soil condition and other factors [7].

Chemical composition have been used as indicator the feed quality for a long time ago. Other parameters such as volatile fatty acid concentration, gas production and methane emission should be considerate as indicator to determine feed quality. Cornell Net Carbohydrate Protein System (CNCPS) is one of the system to determine energy and protein requirement for ruminant based on several carbohydrate and protein fractions [8, 9]. The objectives of this experiments were to evaluate the potency of agro-industrial by products as ruminant feed based on volatile fatty acid concentration, gas production and methane emission.

## **2. Materials and Methods**

### *2.1. Sample Collection and Preparation*

Several agro-industrial by products found in Aceh Province such as sago residues, coconut meal, soybean-ketchup by product, coffee pulp, cacao pod, sago tree, corncob, and rice brand were collected from three different locations. Directly after collection, all samples were stored in freezer at the temperature of  $-20^{\circ}\text{C}$  for further analyse. Prior to analysis, samples from agro-industrial by products were dried for 24 h at the temperature of  $60^{\circ}\text{C}$ , then ground to pass a 1 mm sieve with hammer mill. Samples from eight agro-industrial by products from this study were used for further chemical analysis to determine the concentration of volatile fatty acid, the production of gas and the amount of methane emission.

### *2.2. Volatile Fatty Acid Concentration, Gas Production and Methane Emission*

The following is the procedure to determine the concentration of volatile fatty acid, the production of gas and the amount of methane emission. In vitro study applied the procedure of [10] in which samples were treated in vitro analysis by using the mixture of buffered rumen fluid. Rumen fluid for this study was collected from a rumen fistulated FH cow directly in the morning before feeding at Indonesian Animal Production Research Centre, Bogor. Prior to use, four layer of gauze were used to filter rumen fluid. Samples (0.75 g) was poured into a 125 ml serum bottles and added buffered rumen fluid (75ml) and ratio between fluid and buffer (1:4 v/v). Before incubation process, butyl rubber stoppers and aluminum crimp seals were used to seal the serum bottles. Temperature of incubation in the water bath was kept stable at  $39^{\circ}\text{C}$  for 48 h. The production of gas was collected and periodically recorded at 2, 4, 6, 8, 12, 24, 36 and 48 h after incubation by using a syringe. Determination of methane production was performed by using the method of [11]. Serum bottles

were centrifuged after 48 h incubation to separate between residue and supernatant. Supernatant gotten from 48 h incubation was used for further chemical analysis to determine total amount of VFA and the concentration of ammonia as described by [12].

### 2.3. Data analysis

All data from this study were statistically analyzed by using variance analysis. The model of the statistics is as follow:

$$Y_{ij} = \mu + \alpha_i + \beta_j + \varepsilon_{ij} \quad (1)$$

in which  $Y_{ij}$  is the observed value,  $\mu$  is the overall mean,  $\alpha_i$  is the treatment effect,  $\beta_j$  is the block

$$GP = b (1 - e^{-ct}) \quad (2)$$

in which GP is the production of gas (ml/g DM), b is the potency of gas production (ml/g DM), c is the rate of gas production (/h) and t is the period of incubation (h). Differences between treatments were stated, if  $P < 0.05$ . Duncan's multiple range test was applied for a post- hoc test. The statistical analysis was performed by using SPSS.

## 3. Results and Discussion

### 3.1 Volatile Fatty Acid and $NH_3$ Concentration

Total VFA (mmol/l) and  $NH_3$  (mmol/l) agro-industrial by products are presented in Table 1. The production of VFA in the rumen can be used as indicator and supplement information of feed quality since VFA production in the ruminants was used as source of energy. VFA consisting of acetate, propionate, and butyrate contributed approximately 70% of energy for ruminant animals [13]. In our study, the total of VFA produced from agro-industrial products after incubation from  $83 \pm 2.26$  mmol/l (cacao pod) to  $123 \pm 16.1$  mmol/l (coffee pulp). The production of VFA from agro-industrial by products in this study was not significantly different ( $P > 0.05$ ) from one by-products and others. The production of VFA in rumen was mostly influenced by pH [14]. Total VFA for optimal of microbial growth was 60-120 mmol/l (Waldron et al. 2002). This optimal amount of VFA was in agreement with our study. The study conducted by [15]. Oil palm fronds fermented by *Aspergillus niger* with different carbohydrate soluble produced total VFA 62.15-96.58 mmol/l. Research carried out by [16] to evaluate insects as potential feed ingredients for ruminant feed found that total VFA of insects as protein sources from 103-135 mmol/l.

**Table 1.** Volatile Fatty Acid (VFA) and  $NH_3$  concentration of agro-industrial by products as potential local feed for ruminant animals in Aceh Province after rumen fermentation.

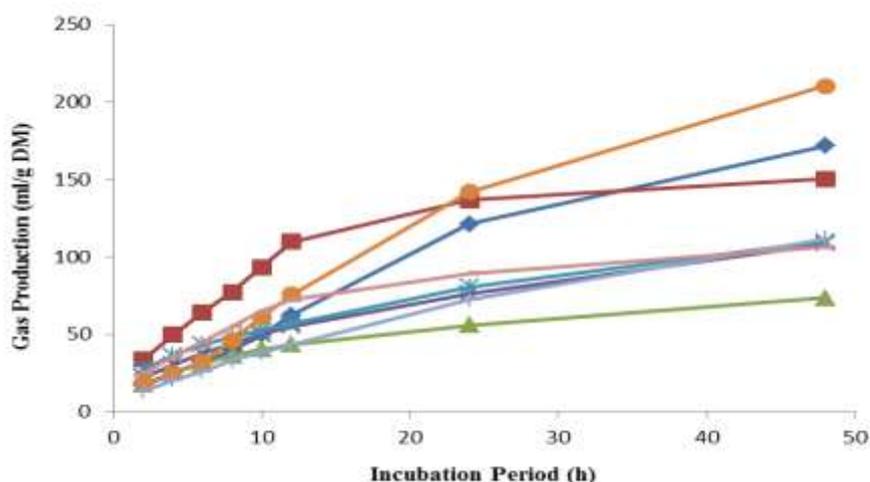
Agro-industrial by Products	VFA (mmol/l)	$NH_3$ (mmol/l)
Sago Residues	$102 \pm 20.8$	$7.73 \pm 0.84^{ab}$
Coconut Meal	$107 \pm 3.00$	$15.71 \pm 2.34^c$
Ketchup Residues	$92 \pm 3.19$	$14.41 \pm 0.67^c$
Coffe Pulp	$123 \pm 16.1$	$14.92 \pm 2.77^c$
Cacao Pod	$83 \pm 2.26$	$8.66 \pm 0.39^{ab}$
Sago Trees	$85 \pm 8.02$	$5.72 \pm 0.20^a$
Corn cob	$97 \pm 7.53$	$10.27 \pm 0.38^b$
Rice Bran	$108 \pm 9.34$	$13.93 \pm 2.55^c$
P – value	1.355	<0.001

Ruminant is able to use non protein nitrogen compounds to satisfy its protein requirement. Rumen bacteria required ammonia as main nitrogen source and protozoa needed N-amino as main nitrogen source. [17] stated that cellulolytic bacteria prefer to utilize ammonia as source of protein. Our study show that concentration of  $\text{NH}_3$  in vitro rumen fermentation (Table 1) was significantly different ( $P < 0.05$ ) amongst agro-industrial by products. The lowest was 5.72 mmol/l (sago trees) and the highest was 15.71 mmol/l (coconut meal). The concentration of  $\text{NH}_3$  in our study was still in the optimal range for animal production as stated by [18] the optimal production of  $\text{NH}_3$  concentration for animal production was 4-21 mmol/l Compared to the research conducted by [15] using oil palm fronds fermented by *Aspergillus niger*, our  $\text{NH}_3$  concentration was higher. [16] informed that insect produced relative high  $\text{NH}_3$  concentration with the amount of 53-88 mmol/l. Insect is one of alternative source protein for animal production as replacement of soybean.

### 3.2 Gas Production

It has been well known that rumen fermentation with the help of anaerobic microbe produced short-chain fatty acids (SCFA),  $\text{CO}_2$ ,  $\text{CH}_4$  and microbial. The concentration of gas which was produced can be used as indicator of acid produced during fermentation and the extent and rate of feed digestion [19]. *In vitro* gas production of agro industrial by products measured periodically up to 48 h is presented in Figure 1. In our study, ketchup residues produced the lowest gas production compared from other agro-industrial products with the amount of 69.90 ml g/DM and sago trees produced the highest gas production with the amount of 210.40 ml g/DM. Gas production was able to be used to predict organic matter digestibility. [20] stated that gas production can be used optimally to predict microbial efficiency if the limitation of batch culture gas production technique was recognized but the role of technique was not able to predict nutrient supply.

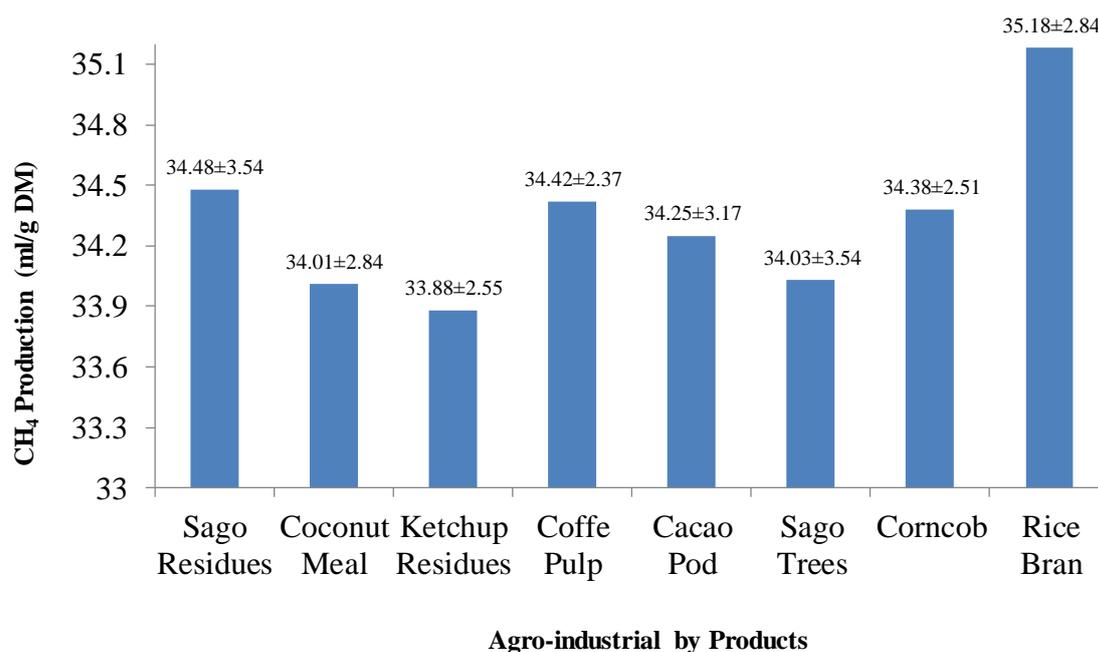
Research conducted [21] to evaluate 3 forage and 16 agro-industrial by products in Ethiopia concluded that based on chemical composition and gas production, some agro-industrial by products in Ethiopia can be utilized as source of feed metabolic energy and protein source and the presence of secondary compound, further study was required. However in our study, we did not measure secondary compound containing in agro-industrial by products in Aceh. Based on the figure, gas production went linearly at the incubation time of 4-14 h and slowly increased at the incubation time of 24-48 h. It happened because gas production total each feedstuffs was inversely proportional with improve of time incubation. [22] stated that the longer time incubation, the lower the fermented substrate.



**Figure 1.** Gas Production of agro-industrial products (◇- sago residues, -■- coconut meal, -Δ- ketchup residues, -x- coffe pulp, -Ж- cacao pod, -●- sago trees, -l- corncob, --- rice bran) as potential local feed for ruminant animals in Aceh Province after rumen fermentation.

### 3.3 Methane Emission

Methane production emitted from enteric fermentation of ruminant animals is one of the factors influencing greenhouse effect. [23] stated that there are several factors affecting methane production in ruminant animals such as intake level, type and feed quality and temperature in the surroundings. Figure 2 presents methane gas emissions of agro-industrial by products as potential local feed for ruminant animals in Aceh Province after rumen fermentation. The results of our study indicated that the methane production was not significantly difference amongst agro-industrial products incubation for 48 h with the range of 33,88 – 35.18 ml/g DM. Lipid was able to methane emission through several mechanisms as example reducing fermentation of organic matter and methanogen activities and protozoa number. The number of protozoa reduced along with the increasing of double bounding in long chain unsaturated fatty acid number [24]. [25] stated that methane production influenced by several factors such as animals, organic content of feed and environmental rumen. Research conducted by [16] indicated that insects produced lower methane compared with other feed sources such soybean meal. The production of methane in the study was about 10-27 ml/g DM at 12 h after incubation. Low methane production in the research due to the content of chitin in insects in which insects containing chitosan which was able to reduce methane emission and the population of methanogen [26].



**Figure 2.** Methane gas emissions of agro-industrial by products as potential local feed for ruminant animals in Aceh Province after rumen fermentation.

### 4. Conclusions

The results from our study concluded that fermentation parameters in this study indicated that local agro-industrial by products in Aceh Province can be used as animal feedstuffs as replacement of conventional feed. Based on fermentation parameters measured in this study, VFA, NH<sub>3</sub>, gas production total were still in optimal range for fermentation process in animal production

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