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Mesophilic batch anaerobic digestion from fruit fragments

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ABSTRACT

Fresh ripe and rotten fruits including oranges, mangosteen, bananas, and rambutan were separated into its fragments, i.e., peel, pulp, and seed in order to determine the rates and yield of their conversion into methane. Methane production from each of the components of the fruit was carried out under mesophilic conditions (35 °C) using 120 ml-glass serum bottles during 60 days of incubation. The effectiveness of the anaerobic digestion was expressed using the value of digestibility. The level of methane yield from the tested fruit fractions was in the order of seed > pulp > peel. The methane yields from the seed, pulp, and peel were in the range of 504.11 ± 21.15 to 657.89 ± 63.58 ml CH_4/g VS, 287.89 ± 38.79 to 468.91 ± 27.62 ml CH_4/g VS, and 0.00 ± 0.00 to 202.75 ± 40.86 ml CH_4/g VS, respectively. The highest digestibility was obtained from the anaerobic digestion of the seed of mangosteen, which was 99.3% and 99.4% from the fresh ripe and rotten mangosteen, respectively. The lowest digestibility was obtained from the mangosteen peel, which was 0.00%. The chemical composition, the presence of flavor compounds, and the physical structure of the fruit fragments affect the methane production.

1. Introduction

World production and trade of fresh ripe fruit have been growing steadily in the last decade. Food and Agricultural Organization of the United Nation (FAO UN) reported that in 2010, the global fresh ripe fruit production was 775 million tons. This number was an increase of 177 million tons over the 2000 base periods [1]. However, around 6.8% of the fruits were wasted during the post-harvest handling process, distribution, and consumption. Fruit is a perishable material and is easily degraded by microorganisms. Mechanical damage, physiological deterioration, and excessive ripeness can all accelerate biological degradation [2,3]. The most common fruit waste disposal practice, particularly in developing countries, is land filling, since it is cheap, easy, and needs little capital investment. However, landfill produces leachate, attracts vectors (e.g. insects, rodents, and birds) and emits greenhouse gases [4,5], which all hinder the future of landfill practices due to the environmental concerns.

Fruits have high moisture and organic contents and are readily biodegradable, therefore, anaerobic digestion is considered as a suitable method for fruit waste treatment [6,7]. Furthermore, anaerobic digestion represents an affordable low cost and low-technology system to supply biogas as a clean energy source [8,9]. Previous studies have shown that fruit wastes can be converted into biogas with reasonably high methane yields. Scano [2] reported that the methane yield from the fruit and vegetable waste that had 8.7% total solid (TS) and 86% volatile solid (VS) was 0.43 Nm^3/kg VS. In another study, the methane yields from sapodilla and pomegranate fruits were 0.327 m^3/kg VS and 0.342 m^3/kg VS, respectively [10]. In this study, four tropical fruits, e.g. oranges, mangosteen, bananas, and rambutan were used as feedstocks for the individually anaerobic digestion to determine the methane yield potential of each fruit. Oranges and bananas are among the major fruits in the world. FAO UN reported that in 2010, the world production of oranges and bananas reached 68.3 million tons and 106 million tons, respectively [1]. Mangosteen and rambutan are typically tropical fruits, which are produced in plenty of amounts in Indonesia. Mangosteen is a tropical evergreen tree and its edible portion is only about 40% of the whole fruit and the rest is peel [11]. Rambutan is a bright-red oval fruit, which has a seed, soft hairy peel, and a translucent aril. According to the Central Bureau Statistics of Indonesia, the productions of mangosteen and rambutan in Indonesia in 2010 were 84,538 tons and 522,852 tons, respectively [12].

In anaerobic digestion process of fruits, characteristics of the substrate, e.g., nutritional value, flavor compound, and physical

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properties of the fruits affect the high value production of methane [13]. In general, fruits have three major fractions, i.e., seed, pulp, and peel. Every single fraction of fruit has a different characteristic. For example, seed, as an ovary in the fruit development, has a high fat, protein, and carbohydrate content. On the other hand, peel contains fiber, e.g., pectin and cellulose. Pulp also contains high carbohydrate, vitamins, minerals, fiber, etc. Due to their nutritional value, seed, peel, and pulp of fruits can be used as a potential substrate for anaerobic digestion to produce methane.

Besides having the high nutritional value, which can be converted into methane using anaerobic digestion, another challenge comes from the substrate characteristics, i.e., flavor compound and physical properties. In nature, most plants are equipped with either physical or chemical defense against degradation by microbia. The physical barrier is the inherent properties of a native cell wall, which is very rigid and complex, making it resistant from enzymatic attack. It involves a sophisticated and integrated defense system combining crystalline cellulose in microfibrils, heteropolysaccharides, and lignin. This is in the case of a lignocellulosic substrate. Lignocellulosic material and lignin are degraded by anaerobic digestion slowly and incompletely [14,15]. Furthermore, several studies showed that some flavor compounds can inhibit the anaerobic digestion process [16,17]. Due to the complexities of the substrate, the fruits were fractionated into seed, pulp, and peel in order to investigate the methane production from each fraction.

2. Materials and methods

2.1. The fruits

Fresh ripe and rotten fruits, including “Pontianak” orange (*Citrus nobilis Lour. Var. microcarpa Hassk.*), mangosteen (*Garcinia mangostana* L.), bananas (*Musa paradisiaca* L.), and rambutan (*Nephelium lappaceum* L.) var. Binjai were used as substrates for the anaerobic digestion. The fruits were obtained from a local market in Yogyakarta. Both the fresh ripe and rotten fruits were peeled and separated into three parts, i.e., peel, pulp, and seed. Rotten fruits were prepared by incubating fresh ripe fruits in open space at room temperature for 4–7 days. The definition of fresh ripe fruits used in this work was fruits that were edible and had bright peel color. Whereas, the definition of rotten fruits is fruits that had damaged peel, moldy, bruised pulp, and easily crushed. Each part of the fruit was chopped and further homogenized using a blender. Thereafter, the samples were put in plastic bags and stored at −20 °C before use. A summary of the chemical compositions of the fresh ripe and rotten fruits is presented in Table 1.

2.2. Anaerobic digestion

Anaerobic digestion was carried out in batch operations using 120 ml-glass serum bottles under mesophilic conditions (35 °C), according to a method previously described by Hansen [18]. Active inoculum was obtained from the biogas digester (the Agricultural Training, Research and Development Station (ATRD), Universitas Gadjah Mada, Yogyakarta, Indonesia). Each bottle contained 0.60 g VS (Volatile Solid) of inoculum and 0.15 g VS of fresh ripe or rotten fruits and an addition of distilled water up to 30 ml. Hence, the ratio of organic content and volume is 0.025 g VS/ml. Blank samples were also prepared containing deionized water and inoculum. All these experiments were performed in triplicate, and the accumulated methane production was determined during 60 days.

2.3. Analytical methods

The total solid, volatile solid, and ash content were determined according to the methods no. 971.28 and no. 940.26 of AOAC by drying the samples using a convection drying oven at 105 °C to achieve a constant weight, and burning the samples at 575 °C [19]. Fat content was determined according to the AOAC method no.963.15. The fat content was extracted from the fruits by petroleum ether as a non-polar solution using a soxhlet extractor and then dried at 105 °C to achieve constant weight [19]. Total nitrogen content was determined according to the Micro-Kjedahl method. The determination of the total nitrogen content was followed by hydrolysis using sulfuric acid and a catalyzer (Na_2SO_4 and HgO (20:1)), distillation, and titration using 0.02 N HCl [19]. Hemicellulose and cellulose were determined according to the method described by Chesson using the thermogravimetric method [20]. In this method, dried blended fruits were hydrolyzed in a two-step acid hydrolysis process with 1 N sulfuric acid to hydrolyze the hemicellulose and then 72% sulfuric acid to hydrolyze the cellulose. After each hydrolysis step, the samples were filtered and dried at 105 °C to achieve a constant weight. In accordance with McKay et al. regarding modification [21], the total sugar and soluble starch were determined by hydrolyzing the samples using 6% HCl to get simple sugars. Then, the simple sugar was reacted with cupric sulfate and ammonium sulfate to form a sugar-copper ammonium sulfate complex. The absorbance of standard sugars in the working reagent was read at 310 nm.

Methane and carbon dioxide production and composition were analyzed using a gas chromatograph (Shimadzu GC-2010, Japan) equipped with a capillary column (RT-Q bond, 30 m, 0.32 nm ID, 10 μm DF, Restek Corporation, U.S.A.), and a thermal conductivity detector (TCD) (Shimadzu, Japan) with an inject temperature of 100 °C. The carrier gas used was helium, operated with a flow rate of 1.5 ml/min at 50 °C using a split ratio of 20. The initial methane production rate was measured as the mean of methane production per day during the first ten days. Analysis of Variance (ANOVA) with a significance level of 0.05 was used to analyze the significance of the means differences of each sample. This test was performed using the Statistical Package for the Social Sciences version 19 (SPSS).

3. Results and discussion

Four tropical fruits were selected as feedstocks for the individual anaerobic digestion, under mesophilic conditions during 60 days of incubation. In this study, the fruits were fractionated into its three components, i.e., peel, pulp, and seed in order to investigate the characteristic of each component and its effect on the methane production. Due to the different chemical and structure of each fraction, the methane production varied significantly.

3.1. Methane production of seed, pulp, and peel of fruits

3.1.1. Oranges

The methane production profile of the oranges is shown in Fig. 1a,b and Table 2. The highest initial methane production rates for both the fresh ripe and the rotten oranges were achieved by anaerobic digestion of the seeds, i.e., 37.59 ± 3.48 ml CH_4/g VS/day and 42.79 ± 2.57 ml CH_4/g VS/day, respectively. The initial methane production rate from the pulp was 27.37 ± 2.79 ml CH_4/g VS/day from the rotten orange and 21.93 ± 7.06 ml CH_4/g VS/day from the fresh ripe orange. Meanwhile, the methane production from the orange peels both fresh ripe and rotten, was very low, compared to that from the seeds and the pulp. No methane was produced during the first 10 days of the digestion. Methane began to be produced after 20 days of digestion (Fig. 1a,b). As shown in the figures, the methane production on the last days of incubation had reached almost a constant level, which means that the maximum methane