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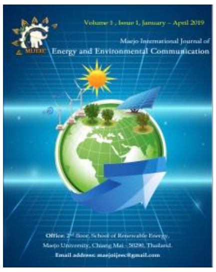

ARTICLE

Evaluation of mango, longan and lychee trees pruning leaves for the production of biogas via anaerobic fermentation

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ABSTRACT

Pruning fruit trees is improving their full health and harvest. Fruit tree pruning leaves waste to represent an abundant amount of organic materials, and these produced during a short period. The basic fuel properties of lignocellulosic biomass from orchards were evaluated on the following fruit tree leaves obtained from pruning operations. Biomass has become a vital source of renewable energy. Biogas is one the renewable energy which can be produced by anaerobic fermentation of biomass. In this study, mango, longan and lychee trees pruning leaves waste was utilized for biogas production. These leaves were examined on proximate analysis and ultimate analysis contents are considered as carbon (C), hydrogen (H), oxygen (O), nitrogen (N) and sulfur (S). In this study, pretreatment was performed using a sodium hydroxide solution (w/v) at different concentrations as above (1, 2, 3, and 4%). For the best feedstock screening, the theoretical biochemical methane potential was confirmed. Mango leaves biomethane content was higher compared to longan and lychee leaves. Finally, this biomass was suggested further large-scale studies. Digestate from biogas system is a highly valuable nutrient and rich fertilizer.

1. Introduction

Pruning is an important horticultural practice that generally promotes vegetative growth. However, the precise characterization of vegetative growth after pruning. After pruning there are lot of wastes are produced. These wastes are excellent biomass source. Biomass ensuing from pruning waste can be used as a source of bioenergy. Mango, longan and lychee are one of the main economic subtropical fruits and most important fruit crop in Thailand. These trees are well distributed Asian countries including Thailand. However, tree plantation produces large amount of wastes from its flowers and leaves which can be a source of biomass to produce biofuel or biogas. Therefore, studying the energy production potential of leaves are very important (Cybulska et al., 2015). This tree yields large amounts of pruning waste, as it has a high propagation capacity and very fast growth, for both the trunk and branches. Additionally, its location on the periphery of land plots facilitates the extraction and transportation of the residual biomass, as it can be done on the same locations as the pruning.

Utilization of agricultural waste to produces energy security and decreasing environment pollution. Also, possible to increase revenue for people in local community from these plants growing between output pending.

Moreover, biomass waste-to-energy conversion reduces greenhouse gas emissions in two ways. Heat and electrical energy are generated which reduces the dependence on power plants based on fossil fuels. The greenhouse gas emissions are significantly reduced by preventing methane emissions from landfills (Dussadee et al., 2014), (Dussadee et al., 2017), (Ennouri et al., 2016), (González-Fernández et al., 2011). Additionally, waste-to-energy plants are highly efficient in harnessing the untapped sources of energy from wastes. Biogas is generated from anaerobic digestion process through biodegradation of organic matter.

The main composition of biogas is 50-70% methane (CH_4), 30-40% carbon dioxide (CO_2), 5-10% hydrogen (H_2), 1-2% nitrogen (N_2), 100-3,000 ppm hydrogen sulphide (H_2S) and other. Methane is the main component of biogas as well as fuel. Biogas is considered as renewable energy and can be an alternative source of as electricity generation, and bio-methane gas production is useful for residential activities such as heating applications (APHA, 2005; Igoni et al., 2008; Govasmark et al., 2011; Haraldsen et al., 2011; Heviánková et al., 2013).

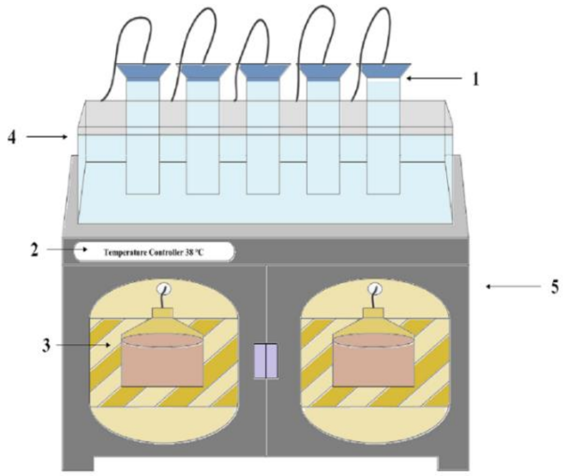

Figure 1. Schematic view of the experimental set up: 1) Gas measuring cylinder, 2) temperature controller, 3) digester, 4) Water bath 5) incubator box

The use of biogas as a renewable energy might be efficient and suitable for improving energy security and decreasing environmental disruption caused by carbon emissions. The properties of lignocellulosic biomass render it resistant to biodegradation. Due to the complexity and variability of biomass chemical structures, the optimal pretreatment method and conditions depend on the types of lignocellulose present (Page et al., 1982; Kirchmann et al., 1991; Metcalf et al., 1991; Pokój et al., 2015; Kinyua et al., 2016). Several structural and compositional properties were found to have impacts on the biodegradability of lignocellulosic biomass, including cellulose crystallinity, accessible surface area, degree of cellulose polymerization, presence of lignin and hemicellulose, and degree of hemicellulose acetylation (Ramaraj et al., 2016a,b,c; Rösch et al., 2013; Saha et al., 2013).

The goal of pretreatment is to alter such properties to improve biomass amenity to enzymes and microbes. These pretreatment methods can be divided into mechanical, thermal, chemical as well as, biological treatments or a combination of these techniques. Our effort is directed towards the possibilities of biogas production increase from mango, longan and lychee trees pruning leaves pre-treated by chemical method using sodium hydroxide. The aim of this work has been to assess the waste leaves obtained from pruning the mango, longan and lychee trees cultivated in the Chiang mai province, Thailand.

2. Methodology

This study was conducted to determine the optimal and efficient pre-treatment condition for enhanced biogas production. The sample was collected from mango, longan and lychee trees garden and was subjected to pretreatment and fermentation processes to produce biogas. The materials were collected from the Maejo university farm, and surround areas, Sansai, Chiang mai, Thailand. The leaves were crushed into small pieces by a grinding machine.

The collected samples were transferred to the lab of Energy Research Center, Maejo University. The experiment produces biogas by batch system using lab-scale digesters fabricated from 6 L water tank by connecting with cylinder 1,000 ml to gas collection, gas measuring and a feed inlet. It was sealed using a rubber stopper with a pipe to extract biogas as shown in Figure 1. Experiment was conducted simultaneously under mesophilic temperature at 37°C for 45 days. Each digester was manually mixed twice a day.

The samples were analyzed for total solids (TS), volatile solids (VS), chemical oxygen demand (COD) (APHA, 2005) and pH by standard methods. Elemental composition (C, H, N, O, and S) was analyzed using the element analyzer Perkin-Elmer. The moisture content of raw materials was determined following the procedure given in ASTM Standard D 4442-07. Effect of pretreatments leaves for biogas production by mono-digestion.

Samples were initially centrifuged at 10,000 rpm for 5 min and determined by the spectrophotometric method. The pH of the sample was adjusted to 4.3 using the CaCO3 and titrated against sulfuric acid following the method by Snoeyink and Jenkins (1980). Total fat, ash, moisture, fiber contents and volatile fatty acids (VFA) were determined using AOAC official method while the pH value was tested by a pH meter (B-711, Horiba, Japan) and the composition of biogas (CH_4, CO_2, and O_2) was measured using a biogas analyzer (BIOGAS 5000, Geotech). Calorific values were estimated according to Chuanchai and Ramaraj (2018).

3. Results and Discussion

Biogas production from the process of biodegradation of an organic matter by anaerobic bacteria in the absence state of oxygen. The product realization after of a process anaerobic digestion namely biogas. The most important biogas that can light a fire is CH_4. The typical biogas composition ranges, irrespective of substrate used in the AD process. The biochemical process of anaerobic digestion: the breakdown of complex organic matter in an anaerobic process involves multiple steps, which are carried out by several groups of microorganisms. The end product of anaerobic degradation of organic compounds is biogas, an energy-rich gas mixture consisting of mainly CH_4 and CO_2. The various steps and microbial groups involved during AD; and the examples of some different groups of extracellular enzymes. Each group contains several enzymes that are specialized in various substrates, such as different proteins. The rate of decomposition during the hydrolysis stage depends significantly on the nature of the substrate. The transformation of cellulose and hemicellulose generally takes place more slowly than the decomposition of proteins (Metcalf et al. 1991; Page et al. 1982; Pokój et al. 2015). Some of the important biochemical processes of anaerobic digestion are presented in the following sections.

Biogas production from process of biodegradation of an organic matter by anaerobic bacteria in the absence state of oxygen. The product realization after of a process anaerobic digestion namely biogas. The main composition of biogas is 50-70% methane (CH_4), 30-40% carbon dioxide (CO_2), 5-10% hydrogen (H_2), 1-2% nitrogen (N_2), 100-3,000 ppm hydrogen sulphide (H_2S) and other. The most important biogas that can light a fire is CH_4. The typical biogas composition ranges, irrespective of substrate used in the AD process, are noted in Table 1.

Table 1. Typical composition of biogas

Substance	Formula	Percentage (%)
Methane	CH_4	50-80
Carbon Dioxide	CO_2	20-50
Hydrogen	H_2	5-10
Nitrogen	N_2	1-2
Water Vapour	H_2O	0.3
Hydrogen Sulphide	H_2S	Traces

The breakdown of complex organic matter in an anaerobic process involves multiple steps, which are carried out by several groups of microorganisms. The end product of anaerobic degradation of organic compounds is biogas, an energy-rich gas mixture consisting of mainly CH_4 and CO_2. Figure 2 shows the schematics of various steps and microbial groups involved during AD. Contains examples of some different groups of extracellular enzymes. Each group contains several enzymes that are specialized in various substrates, such as different proteins. The rate of decomposition during the hydrolysis stage depends greatly on the nature of the substrate. The transformation of cellulose and hemicellulose generally takes place more slowly than the decomposition of proteins (Adekunle and Okolie, 2015). Some of the important biochemical process of anaerobic digestion are presented in the following sections.

Hydrolysis: Biodegradation from a complex organic compound become organic monomer by extracellular enzyme from anaerobic bacteria. The cleavage of chemical bonds by the addition of water such as carbohydrate polysaccharides (complex sugars) are broken down into monosaccharides. One example is the breakdown of lactose into galactose and glucose, Triglycerides are split into three fatty acids and glycerol by the addition of three water molecules, proteins, peptide bonds are broken to separate amino acids.

Acidogenesis: The Second step in the anaerobic digestion process. Acidogenesis is the volatile fatty acid production from acidogenic bacteria activities use organic monomer nutrients for growth. The acidogenic bacterial activity is organic monomer into volatilefatty acid molecule not more than 5 atoms such as acetic acid, formic acid, propionic acid, isobutyric acid, valeric acid, isovaleric acid, butyric acid. Acetogenesis: The third step in the anaerobic digestion process. Acetogenesis is an acetate and formate production process from acetogenic bacteria by acetogenic bacteria consume precursors and produce acetate and formate. The acetate and formate is an important component in to create CH_4. Methanogenesis: The final step of anaerobic digestion process. Methanogenesis is the formation of methane from methanogens bacteria by use an acetate and formate in the formation of methane.

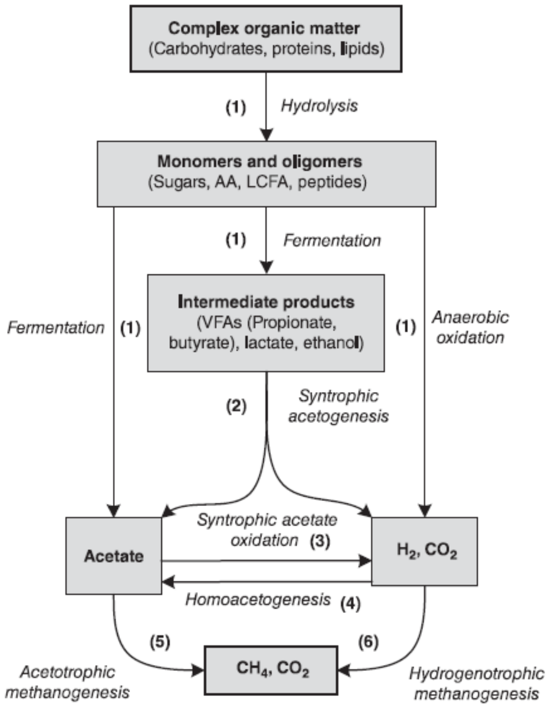

Figure 2 Stages of anaerobic digestion 1. Fermentative bacteria; 2. Acetogenic bacteria; 3. Syntrophic acetate oxidizing bacteria; 4. Homoacetogens; 5. Acetotrophic methanogens; 6. Hydrogenetrophic methanogens. Notes:

AA: amino acids; LCFA: long-chain fatty acids; VFAs: volatile fatty acids (Khanal, 2008).

Hydrolysis: Biodegradation from a complex organic compound become organic monomer by an extracellular enzyme from anaerobic bacteria. The cleavage of chemical bonds by the addition of water such as carbohydrate polysaccharides (complex sugars) are broken down into monosaccharides. One example is the breakdown of lactose into galactose and glucose, and Triglycerides are split into three fatty acids and glycerol by the addition of three water molecules, proteins, peptide bonds are broken to separate amino acids. Acidogenesis: The Second step in the anaerobic digestion process. Acidogenesis is the volatile fatty acid production from acidogenic bacteria activities use organic monomer nutrients for growth. The acidogenic bacterial activity is organic monomer into the volatile fatty acid molecule, not more than 5 atoms such as acetic acid, formic acid, propionic acid, isobutyric acid, valeric acid, isovaleric acid, butyric acid.

Table 2. Characteristics of mango, longan and lychee trees

Parameters	Mango leaves	longan leaves	lychee leaves
Proximate analysis (wt. %)			
Moisture (%)	10.78±0.50	8.65±0.20	9.88±0.35
Ash (%)	9.4±0.30	16.88±0.41	13.60±1.32
Ultimate analysis (wt. %)			
Carbon (%)	40.89±0.15	42.15±0.33	39.88±0.87
Hydrogen (%)	5.97±0.15	5.69±0.21	6.02±0.33
Nitrogen (%)	0.43±0.02	1.45±0.02	0.66±0.89
Sulfur (%)	0.46±0.03	1.02±0.01	1.67±1.33
Oxygen (%)	49±0.22	49.73±0.15	49.44±0.30
Biochemical analysis			
TS (%)	94±0.33	33.26±0.41	55.04±0.11
VS (%)	88±0.31	71.70±0.11	69.88±0.24
pH	8.1±0.22	7.82±0.42	7.56±0.10
COD (mg/L)	341±0.25	7564±0.51	6740±2.51
VFA (mg/L)	9200±0.53	8440±0.22	9301±1.43

Acetogenesis: The third step in the anaerobic digestion process. Acetogenesis is an acetate and formate production process from acetogenic bacteria by acetogenic bacteria consume precursors and produce acetate and formate. Acetate and formate is an essential component to create CH_4. Methanogenesis: The final step of the anaerobic digestion process. Methanogenesis is the formation of methane from methanogens bacteria by using acetate and formate in the formation of methane.

Since biogas production from an anaerobic digester consists the bacterial activity; thus factors biogas production depends on the ability of bacteria to thrive inside the digester such as temperature (Pokój et al., 2015; Ramaraj et al., 2016a,b,c; Rösch et al., 2013). Anaerobic processes, like most other biological processes, are strongly dependent on temperature. Although anaerobic microorganisms, especially methanogens, are viable at different temperatures, methanogens are accordingly classified as psychrophilic, mesophilic, and thermophilic. The anaerobic conversion rates generally increase with temperature up to 60°C.

There are three anaerobic digestions in temperature ranges; mesophilic digestion occurs between 25-45°C, psychrophilic digestion occurs at below 25°C or room temperature, and thermophilic digestion occurs above 45°C. Characteristics of mango, longan and lychee trees Table 2. The properties of lignocellulosic biomass render it resistant to biodegradation. Due to the complexity and variability of biomass chemical structures, the optimal pretreatment method and conditions depend on the types of lignocellulose present. Several structural and compositional properties were found to have impacts on the biodegradability of lignocellulosic biomass, including cellulose crystallinity, accessible surface area, degree of cellulose polymerization, presence of lignin and hemicellulose, and degree of hemicellulose acetylation (Van Soest et al., 1991; Tang et al., 2011; Saha et al., 2013; Teghammar et al., 2013; Vázquez-Rowe et al., 2015; Tsai et al., 2016; Unpaprom et al., 2017; Vu et al., 2018; Tran et al., 2019; Manmai et al., 2020). The goal of pretreatment is to alter such properties to improve biomass amenity to enzymes and microbes.

These pretreatment methods can be divided into mechanical, thermal, chemical as well as, biological treatments or a combination of these techniques. Co-digestion is yet another method used to enhance biogas production. This method entails planning loading of the digester so that an advantageous blend of different substrates serves as organic load. The effects of different pretreatment techniques on the chemical composition and physical characteristics of lignocellulosic biomass (Miller, 1959; Orth et al., 1993; Zhang et al., 2008; Yan et al., 2016). Accordingly, the results of this study verified that high-calorific biogas was obtained in this study system after methane was enriched through biological biogas purification. This study investigated the potential of leaves as a feedstock for biogas production. The results indicated that leaves contained rich amount of organic substances and these substances are suitable to use in anaerobic fermentation process to sustain microbial life and transform nutrients into biogas. It was found out that leaves and pig manure fermentation can produce as much as 74 L of biogas a day. In the laboratory scale part, 60.7% concentrated CH_4 was generated. While the scale up part, biogas production by leaves with by 2%NaOH for pretreatment yielded a 3,325 L of biogas with a methane concentration of 68.1%. Additionally, it was observed that the biogas production can be enhanced in a biogas digester when the substrate temperature was increased in 37°C for 8 h. Moreover, CO_2 removal in the biogas was found to be efficient through the use of different concentrations of NaOH (1, 2, and 3%), which resulted to the purification of methane to 78% from 68.10%.

Biogas and bioenergy technologies have been proven the environmentally safer with fewer or lowest health impacts, economically effective and helpful in energy conservation. The higher calorific values (HCV) and lower calorific values (LCV) of pure methane were 39.82 and 35.87 MJ/m^3, respectively. HCV and LCV of produced biogas were determined according to the following formula:

$HCV_{biogas} = 0.3989 \times MC = 0.0213\ (R2 = 1)$ …… Eq.1

$LCV_{biogas} = 0.3593 \times MC = 0.0192\ (R2 = 1)$………Eq.2

Where; MC is the methane content in biogas (%).

The calorific values of the agriculture residue samples were determined and presented in Table 2. Calorific values were set as the quantity of heat produced by complete combustion of a unit of a combustible compound. Especially for the main indicators, which are volume of CH_4 in biogas, CH_4:CO_2 index and calorific value of biogas. As shown in Table 2, CH_4, CO_2, and calorific value were recognized as significantly influenced.

Table 2. Biogas composition yield and calorific values of from leaves

Parameters	Leaves		
	Mango	**Longan**	**Lychee**
CH_4 (%)	78	75	72
CO_2 (%)	21	23	25
O_2 (%)	0.5	1.9	2.25
H_2S (ppm)	55	145	162
HCV MJ/m^3	31.11	29.92	28.72
LCV MJ/m^3	28.03	26.95	25.87

The biogas economy is related to factors such as agricultural waste availability and logistics, process efficiency, and end-product properties. Recently, AD technology has been demonstrated, and has robust commercial availability. There is a wide variety of lignocellulosic waste with low cost and high availability that can be treated for biogas production. Biogas is a key player in the Asia bio-based economy because it provides strategic perspectives for global producers, especially when the price of oil is reduced. The Asia renewable energy directive sectors call for increase in the use of green fuels.

4. Conclusion

This study investigated the potential of mango, longan and lychee trees leaves as a feedstock for biogas production. The results indicated that these leaves contained rich amount of organic substances and these substances are suitable to use in the anaerobic fermentation process to sustain microbial life and transform nutrients into biogas. Biogas and bioenergy technologies have been proven environmentally safer with fewer or lowest health impacts, economically productive and helpful in energy conservation. The results suggested that digestion of pruning leaves was a promising approach for improving biogas production. Furthermore, the digestate has high nutrient concentrations that can potentially use as fertilizer.

Nomenclature and Abbreviation

HCV	Higher calorific values
LCV	Lower calorific values
TS	Total solids
VS	Volatile solids (VS),
COD	Chemical oxygen demand
TS	Total sugar
RS	Reducing sugar
CH_4	Methane
CO_2	Carbon dioxide

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