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Biodegradability simulation of coffee wastewater using instant coffee

Elida Novita[a]*

Agricultural Engineering, Jember University, Jember 68132, Indonesia

Abstract

Water minimization in wet process of robusta coffee can improve coffee bean quality while reduce the volume of coffee wastewater. However, this process increases the concentration of organic material in wastewater up to 100% that requires specific handling. High organic matter content in wastewater has potential to be utilized as a source of renewable energy through anaerobic process. The objective of this study is to determine the potential biodegradability of coffee wastewater that has high concentration of organic material into biogas using a composite of microorganisms derived from specific anaerobic reactor. Fluctuated coffee wastewater concentration in coffee cherry processing and limited time of coffee harvested cause limited time to get coffee wastewater with certain concentration. As an approach, this study used instant coffee solution as coffee synthetic wastewater by considering some different characteristics between them. Results of study showed combination of floccular and granular sludge that has broad tropic microorganism composite is the best type of bacteria to convert organic matter into biogas. Moreover, it can overcome acidic environment in coffee wastewater. Since accumulated methane gas reach highest percentages in high concentration of organic matter, it is better to degrade with low rate handling anaerobic.

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Keywords: coffee wastewater, instant coffee, sludge, water minimization, anaerobic.

1. Introduction

Coffee is a global important commodity which is produced in the tropics such as Indonesia. Coffee in Indonesia included 5 main crops commodities and 10 main export commodities. Since 2008, Indonesia is the third exporter

* Corresponding author.
E-mail address: elide_novita.ftp@unej.ac.id

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countries (ICO, 2010; Mutakin et al., 2008). There are two varieties which internationally traded; Arabica (*Coffea Arabica*) and Robusta (*Coffea Canephora).* Processing coffee cherries into green coffee (traded coffee beans) are generally divided into wet process and dry process. Most coffee producer in Indonesia is smallholder robusta coffee which used to dry process for coffee cherry. In the dry process, coffee cherries are laid out in the sun after picked to dry, either on large concrete or brick patios or on matting raised to waist height on trestles. There was a natural method, friendlier to environment but it is hard to control the quality consistently. In the wet process, the fruit covering the seeds/beans is removed before they are dried. The wet method requires specific equipment and substantial quantities of water. The process is more sophisticated than the dry process and leads to better quality coffee bean (Clifford & Wilson, 1985). This method is generally used for Arabica coffee, but is starting to use for robusta coffee with some modifications.

Indonesia Coffee and Cocoa Research Institute (ICCRI) introduces wet grinding process which is a wet process modification to apply at smallholder coffee producer for increasing the quality of coffee bean. In wet grinding, farmers remove the outer skin of coffee cherries mechanically, using pulping machines. Then, stored form fermentation for about 24 hours. Coffee processed by the wet method is called wet processed or washed coffee of coffee bean is better than the dry quality; though produce wastewater and solid waste that can pollute the environment. Through the wet processing, more than 80% of harvest volume left as organic waste. Some parts of the organic waste can be recycled into valuable products, economically (Pelupessy in Mattson & Sonesson, 2003).

Coffee has been shown to have definitely beneficial for Indonesian farmer. As annual crops, coffee in Indonesia harvested in April until September. At the peak harvest, June to August, huge waste from coffee processing can lead to serious problem if untreated. Coffee wastewater and their solid waste could reduce the quality of water body and soil lead to harmful for environment. One effort to overcome this problem is water process minimization. Water minimization based on pollution prevention concept can be done to reduce coffee wastewater volume (Mburu, 2010; Novita et al., 2010). Even though this concept is familiar with application on large Arabica plantation but it is still limited for smallholder coffee, especially for robusta processing.

Water minimization in the wet process of robusta coffee can improve coffee bean quality while reduce the volume of coffee wastewater. However, this process increases the concentration of organic material in wastewater up to 100% that requires specific handling. COD concentration of robusta coffee wastewater processing reaches 7000 mg/L and with minimization, it increase up to 14 000 mg/L moreover up to 26000 mg/L. (Novita et al., 2010). Mburu et al. (1994); Von Enden & Calvert. (2002) explained that coffee wastewater containing high concentrations of pollutants because the content of organic material results of the process of stripping the flesh of the fruit (mesocarp when peeled) and mucilage. Pulp and mucilage content of protein and sugar in pectin polysaccharides formed such as carbohydrates.

Pulp and mucilage are composed of a number of proteins and sugars in the form of pectin polysaccharides such as carbohydrates. Selvamurugan et al. (2010) has reported that the high sugar content causes fermentation on water stripping process rapidly by enzymes-contained bacteria from coffee fruit. As for other components of the stripping process water is acidic and toxic chemicals such as polyphenols (tannins and caffeine). Another substance that can be found in coffee effluent is toxic chemicals such as tannins, alkaloids (caffeine) or polyphenols. These components when discharged into the environment will lead to the degradation process of organic materials difficult to degrade, biologically (Selvamurugan et al., 2010).

Angelidaki et al. (2007), reported that high organic matter content in the wastewater has the potential to be utilized as a source of renewable energy through anaerobic process. Anaerobic digestion is the conversion of organic compounds in the absence of any inorganic electron acceptors to the most reduced and oxidized form of carbon, namely to methane and carbondioxide, while partial conversion of the organic compounds to intermediates is referred to as transformation or primary biodegradation. Anaerobic biodegradation is a highly complex process, involving a consortium of several microbial groups and interrelated biochemical processes.

Therefore it is necessary to do the assessment of anaerobic biodegradability rate in coffee wastewater treatment to find out the potential of methane formation and types of microorganism composite which can produce highest methane as renewable energy. Gunaseelan (1997) suggests the analysis of biochemical methane assays (BMP) can be performed to determine the output of CH_4 (methane) from organic substrates and to monitor the level of anaerobic toxicity. BMP is a valuable, rapid and inexpensive method to determine the potential and the rate of the conversion of biomass and waste into CH_4. The objective of this study is to determine the potential biodegradability of coffee wastewater that has a high concentration of organic material into biogas using a composite of microorganisms derived from specific anaerobic reactor.

Fluctuated coffee wastewater concentration in coffee cherry processing and limited time of coffee harvested cause limited time to get coffee wastewater with certain concentration. This specific concentration with unfluctuated coffee wastewater is needed to get the information about how far highly organic concentration can be overcome in anaerobic treatment. As an approach, this study used instant coffee solution as coffee synthetic wastewater by considering some different characteristics between the two. Daoming & Forster (1994) used instant coffee as simulated coffee wastewater to determine inhibitory effects in the thermophilic anaerobic treatment.

2. 2. Materials and Method

2.1. Preparation

This study is part of main research about coffee processing design based on cleaner production using wet process for smallholder Robusta coffee industry. Synthetic wastewater coffee made from instant coffee solution with consideration of material availability and the ease making coffee wastewater in specific range COD concentration. Preparation of solution concentration adjusted to the range of concentrations of soluble COD obtained from the minimization of water treatment. Temperature and pH are maintained at optimum conditions of 37°C and neutral acidity (pH: 6.5 to 7.0) except at 1st batch. The compositions of synthetic wastewater at different levels of COD concentrations are presented in Table 1.

Table 1. Syntetic wastewater composition in 100 ml test volume

Instant coffee	7.65 – 22.94 g for making coffee solution with range COD concentration between 10 g/L until 30 g/L.
1st batch	Inoculum types : Mixed of granular and floccular sludge (20:20 in 40 % volume)
	Blank : water
	Control: cellulose (1g/100 mL)
	Substrate: instant coffee solution with COD concentration: 10 g/L, 20g/L and 30 g/L prior in certain pH
	Each treatment carried out in triplicate
	Data collection: methane accumulated measured by GC until stationery phase (61 days)
2nd batch	Inoculum types : granular sludge (30 % volume)
	Blank : water
	Control: cellulose (1g/100 mL)
	Substrate: instant coffee solution with COD concentration: 10 g/L, 15 g/L, 20g/L and 30 g/L prior in neutral pH
	Each treatment carried out in triplicate
	Data collection: methane accumulated measured by GC until stationery phase (52 days)
3rd batch	Inoculum types: granular sludge (30 %)
	Substrate:Nicotinic acid in anaerobic medium (1g/L, 5g/L and 10g/L) and Phenol in anaerobic medium (1g/L, 5g/L and 10g/L)
	Each treatment carried out in triplicate
	Data collection: methane accumulated measured by GC until stationery phase (34 days for nicotinic acid and 29 days for phenol)

The inoculums are from anaerobic reactor of brew industry for granular sludge from UASB reactor and from CSTR reactor of floccular sludge of water reclamation plant. These inoculums have a “broad trophic” microbial composition in order to ensure that the anaerobic conversion of substrate is not limited. So, both have mesophilic and thermophilic inoculum but with different composition. The amount of inoculums for first batch is about 40%

composed of granular and floccular sludge. The sludge taken from CSTR has typically 14 to 19 % VS and from UASB has typically 14-20 % VS. Second batch composed of granular sludge around 30 %, also treat for the third batch. Furthermore, granular sludge mostly consists of microbial biomass.

Anaerobic basic medium was made for blank solution. According to Angelidaki & Sanders (2004), the basic medium is prepared from the following stock solutions (chemicals given below are concentrations in g/l, in distilled water).

Table 2. Basic anaerobic medium

A	NH_4Cl, 100; NaCl, 10; $MgCl_2$ $6H_2O$, 10; $CaCl_2$ $2H_2O$, 5.
B	K2HPO4 3H2O, 200
C	Resazurin 0.5
D	Trace-metal and selenite solution: $FeCl_2$ $4H_2O$, 2; H_3BO_3, 0.05; $ZnCl_2$, 0.05; $CuCl_2$ $2H_2O$, 0.038; $MnCl_2$ $4H_2O$, 0.05; (NH_4) $6Mo_7O_{24}$ $4H_2O$, 0.05; $AlCl_3$, 0.05; $CoCl_2$ $6H_2O$, 0.05; $NiCl_2$ $6H_2O$, 0.092; ethylenediaminetetraacetate, 0.5; concentrated HCl, 1 ml; Na_2SeO_3 $5H_2O$, 0.1
E	vitamin mixture (components are given in mg/l): Biotin, 2; folic acid, 2; pyridoxine acid, 10; ridoflavin, 5; thiamine hydrochloride, 5; cyanocobalamine, 0.1; nicotinic acid, 5; P-aminobenzoic acid, 5; lipoic acid, 5; DL-pantothenic acid.

To 974 ml of distilled water, the following stock solutions should be added (A), 10 ml; (B), 2 ml; C), 1 ml; (D), 1 ml and (E), 1 ml. The mixture is gassed with 80% N_2 - 20% CO_2. Cysteine hydrochloride, 0.5 g and $NaHCO_3$, 2.6 g, are added and the medium is dispensed to serum vials and autoclaved if necessary. Before inoculation, the vials are reduced with Na_2S $9H_2O$ to a final concentration of 0.025%. Samples are incubated at 37^oC.

The biodegrability assay is performed in closed vessels (experimental vessels sized 150 ml volume) (Fig.1).

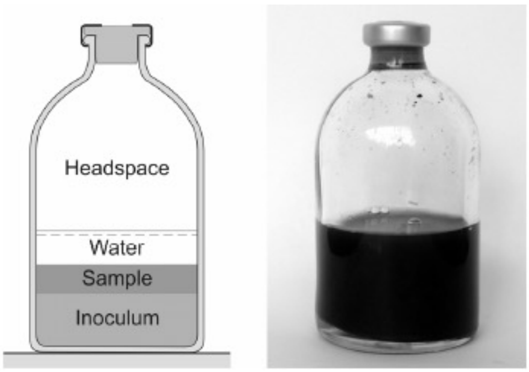

Figure 1. Example of an Assay Vessel for Anaerobic Biodegradability Test (Angelidaki et al., 2007).

2.2. Blank and control

The background methane production from the inoculum, determined in blank assays with medium and no substrate, is subtracted from the methane production obtained from the sample assays. The blank and the control carried out in triplicate. The control vessels contain a cellulose standard substrate that usually used for crops or municipal biowaste (Angelidaki et al., 2007).

2.3. Data collection

The methane accumulated in the headspace of the closed vessel will be measured by gas chromatography (GC). For that, a sample volume would be collected with a gas-tight syringe and injected into the GC. The obtained peak area should be compared to that obtained by injecting the same volume of a standard gas mixture of the known composition. The standard gas mixture should be injected at the atmospheric pressure because if the gas sample is taken with a gas tight over pressurised syringe, and compared with a gas standard injected under atmospheric pressure the methane percentage will be more than 100%. The volume of methane produced is obtained by multiplying the headspace volume by the % of CH_4 in the headspace as determined by GC analysis.

2.4. Analysis method

Anaerobic biodegradability assays are used not only to establish anaerobic biodegradability, to determine the ultimate methane potential of wastes, but are also used for determination of the rate of this biodegradation in general. Methods based on product formation measure either the gaseous end product (biogas) or liquid phase production of intermediates such as volatile fatty acids. Most methods in the literature are based on monitoring biogas production. Biogas production is measured either as volume increase under constant pressure (volumetric methods), or measurement of pressure increase in constant volume (manometric methods), or measurement of methane formation by gas chromatography. Gas chromatography is used to measure content of methane and carbon dioxide of the biogas that ends up in headspace of closed vials. Using GC with thermal conductivity detection (TCD) where both methane and carbon dioxide are measured. This method using a reference gas e.g. nitrogen in the headspace and regular sampling the volume of biogas can be estimated based on the molar fractions of CH_4 and CO_2.

VFA (Volatile Fatty Acid) in synthetic wastewater are determined by GC using a FID (flame ionization detector) and a polar capillary volume (DB-FFAP). The samples are filtered through a 0.45 µm or 0.22 µm membrane filter. Then 0.9 mL of the sample is transferred into a GC vial to which 0.1 ml of 10% v/v formic acid is added. The sample should cap immediately, mixed and kept in the fridge before analysis.

3. Results and Discussion

3.1. Chemical composition of synthetic coffee wastewater

Chemical composition of coffee wastewater is determined by coffee pulp and coffee bean composition. About 50% of the dry matter of coffee bean green (unroasted) consists of carbohydrates, mainly polysaccharides. The proteins represent 10 to 12% and lipids 10 to 18%. Coffee beans are valued for their bioactive compounds: the chlorogenic acid (5 to 9% dry mass) properties antioxidant and caffeine (0.5 to 3.5%) with stimulating properties.

Table 3. Composition of green and roasted coffee and instant coffee

Component	Robusta		Instant
	Green	Roasted	
Minerals	4.0 – 4.5	4.6 -5.0	9.0 – 10.0
Caffeine	1.6 – 2.4	Appr 2.0	4.5 – 5.1
Trigonelline	0.6 – 0.75	0.3 – 0.6	-
Lipids	9.0 – 13.0	11.0 – 16.0	1.5 – 1.6
Total Chlorogenic Acids	7.0 – 10.0	3.9 – 4.6	5.2 – 7.4
Aliphatic acids	1.5 – 2.0	1.0 – 1.5	-
Oligasaccharides	5.0 – 7.0	0 – 3.5	0.7 – 5.2
Total polysaccharides	37.0 – 47.0	-	Appr 6.5
Amino acids	2.0	0	0
Proteins	11.0 – 13.0	13.0 – 15.0	16.0 – 21.0
Humic acids	-	16.0 – 17.0	15.0

Source: (Clifford & Wilson, 1985)

Table 4. Organic components present in coffee pulp

Components	% dry weight
Tannins	1.80 – 8.56
Total pectic substances	6.5
Reducing sugars	12.4
Non reducing sugars	2.0
Caffeine	1.3
Chlorogenic acid	2.6
Total caffeic acid	1.6

Source: Braham & Bressani (1979)

During roasting, the composition changes dramatically, a lot of chlorogenic acids disappear and lactones are shaped. The water content decrease, the polysaccharides are degraded, pigments are shaped (polycondensed furans) and a complex aroma develops, consisting of alcohols, the phenols, with aldehydes, pyrrole and furan derivatives, hydrocarbons, thiophenes etc (Clifford & Wilson, 1985). Clarke & Macrae. (1989), it was identified over 800 volatile aromatic compounds in roasted coffee, 42 of which are phenolic in nature. They come mainly from the thermal degradation of chlorogenic acid and lignin. Some soluble substances have already been extracted at the point of instant coffee manufacture. The percentage removed could vary according to brand and may up be up to 50%, so that more of the normally insoluble substances are rendered at 100°C. Clearly, the composition of different instant coffees varies considerably, especially in the relative proportions of the various constituents. The organic component in instant coffee which used in this study is phenols, little parts of polysaccharides, proteins and humic acids that control COD concentration (Table 5).

Table 5. Composition of synthetic coffee wastewater

Components	Coffee Solution with Varieties of COD		
	10 g/L	20 g/L	30 g/L
Protein (gr)	0.077	0.115	0.153
Carbohydrate (gr)	0.459	0.688	0.918
Sugars (gr)	0.076	0.114	0.153
Fibre (gr)	0.382	0.574	0.765
Polyphenol & melanoidin (mg)	275.314	412.970	550.620
Caffeine (mg)	53.533	80.299	107.065
pH solution	5.28	5.15	5.03

3.2. Biodegradability test (BMP test)

The biodegradability test (BMP test) of coffee wastewater carried out in 3 stages as Batch 1 that Biodegradability test (BMP test) at various concentrations of synthetic coffee wastewater which use mix granular and floccular sludge without pH adjustment. Batch 2 is Biodegradability test (BMP test) at various concentrations of synthetic coffee wastewater which use granular sludge with pH adjustment (6.5 to 7.0), and Batch 3 is Biodegradability test (BMP test) on nicotinic acid and phenol use granular sludge with pH adjustment (6.5 to 7.0). Nicotinic acid and phenols are organic components contained in instant coffee and especially increase after roasting.

Granular sludge is derived from microorganism composite at UASB (Upflow Anaerobic Sludge Blanket) reactor. Floccular sludge is heterogeneous composite microorganisms which are suspended in the form of activated sludge from CSTR (Continuous Stirred Tank Reactor). According to Liu et al. (2002), UASB and CSTR are part of the anaerobic system that can be used to handle organic waste. UASB is developed based on methanogenesis bacterial activities in fast flowrate. The rapid upward flow in UASB reactor and methanogenesis bacterial activity patterns to form some suspended layer urge development of methanogenic consortium formed themselves into granules. These granules more viscous than water and they flow through wastewater toward into the upper of reactor. UASB was developed especially for high concentration of wastewater. High density of granular microorganism can facilitate the separation process of wastewater effluent that has been purified by biomasss. Even though, particulate solids in wastewater sometimes can intrude the system.

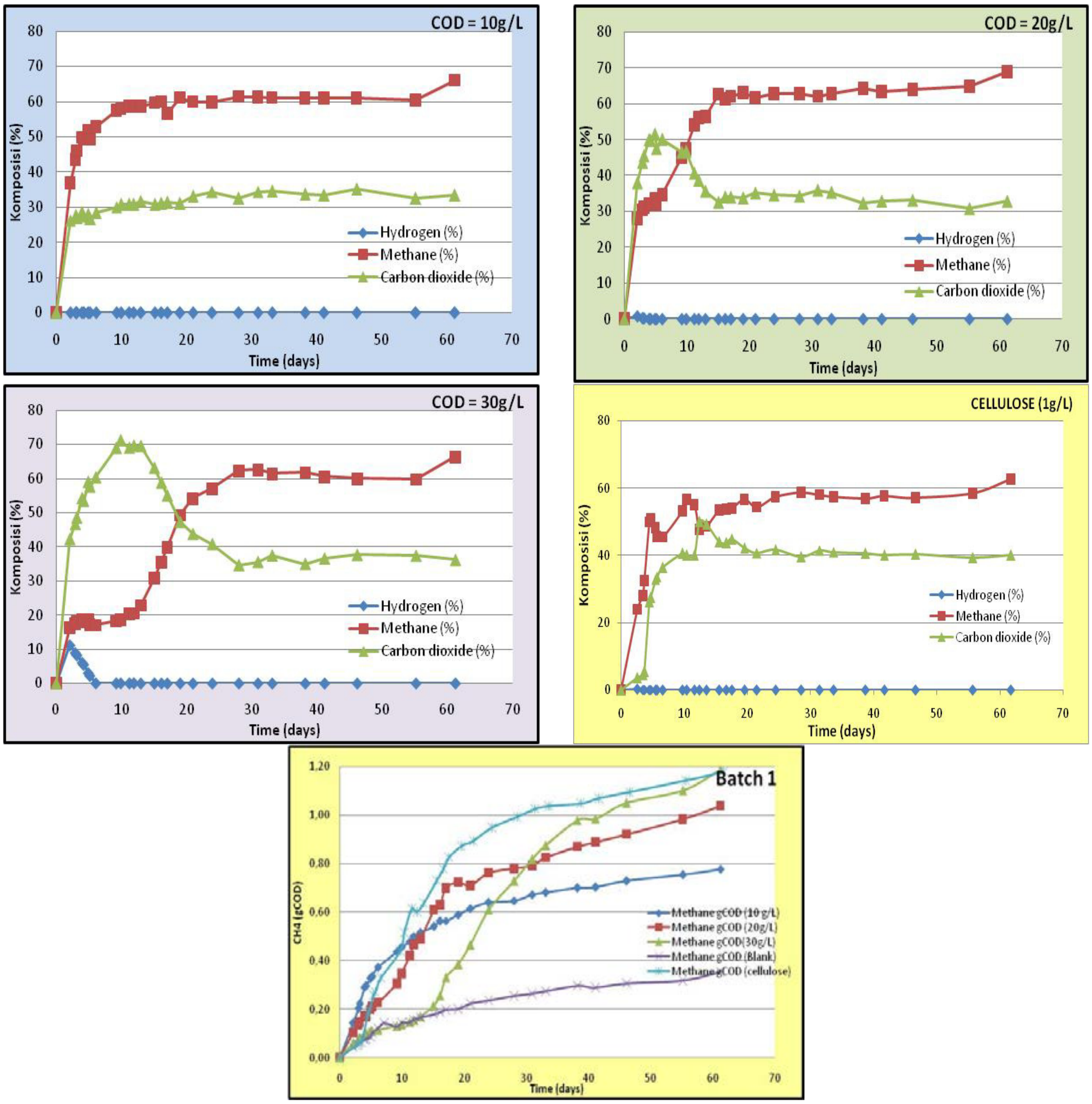

Figure 1. Biogas Composition Generated at 1st Batch Anaerobic Process.

CSTR was adapted from aerobic reactor for organic waste processing using activated sludge from secondary treatment sludge. The purpose of mechanical agitation existences in aerobic reactor is to keep the particulate in suspended form and to make oxygen dissolve in easier. On contrary, mechanical agitation in anaerobic tank could affect rate of growth of microorganism consortium especially in asidogenesis and methanogenesis environments. The dynamical activities of bacteria tend to not uniform in CSTR. This is the weakness of floccular sludge from CSTR.

Biodegradability test at 1^{st} batch was done at various COD concentration to find out ability of combine microorganism in synthetic wastewater degradation. Cellulose biodegradability test also done as a part of carbohydrate component performed (control test) to degrade. Test also carried out without pH adjustment to determine the ability of combined microorganism to degrade wastewater (Figure 2).

The ability of combined granular and floccular sludge were found to be moderately degradable at 10 g/L COD concentration and tends to decrease with increasing concentration of COD. Even though, without pH adjustment, microorganism could adapt in acidic condition except at high concentration of COD of 30 g/L. They need more time to cope acid component. Percentage of H_2 higher at the first lag and will decrease after 8 days. Percentages of CO_2 become the highest component at the early 10 days while CH_4 gas showed increasing after 10 days. It seem high methanogenic activities from granular sludge and asetogenic activities from floccular sludge can maintain the process to degrade highly organic concentration of wastewater (30 g/L), though need some periods to settle.

Stability of methane formation process depends on COD concentration. The higher COD concentration requires longer time to achieve the stability phase. Samples with concentration at 10 g/L, 20 g/L and 30 g/L require consecutive 10 days, 15 days and 25 days, reach equilibrium formation of CH_4. It causes cumulative percentage of CH_4 at highest concentration of COD greater than others do. Even though degradation process needs 60 days, lots of organic components convert into useful biogas. Hydrolysis of carbohydrate, lipids and proteins into monomer and simple oligomer at high concentration of COD (30 g/L) greater than other samples. The results obtained by synthetic coffee wastewater treatment of average composition of methane and carbondioxida, respectively, 60 – 70% and 30% - 40%. Borjesson & Berglund (2006), the composition of biogas is mainly composed of CH_4 (60-70%) and CO_2 (30-40%), with moisture content and some of the gases nitrogen, hydrogen sulfide and ammonia. This happens because hydrolysis process of the amount of carbohydrates, fats and proteins into simpler monomers and oligomers can continued to other phase of asidogenesis and methanogenesis.

Biodegradability test generally takes place even without neutralization. Dinsdale et al. (1997), use UASB reactors with pre-acidification stage explain neutralization is not needed at asidogenesis phase for efficiency. Effluent from asidogenic phase with pH 5.2 does not require neutralization with alkali before methanogenic phase. Floccular sludge within asidogenic phase help overcome acid condition of wastewater for methanogenic phase of granular sludge to degrade into CH_4. This acidification phase take a role to receive high loading rate of organic component.

Biodegradability test at 2^{nd} batch only use granular sludge from UASB reactor to degrade wastewater. The time of synthetic wastewater to degrade at the same concentration is shorter than the first batch. But, granular sludge seems not capable enough to degrade high concentration of synthetic wastewater (30 g/L). Anaerobic process does not take place when optimum composition of CO_2 (60 – 70%) is greater than CH_4 (20-30%). The percentages of CH_4 reached peak point on 2^{nd} day and then tend to stable up to 40 days. Granular sludge only contains methanogenic bacteria that work optimum at methanogenesis phase.

Since anaerobic digestion can be devided in 4 phases according to the characteristic of microorganisms and important conversions taking place; hydrolysis, acidogenesis, acetogenesis and methanogenesis. Malina and Pohland. (1992), a complex culture of different microorganism is part of of anaerobic digestion process allowing the transformation of the original substrate into methane gas and other by products. Pereira. (2009), complex suspended compounds and colloidal matter are converted into their monomeric or dimeric components, such as aminoacids, single sugars and long chain fatty acids (LCFA). Acidogenic bacteria excrete enzymes for hydrolysis and convert soluble organics into volatile fatty acids and alcohols. Acetogenic bacteria then convert volatile fatty acids and alcohols into acetic acid or hydrogen and carbon dioxide. Methanogenic bacteria use acetic acid, hydrogen and carbon dioxide to produce methane.

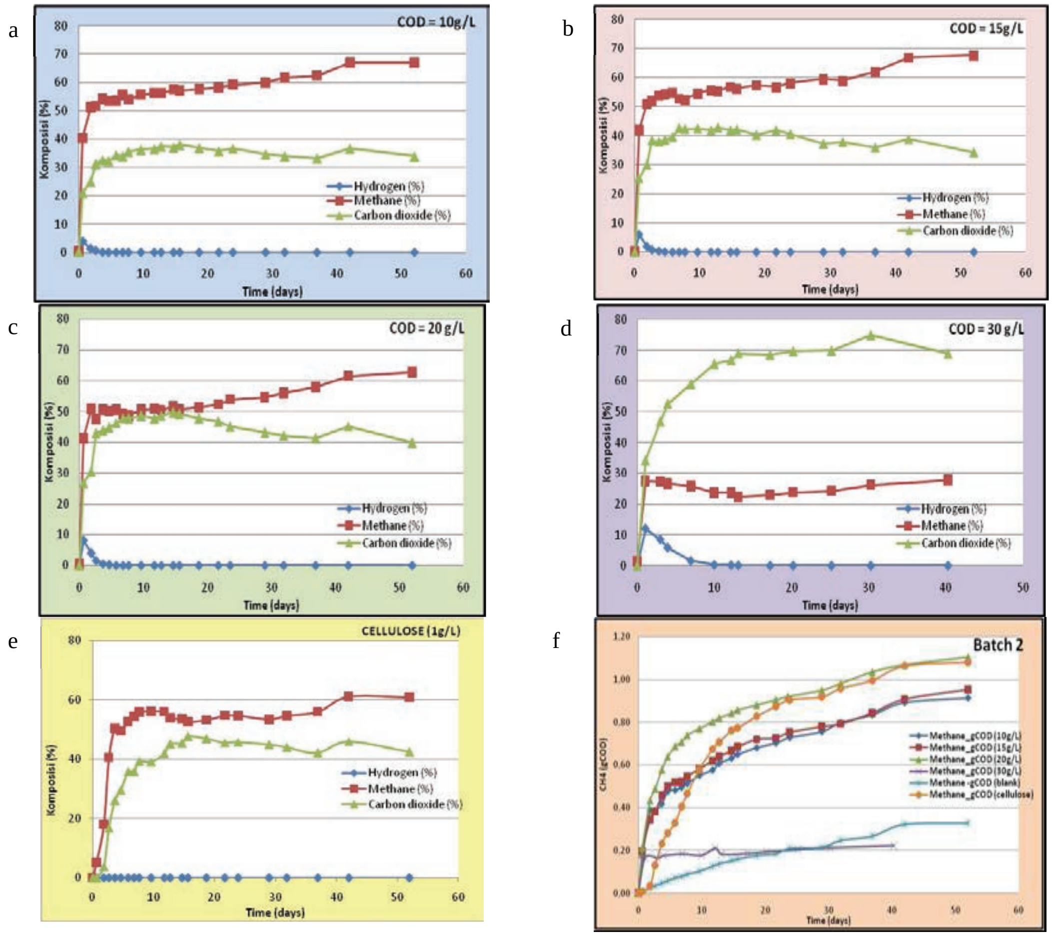

Figure 2. Biogas composition generated at 2nd batch anaerobic process (a) 10 g/L COD (b) 15 g/L COD (c) 20 g/L COD (d) 30 g/L COD (e) Cellulose 1g/L as control (f) Methane Production

Methane starts to highly produce at 23rd days in 1st batch and at 3rd days. After 23rd days, H_2 seems disappeared while methane (CH_4) percentages increased until stabilized at 61st days (70 % percentage of methane). 2nd batch only need 3 days to reach higher conversion of organic component into biogas. Methanogenic bacteria which the most composition within granular sludge works optimum in the 3 days and stabilizes until 40th days. Degradation processes lead to production of carbondioxide higher than the production of methane causes granular sludge not work optimally since high concentration of organic components. VFA composition showed environment condition inside anaerobic vessel. High composition of carbondixide and VFA at 2nd batch can cause the degradation process fail (Table 6). The combination of inoculums to degrade coffee wastewater with high concentration of organic components eventhough they need more time. Adaptability and “broad tropic” of microorganism composition in inoculums can overcome the acidity and complex organic in coffee wastewater.

Nevertheless, granular bacteria still showed their capability to degrade the cellulose component as a complex organic at 1 g/L concentration. Pereira (2009), time to reach the anaerobic degradation with COD conversion of organic component into methane is influenced by the quality of composite bacterial and environmental quality. Methanogenic bacteria in granular sludge could not expect to work optimum within asidogenesis phase when it is suitable for floccular sludge, eventhough not in acidic environment.

Nicotinic acid and phenol are chemical components which contained in coffee especially instant coffee that increased in roasting. BMP test or biodegradability analysis of nicotinic acid and phenol are needed for knowing their influence on degradation of synthetic coffee wastewater. Phenol or phenolics are chemical component that has a hydroxyl functional group (-OH) which bounded in aromatic hydrocarbon group. Simple component of phenolics is phenol (C_6H_5OH). Phenolics generally found in plants in esthers or glicosides form which bounded with other components such as flavonoid, alcohol, etc. Total phenolic acids in coffee berry reached 97 mg/100 mg in the form of caffeic acid (Musatto et al., 2011). Azhar & Stuckey (1994) state that components contained in coffee phenolics are phenol, quinic acid and caffeic acid.

Table 6. Average of biogas and VFA comparison at high concentration of COD

Components	Unit	1 st batch	2nd batch
CH_4	%	57,01	27,38
CO_2	%	40,85	46,96
H_2	%	0,00	8,54
acetic acid	ppm	**389,92**	**756,46**
propionic acid	ppm	**80,84**	**117,98**
iso-butyric acid	ppm	27,89	31,40
butyric acid	ppm	**688,33**	**987,97**
iso-valeric acid	ppm	42,79	51,17
valeric acid	ppm	21,24	24,67
hexanoic acid	ppm	22,98	43,83

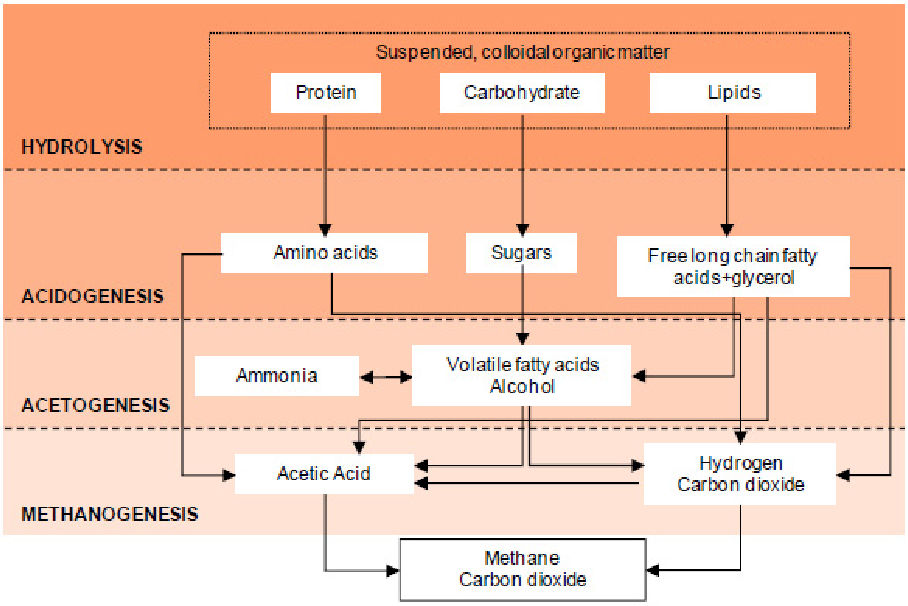

Figure 3. Simplied Schematic Representation of The Anaerobic Degradation Process. Source: Pereira (2009)

Nicotinic acid is one components of nitrogenous compounds contained in coffee as degradation results of organic components (Azhar & Stuckey, 1994). Nicotinic acid is part of the B vitamins known as Niacin (Vitamin B3). Generally, in a cup of coffee will contain only 0,5 mg of nicotinic acid.

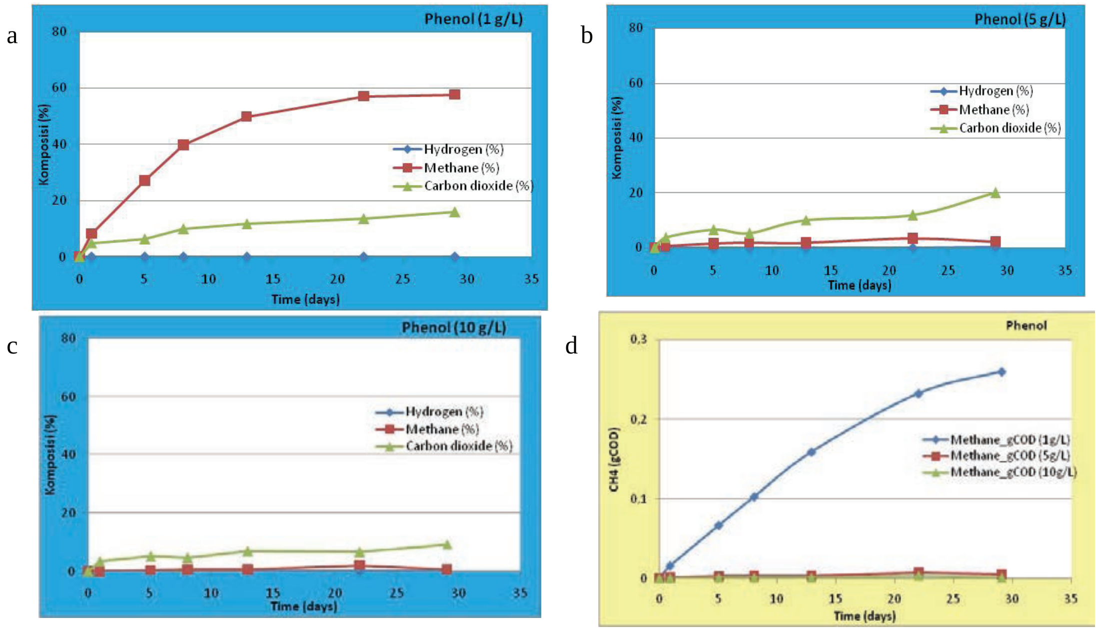

Figure 4. Biogas composition generated at phenol degradation (a) 1 g/L COD (b) 5 g/L COD (c) 10 g/L COD (d) Methane Production

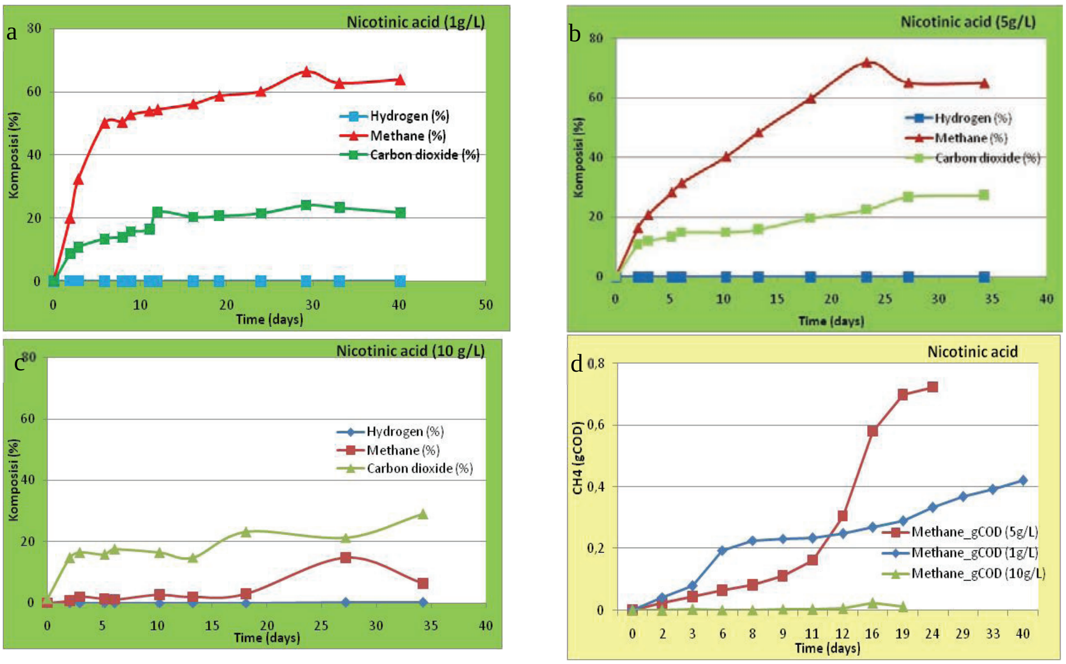

Figure 5. Biogas composition generated at nicotinic acid degradation (a) 1 g/L COD (b) 5 g/L COD (c) 10 g/L COD (d) Methane Production

Anaerobic process work optimum at phenol concentration 1g/L (1000 ppm). Process stability is achieved after 28 days. Phenolic component is an inhibitor of anaerobic wastewater treatment at the early process. However, after acclimatization process is exceeded, 61% phenol was degrading. Anaerobic decomposition by granular sludge is not achieved at high concentration of phenol (5 g/L and 10 g/L). It happened because of phenol aromatic components is hard to degrade only with granular sludge. Granular sludge with methanogenic bactery composites works optimally to disentangle simple monomer organic component such as acetic acid.

Nitrogenous compounds of nicotinic acid degradation can take place until concentration of 5 g/L (5000 ppm). Stability of process is achieved at average of 23 days. Azhar & Stuckey. (1994) explain that nicotinic acid generally can degrade rapidly, especially after 20^{th} days. Nicotinic acid is non volatile components decomposed from trigonelline in instant coffee. Nicotinic acid initially decomposed quite rapidly before reaching a plateu at 20^{th} days, but it did not give high CH_4 evolution. Nitrogenous compound easy to decompose during roasting process with high temperature and produces the aromatic components of coffee. Since phenol and nicotinic acid in instant coffee solution are less than 1 g/L, these components do not affect anaerobic degradation of synthetic coffee wastewater.

Conversion of organic components in synthetic coffee wastewater into methane, carbondioxida and volatile fatty acid also reduce their COD (chemical oxygen demand) concentration. The percentage of methane conversion (Table 7) at 1^{st} batch which has combined sludges gives opportunity to reduce organic complex in coffee wastewater. Type of bacteria inoculum becomes the dominant factor influences the efficiency of anaerobic process to produce biogas.

Based on results, anaerobic degradation can be considered into 2 phases, asidogenesis and methanogenesis. Asidogenesis is conversion of organic components into acid products by anaerobic and facultative bacteria which have rapid growth while methanogenesis is conversion from acid products into methane by strict anaerobic bacteria that have slow growth rate. Acidic acid, butyric acid and propionic acid are the three main groups of free fatty acids which produced by anaerobic fermentative bacteria that live in glucose medium. Methanogenic bacteria (methanotrix and methanosarcina) usually use single hydrogen and carbon components such as CO_2, CO and HCOOH as substrate to produce methane and carbon atoms (acetic acid). But these type of bacteria do not use combined of H_2 and CO_2 but acetic acid, CO and methanol as a substrate to produce methane.

Table 7. Percentage of conversion

COD concentration (g/L)	Conversion (%)
1^{st} Batch	
10	64,47
20	68,41
30	60,90
2^{nd} Batch	
10	68,75
15	67,60
20	52,89
30	56,56
Phenol	
1 g/L	57,71
5 g/L	2,34
10 g/L	0,54
Nicotinic acid	
1 g/L	63,96
5 g/L	65,04
10 g/L	6,32

Percentages of organic component removal of COD in coffee wastewater showed the best degradable at maximum concentration of 20 g/L. The degradation process still happened despite of in a long progress at the COD concentration more than 20 g/L. Malina & Pohland (1992) suggested that wastewater with high COD concentration more than 20 g/L must be processed by low rate handling anaerobic. Handling high rate anaerobic only suitable for wastewater with COD less than 20 g/L. With the right handling on anaerobic treatment, more than 80 – 90% of BOD can be reduced while COD removal can reach 1.5 times than BOD removal.

4. Conclusions

Water minimization in coffee processing increase high organic matter content but it is potential to produce biogas with anaerobic digestion. Biogas production in anaerobic fermentation is proportional to the rate of substrate utilization, environmental conditions such as pH and temperature and also the composite of diverse bacterial within the process. Combination of floccular sludge and granular sludge that has broad tropic microorganism composite is the best kind of bacteria to convert organic matter into biogas. Moreover, they can overcome acidic environment in coffee wastewater. Since accumulated methane gas reachs highest percentages in high concentration of organic matter, it is better to degrade with low rate handling anaerobic

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