EFFECT OF EFFECTIVE MICROORGANISMS ADDITION ON METHANE PRODUCTION FROM COFFEE HUSKS

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ABSTRACT

Coffee husk is a lignocellulosic material that is abundant and can be used to produce biogas. This study compares the production of biogas produced from coffee husk substrate using cow dung and a mixture of cow dung and effective microorganisms. This experiment was carried out for 30 days in an anaerobic batch reactor with a reactor working volume of 3.6 L at mesophilic temperature. The parameters tested in this study were the lignocellulosic content of coffee husks, total solids (TS), volatile solids (VS), volatile fatty acids (VFA), chemical oxygen demand (COD), and the content of biogas produced from both variables. The lignocellulosic compositions obtained from this study were cellulose 65.90%, hemicellulose 24.95%, lignin 0.21%, pectin 0.42%, protein 0.81%, tannins 1.05%, caffeine 0.09%, and polyphenols. The values of Total Solids and Volatile Solids for the two variables are K-KS of 16.78% and 33.98% and K-KSEM of 24.87% and 48.42%, respectively. The total VFA for the two variables is 2.06% (v/v) for K-KS and 2.36% (v/v) for K-KSEM. The COD values for the K-KS and K-KSEM variables were 78.05% and 81.42%, respectively. The composition of biogas for K-KS is CH_4 12.35%, CO2 21.68%, and H_2 0.32%, while for K-KSEM it is CH_4 19.64%, CO_2 2.82%, and H_2 0.35 %. The methane yields for the two variables, K-KS and K-KSEM, were 0.76 Nm^3/kgCODremoval and 1.43 Nm^3/kgCODremoval, respectively.

PENGARUH PENAMBAHAN *EFFECTIVE MICROORGANISMS* TERHADAP PRODUKSI BIOGAS DARI KULIT KOPI

Abstrak- Kulit kopi merupakan bahan berlignoselulosa yang keberadaannya melimpah dan dapat dimanfaatkan untuk menghasilkan biogas. Penelitian ini membandingkan produksi biogas yang dihasilkan dari kulit kopi dengan menggunakan kotoran sapi dan campuran kotoran sapi dan *effective microorganism*. Percobaan ini dilakukan selama 30 hari di dalam reaktor *batch* anaerob dengan volume kerja reaktor 3,6 L pada suhu mesofilik. Parameter-parameter yang diuji dalam penelitian ini yaitu kandungan lignoselulosa kulit kopi, total padatan (TS), volatil padatan (VS), asam-asam lemak volatil (VFA), *chemical oxygen demand* (COD), dan kandungan dari biogas yang dihasilkan dari kedua variabel. Komposis lignoselulosa yang didapatkan dari penelitian ini yaitu selulosa 65,90%, hemiselulosa 24,95%, lignin 0,21%, pektin 0,42%, protein 0,81%, tanin 1,05%, kafein 0,09%, dan polifenol 0,81%. Nilai *Total Solids* dan *Volatile Solids* untuk kedua variabel yaitu K-KS sebesar 16,78% dan 33,98% dan K-KSEM sebesar 24,87% dan 48,42%. Adapun total VFA untuk kedua variabel adalah 2,06% (v/v) untuk K-KS dan 2,36% (v/v) untuk K-KSEM. Nilai COD untuk variabel K-KS dan K-KSEM masing-masing adalah 78,05% dan 81,42%. Komposisi biogas untuk K-KS yaitu CH_4 12,35%, CO_2 21,68%, dan H_2 0,32%, sedangkan untuk K-KSEM yaitu CH_4 19,64%, CO_2 2,82%, dan H_2 0,35%. Yield metana untuk kedua variabel yaitu K-KS dan K-KSEM masing-masing adalah 0,76 Nm^3/kgCODremoval dan 1,43 Nm^3/kgCODremoval.

Kata Kunci: *anaerobic digestion*, kotoran sapi, cairan rumen, *effective microorganism (EM)*, metana

INTRODUCTION

The increase in world crude oil prices has made many countries look for alternative energy sources to replace fossil fuels. Solar energy is considered an effective energy source because it is environmentally friendly. Some of the disadvantages of alternative energy sources such as solar, hydro, and wind are that they are very expensive to operate. The increase in world crude oil prices has made many countries look for alternative energy sources to replace fossil fuels. Solar energy is considered an effective energy source because it is environmentally friendly. Some of the disadvantages of alternative energy sources such as solar, hydro, and wind are that they are very expensive to operate. Biogas is an environmentally friendly alternative renewable energy source that can be used such as heating, electricity, or vehicle fuel to replace fossil fuels (Machunga-Disu and Machunga-Disu, 2012). The composition of the biogas produced varies and depends on the physical and chemical properties of the substrate used (Chenxi and Bruce, 2011).

Methane production from various biomass wastes through anaerobic digester technology is growing worldwide because it is cheap and environmentally friendly (Fantozzi, 2009; Tekin et al, 2000; Fantozzi, 2011; Bouallagui et al, 2005; Gallert et al, 2003; Hamdi, 1996; Lopes et al, 2004). Methane production is the most efficient technology for energy generation from waste biomass in terms of energy output/input ratio (28.8 MJ/MJ) compared to technologies used for both biological and thermochemical (Deublein, 2008). The use of agricultural waste for biogas production has increased in recent years. Agricultural biomass waste used is corn, sugar cane, and non-food plant parts such as leaves, stems, coffee grounds, and husks. The use of microorganisms is used to increase the yield and stability of the final biogas product. In addition, it can help reduce environmental pollution (Pandey et al, 2000; Brand et al, 2000).

Coffee is the second largest traded commodity in the world and also produces by-products and residues. Lignocellulosic components present in coffee grounds include: cellulose (63%), hemicellulose (2.3%), lignin (17%), protein (11.5%), tannins (1.80-8.56%), pectin (6.5%), reducing sugar (12.4%), non-reducing sugar (2.0%), caffeine (1.3%), chlorogenic acid (2.6%) and caffeic acid (1.6%) (Mussatto et al, 2011; Franca et al, 2009). Waste from coffee husks is a source of contamination that can cause serious environmental problems, especially in coffee-producing countries. Not only because of the formation of gases produced due to decay but also because of the high content of caffeine, phenol, and tannins which are toxic in biological processes (Fan et al, 2003). The use of cow dung for biogas production has been widely practiced (Chanakya et al, 1997). However, the cost of a digester for biogas production using cow dung is not profitable because the quantity of biogas is relatively low compared to some other types of organic waste such as food waste (Moller et al, 2004). Cow dung contains little cellulose, lignocellulose, lignin, and organic components that are good for bacterial growth in biogas production (Corro et al, 2013). Cow dung contains bacteria and microorganisms including Clostridium, Bacteroides, Bifidobacterium, Enterobacteriaceae (E. Coli), and Ruminococcus (Alfa et al, 2014).

Effective Microorganisms (EM) is a mixture of various organisms described as anaerobic multicultural that live with beneficial microorganisms in an anaerobic process (Jochen, 2008). The main species involved in EM include *lactic acid bacteria, photosynthetic bacteria, yeast, and Candida utilis. Lactic acid bacteria consist of Lactobacillus plantarum, Lactobacillus casei, and Streptoccus lactis. Photosynthetic bacteria consist of Rhodopseudomonas palustrus and Rhodobacter spaeroides. Yeast consists of Saccharomyces cerevisiae. Candida utilis consists of Actinomycetes, Streptomyces albus, and Streptomyces griseus.* This is the basis of the use of Effective Microorganisms can to secrete organic acids, enzymes, antioxidants, and metal acid chelates *(Ke B et al, 2009).* Effective microorganisms can also reduce the growth of pathogenic bacteria that produce H_2S gas in the anaerobic digester process (Higa, 1994).

This study aims to compare biogas production using coffee husk as a substrate by adding rumen fluid and a mixture of rumen fluid and cow dung into the digester.

MATERIALS AND METHODS

The equipment in this research is a batch reactor as shown in Figure 1, autoclave, hot plate & stirrer, water bath, Spectrophotometer, analytical balance, Incubator, furnace, oven, vacuum pump (Weich), vortex, manometer, gas chromatography (Hewlett Packard), COD tube, COD reactor, and gas chromatography (GC-2010 Plus-SHIMADZU).

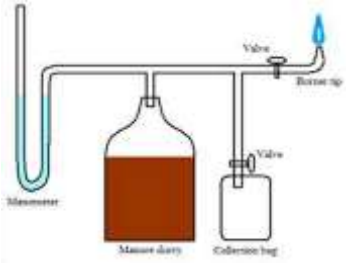

Figure 1. The equipments of anaerobic process from rice straw waste

The materials that will be used in this study are coffee skin, cow dung, and effective microorganisms (EM). Three kilograms of cow dung were taken, and then put in an airtight container. The cow dung obtained was diluted with aquadest in a ratio of 1:3, then filtered using gauze and then put into the digester in accordance with a predetermined volume of 15% of the working volume of the reactor. Effective microorganisms are purchased at a farm store. In addition, glucose, ethanol, $NiCl_2.6H_2O$, $MnCl_2.4H_2O$, $K_2Cr_2O_7$, NaOH, H_2SO_4, CH_3COONa, NH_4Cl, KH_2PO4, $CaCl_2.2H_2O$, $MgCl_2.6H_2O$, Fe-EDTA, $CoCl_2.6H_2O$, and yeast extract were also used.

Analysis of Lignocellosic

In this study, the analysis of cellulose, hemicellulose, and lignin used the Chesson method

Hemicellulose

The hemicellulose content was analyzed using the Chesson method (Isroi, 2013), namely by mixing 1-2 grams of a sample with 150 mL of distilled water, then heated at 100 ^oC for 2 hours, then filtered using filter paper and finally rinsed with distilled water, then The solids were dried in an oven at 105 ^oC to constant weight (a). Then the sample was mixed with 150 mL H_2SO_4 1 N, then the sample was heated at 100 ^oC for 1 hour, filtered with filter paper, and finally rinsed with distilled water. Then the solids were put into the oven at a temperature of 105 ^oC to constant weight (b). The hemicellulose content was calculated using equation (1).

Hemicellulose content (%) = (b-c)/a x 100% ....(1)
Noted:

a) Dry weight reduction of lignocellulosic biomass samples.
b) Reduction of the dry weight of reflux sample residue using hot water.
c) The Reduction dry weight of sample residue after refluxing using 0.5 M H_2SO_4.

Cellulose

Cellulose content was analyzed by the Chesson method. The dried sample in hemicellulose analysis (b) was mixed with 10 mL of 72% (v/v) H2SO4 solution at room temperature for 4 hours, then H2SO4 was diluted to a concentration of 0.5 M. Then the sample was refluxed at 100 oC for 2 hours. The cellulose content was calculated using equation (2).

Cellulose Content (%) = c−d/a 100% ….(2)
Noted:

a) The Reduction in dry weight of lignocellulosic biomass samples.
c) The Reduction in the dry weight of the sample residue after refluxing using 0.5 M H_2SO_4.
d) The Reduction in the dry weight of the sample residue after being mixed using 72% H_2SO_4 after which it was diluted to 4% H_2SO_4.

Lignin

Lignin content was analyzed by the Chesson method. The dried sample in the cellulose analysis (c) was filtered and then washed with distilled water. Next, the solids were put into an oven at a temperature of 105 oC to constant (d). Cellulose content is calculated using equation (3).

Lignin content (%) = d-e/a x 100% ….(3)
Noted :

a) The Reduction in dry weight of lignocellulosic biomass samples.
d) The Reduction in the dry weight of the sample residue after being mixed with 4% H_2SO_4.
e) Ash from sample residue

Biogas Production

Biogas production is produced through an anaerobic process in a batch reactor with a working volume of 3.6 L. This study compares the yield and quality of biogas by adding effective microorganisms (EM). The parameters measured in this study were total solids, volatile solids, chemical oxygen demand (COD), volatile fatty acids (VFA), and biogas composition.

Analysis TS

Cawan porselein dipanaskan selama 1 jam pada suhu 550 ^oC di dalam furnace, kemudian didinginkan didalam desikator, setelah dingin cawan kosong ditimbang (Wdish). sebanyak 10 ml sampel dimasukkan ke dalam cawan yang telah ditimbang sebelumnya kemudian ditimbang kembali (Wsample). cawan berisi sampel dimasukkan ke dalam oven kemudian dipanaskan selama 12 jam pada suhu 105 ^oC. Kemudian cawan didinginkan dalam desikator dan ditimbang kembali sampai beratnya tetap (Wtotal).

$$\% \text{ total solids} = \frac{Wtotal - Wdish}{Wsampel - Wdish} \text{ x } 100$$

Noted:
Wdish = weight of the cup
Wsample = weight of sample and cup
Wtotal = weight of dry sample and cup
(EPA Method 1684, 2001)

Analysis VS

The cup containing the sample whose TS has been weighed is then reheated in the muffle furnace at a temperature of 550 ^oC for 2 hours. After that, the porcelain cup was cooled to room temperature and the weight was re-weighed.

Ash [mg/l] = a × (1000/v)
a = the difference in weight of the evaporating dish after being heated at 550 °C with the weight of the empty dish
v = sample volume
VS [mg/l] = TS [mg/l] - Ash [mg/l]
(EPA Method 1684, 2001)

Analysis COD

COD was measured by adding a digestion solution ($K_2Cr_2O_7$) with 3.5 mL of H_2SO_4 solution in a COD tube, then homogenized (the solution became hot), allowed to settle, then added 2.5 mL of distilled water as a blank, homogenized, then heated at 148 °C for 2 hours using a COD reactor, let it come to room temperature and measure it with a spectrophotometer at a wavelength of 620 nm.

Analysis VFA and CH_4

For analysis of VFA content, the slurry sample was taken through a sampling valve digester using a syringe and a hose and then accommodated into 1.5 mL eppendoff, then homogenized with a centrifuge to separate the filtrate and precipitate. The resulting filtrate was analyzed using Gas Chromatography (GC) HP-6890 at an oven operating condition with an initial temperature of 170 °C for 18.57 minutes. Injector operating conditions using Helium as carrier gas at an initial temperature of 275 °C at a pressure of 17.21 psi. The content biogas such as methane gas (CH4) using gas chromatography (Hewlett Packard, USA).

RESULT AND DISCUSSION

Cellulosic Contents of Coffee husks

Table 1. Chemical composition of coffee husks

Coffee husk components	Percent
Cellulose	65.90%
Hemcellulose	24.95%
Lignin	0.21%
Pectin	0.42%
Protein	0.81%
Tannin	1.05%
Caffeine	0.09%
Polyphenol	0.81%

Based on the results of the analysis of the coffee skin components using the gravimetric method, the cellulose content was 65.90%, hemicellulose 24.95%, lignin 0.21%, pectin 0.42%, protein 0.81%, tannins 1.05%, caffeine 0 .09%, and 0.81% polyphenols. Based on the lignocellulosic content obtained, the coffee rind has a higher potential to be used as a substrate for biogas production, but coffee rind has a composition of toxic substances such as tannins, pectins, polyphenols, and caffeine so that it interferes with the activity of microorganisms in degrading the coffee husk substrate (Corro, 2013).

Total Solids (TS) dan Volatile Solids (VS)

Based on the results of the TS and VS analysis in Figures 2 and 3, it can be seen that the TS and VS values in KK-R and KK-RKS decreased significantly during 30 days anaerobic fermentation time. The decrease in TS and VS values in K-KSEM was greater than in K-KS, namely by 24.87% and 48.42% and 16.78% and 33.98%, respectively. The values of TS and VS are influenced by the increase in the growth of microorganisms from degraded organic compounds. After day 20, the values of TS and VS decreased significantly for both treatments. Total Solid (TS) and Volatile Solid (VS) values in each treatment tended to decrease. The significant decrease in TS and VS was caused by the increased growth of microorganism cells and supported by an adequate supply of nutrients so that these microorganisms were able to degrade organic compounds.

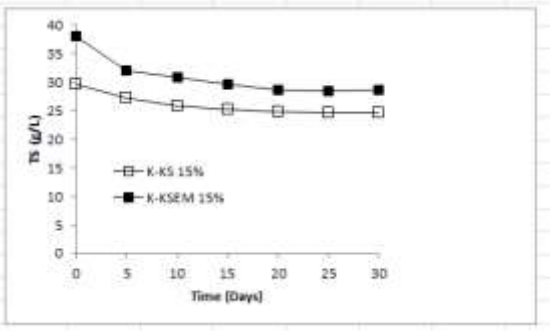

Figure 2. Total Solids profile for anaerobic digestion from K-KS and K-KSEM

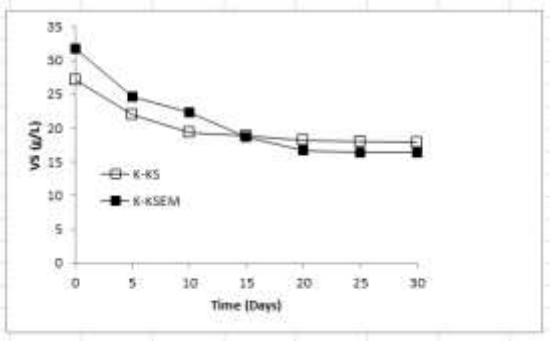

Figure 3. Volatile Solids profile for anaerobic digestion from K-KS and K-KSEM

***Chemical Oxygen Demand* (COD)**

Figure 4 shows the results of COD analysis for both treatments. The decrease in COD in each of the K-KS by 78.05% and K-KSEM by 81.42%. Based on the results of the analysis, COD tends to decrease every 5 days for both treatments. This

decrease in COD indicates that methane gas products are formed.

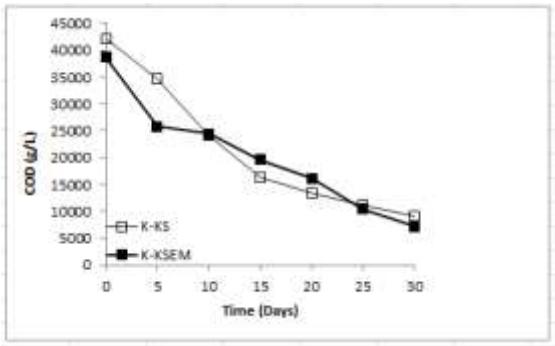

Figure 4. COD profile for anaerobic digestion from K KS and K-KSEM

Volatile fatty acids (VFA)

In this study, volatile fatty acids such as acetic, propionic, and butyric acids were obtained from the analysis using gas chromatography (GC). Volatile fatty acids such as acetic, propionic, and butyric acids are the main products in the methane formation process. The results of the analysis of Total VFA (acetic, propionic, and butyric acids) can be shown in Figure 5.

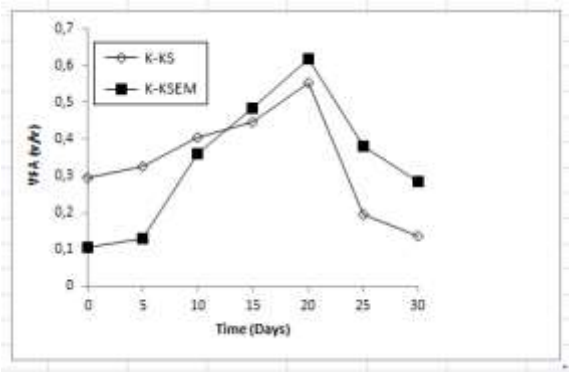

Figure 5. Total VFA for anaerobic digestion from K-KS and K-KSEM

The resulting VFA was obtained from the total acetic acid, propionic acid, and butyric acid produced. The increase in the production of volatile acids (acetic acid, propionic acid, and butyric acid) in each treatment indicated an increase in the growth of acetogenic bacteria, while the decrease in volatile acids on certain days indicated that the resulting VFA was obtained from total acetic acid, propionic acid, and the resulting butyric acid. The increase in the production of volatile acids (acetic acid, propionic acid, and butyric acid) in each treatment indicated an increase in the growth of acetogenic bacteria, while the decrease in volatile acids on certain days indicated the process of methane formation. The concentration of these volatile acids indicates the production of biogas produced (Buyukkamaci, 2004).

Based on the results of the analysis of VFA, it was found that the VFA concentration was higher in K-KSEM by 2.36% (v/v), while in K-KS it was 2.06% (v/v). From the results of the analysis of methane, it was found that the highest concentration of methane produced in the KK-RKS was 31085 ppm. This value is greater than the concentration of methane produced in KK-R, which is 11077 ppm. The production of VFA in KK-RKS is proportional to the production of methane produced. At the methanogenesis stage, methanogenic archaea bacteria such as Methanosarcina Sp and Methanothrix Sp convert H_2 and acetic acid into CO_2, CH_4, and water, and convert H_2 and propionic acid to CH_4 with the bacteria Methanobacterium, Methanococcuss). Acetic acid and propionic acid are the main products in anaerobic biogas production (Wright et al, 2011).

Biogas Composition

The composition of biogas (CH_4, CO_2, H_2) was analyzed for 30 days of anaerobic fermentation. Table 2 shows the comparison of biogas composition between K-KS and K-KSEM

Table 2. Comparison of biogas composition in K-KS and K-KSEM

Compounds	K-KS (%)	K-KSEM (%)
CH_4	12,35	19,64
CO_2	21,68	2,82
H_2	0,32	0,35

The content of methane gas in K-KSEM is higher than in K-KS, which is 19.64% with impurities (CO_2) of 2.82%, and H_2 of 0.35%, while in K-KS the composition of biogas namely CH_4 of 12.35%, CO_2 of 21.68%, and H_2 of 0.32%. The purity of biogas is influenced by the impurity level of CO_2 gas produced and will reduce the calorific value of methane gas which is shown in red in the resulting fire (Burke (2001).

Methane Yield

The highest methane yield was produced in KK-KSEM of 1.43 Nm^3/kgCODremoval, while K-KS produced methane yield of 0.76 Nm^3/kgCODremoval.

CONCLUSION

The addition of effective microorganisms in biogas production on coffee skin substrate is more effective than using only cow dung microorganisms. In addition to increasing the methane yield, it also improves the quality of the biogas produced. This is indicated by the carbon dioxide gas produced which is an impurity, which

is 2.82% less in K-KSEM compared to K-KS which is much larger, which is 21.68%.

In the mixture of rumen and cow dung, the methane composition produced was 22.30% CH_4 with CO_2 impurities of 13.75%, while in the rumen fluid the methane composition produced was 18.40% CH_4 with CO_2 impurities of 14.11%.

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