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Effect of EM(Effective Microorganism)Addition on the Quality of Methane Production from Rice Straw

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Abstract : Utilization of agricultural wastes for biogas production is one of the most demanding technologies in sustainable energy production concerning to the sustainability of environmental issues. Rice straw is one of abundant agricultural wastes in Indonesia that can be used as the source of lignocellulose for biogas production. One of the methods to increase the methane quality is by adding a Effective Microorganism (EM). EM contains of 80 microorganisms species which can produce organic acids and enhance decomposition of organic material. This study tested the effect of EM addition on the methane quality of a rice straw-cow dung mixture (RCE) with variable rice straw-cow dung mixture only (RC) as a control. This experiments were conducted in anaerobic batch reactor over 30 days with a working volume of 3.6 L at the mesophilic temperature. Several parameters were measured to determine the effect of EM addition on methane quality such as volatile fatty acids (VFAs), chemical oxygen demand (COD), and methane yield. In addition, total solid (TS), volatile solid (VS), CH_4,CO_2 and H_2S were analyzed. COD of RC and RCE were 62.34% and 40.54% respectively. Yield of methane production for RC and RCE were 0.195 $Nm^3/kgCOD_{removal}$ and 0.234 $Nm^3/kgCOD_{removal}$ respectively. When the addition of EM was done, the quality of methane increased from 38.54% to 41.24%. With cow dung microorganism, the composition of biogas was 38.54% CH_4, 9.41% CO_2 and 0.39%H_2. With a mixed cow dung microorganisms-EM, the composition of biogas was 41.24 %CH_4,8.7% CO_2 and 0.36%H_2.
Keywords : Rice straw, Cow dung,EM,Methana, Volatile fatty acid.

Introduction

Fossil fuels had for long become the major source of global energy which are generally used as sources of energy in engines combustions and in some intances as raw materials for the petrochemical industries. Although, fossil fuel did play an important role ineconomy sector. There are some problems caused by the extensive usage of fossil fuel; for example, global warming, oil spills, environmental pollution, and gas flares[1]. The production of biogas from renewable resources is becoming a prominent feature of the most developed and developing countries in the world. Lignocellulosic biomass,the abundant and cheap non-food materials are availablefor the production of biogas. Lignosellulosic feedstock can be used as a source of energy supply in the future[2]. Lignosellulosic agricultural residuesare one of potential material for biogas[3]. Among all the alternative wastes, rice straws are one of the most abundant, cheap, renewable energy sources and easily available. As reported by previous study, biogas production with cowdung microorganism without EM addition yields biogas with methane concentration of 11.27%.

Effective Microorganisms (EM) are mixture of organism groups or multiculture of coexisting benefical anaerobic microorganismwhich have reviving action on humans, animals, and the natural environment[4].The main species in EM included lactid acid bacteria, photosynthetic bacteria, yeast, and *Candida utilis*. Lactid acid bacteriawere represented by*Lactobacillus casei, Lactobacillus plantarum*and*Streptoccus lactis*. Photosynthetic bacteria were represented by*Rhodopseudomonas palustrus* and *Rhodobacter spaeroides*. Yeastwere represented by*Saccharomyces cerevisisae. Candida utilis* were represented by*Actinomycetes,Streptomyces albus* and*Streptomyces griseus*. Innoculant which contained 90% *Lactobacillus sp.*produced lactid acid which can accelerate the structural alteration of organic materials such as lignin and cellulose. The EM is used as basis because they contain various organic acids due to the presence of lactid acid bacteria, which secrete antioxidants, organic acids,enzymesand metallic acid chelates[5]. One of the major benefits of using EM was the reduction in sludge volume. Theoretically, the present of organism in EM should be decomposedbecome the organic matter by converting it to carbondioxide (CO_2), methane (CH_4) or use it for growth and reproduction. Effective microorganism also can reduce the growth of pathogen bacteria which produce H_2S in anaerobic digestion[6].Therefore, in the present work we investigated the effects of EM addition to enhance the quality of methane produced in the production of biogas from rice straw.

Materials and Methods

Collection of rice straw

Rice straw was obtained from Sumenep, Madura, East Java, Indonesia. Before introducing into the reactor, rice straw was heated in the thermal pretreatment by sun for 3-4 days.Furthermore, the dry rice straw was grounded into a powder and sieved with100 mesh to obtain the desired size of rice straw.

Cow dung was taken from slaughter houses (RPH) PegirianAmpel, Surabaya, Indonesia. Cow dung of 1111.14 g was diluted in water with a ratio of cow dung and water is 1:2 and then filtered with gauze to separate liquid from solids. Effective Microorganism (EM, 1 L volume) waspurchased from Songgolangit Persada PT.(Surabaya, Indonesia) with the trademark of EM-4.One litre of EM-4 contains *Lactobacillus caseii* of1.5 x 10^6 CFU/mL, *Saccharomyces cerevisiae* of1.5 x 10^6CFU/mL, and *Rhadopseudomonas palustris*of 1.0 x 10^6CFU/mL. This product was registered in the Agriculture Ministry of Republic of Indonesia under the number of No.D.11064101 FTC with certification label of No.IDM 000 073 421.

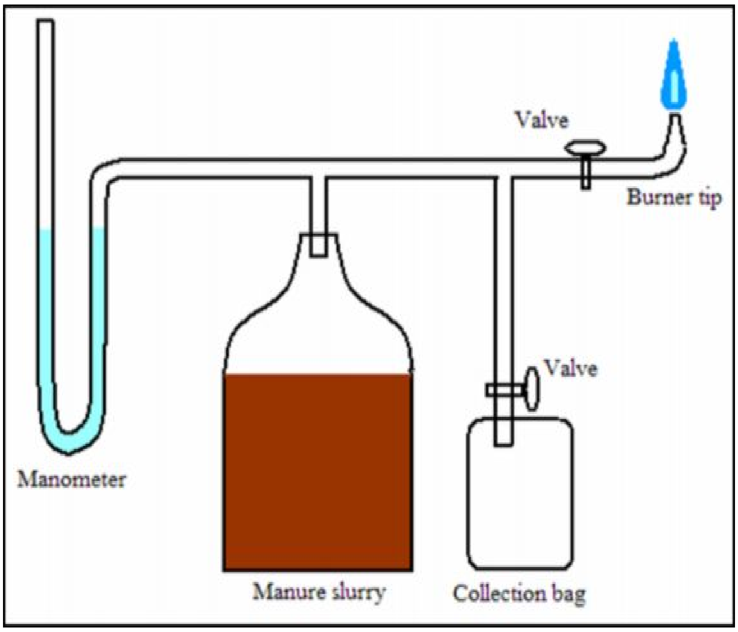

Figure 1 : The equipments of anaerobic process from rice straw waste

The Anaerobic Process

The anaerobic process was conducted in 6 L batch reactor with a working volume of 3.6 L. Control variables in this study was cow dung microorganism (RC). Figure 1 shows a set of batch type biogas reactor.

The comparison of composition of rice straw and cow dungwas obtained from a previous studies.A previous studyreported that the composition of cow dung microorganisms varied, which were 5, 10, and 15% of a working volume of reactor[7]. Biogas produced at thecomposition of cow dungwith15%of working volume was greater thanthat of 5and 10%. Then this results become a basis to use cow dung compositionwith 15% of working volume as a control variable in this study. As the dependent variable, we added a mixture of cow dung and EM with 15% into reactor. The effects of EM on the quality of methane in biogas obtained during the fermentation process were studied.

Starter is introduced into a 500 ml erlenmeyer with a volume of 540 ml, then added CH_3COONa of 2 g/L, rice straw of 4 g/L, $NH_4Cl$of 4 g/L, $KH_2PO_4$of 0.016 g/L, $CaCl_2$of 0.025 g/L, $MgCl_2.6H_2O$ of 0.025 g/L, FE-EDTA of 0.03 g/L, $CoCl_2.6H_2O$of 0.005 g/L,$NiCl_2.6H_2O$ of 0.005g/L, $MnCl_2.4H_2O$of 0.005 g/L, and yeast extract of 0.1 g/L[8].

The parameters measured in this study were chemical oxygen demand (COD), volatilefatty acids(VFAs), total solids (TS), volatile solids (VS), production of CH_4, composition and heating value of biogas.

Analytical Procedure

Analysis of cellulosic level was done to determine the initial concentration of cellulose, hemicellulose, and lignin. Cellulosic content of rice straw was measured using a gravimetric method[8].

Analysis of COD, TS and VS

Analysis of COD, TS and VS was done every three days during the fermentation process. COD was measured by addition of $K_2Cr_2O_7$as as oxidizing agent to determine the amount of oxygen (mg O_2) required to oxidize organic substances contained in 1Lsample of water. By determining the amount of $K_2Cr_2O_7$, COD can be calculated[9].

Total solidswereidentifiedas the amount of solids in the organic material.The value of TS influencedonthe length of digestive processgreatly. TS analysis was done by introducing a 10 mL sample into the evaporating dish, followed by heatingin an oven at a temperature of 130 ^{o}C for 4 hours, and then weighingthe cooled dish. Total solid can be determined by calculating the weight difference of the samples before and after heating. Volatile solid (VS) is a solid part which turns into a gas phase on acidification and methanogenesis stages. VS indicated that how much organic material that can be converted into biogas.

Analysis of VFAs and biogas (CO_2, H_2, H_2S, and CH_4)

Methane wasanalyzed using a gas chromatography (Hewlett Packard, USA) with a flame ionization detector (FID). The chromatograph used a Agilent 19095P-Q04 HP Plot Q column roomates allowed to determine methane (CH_4) in a the mixture as a function of digestion time. The temperature of FID, oven and injector ports were 280^{o}C, 150^{o}C, and 275^{o}C respectively. The flowrate of 30 ml/min of Helium was used as carrier gas. The volume percentage of CH_4 was calculated by interpolating values from the calibration curve obtained with ultra pure CH_4. Biogas samples were analyzed by collecting the gas in venojeckand injecting to the column by syringe. The concentration of VFAs was analyzed using a gas chromatography (Hewlett Packard, USA)equipped with flame ionization detector and poraplot-Q041 direct which was working at 275^{o}C and flow rate of 45 mL/min.

H_2 and CO_2wereanalyzed using a gas chromatography (GC-2010 plus,Shimadzu, Japan) equipped with a thermal-conductivity detector (TCD). Nitrogen was used as carrier gas. With resident time of 6 minutes,inject volume was 0.2 mL. The temperature of Pre TCD, SPL-1, and column were 200 °C, 40 °C, and 30 °Crespectively.Theelectric current was 30 mA with pack column asmolecular sieve 13X (30 mx 0.53 mm i.d.; x 20 μm). Split was injection mode with column flow of 1.77 mL/min, linear velocity of 15 cm/s, purge flow of 3 mL/min.

Results and Discussion

Cellulosic Contents of Rice Straw

The chemical composition of rice straw from Madura Island used in this research has special composition as listed in Table1.

Table 1 :Chemical composition of rice straw

Compounds	In this study	Garrote et al. [9]
Cellulose	62.80 %	32-47%
Hemicellulose	26.07%	19-27%
Lignocellulose	3.74%	5-24%

Table 1 shows that the composition of rice straw used in this study was different with the result reported by Garrote et al. According to the previous study[10], the differences in chemical composition can be caused by differences in ash content and effect of relativity extraction by hot water at the time of analysis. Differences in chemical composition is also caused by the origin, type and maturity of raw materials which could affect the composition of the biomass.

Total Solid (TS) and Volatile Solid (VS)

Samples of liquid were taken from the reactor to be analyzed according to TS and VS parameters. Analysis of TS and VS were done every 3 days for 30 days during the anaerobic process by standard methods[11]. Figures 2 and 3 show TS and VS profiles of rice straw as fermentation substrate to the length of time (days).It canbe seen thatTS and VS for the both variables of RCand RCEdecrease significantly at the beginning of the process of anaerobic digestion due to degradation process of organiccompounds into monosaccharide,amino acids, alcohols,fatty acids, and many simpler organic compounds. The degradation process of organic compounds is assisted by strick bacteria such as *Bactericides, Clostridia* and facultative anaerob such as *Streptococcus*[12]. The content of TS and VSdecreased significantlywith the increasing growth of microorganism cellswhereas also supported by sufficient nutrients supply so that organic compounds were degraded by the microorganisms. After the 15th day,TS and VS decreasedfairly constant for all variables. It indicated that the amountof nutrients reduced while the number of bacteria remained and an eventually number of bacteriadecreased(bacterial death phase). The content of TS and VS in RCwere compared significantlyto other variables, i.e.19.11% and 45.87%respectively. Meanwhile, TS and VS inRCE were 18.39% and 27.93% respectively.

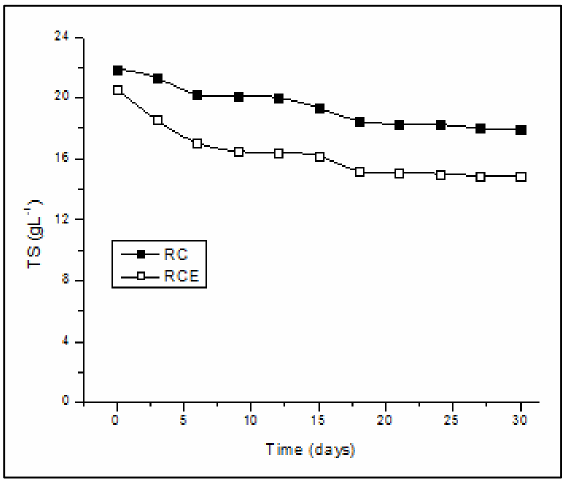

Figure 2 : Total solid (TS) profile for anaerobic digestion from cow dung microrganism and a mixed cow dung- EM for 30 days

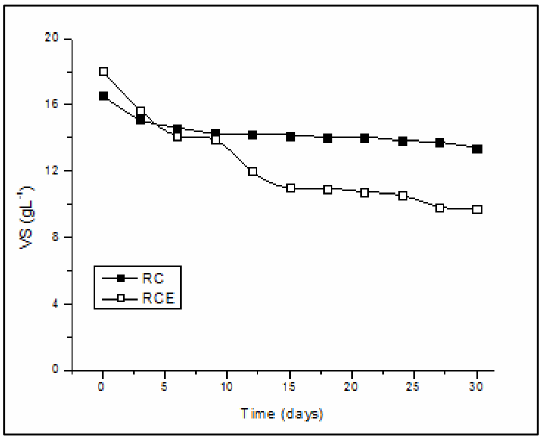

Figure 3 :Volatile sollid (VS) profile for anaerobic digestion from cow dung microrganism and a mixed cow dung- EM for 30 days

Production of volatile fatty acids (VFAs)

Samples of digester slurry were taken through the sampling valve, and then the samples were accommodated into eppendoff to separate precipitate to obtain filtrate. This filtrate was analyzed using a gaschromatography (GC) to determine VFAs content. VFAs analysis was done every 3 days for 30 days. In the study, VFAswere found in the form of acetic, propionic, and butyric acid because these acids were the main products in the process of methane formation. The results of VFAs analysis can be shown in Figure 4.

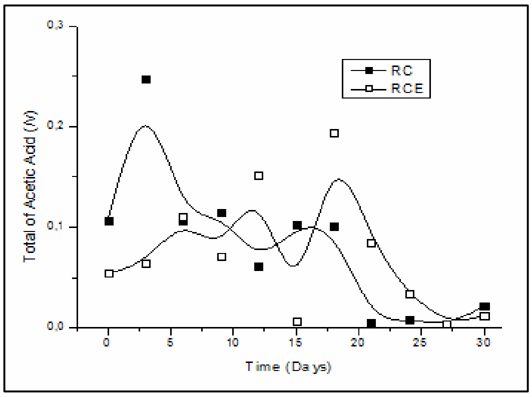

Figure 4 :The total production of acetic acid from cow dung microorganism for 30 days, symbols (□) acetic acid,(○)propionic acid, (Δ) butyric acid

Figure4 shows the total VFAs in each variables. The VFAsproducedwere obtained from the amount of acetic acid formed and the conversion calculation of propionic acid and butyric acid to acetic acid. Acetate can not only be produced through the fermentation of soluble organic compounds, but also through acetogenesis. Acetogenesis occurred at the stage of acid formation. At this stage, many acids and alcohols, such as butyrate, propionate, and ethanol produced during the establishment phase of acid can be degraded into acetic acid which can be used as a substrate by bacteria for methane production.It indicatedthatthe highestconcentration of acetic acid in RCwas 0.88% (v/v). The concentration of acid showed the quantity of methane producedfrom the conversion of acetic acid. Based on the analysis of methane,the highest concentration of methane produced in RCEwas 68718.30 ppm. This value was greater than the concentration of methane produced in RC, i.e50639.7 ppm. The production of acetic acid in RCwas comparable with the production of methane.

The increasing of acetic acid productionat each variablesindicated the increaseofacetogenic bacteria growth.While the decline on acetic acid concentration in a particular day indicated the formation process of methane from acetic acid. The concentration of volatile acids was observed as the quantity of biogas produced[13]. Datta et al.[14]reported that methane can be produced from two processes,i.e. acetic acid as feedstock and the other process which H_2 and CO_2 was used. At the conversion of H_2 and CO_2 into methane, formic acid, carbinol, and CO were also converted into methane. VFAswere the main product during the process of anaerobic methane digestion so that the most widely produced through acetic acid[15]. Acetate was an intermediate product in the anaerobic fermentationconverting 70%[16]of total product of methane during slurry fermentation. VFAswere converted into acetic acid and hydrogen by acetogenic bacteria. Acetic acid and propionic acid were the main product in biogas production by anaerobic process[17].

At the stage of acidogenesis, acidogenic bacteria (acid-producing bacteria) breaks down sugar molecules into VFAs(including butyrate, propionate, and acetate), CO_2, NH_3, H_2S, and H_2[18]. This type of reaction occured is shown in the equation from 1 to 5.

$$CH_3CH_2COOH + 2H_2O \longrightarrow CH_3COOH + CO_2 \quad (1)$$

$$CH_3CH_2COOH + ½\, H_2O \longrightarrow 7/4\, CH_4 + 5/4\, CO \quad (2)$$

$$2CH_3(CH_2)_2COOH + 2H_2O \longrightarrow 4CH_3COOH + CH_4 \quad (3)$$

....

$$CH_3COOH \longrightarrow CH_4 + CO_2 \quad (4)$$

$$4\, H_2O + CO_2 \longrightarrow CH_4 + 2H_2O \quad (5)$$

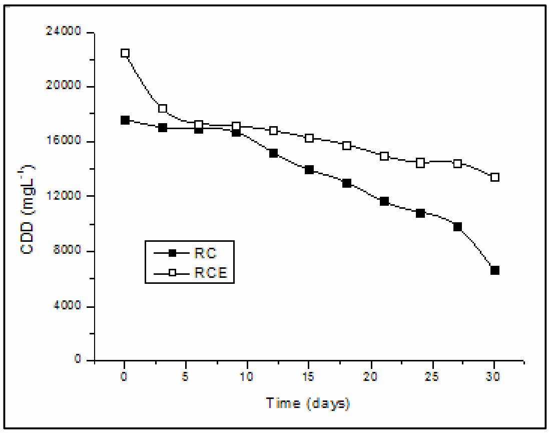

Figure 5 : COD profile of cow dung microorganism and a mixed cow dung-EM

Chemical Oxygen Demand (COD)

COD analysis was done every 3 days for 30 days. The results of the analysis are shown inFigure 5. COD decreased in rice straw substrate for each variable.COD in JP-KS of 15% and RCE of 15% were 62.34% and 40.54% respectively.COD in RC 15% was greater than that of RCE.As reported by Castrillon et al.[19],COD removal in cow dung was generally in the range of 51-79%. The COD removal indicated that biogas was produced.

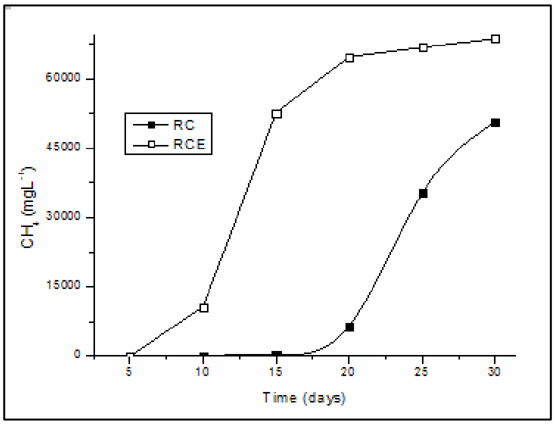

Figure6: CH_4 profile of cow dung microorganism and a mixed cow dung-EM.

Biogas Composition

Biogas composition (included CH_4, CO_2, H_2, and H_2S) was analyzed after 30 days of anaerobic digestion. The gas composition effected onthe heating value of biogas. Table 2 showed the comparation of biogas composition between every microorganism variables. The composition of biogas for cow dung microorganism was 38.54% CH_4,9.41 % CO_2,and 0.39%H_2.Witha mixed cow dung-EM, the composition of biogas was41.24%CH_4, 8.7% CO_2,and 0.36%H_2.

Table 2 :The comparation of biogas composition from cow dung microorganism and a mixed cow dung-EM.

Compounds	RC, %	RCE, %
CH_4	38.54	41.24
CO_2	9.41	8.7
H_2	0.39	0.36

Heating Value

Table 3 show that the heating value with a mixed cow dung microorganism-EMwashigher thanthat of cow dung microorganisms. Heating value in the cow dung microorganisms was13790kg/kJ, while with a mixed cow dung microorganism-EMwas15010kg/kJ.

Table 3. The heating value of methane based on biogas composition

	RC	RCE
Heating Value (kJ/kg)	13790	15010

Methana Yield

The calculation of methana yield was according to methane volume in normal m^3 (Nm^3) per $kgCOD_{removal}$. Normal m^3 is a gas volume unit at Standard Temperature and Pressure (STP) (Eg. 25^0C and 101,325 kPa as reference). Actually, methane of 350 mLwas produced from 1 g of COD[20].The methane yield with a mixed cow dung microorganisms-EM was higher than that of cow dung microorganisms. With cow dung microorganisms,methane yieldwas0.195 $Nm^3/kgCOD_{removal}$. While with a mixed cow dung microorganisms-EM,methane yield was 0.234 $Nm^3/kgCOD_{removal}$.

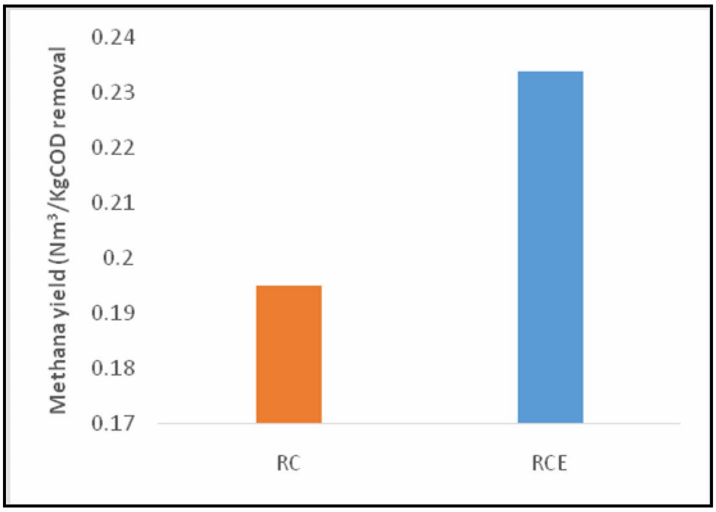

Figure 8 :The methane yield ofcow dung microorganism and a mixedcow dung microorganism-EM

Conclusion

The methana yield with cow dung microorganisms- EM was higher than that of cow dung microorganisms. It can be concluded thatEM (effective microorganism)affected on the quality of methane with the increase of methane from 38.54% to 41.24%.With cow dung microrganism,the composition of methane produced was 38.54%CH_4, 9.41% CO_2,and 0.39%H_2. With a mixed cow dung microorganisms - EM, the composition of methane produced was 41.24 %CH_4, 8.7% CO2, and 0.36%H_2.

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