

Effect of iron and magnesium addition for ethanol production from the conversion of palm oil mill effluent by anaerobic processes

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Abstract. Nowadays, crisis of the energy is the main problem in the world. Currently, most the energy resource derived from the fossil material that cannot be refurbished. Ethanol is an alternative fuel that content as a fossil fuels. Wastewater with the high concentration of the organic can be used for the ethanol production to replace foodstuff as a raw material. In this study, palm oil mill effluent (POME) with the concentration of COD is 24,500 mg/L has been used as a substrate. The purpose of this study was to determine the effect of the metal addition in the substrate metabolic pathways. Circulating batch reactor (CBR) is used with the flushing N_2 1L/min for 24 hours and continued operates for 72 hours by internal biogas. The additional variation concentration of Fe(II) ion are 0.5; 1.0 and 2.5 mg/L, and Mg(II) are 0.5 and 1.5 mg/L were added by combination. The results showed that the combination of Fe (II) 2.5 mg/L and Mg(II) 1.5 mg/L produced the highest ethanol concentration is 715.8 mg/L and degree of acidification (DA) 0.284-0.357. Another combination of Fe(II) and Mg(II) provide results for the ethanol production 463.7-689.7 mg/L with the rate of ethanol production is 1.09–26.5 mg/L/hour.

Keywords: anaerobic process, circulating batch reactor, palm oil mill effluent

1. Introduction

The limitations of fossil fuel being a serious problem while the world energy demand were increase every year. Biofuels such as biogas, bioethanol, biodiesel, bio-methanol, bio-oil, bio-hydrogen and bioelectricity are renewable energies with high quality fuel properties and can be utilized for various energy services [1]. Bioconversion of the wastewater with the high concentration of organic by anaerobic process can be produced biofuels which is can instead of fossil fuels [2]. Renewable resources including agricultural and industrial residues have been studied through microbial bioconversion processes.

Indonesia is the world's largest producer and exporter of crude palm oil (CPO). CPO's are one of the main commodities on agro industrial sector in Indonesia. The land area of palm trees increased every year to meet the Government target for crude palm oil production to 40 million metric tons per year in 2020 [3]. In 2014, the production of palm oil in the world reached 62.34 million tons, mostly from Indonesia (30.5 million tons) and Malaysia (19.9 million tons) [4]. The process of palm oil can produce the wastewater with the high concentration of organic. Nowadays, the potential of the wastewater are not utilized optimally, for example the treating of the wastewater released the methane into the environment about 32.9 kg/ton palm oil produced [5].



Wastewater from the palm oil mill effluent has a very high concentration of organic. BOD and COD concentration of the wastewater are 25,000-65,714 and 44,300-102,696 mg/L respectively [6]. Characteristic of the palm oil mill effluent shown in Table 1.

Table 1. Characteristic of raw palm oil mill effluent (POME) [6].

Parameter	Unit	Range
pH	-	4-5
Biological oxygen demand (BOD)	mg/L	25,000-65,714
Chemical oxygen demand (COD)	mg/L	44,300-102,696
Total solids (TS)	mg/L	40,500-72,058
Suspended solid (SS)	mg/L	18,400-46,011
Volatile solids (VS)	mg/L	34,000-49,300
Oil and grease (O and G)	mg/L	4,000-9,341
Ammonia nitrogen (NH ₃ -N)	mg/L	35-103
Total nitrogen (TN)	mg/L	750-770

Anaerobic treatment using microbe can produce the biofuels. In general, this processes divided into four main stages, namely hydrolysis, acidogenesis, acetogenesis and methanogenesis [7]. In every stages depend on the condition such as pH, microbe, temperature and so on. The presence of micro nutrients (metal addition) can increase the activity of alcohol dehydrogenase enzyme [8]. During the first stage, organic compounds (carbohydrates, proteins and lipids) are hydrolyzed and utilized by hydrogen producing bacteria to produce ethanol, H₂ and volatile fatty acids (VFAs) [9].

The aim of this study was to determine the effect of metal addition for the formation of ethanol and other product in acidogenic step. Ethanol and other intermediates products are formed during acidogenic fermentation by anaerobic mixed culture bacteria. The alcohol dehydrogenase (ADH) is the main enzyme for the ethanol production [10].

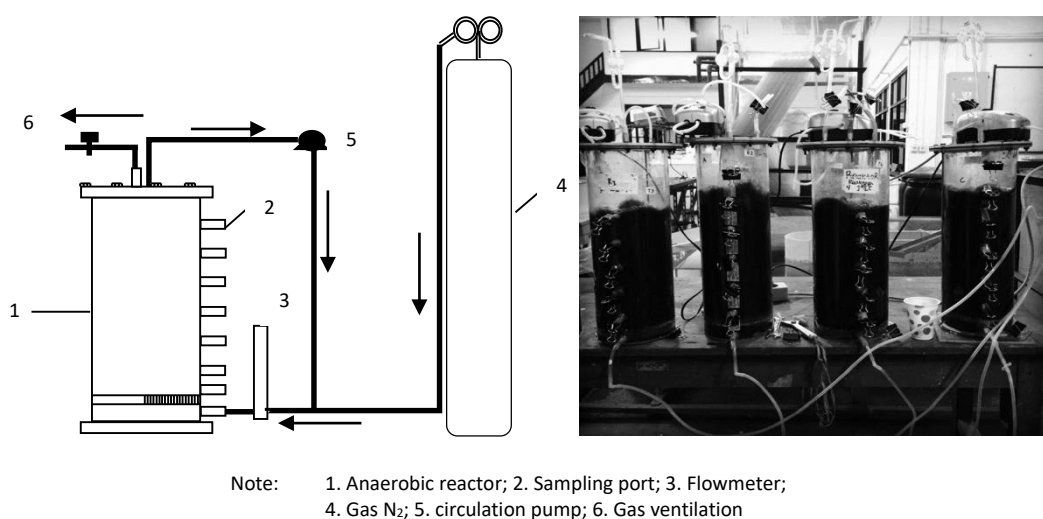
2. Research Method

2.1. Organic wastewater and biomass

The fresh raw palm oil mill effluent (POME) was collected from the influent basin of PT. Condong, Garut, Indonesia. In this study, biomass was taken from the sludge of palm oil mill effluent mixed by cow rumen with the ratio of 50:50 and has been acclimated to the POME. The acclimated biomass concentration used for the anaerobic batch reactor in terms of mixed liquor suspended solids (MLSS) and mixed liquor volatile suspended solids (MLVSS), were about 5.2 g SS/L and 3.98 g VSS/L respectively.

2.2. Analytical methods

Biological oxygen demand (BOD), chemical oxygen demand (COD), TSS, oil and grease, total nitrogen and pH were analyzed according to standard methods [11]. Analysis of volatile fatty acids by using HPL Ctype Hitachi inter faceD-7000 HSM with analytical column Aminex HPX-87H, ethanol by using GC Shimadzu17-A with column Analytic DB-Wax.

**Figure 1.** Anaerobic batch reactor.

2.3. Bioreactor and experimental design

Anaerobic batch reactor with the working volume of 5L were operated with the flushing N₂ 1L/min for 24 h and continued operates for 72 h by internal biogas (Figure 1). The reactor filled with a mixture of consisted of 20% (v/v) mixed culture bacteria and 80% (v/v) of POME as a substrate. The additional variation concentration of Fe(II) ion are 0.5; 1.0 and 2.0 mg/L, and Mg(II) are 0.5 and 1.5 mg/L were added by combination as shown in Table 2. Sample was collected every 6 h and analyze for ethanol and volatile fatty acid.

Table 2. Combination of magnesium (Mg) and iron (Fe) addition.

Metal addition (mg/L)		Reactor ID
Mg	Fe	
0	0	Control
	0.5	A
	1	B
	2.5	C
0.5	0	D
	0.5	E
	1	F
	2.5	G
1.5	0	H
	0.5	I
	1	J
	2.5	K

2.4. Calculation

The determination of the optimum conditions based on the efficiency and the formation of ethanol and a few other variables, namely :

$$\text{Degree of acidification (DA): } DA = \frac{(\text{TAV})}{\text{influent of soluble COD}} \frac{[\text{mgCOD/L}]}{[\text{mgCOD/L}]} \quad (1)$$

$$\text{Degree of ethanofication (DE) : } DE = \frac{(\text{Ethanol})}{\text{influent of soluble COD}} \frac{[\text{mgCOD/L}]}{[\text{mgCOD/L}]} \quad (2)$$

Acidogenic product equation Van Haandel and Van Der Lubbe (2012) :

$$\text{COD}_{\text{theoretical}} = 8 \cdot (4x + y - 2z) / (12x + y + 16z) \text{ gCOD} \cdot \text{g}^{-1} \text{C}_x\text{H}_y\text{O}_z \quad (3)$$

where : x = C; y =H and z = O

Formation rate product (mg COD/L/hour) :

$$= \frac{[\text{Product concentration}_{\text{end}} - \text{Initial concentration}_{\text{in}}]}{\text{time}} \frac{[\text{mgCOD/L}]}{[\text{hour}]} \quad (4)$$

3. Results and Discussion

Palm oil mill effluent (POME) was characterized as shown in POME also contains considerable amounts of nutrients such as nitrogen, potassium Table 3., magnesium, calcium, cadmium, copper, chromium and iron. The high concentration of carbohydrate, nutrients, lipids and minerals also found in POME [12].

Table 3. Characteristic of palm oil mill effluent (POME).

Parameter	Unit	Results
BOD	mg/L	12,450
Soluble COD	mg/L	17,300
Total COD	mg/L	24,500
TSS	mg/L	15,000
Oil and Grease	mg/L	185
Nitrogen Total	mg/L-N	250
Ethanol	mg/L	39.05
Volatile fatty acids	mg/L	361.07
pH	-	5.85

Ethanol production during the process shown in Figure 2. Ethanol was start to produce after 12 hr for each reactor with the combination of metal addition. Highest ethanol production occurs at the reactor with combination of 2.5 mg/L-Fe + 1.5 mg/L-Mg is 715.8 mg/L and the maximum rate of ethanol production is 33.33 mg/L/hr. The concentration of ethanol increased until 42 hr, then after 42 to 72 hr ethanol concentration are stable at 628.8 to 678.1 mg/L by the rate of ethanol production is 5.42 to 25.57 mg/L/hr. In other variations, typical of ethanol production is the same, ethanol concentration was increased until 42 to 48hr and then stable until the end of experiments. The highest ethanol concentration for each combination 0.5 mg/L-Mg + 0.5 mg/L-Fe; 0.5 mg/L-Mg + 1.5 mg/L-Fe; 0.5 mg/L-Mg + 2.5 mg/L-Fe; 1.5 mg/L-Mg + 0.5 mg/L-Fe and 1.5 mg/L-Mg + 1.5 mg/L-Fe are 463.7; 572.3; 689.7; 480.6 and 555.8 mg/L respectively. Ethanol production for the reactor without metal addition (control) has lower concentration than the reactor with metal addition, either individual addition of metal and combination. The highest concentration of ethanol in control is 312.7 mg/L. Highest concentration of ethanol production at the reactor with the individually metal addition are 292.3; 310.6; 422.4; 532.2 and 674.5 mg/L for the metal addition of 0.5 mg/L-Mg; 1.5 mg/L-Mg; 0.5 mg/L-Fe; 1.5 mg/L-Fe and 2.5 mg/L-Fe respectively. Iron (II) addition can enhance of ethanol production. The addition of Fe (II) 200 mg/L will produce ethanol with the same amount without addition of Fe (II), addition 400 mg/L will produce ethanol 122 times lower than without addition of Fe (II) and addition of 600 mg/L will not produce the ethanol [13]. Nitrogen flushing can shift metabolism pathway to be acetyl CoA production then it will be oxidized to be ethanol by alcohol dehydrogenase enzyme. Addition of Fe (II) 10 times that of the control can increase the activity of alcohol dehydrogenase enzyme and can enhance ethanol production [8].

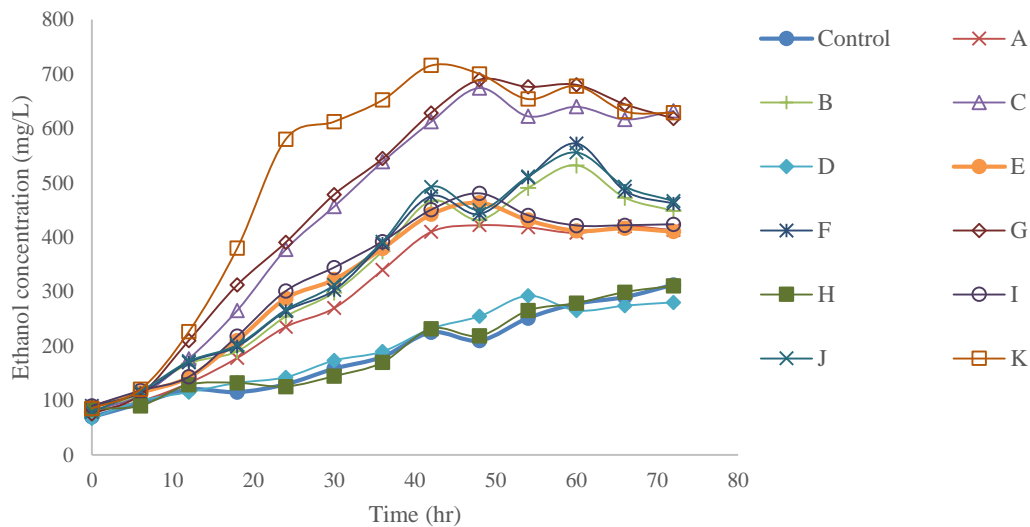


Figure 2. Ethanol formation from the variation of metal addition.

Volatile fatty acids (VFA's) such as acetate, propionate, butyrate, valerate and biogas (H_2 , CO_2 and CH_4) was produced during the experiments. Acetate and propionate is acidogenic product most widely produced in the anaerobic processes. Degree of acidification (DA) was calculated by the comparison of VFA's production and soluble COD in the influent. In appropriate concentration of metal addition, micronutrient such as Fe, Mn, Zn, Co, Cu, Mo, Se and Ni are required by enzyme as co-factor, but at the concentration does not exact this is can be toxic for microorganism to convert the substrate [14]. In Table 4 shows the acidogenic product in degradation of wastewater by anaerobic processes.

Table 4. Acidogenic product in degradation of wastewater by anaerobic processes.

Parameter	Unit	Metal addition (mg/L)						
		Mg (II)	0.5	0.5	0.5	1.5	1.5	1.5
		Fe (II)	0.5	1.5	2.5	0.5	1.5	2.5
Ethanol *	mg/L		463.7	572.3	689.7	480.6	555.8	715.8
Acetate *	mg/L		1885.7	1765.4	1729.0	1766.2	1810.1	1730.2
Propionate *	mg/L		1007.8	1229.5	1182.0	1753.7	1410.0	1170.2
Butirate *	mg/L		533.5	452.4	801.8	653.2	662.7	702.6
Valeric acid *	mg/L		201.8	236.0	242.8	219.7	235.8	188.0
Ethanol formation rate *	mg/L/hr		14.80	17.62	27.08	21.60	28.81	33.33
Degree of Acidification (DA)*	-		0.284	0.292	0.323	0.357	0.332	0.305
Degree of Ethanofication (DE)*	-		0.056	0.069	0.083	0.058	0.067	0.086

* maximum results

4. Conclusion

The addition of Mg (II) and Fe (II) are influenced to the ethanol production. The addition of 1.5 mg/L-Mg + 2.5 mg/L-Fe has produced ethanol with the highest concentration is 715.8 mg/L. Volatile fatty acid such as acetate and propionate are the highest product formation from the anaerobic process.

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