Biodiesel Production from Chlorella vulgaris via Homogenous Acid Catalyzed In situ Transesterification with Microwave Irradiation

Ummu Kalsum*1, Mahmuddin2, Mahfud Mahfud3 & Ahmad Roestiyadi4
1Chemical Engineering Department, Sepuluh November Institute of Technology, Surabaya, Indonesia
2Mechanical Engineering Universitas Muslim Indonesia Jl Urip Sumohardjo Kmr 05, Makassar, Indonesia 90231.
Email: umkal29@gmail.com

Abstract. Acid catalyzed In situ transesterification under microwave irradiation is one of method for producing biodiesel from microalgae which is an alternative to replace fossil fuel energy. The present study investigated the the effect of microwave irradiation on the acid catalyzed in situ transesterification of Chlorella vulgaris into fatty acid methyl ester. In situ transesterification carried out using sulfuric acid catalyst with different variables such as catalyst concentration, reaction time, biomass to methanol (wt/vol) ratio and microwave power. Based on the experiment results, the optimal yield was 31.56% with reaction time was 70 minutes. From GC-MS analysis obtained that Chlorella vulgaris methyl ester profile is a medium chain fatty acid, MCFA which consist of Saturated Fatty Acid (SAFAs), Monounsaturated Fatty Acid (MUFAs) and Poly Unsaturated Fatty Acid (PUFAs).

Keywords; Chlorella vulgaris; microwave; acid catalyzed; in situ transesterification

1. Introduction

The transportation and energy sectors are the major cause sources, responsible for the decreasing of fossil fuels and increasing of green house gas (GHC) emissions in environment. GHC as the side effect of fossil fuels energy use has the largest contribution to global warming and environmental damage. Green house gas emissions not only contributes to global warming but also more environmental effects and aspects in human life. In facing this problem, clean energy procurement and renewable energy resources are big challenges for human as far as their life. In Indonesia, fossil fuel energy still dominate in all aspects. It needs an energy treatment paradigm that purpose diversification and conservation of energy, therefore new and renewable energy role will be a main support for future energy. Biodiesel from microalgae is one of a renewable energy which produced from renewable resources and expected can give contribution to replace fossil fuel energy, decrease gas house emission and increase a protection from energy supply. It states as the third generation biofuel and has higher potential as a substitution of diesel fuel than biodiesel produced from raw material performance and fuel aspect (Dinh LTT et al, 2009).

Biodiesel from microalgae produced by transesterification algal oil and methanol using base or acid catalyst (Canalci 1999; Chisti 2007; Demirbas 2009). Several researches have been done to eliminate extraction process with in situ transesterification method or direct transesterification. In situ transesterification process different with conventional process which is oils saved in material contact directly with alcohol alkalized or alcohol acidified that as pre extraction reaction with alcohol. In this process, extraction and transesterification take place simultaneously and alcohol act as a solvent and reagent. In situ transesterification for biodiesel production from microalgae can be carried out using both homogenous and heterogeneous acid and base catalyst such as KOH, NaOH, HCl and H2SO4. E.A. Ehime et al (2010), Johnson and Wen (2009), Haas and Wagner (2011) have conducted in situ transesterification using acid catalyst with conventional heating method. The use of base catalyst in in situ transesterification of Chlorella vulgaris also reported with maximum FAME 77%, methanol to microalgae molar ratio 600:1 and reaction time on 75 minutes.
Heating method development have been done for biodiesel production by in situ transesterification using microwave as energy source. Microwave oven works by the exposure of microwave with frequency 2450 MHz and length 12.24 cm. Microwave will be absorbed by materials through dielectric heating. Some molecules include water are the electric dipoles which have positive and negative poles and cause the molecules rotate and generate heat (Shinmen, Zhuge., 2008). The heating system by microwave irradiation causes dielectric properties of polar mixtures and ionic components of oils interact molecularly to produce intermolecular mixture. The agitation causes the increasing of alcohol and oil molecules collide each other, therefore the interaction between microwave and materials in molecular scale causes reaction time to be faster.

Heat transfer from microwave into materials take places through dipolar polarization, ionic conduction and interfacial polarization that lead to localization and rapid superheating from materials. In the in situ transesterification, since microwave radiation exposed into material mixture, alcohol molecules produce a dipole moment that try to align with the applied electric field. The dipol moments moves constantly following the electric dipoles and the continues movement results in friction and release the thermal energy. If energy interacting with molecules at a rapid rate, heat generated can be resulted at microscopic level in short time. In this condition, the temperature of molecules may higher than bulk temperature saved in the reaction mixture and there will be a continues superheating localization (Guerra., 2014).

In this work, we have conducted an experiment for producing biodiesel from microalgae using H$_2$SO$_4$ catalyst. Some parameters such as reaction time, ratio methanol-microalgae, catalyst concentration and microwave power have been studied to determine he optimum condition of process.

2. Methodology

2.1 Materials

Process stage involves characterization of microalgae by lipid extraction and in situ transesterification of microalgae using sulfuric acid and microwave as a heating source. *Chlorella vulgaris* was purchased from Balai Budi Daya Air Payau (BBAP) Jepara Jawa Tengah. Methanol (>99% purity), n-hexane (98%) and H$_2$SO$_4$ were purchased from Brataco Chem.

2.2 Lipid extraction of microalgae

Ten grams of dry algae were extracted by soxhlet using n hexane solvent. Soxhlet extraction carried out for 3.5 hours with 26 circulation. parameters such as reaction time, ratio methanol-microalgae, catalyst concentration and microwave power have been studied to determine he optimum condition of process.

2.3 In situ transesterification of microalgae

In situ transesterification conducted in a microwave reactor equipped with a reflux condensor with design as in figure 1. Microwave reactor is a microwave type Electrolux EMM2007X with maximum power 800 Watt. Reflux condensor function is to provide condensation of solvent mixture evaporation and magnetic stirrer was used to keep the uniform of mixing during the experiment.
Figure 1. Microwave reactor design for in situ transesterification: 1. Reactor 2. Stative clamp 3. Microwave 4. Condensor 5. Power setting 6. Time Setting 7. Magnetic stirrer.

Ten grams of microalgae powder were mixed with the mixed homogenous solution of methanol and sodium sulfate catalyst. The mixture was then exposed with microwave irradiation with power 450 watt: reaction times of 10 to 50 min; catalyst concentration 20% and dry algae to methanol (wt/vol) ratios of 1:10. After filtration, separation and purification stage, the samples were weighted and analysis by GC MS.

3. Results

3.1 Extraction of Microalgae Lipids

From the extraction process by soxhlet obtained that total lipid of *Chlorella vulgaris* can be extracted from 10 grams of dry algae powder was around 2.36 g/gr microalgae. The total lipid obtained after extraction for 3.5 hours with 29 circulations. GC MS analysis on microalgae lipids shows that fatty acid components of *Chlorella vulgaris* consist of group of Saturated Fatty Acid: palmitic acid (C16:0); stearic acid (C18:0), Mono Unsaturated Fatty acid; oleic acid (C18:1) and Poly Unsaturated Fatty Acid; linoleic acid (C18:2).

3.2 Acid Catalysed In Situ Transesterification of Microalgae and Effect of algal biomass to methanol ratio

Figure 2 shows the correlation between alcohol volume to crude biodiesel which transesterified using H$_2$SO$_4$ 30%, microwave power 450 W for 30 minutes. From the graph shows that a significant increase of yield occurred on the use of methanol 50 ml to 150 ml. The optimum yield was achieved on ratio 1:15 that indicates the increase of yield with the increase of biomass: methanol ratio. However, on ratio 1:20 methyl ester yield decreased immediately on 22.93%. It indicates that the increasing of ratio biomass to methanol in the in situ transesterification with H$_2$SO$_4$ 30% under irradiation microwave is ineffective and does not lead to an increasing of yield.
Figure 2. Effect of alcohol volume on crude biodiesel yield (H₂SO₄ 30%, power 450 Watt, reaction time 3 minutes).

3.3 Effect of time and catalyst concentration on Yield of Crude Biodiesel.

Figure 3 shows the effect of time to the formation of microalgae methyl ester by in situ transesterification with various catalyst concentration.

Figure 3. The effect of reaction time to crude biodiesel yield power 450 watt, ratio biomass: microalgae 1:15 (w/v), H₂SO₄ 30%, 50% and 100%.

The graph shows that yield of crude biodiesel increased significantly with the increasing of reaction time. In the use of H₂SO₄ 30%, yield of crude biodiesel increased until 70 minutes with was 31,56% and after that it went to 30,98% with a slightly decrease in 90 minutes. In contrast, the moderately increase occurred in the use of H₂SO₄ 50% during reaction with was 24,97% of yield reached in 90 minutes. The use of higher catalyst concentration also indicates the increasing of yield with the increasing of time as showed in H₂SO₄ 100%, however after 50 minutes yield decreased rapidly to 23,49% in 90 minutes. From the graph shows that the optimum reaction time was in 70 minutes with 31,56% as the highest of yield and the use of 30% of catalyst concentration is more effective than the use of higher catalyst concentration.

3.4 Effect of microwave power on yield of crude biodiesel.

From the experiment shows that the increasing of power has a big influence on the in situ transesterification process. In the first 30 minutes of reaction, the usage of 300 Watt, 450 Watt and 600 Watt indicates the significant increase with the increasing of yield. The increasing of yield occurs until 70 minutes with 26,32%, 31,56%, 24,96% of yield respectively. After this time, yield of both powers 450 Watt and 600 Watt decreased in 90 minutes. However, in the usage of 300 Watt, yield of crude biodiesel increased significantly from 30 minutes to 70 minutes and continue to increase with the slightly increase in 90 minutes. The study on the effect of microwave powers to yield of
microalgae crude biodiesel obtained 450 watt as the optimum power with 31.56% of yield. It was the highest yield that achieved after 50 minutes of reaction using 30% H$_2$SO$_4$.

![Figure 4. The effect of microwave powers on yield of microalgae crude biodiesel H$_2$SO$_4$ 30%; ratio biomass:methanol 1:15 (w/v)).](image)

4. Discussion

4.1 Characterization of Microalgae Lipids

Characterization of algae lipids aims to specify fatty acid compositions of microalgae which carried out using soxhlet extraction. The soxhlet extraction process using n hexane as a non polar solvent that useful to break the hydrophobic component of microalgae. This process occurs by interaction between non polar solvent with neutral lipid components in microalgae (Mubarak et al, 2015). The extraction method rely on the ability of solution to penetrate microalgae cell structure and attract the lipid components from the cell matrix. From GC MS analysis shows that the fatty acid components are various from C$_{14}$ to C$_{18}$ that known as saturated fatty acid (SAFA), unsaturated fatty acid (MUFA) and polysaturated fatty acid (PUFA). Lipid components are dominated with unsaturated fatty acid (MUFA) which is oleic acid as the prominent fatty acid.

4.2 Effect of Biomass to Methanol Ratio on Yield of Crude Biodiesel

Algal biomass to methanol ratio has a significant effect on in situ transesterification reaction. In the in situ transesterification process methanol has a double function as a solvent to extract microalgae lipids or oils (Mulbry et al, 2009) and also as a reactant during transesterification process. From the stochiometry ratio indicates that transesterification needs 3 moles of alcohols and 1 mole of triglycerides to form 3 moles of methyl esters and 1 mole of glycerol. Since the equilibrium needs to be shifted, an excess alcohol required to push reaction to the right thereby algal biomass to methanol ratio used to be higher. A large amount of methanol facilitates contact between methanol and microalgae maximally and results the greater of yield. The use of excess methanol increases the absorption capacity of material microwave that improve excitation of molecule in materials and induce a rise of temperature. The decreasing of yield in algal biomass-methanol ratio 1:20 indicates that the increasing of ratio biomass to methanol in in situ transesterification with H$_2$SO$_4$ 30% under irradiation microwave is uneffective.

4.3. Effect of time on Yield of Crude Biodiesel

Reaction time is one of factors that effect on in situ transesterification under microwave irradiation. Figure 3 displays the correlation between reaction time and yield of crude biodiesel. The graph shows that yield of crude biodiesel increased with the increasing of time during reaction using various catalyst concentration. One factor which causes the increasing of yield during in situ transesterification process is the increasing of contact time between biomass with methanol. The length of contact time between both materials causes an increasing of damaged cell walls and
triglycerides released into the solvent and also present a sufficient contact time for reactant to interact and produce biodiesel (P.D. Patil et al, 2011).

The same opinion also reported by Guerra et, 2013 who argue that microwave accelerates organic synthesis with the increasing of rate of reaction. It states by the Arrhenius Law’s that with the longer of reaction time, the contact between molecules will be greater. However, the longer of reaction time also can be unefficient and leads to the over heating on the reaction, loss of solvent in large quantities, by product formation and loss of energy during the process (Patil et.al, 2011).

4.4. Effect of Catalyst Concentration on Yield of Crude Biodiesel

Catalyst on in situ transesterification needs to accelerate rate of reaction and reaction time. The use of catalyst can refer the reaction therefore it may diminished by product and decreased energy of activation. In this research, the use of catalyst studied on 30%, 50% and 100% with different reaction time and microwave powers. The highest yield was 31.56% resulted from in situ transesterification using 30%. Profile the effect of catalyst concentration indicates a decline trend on the higher concentration of H\textsubscript{2}SO\textsubscript{4}.

Ramadhan et al., 2005 argues that the higher concentration of acid can affect negatif effects such as colors become more dense and intermediate reaction between excess of H\textsubscript{2}SO\textsubscript{4} with methanol that cause a decreasing amount of methyl ethyl ester formed. The same argumentation also stated by Ejikeme et al., 2010 that the high of acid concentration gives a negative effect by the formation of ether through alcohol dehidration.

4.5. Effect of Power on Yield of Crude Biodiesel

The effect of the usage different power on yield biodiesel shows the significant influence in in situ transesterification process. From the experiment represents that the application of low power result the increasing of yield significantly. In this application, reaction mixture can interact maximally because of lack of solvent evaporated during the reaction. The different phenomenon occurs on the higher microwave power usage. The application of power 600 watt on the in situ transestrification leads to the rising of temperature which produce a higher of yield with the increasing of time. The microwave power may acts as a driving force and produce a heating localization to ease in disrupting the matrix of microalgae, therefore soluts are easier to diffuse out and dissolve in the solvent. Furthermore, energy transferred by microwave into material through dipolar polarization, ionic conduction and interfacial polarization which cause heating localization in materials and accelerate the reaction (Quitan et al., 2011).

4.6. Fatty acid Methyl Ester Analysis

From the GC MS Analysis of Fatty Acid Methyl Ester obtained that FAME composition of Chlorella vulgaris consist of esters of lauric acid (C12:0), miristyc acid ester (C14:0), palmitic acid ester (C16:0), palmitoleat acid (C16:1), stearic acid (C18:0), oleic acid ( C18:1n9), vecenic acid (C18:1n7), linoleic acid (C18:2), linoleidic acid (C18:2), arachidonic acid (C20:4) and eicosapentanoic acid (C20:5)

Fatty Acid Methyl Ester of Chlorella vulgaris is a medium chain fatty acid, MCFA which are consist of saturated fatty acid, SAFAs and unsaturated fatty acid, MUFAs and PUFAs. Saturated fatty acid, SAFAs dominates the composition of FAME with the percentage around 65,95%; MUFAs, 9,94% and PUFAs, 15,84%.

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