

The effects of (sulfuric acid concentration, time, temperature) in hydrolysis and (type, weight of yeast) in fermentation process to bioethanol yield in the making of bioethanol from peat

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Abstract

The Indonesian Government's policy regarding the development of alternative energy sources as a substitute for fuel oil has been established bioenergy as an alternative fuel. Bioenergy is energy derived from biofuels. Common biofuel is bioethanol. Bioethanol burning is cleaner than fossil fuels which means reducing greenhouse gas emissions. Therefore, further research is needed on biomass which can be used as raw material for bioethanol production and also the process stages that affect on bioethanol production. The material that can be a substrate for bioethanol production is lignocellulose material. One of the lignocellulosic biomass is peat. This study aims to review the effect of the stages of the process in making bioethanol from peat, namely the delignification process, hydrolysis and fermentation of ethanol produced.

Keywords: Peat Soil; Delignification; Acid Hydrolysis; Bioethanol.

1. Introduction

Peat is the surface organic layer of a soil, consisting of partially decomposed organic material, derived mostly from plants, that has accumulated under conditions of waterlogging, oxygen deficiency, acidity and nutrient deficiency. In the taxonomic key to the soil, peat soil is a type of histosol, which is a soil formed from sediment of organic matter derived from the accumulation of plant residues with a minimum depth of 40 cm [1].

Indonesia ranks the fourth largest peat swamp land after Russia, Canada and the United States with an area of 14.9 million ha. Whereas in South Sumatra itself, the area of peat land reaches 1.2 million ha [2].

The function of peat ecology is as a carbon sink, water storage, climate regulators and sources of biodiversity. Conversion of the peat land into agricultural land and plantations will result in changes in its ecological function so it will cause the environmental impacts, especially the increase in CO₂ emissions released by peat lands. This is believed to be one of the factors causing global warming, climate change and rising sea levels [3]. Therefore, peat soils need to be used in other ways, one of which is to produce bioethanol.

Peat soil has a lignocellulose composition which is a potential raw material for the manufacture of ethanol from cellulose fibers which have low emission levels. Lignocellulose components in peat soils are 0.2-10% cellulose, 1-2% hemicellulose, and 64-74% lignin. Cellulose is a material that is rich in carbon. The carbon contained in cellulose can be utilized in the process of microbial fermentation. In this case, cellulose can be used to produce ethanol by fermentation using *Saccharomyces cerevisiae*.

1.1. Peat soil

Peat is formed by the accumulation of tropical vegetation residues that are rich in lignin and cellulose content [4]. Peat contains organic material that cannot be directly utilized because it is still in the form of complex compounds. One of the complex compound is cellulose. Cellulose is a linear polymer that is larger than 1000 long glucose subunits with 1.4b-β bonds [5].

Peat soils formed from tropical swamp forest vegetation with heterogeneous composition, consisting of logs, twigs and coarse roots that are still similar to the original plants.



Fig. 1: Peat Soil.

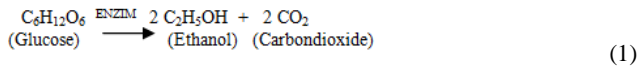
1.2. Bioethanol

Bioethanol is ethanol derived from biological sources. Bioethanol is sourced from potential carbohydrates as raw materials such as sugar cane, sap, sorghum, cassava, arrowroot, sweet potato, sago, corn, straw, corncob and wood. After going through the fermentation process, ethanol is produced (www.energi.lipi.go.id) [6].

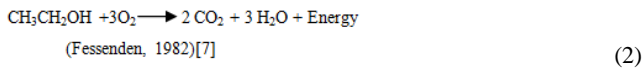
Ethanol is an organic compound consisting of carbon, hydrogen and oxygen, so that it can be seen as a hydrocarbon compound that

has a hydroxyl group with the formula C_2H_5OH . Ethanol is a liquid, colorless, odor-specific substance, flammable and volatile, can mix in water in all comparisons. Ethanol combustion is cleaner than fossil fuels which mean reducing greenhouse gas emissions. This is the most significant advantage of ethanol for the environment compared to fossil fuels.

a) Bioethanol Formation



b) Bioethanol Combustion



1.3. Making bioethanol

The stage of making bioethanol is carried out through a process of delignification, hydrolysis, fermentation and purification. Raw material preparation is done to get glucose. Glucose is obtained through 2 stages, namely delignification and hydrolysis. At the delignification stage it will produce cellulose. Cellulose will be processed further by the process of hydrolysis so that glucose will be produced.

1.3.1. Delignification

Lignocellulose ingredients generally consist of cellulose, hemicellulose and lignin. Cellulose is naturally bound by hemicellulose and protected by lignin. This lignin-binding compound causes lignocellulose materials to be difficult to hydrolyze [8].

Lignin is a complex molecule composed of phenyl propane units that are bound in a three-dimensional structure. Lignin is the strongest material in biomass. Lignin is very resistant to degradation, biologically, enzymatically and chemically.

In making ethanol from the peat, the cellulose is used so that lignin in the wood must be removed. The process of separating or removing lignin from cellulose fibers is called delignification or pulping. Delignification is carried out using alkaline solutions (chemically) because this solution can damage the structure of lignin in the crystalline and amorphous parts and separate part of hemicellulose.

1.3.2. Hydrolysis

Hydrolysis aims to break down the lignin bond, eliminate lignin and hemicellulose content, damage the crystal structure of cellulose and increase the porosity of the material [9]. The Damage of cellulose crystal structure will make it easier to break down cellulose into glucose. In addition, hemicellulose also decomposes into simple sugar compounds: glucose, galactose, manose, hexose, pentose, xylose and arabinose. Furthermore, these simple sugar compounds which will be fermented by microorganisms produce ethanol [10].

The parameters of acid concentration, temperature and hydrolysis time are very crucial in the process of hydrolysis so as to minimize the product in the hybrid which can ultimately increase the yield of ethanol in the fermentation process [11]

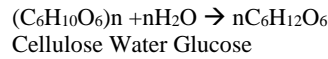
a) Acid Hydrolysis

In the method of acid hydrolysis, lignocellulose biomass is exposed to acids at certain temperatures and pressures for a certain time, and produces sugar monomers from cellulose and hemicellulose polymers. Some of the acids commonly used for acid hydrolysis include sulfuric acid (H_2SO_4), perchloric acid, and HCl. Sulfuric acid is the most studied acid and is used for acid hydrolysis. Acid hydrolysis can be classified into: concentrated acid hydrolysis and dilute acid hydrolysis (Taherzadeh & Karimi, 2007) [12].

Cellulose hydrolysis using acids was first commercialized in 1898 (Hamelinck, Hooijdonk, & Faaij, 2005) [13]. The main advantages

of hydrolysis with dilute acids are, no need for acid recovery, and no acid loss in the process (Iranmahboob et al., 2002) [8]. Generally, the acid used is H_2SO_4 or HCl (Mussatto and Roberto, 2004) [11] in the range of 2-5% concentration (Iranmahboob et al., 2002; Sun and Cheng, 2002), [8,9] and reaction temperature $\pm 160^\circ C$.

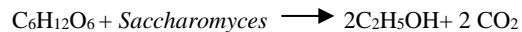
The breakdown of sugar molecules, complex carbohydrates and cellulose into the form of monosaccharide molecules is done by boiling solutions containing carbohydrates with acidic solutions:



1.3.3. Fermentation

Fermentation is the process of breaking organic compounds, especially sugar, fat) by microorganisms in anaerobic conditions to produce simpler organic products (Abercrombie, 1993) [14].

Ethanol fermentation is a biological process that involves microorganisms to convert organic matter into simpler components. Microbes that are often used in the fermentation process are *Saccharomyces cerevisiae*.



1.3.4. Purification (distillation)

To separate ethanol from fermentation can be done by distillation. Distillation is a separation method based on boiling point differences. Distillation separates components - volatile components of a liquid mixture by evaporating them (separating the heat agent) followed by steam condensation that forms and holds the condensate produced. The vapors released from the mixture are referred to as free vapor, the condensate that falls as a distillate and the part that does not evaporate is called residue [15]. This process is carried out to extract alcohol from fermentation. Distillation can be carried out at $80^\circ C$ because the boiling point of the alcohol is $78^\circ C$ while the boiling point of the water is $100^\circ C$.

1.4. Previous research

Making bioethanol from peat soil with hydrolysis and fermentation processes, by Dr. Kiagus Ahmad Roni, ST., MT., Merisha Hastarina, ST., M.Eng. dan Rully Masriatini, ST., MT., 2017, [16] University of Muhammadiyah Palembang. The method used is dilute acid hydrolysis followed by fermentation on peat soil. Hydrolysis uses sulfuric acid (H_2SO_4) and ferments using bread yeast (*Saccharomyces cerevisiae*).

1.5. Research design

1.5.1. Peat soil sample preparation

Peat soil is taken from the peatland and then washed with water to separate the soil and peat. After washing, the peat is dried in the sun and in an oven at $70^\circ C$ to remove the water content. Then the size of the blender is reduced by 3 mm or 3-6 mesh.

1.5.2. Delignification process

1) Addition of NaOH to Peat Soil

A total of 100 gr of peat soil that has been dried in the treatment using 200 ml of NaOH solution 10% for 30 minutes at a temperature of $120^\circ C$.

2) Washing Peat Soil Deposits with Aquadest

From the results of the delignification of peat soil, the peat dregs are found to be wet with pH 9. The peat soil is then washed with aquadest until the pH of the peat soil becomes neutral (pH 6.5 - 7.5).

1.5.2. Sulfuric acid hydrolysis

At this stage peat soil dregs that have been washed with distilled water to neutral levels, then hydrolyzed with 200 ml of H_2SO_4 solution with a concentration (1%, 1.5%, 2% and 2.5%) for (30 minutes, 60 minutes, 90 minutes and 120 minutes) with temperatures (100, 120, 140, 160 and 180 °C) then filtered. In this process, we get acidic peat soil with pH 2. Then add NaOH until it reaches pH 5.

1.5.3. Bioethanol manufacturing procedure

The process of making bioethanol is done by fermentation. At this stage peat soil dregs will be fermented with as much bread yeast as possible. Preparing both types of yeast are yeast bread and yeast tape with a variation of yeast weight of 5%, 10%, 15%, 20% and 25% (from the weight of the feed) and with as much urea 10 gr as nutrients. In this process the fermented peat soil will be left for 3 days.

2. Research methods

2.1. Research implementation

Peat soil samples from the Talang Keramat Palembang area were washed with water, dried in the sun and dried in oven at 70 °C for 20 minutes to remove the water content. Then, the dried peat in the treatment using NaOH solution then filtered and washed with aquadest to neutralize the pH. Then hydrolysis using H_2SO_4 acid and continued with fermentation using yeast bread and yeast tape.

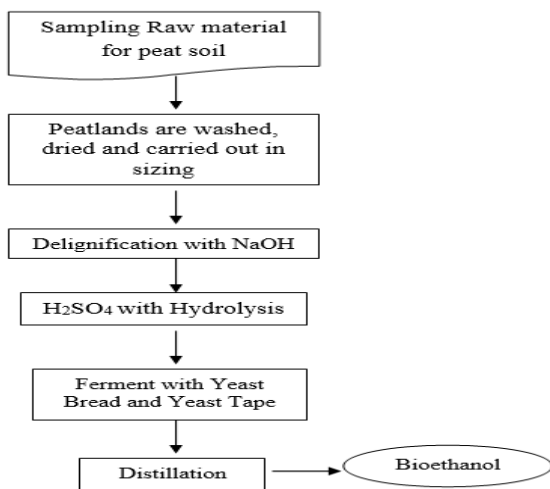


Fig. 2: Flowchart of Bioethanol Making Process from Peat.

3. Results and discussion

3.1. Preparation stage

Peat soil is taken from peatlands, and weighing 100 grams, then dried in an oven with a temperature of 70 °C for 20 minutes and then smoothed to 3 mm or 3-6 mesh.

3.2. Treatment stage

3.2.1. Addition of NaOH to peat soil

A total of 100 grams of peat soil that has been dried in the treatment using 200 ml of NaOH solution 10% for 30 minutes at a temperature of 120 °C then filtered. In the above process can produce peat soil with a wet level with a pH of 9.

3.2.2. Washing peat soil deposits with aquadest

In this process the treated peat soil using 200 ml of NaOH solution and obtained wet peat soil with pH 9 will be washed with distilled water 10 times to the level obtained on peat soil to neutral (pH 6.5-7, 5).

3.2.3. Sulfuric acid hydrolysis

At this stage the peat soil has been washed with distilled water to a neutral level, then the peat soil is hydrolyzed with 200 ml of H_2SO_4 solution with concentration (1%, 1.5%, 2% and 2.5%) during (30 minutes, 60 minutes, 90 minutes and 120 minutes) with temperatures (100, 120, 140, 160 and 180 °C) then filtered. In this process, you will get acidic peat soil with pH 2. Then add NaOH until it reaches pH 5.

3.2.4. Fermentation

The process of making bioethanol is done by fermentation process. In this process peat soil that has gone through the delignification stage, hydrolysis with acid will be fermented with yeast bread and yeast tape with a variation of yeast weight of 5%, 10%, 15%, 20% and 25% (from the weight of the feed) and accompanied by as much urea 10 gr as nutrients. Fermentation will be carried out for 3 days (72 hours).

3.2.5. Distillation

After the fermentation stage, the purification stage (distillation) is carried out. At this stage the hydrolyzed peat soil that has been fermented will be distilled with a temperature of 80 °C. In this process it will get the solution resulting from the fermentation of peat soil as bioethanol.

3.3. Effect of sulfuric acid concentration and hydrolysis time on % bioethanol yield

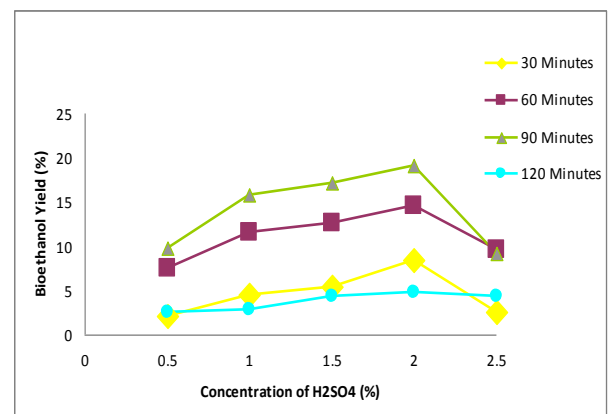


Fig. 3: Graph of the Effect of Hydrolysis Time and Acid Concentration on % of Bioethanol Yield.

In Figure 3 above shows a graph of the effect of hydrolysis time and acid concentration on % Bioethanol yield that produced from fermentation. For the length of the hydrolysis time that produces the highest yield of ethanol is at 90 minutes hydrolysis time is 19.16%, while the hydrolysis time is 120 minutes, the resulting ethanol yield tends to decrease at 11.91%. This is due to the fact that equilibrium has occurred for 90 minutes. For variations in sulfuric acid, 2% sulfuric acid concentration gives the highest yield.

3.4. Effect of hydrolysis temperature and yeast weight on % of bioethanol yield

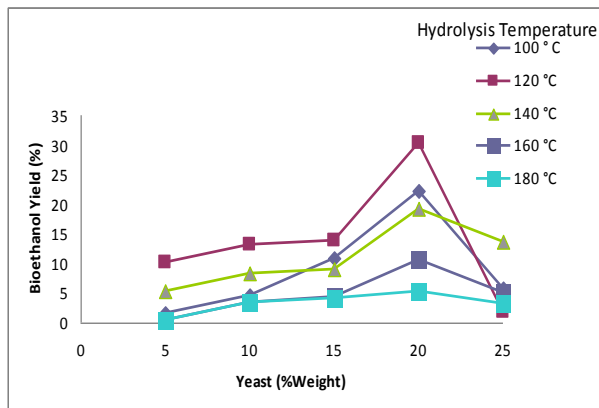


Fig. 4: Chart Effect of Hydrolysis Temperature and Weight of Yeast of Bioethanol Yield.

From the picture above shows the percent of bioethanol yield is influenced by the temperature at the time of hydrolysis and also the weight of yeast used when fermentation. It can be seen that the highest yield of bioethanol is at temperatures of 120 ° C and 20% by weight of yeast, which is as much as 30.2%. Subsequent temperature addition at temperatures of 140 ° C, 160 ° C and 180 ° C can reduce the yield of bioethanol because it has passed the optimum point. At temperatures that are too high, glucose will degrade into furfural and hydroxymethylfurfural.

Whereas the optimum weight of bread yeast is at 20% by weight. From the variation of% bread yeast weight 5% -20% shows a significant increase in bioethanol yield. The more yeast added, the higher the yield of bioethanol.

3.5. Effect of hydrolysis temperature and yeast weight on% of bioethanol yield

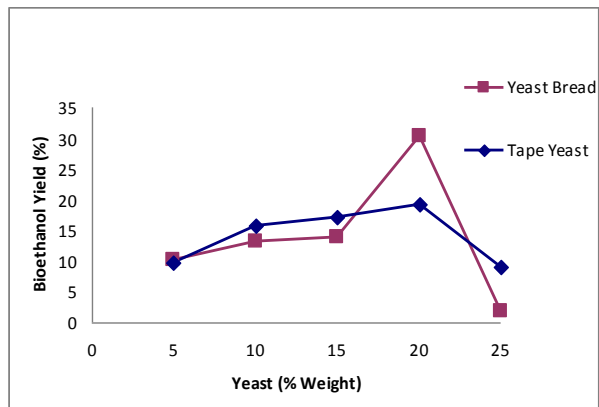


Fig. 5: Type Comparison Graph Yeast Used when Fermented

From the picture above can be seen the comparison of the yield of ethanol produced from fermentation using two different types of yeast. The highest bioethanol content by fermentation using bread yeast is 30.2%. While the highest percentage of bioethanol yield by fermentation using yeast tape is 19.16%. The percentage of yield obtained from this study shows that the manufacture of bioethanol from peat by fermentation using bread yeast will be better than using yeast tape. Although both yeasts contain *Saccharomyces cerevisiae* but yeast tape does not only contain one type of yeast but also contains other microorganisms (bacteria and fungi), namely *Aspergillus*, *Saccharomyces*, *Candida*, *Hensula* and *Acetobacter*. So that the contents of other microorganisms can disrupt *Saccharomyces cerevisiae* activity in the fermentation process.

4. Conclusion

- 1) Peat soil can be used as raw material for making bioethanol because peat contains cellulose which can be converted into glucose by using acid in the process of hydrolysis and yeast in the fermentation process.
- 2) At the stage of the hydrolysis process, the optimum operating conditions occur at 90 minutes hydrolysis time and 2% sulfuric acid concentration by yielding a yield percentage of 19.16%.
- 3) In the process of hydrolysis with temperature variation the optimum conditions are obtained at a temperature of 120 ° C with the yield of bioethanol produced at 30.2%.
- 4) In the variation of heavy use of yeast, it was found that the optimum conditions for both yeast bread and yeast tape were at 20% by weight. Of the two types of yeast used, it was found that the use of bread yeast yielded a higher yield of bioethanol yield of 30.2% while the yield percentage produced by fermentation using yeast tape is 19.1%.

References

- [1] Soil Survey Staff. 2010. Soil Taxonomy a Basic System of Soil Classification for Making and Interpreting Soil Surveys Eleventh Edition. United States Department of Agriculture. Washington DC.
- [2] Fahmuddin, Markus, Ali, Masganti. 2014. Lahan gambut Indonesia (Pembentukan, Karakteristik, dan Potensi Mendukung Ketahanan Pangan). IAARD Press, Jakarta.
- [3] Page, S., Rieley, J. O., Wust. 2006. Peatlands: Evolution and Records of Environmental and Climate Changes. Elsevier BV
- [4] Brady, M.A. 1997. Effects of vegetation changes on organic matter dynamics in three coastal peat deposits in Sumatra, Indonesia. In: Rieley, J.O. and Page, S.E. (Eds.): Biodiversity and Sustainability of Tropical Peatlands. Samara Publishing, Cardigan, pp. 113-134.
- [5] Waluyo. 2008. Fluktuasi Genangan Air Lahan Rawa Lebak dan Manfaatnya bagi Bidang Pertanian di Ogan Komering Ilir, Jakarta. www.energi.lipi.go.id. (accessed May 01, 2018)
- [6] Fessenden, R.J & Fessenden, R.J. 1982. Kimia Organik. Erlangga, Jakarta.
- [8] Iranmahboob, J., Nadim, F., Monem, S., 2002. Optimized Acid-Hydrolysis: A Critical Step For Production Of Ethanol From Mixed Wood Chips. Biomass and Bioenergy 22, pp. 401-404. [https://doi.org/10.1016/S0961-9534\(02\)00016-8](https://doi.org/10.1016/S0961-9534(02)00016-8).
- [9] Sun Y., Cheng J., 2002. Hydrolysis of Lignocellulosic Materials for Ethanol Production: A Review. Bioresource Technology. [https://doi.org/10.1016/S0960-8524\(01\)00212-7](https://doi.org/10.1016/S0960-8524(01)00212-7).
- [10] Mosier, N., C, Wyman, B. Dale, and R. Elander, Y., 2005. Feature of Promising Technologies for Pretreatment of Lignocellulosic Biomass. Bioresource Technology. <https://doi.org/10.1016/j.biortech.2004.06.025>.
- [11] S.I. Mussatto, I.C. Roberto, Alternatives for detoxification of diluted-acid ligno-cellulosic hydrolyzates for use in fermentative processes: a review, Bioresour. Technol. 93 (2004) 1–10. <https://doi.org/10.1016/j.biortech.2003.10.005>.
- [12] Taherzadeh, Mohammad J., 2007. Acid- Based Hydrolysis Processes for Ethanol From Lignocellulosic Materials. Bio Resources 2(3), pp. 472-499.
- [13] Hamelinck, C.N.G., 2005. Ethanol from Lignocellulosic Biomass: Techno-economic Performance in Short, Middle, And Long Term. Biomass and Bioenergy 28, pp. 384- 410. <https://doi.org/10.1016/j.biombioe.2004.09.002>.
- [14] Abercrombie, M., Hickman, M., Jhonson, M.L. dan Thain, M. 1993. Kamus Lengkap Biologi. Penerjemah: T. Siti Sutarni dan Nawangsari Sugiri. Jakarta: Erlangga.
- [15] McCabe, Warren. 1993. Unit Operation of Chemical Engineering. Mc. Grow Hill, Singapore
- [16] Roni, K.A., Merisha Hastarina, ST., M.Eng. dan Rully Masriatini, ST., MT., 2017, "Pengaruh Berat Ragi dan Waktu Fermentasi terhadap Yield Etanol dari Tanah Gambut ", Laporan Penelitian, LP2M, Universitas Muhammadiyah Palembang.