

Effect of delignification and treatment of fermentation on bioethanol percent levels from coconut fabrics

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Abstract

Indonesia's dependence on fossil fuels continues to increase, so the government strives to develop alternative energy to reduce dependence on fuel oil. Bioethanol is an alternative energy that is renewable. The production of bioethanol from coconut fiber waste is very potential because this waste is found and underutilized. Making bioethanol uses a fermentation process with *Saccharomyces cerevisiae*. The effect of delignification and fermentation treatment affects the density, refractive index and bioethanol content. Delignification was carried out using variations of NaOH and KOH while the fermentation treatment was in a variety of time, temperature and nutrition, the time variation lasted for 2 days, 3 days, 4 days, 5 days and 6 days, temperature variations took place at 28 ° C, 29 ° C, 30 ° C, 31 ° C and 32 ° C and variations in nutrition by using Diammonium phosphate and urea as much as 1 gr, 1.5 gr, 2 gr, 2.5 gr, 3 gr. This research is expected to provide a reference for the development of alternative energy from plants.

Keywords: Bioethanol; Coconut Fiber; Fermentation; *Saccharomyces Cerevisiae*.

1. Introduction

Indonesia's dependence on fossil energy is increasing year by year. This is evidenced in 2017 the Indonesian nation imports 140 million barrels of fuel oil. This of course has an impact on Indonesia's domestic economy. In the long run, the government is trying to reduce fuel consumption, among others by diverting fuel to Biofuel, both for households (kerosene) and transportation (gasoline). For this reason, recently the government is seeking the development of alternative energy to reduce our dependence on fuel oil [1].

Bioethanol is an environmentally friendly alternative energy. Bioethanol can be produced from various raw materials that are widely available in Indonesia, so the potential to be processed and developed. Bioethanol is a renewable energy source so there is no need to worry about the depletion of energy supply.

Coconut coir is a large amount of waste, so it needs special handling to handle the waste. Coconut fiber waste is crude fiber. Therefore, the waste of coconut husk cannot be disposed of directly into the final disposal, because decay will be very long.

Fermentation is defined as a process to convert glucose to ethanol molecules or better known as bioethanol (alcohol) by using yeast or bacterial microorganisms [2]. This fermentation, besides producing ethanol, also produces other substances, namely carbon dioxide.

From the background, which has been explained by the lack of attention to waste of coconut husk, our desire to conduct research on the manufacture of bioethanol from coconut fiber is our desire. The problem that we raised in this study was: how the effect of fermentation treatment used in the fermentation process, how the effect of fermentation treatment on the percentage of bioethanol produced, how the effect of delignification on the percentage of bioethanol produced, and what percentage of optimum levels were obtained.

The purpose of this study was to determine the effect of delignification and treatment of fermentation on the percentage of bioethanol produced. Add knowledge about the process of processing coconut husk waste into bioethanol and can increase the value of coconut fiber waste. The benefits expected by researchers are to reduce environmental pollution caused by a lack of public knowledge about waste coconut husk that is not utilized and provide a reference to seek the development of alternative energy from vegetable to reduce our dependence on fuel oil.

2. Literature review

2.1. Coconut

Fiber Coconut fiber is a fibrous material that makes up the thick mesocarp (middle layer) of the coconut fruit (*Cocos Nucifera*). Coconut fruit has various types, but the one that has the most fiber in the coconut husk is the type of coconut with viridis varieties.



Fig. 1: Coconut Fiber Waste.

Fiber is very potential to produce bioethanol because it has high levels of Lignin, Cellulose and Hemicellulose in it. Both components, after pretreatment and enzymatic hydrolysis will be converted into sources of sugar that can be fermented into bioethanol [3]. The following is the composition of Coconut Fiber Chemistry.

Table 1: Composition of Chemical coconut Fiber

Compounds	Composition (%)
Cellulose	46.44
Hemicellulose	0.25
Lignin	45.84
Water	5.25
Abu	2.22

Source: Sukadarti, et al, 2010. [4].

2.2. Bioethanol

Bioethanol is a biochemical liquid from the results of the fermentation process of sugar from carbohydrates using the help of microorganisms. The raw material for making bioethanol consists of ingredients that contain carbohydrates, glucose and cellulose. Bioethanol is often written with the formula EtOH. The molecular formula for ethanol is C_2H_5OH or the empirical formula C_2H_6O or the formula for the formation of CH_3-CH_2-OH . In general, the acronym of (Bio) Ethanol is EtOH (Ethyl- (OH)). Bioethanol (C_2H_5OH) is one of the biofuels that are present as alternative fuels that are more environmentally friendly and are renewable. An alternative fuel that is processed from plants that has the advantage of being able to reduce CO emissions₂ by 18%, compared to the emissions of fossil fuels such as kerosene. Bioethanol can also be interpreted as a chemical that has similar properties to premium oil, because there are elements such as carbon (C) and hydrogen (H). Bioethanol has better characteristics compared to petrochemical-based gasoline [5].

The following is a table Isik properties of ethanol based on SNI 06-3565-2014:

Table 2: Physical Properties ethanol

Parameter	Ethanol
Chemical formula	C_2H_5OH
Molecular weight	46.07
Density (g / ml)	0.7851
Boiling Point(°C)	78.4
Flash Point (°C)	13
Freezing Point (°C)	-112.4
Bias Index	1.3633
Evaporation Heat (cal / gr)	204
Viscosity at 20°C (Poise)	0.0122

Source: National Standardization Agency For.

2.3. Preparation

Materials prepared for use in making bioethanol are coconut fiber, NaOH, H_2SO_4 , Saccharomyces Cerevisiae, urea and aquadest.

- 1) Extraction of coconut milk Coconut milk is obtained from traders of coconut milk. Then the coconut fiber is dried and chopped.
- 2) Preparation of Samples 100 gr of coconut husk smoothed using a blender. Then delignified with NaOH and KOH and hydrolyzed with H_2SO_4 .
- 3) Preparation of Bioethanol Samples that have been made are added to yeast Saccharomyces Cerevisiae and Diammonium Phosphate / Urea. Then let stand for 2, 3, 4, 5 and 6 days.

2.4. Delignification

Lignin is one of the woody parts of plants. Lignin contains complex substances and is a combination of several compounds, namely carbon, hydrogen and oxygen. Besides lignin, the other part of coconut fiber is cellulose. Cellulose is a polysaccharide which contains sugar substances. In making ethanol from the coconut fiber, the cellulose is used so that the lignin in the coconut husk must be removed.

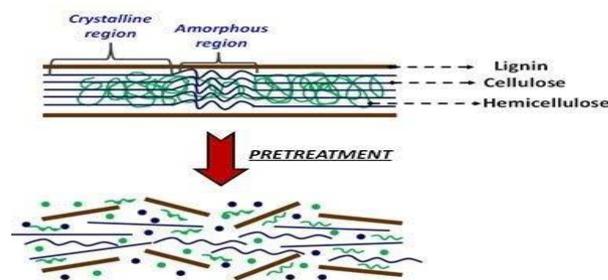


Fig. 2: Delignification Process by Chemical Method.

In this study NaOH was used as a delignification because NaOH can damage the structure of lignin in crystalline and amorphous. NaOH can extract hemicellulose by breaking the amorphous structure in hemicellulose.

2.5. Hydrolysis

Hydrolysis includes the process of breaking the lignin bond, eliminating the content of lignin and hemicellulose, damaging the crystalline structure of cellulose and increasing the porosity of the material [6]. Damage to cellulose crystals will facilitate the breakdown of cellulose into glucose. In addition, hemicellulose also decomposes into simple sugars: glucose, galactose, manose, hexose, pentose, xylose, and arabinose. Furthermore, the sugar compounds will be fermented by microorganisms to produce ethanol [7].

2.6. Acid hydrolysis

Some commonly used for acid hydrolysis include sulfuric acid (H_2SO_4), loic acid and HCl. Sulfuric acid is the most studied acid and is useful for acid hydrolysis. Acid hydrolysis can be classified into: concentrated acid hydrolysis and dilute acid hydrolysis [8].

Hydrolysis includes the process of solving polysaccharides in lignocellulosic biomass, namely: cellulose and hemicellulose into the sugar monomers of the feeder. Cellulose hydrolysis perfectly produces glucose, while hemicellulose produces several pentose (Csugar monomers₅) and hexose (C_6).

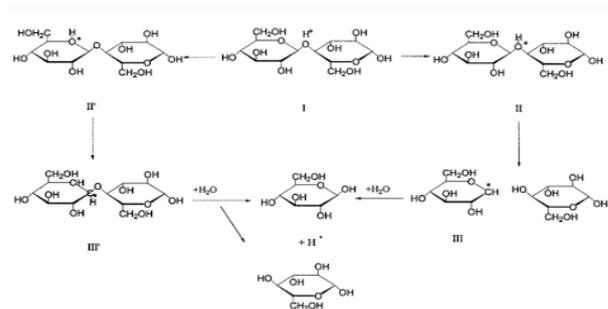


Fig. 3: Mechanism of Cellulose Hydrolysis with Acid.

2.7. Fermented

Fermentation is a chemical process that takes place by the presence of microorganisms that catalyze the reaction. Types of microorganisms that can be used include yeast, bacteria or fungi to produce compounds such as ethanol, butanol, glycerol, acetic acid, or citric acid [9].

Alcohol fermentation is the process of decomposing carbohydrates into ethanol and CO_2 produced by the activity of a type of microbe called yeast in anaerobic conditions [10]. The fermentation process usually does not cause foul odor and usually produces carbon dioxide gas. Fermentation results are influenced by many factors. Like food or substrate, microbial types and surrounding conditions.

The microbe that can be used is *Saccharomyces cerevisiae*. Changes in the fermentation using *Saccharomyces cerevisiae* usually expressed in the following equation:



2.8. Nutrusi

2.8.1. Diammonium phosphate

Diammonium Phosphate is one of a series of water-insoluble in ammonium phosphate salts produced when ammonia reacts with phosphate acid. Diammonium phosphate is an excellent source of P and nitrogen (N) for nutrients in fermentation. Microbes use NH_4^+ as the main nitrogen source, and many organisms have the ability to produce NH_4^+ from amines ($R-NH_2$) or from amino acids ($RCHNH_2COOH$).

2.8.2. Urea

Microbes need nutrients to meet energy needs and for cell building materials, for the synthesis of protoplasm and other cell parts. For life purposes, microbes need organic and inorganic materials taken from their environment. Food ingredients are called nutrients, while the process of assimilating food is called nutrition.

Nutrients are inorganic and organic substances which are in solution across the cytoplasmic membrane. In order to get nutrients from food, cells must be able to digest the food, which converts large and complex proteins, carbohydrates and lipids into small, simple molecules that dissolve immediately so they can enter the cell. The process of assimilating food is what is called nutrition.

Urea is an organic compound consisting of elements of carbon, hydrogen, oxygen and nitrogen with the formula CON_2H_4 or $(NH_2)_2CO$. In the fermentation process of making bioethanol, urea fertilizer functions as a nutrient for the bacteria *Saccharomyces Cerevisiae* which works in the fermentation process because urea fertilizer contains the N element needed for food, the bacteria is around 41-45% N.

2.9. Destillation

Distillation is a mixture separation technique in the liquid phase homogeneous by evaporation and condensation, so that distillate (distillation product) is obtained which relatively contains more volatile (volatile) components than the original solution which is more difficult to evaporate [9].

To separate alcohol 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 agent heat) followed by steam condensation that forms and holds the condensate produced. The steam released from the mixture is referred to as free vapor, the condensate which falls as a distillate and the part that does not evaporate is called residue. This process is carried out to extract alcohol from fermentation. Distillation can be done at 80 ° C because the alcohol point is 78 ° C while the boiling point of water is 100 ° C.

2.10. The previous on research

Effect of Temperature and Addition of Nutrition on Fermentation Process for Bioethanol Production from Coconut Fiber [11]. Malang National Institute of Technology. The method carried out is by fermentation temperature variation and addition of diammonium phosphate nutrients, the best bioethanol content was obtained at 29 ° C with the addition of diammonium phosphate 7 g nutrients.

3. Methods

3.1. Tools and materials

Tools : Erlenmeyer 500 ml, Measuring cup, glass cup 500 ml, Aluminum foil, Strainer, Filter paper, Stirrer, pH meter, Incubation, Distillation, Refractometer and Piconometer.

Materials: Coconut Coir, *Saccharomyces cerevisiae*, H_2SO_4 , NaOH and Urea (CH_4N_2O).

3.2. Research methodology

3.2.1. Preparation process

Material prepared for use in making bioethanol is coconut fiber, NaOH, KOH, H_2SO_4 , *Saccharomyces Cerevisiae*, Diammonium Phosphate, Urea and aquadest. Then the coconut husk is mashed then weighed as much as 100 grams, and processed by Delignification, hydrolysis, fermentation and distillation.

3.2.2. Delignification process

Coconut husk which has been mashed is taken as much as 100 grams then added NaOH / KOH solution and heated for 2 hours at a temperature of 100 ° C. After heating the coconut husk is washed to pH 6-7.

3.2.3. Hydrolysis process

Washed and delignified coconut husk was added with H_2SO_4 solution with various concentrations of 1N, 2N, 3N, 4N and 5N. Then heated for 4 hours at a temperature of 100 ° C. After heating the coconut husk is washed to pH 4-5.

3.2.4. Procedures manufacturing of bioethanol

Production is carried out in the fermentation process. Coconut coir which has been hydrolyzed and washed is fermented with yeast *Saccharomyces Cerevisiae* with a variation of time of 2 days, 3 days, 4 days, 5 days and 6 days and with variations in temperature 28 ° C, 29 ° C, 30 ° C, 31 ° C and 32 ° C. The best time and temperature of fermentation was added with a variation of diammonium phosphate and urea nutrition as much as 1gr, 1.5 gr, 2 gr, 2.5 g, 3 gr.

3.2.5. Distillation process

The fermentation results that have been obtained are distilled with a temperature of 80 ° C and a result of 50 ml is obtained.

3.2.6. The analysis process

Analysis used to view bioethanol content is analysis with refractometer, piconometer and Gas Chromatography. Refractometer is used for bioethanol refractive index analysis, piconometer is used for bioethanol density analysis, while Gas Chromatography is used for bioethanol content analysis and bioethanol yield.

3.2.7. Research flow chart

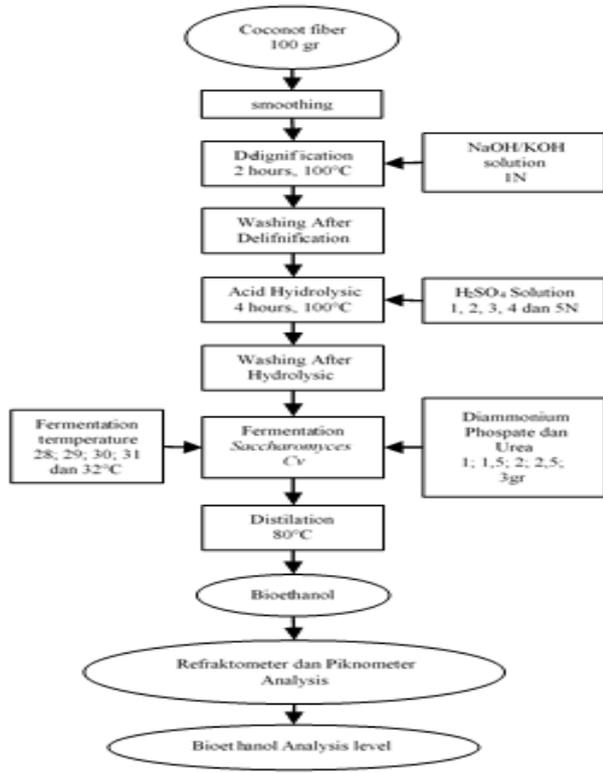


Fig. 4: Bioethanol Making Flow Chart.

4. Results

4.1. Changes of H₂SO₄ concentration to density, bias index and ethanol levels

The results of testing the effect of H concentration₂SO₄ on the process of making bioethanol from coconut fiber waste which was fermented by 9.6 gr, the amount of yeast is 0.96, the amount of nutrition is 1 gram, and the duration of fermentation is 3 days.

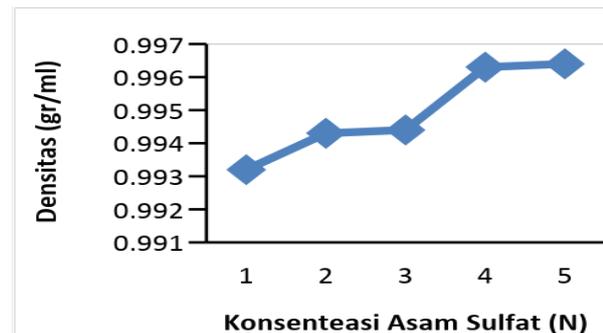


Fig. 5: Relationship between H₂SO₄ Concentration and Density.

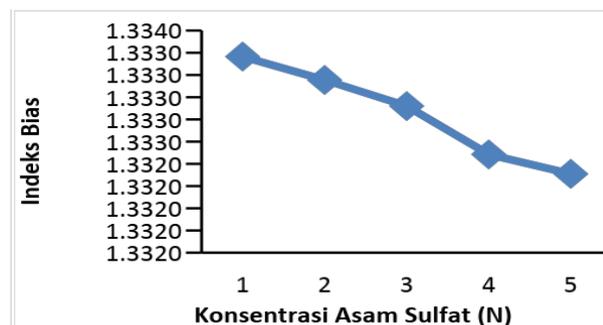


Fig. 6: Relationship between H₂SO₄ Concentration and Bias Index.

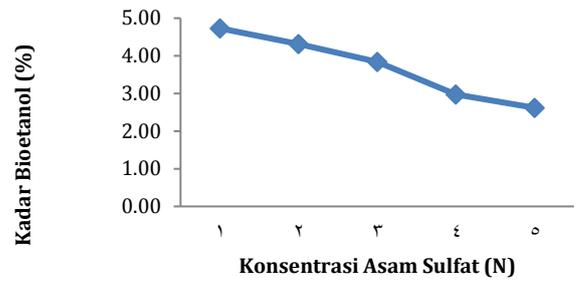


Fig. 7: Relationship between H₂SO₄ Concentration and Bioethanol Levels.

From the graph above it can be seen that the best density is at H concentration₂SO₄ 1N of 0.9932 gr / ml, which is the closest to ethanol density of 0.789 gr / ml. The best refractive index is at H concentration₂SO₄ 1N namely 1.3334 which is the closest to the refractive index of ethanol, which is 1.361. While for the best bioethanol content is also at H concentration₂SO₄ 1N which is 4.73% so that it can be concluded that the best bioethanol content is at H concentration₂SO₄ 1N namely at the lowest acid [11].

4.2. Effect of fermentation time on density, bias index and ethanol levels

The results of testing the effect of fermentation time on the process of making bioethanol from coconut fiber waste with a concentration of H₂SO₄ 1N the weight of fermented coconut husk was 9.6 g, the amount of yeast was 0.96 1 gram of nutrition, and 3 days of fermentation time.

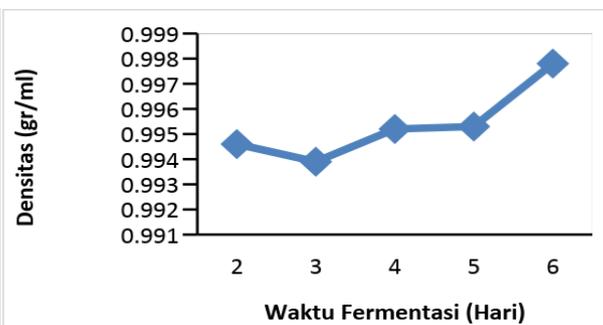


Fig. 8: Relationship between Fermentation Time and Density.

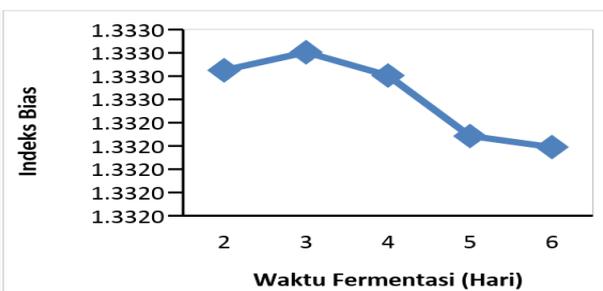


Fig. 9: Relationship between Fermentation Time and Bias Index.

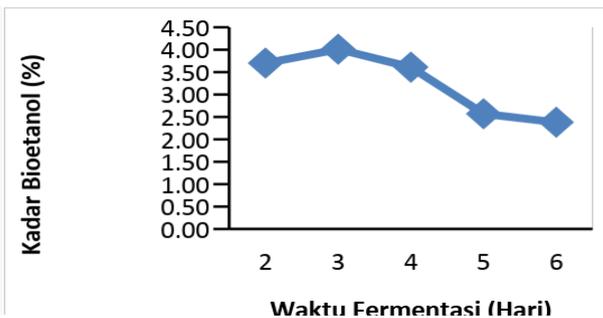


Fig. 10: Relationship between Fermentation Time and Bioethanol Content.

From the graph above it can be seen that the best density is at the 3rd Fermentation Time of 0.9939 gr / ml, which is the closest to the density of ethanol which is 0.789 gr / ml. For the best refractive index, the third day of fermentation is 1.33301 which is closest to the refractive index of ethanol, which is 1.361. Whereas the best bioethanol content is also at the third day of fermentation which is 4.01% so that it can be concluded that the best bioethanol content is at the third fermentation time of the day. It can be concluded that the length of fermentation affects the value of ethanol which is as long as fermentation, the bioethanol content increases to a certain extent and then decreases [12].

4.3. Effect of fermentation temperature on density, bias index and bioethanol level

The results of testing the effect of fermentation temperature on density, refractive index and bioethanol content in the process of making bioethanol from coconut fiber waste with a concentration of H₂SO₄ 1N, the weight of fermented coconut husk 9.8 gr , fermentation time is 3 days, the amount of yeast is 0.98 gr and the amount of urea 1.5 can be seen in figure 5 to 7. The results of testing the effect of fermentation temperature on density, refractive index and bioethanol content in the process of making bioethanol from coconut fiber waste with a concentration of H₂SO₄ 1N, the weight of fermented coconut husk 9.8 gr, fermentation time 3 days, the amount of yeast 0.98 gr and the amount of urea 1.5 can be seen in figure 5 to 7.

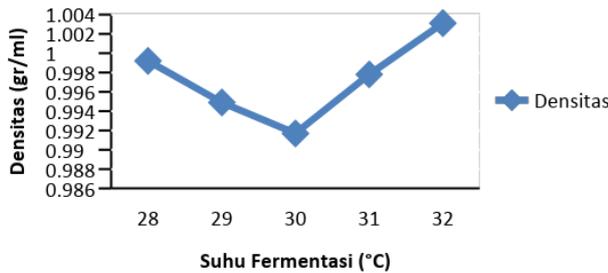


Fig. 11: Relationship between Fermentation Temperature and Density.

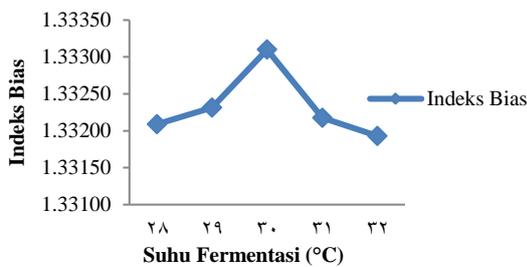


Fig. 12: Relationship between Fermentation Temperature and Bias Index.

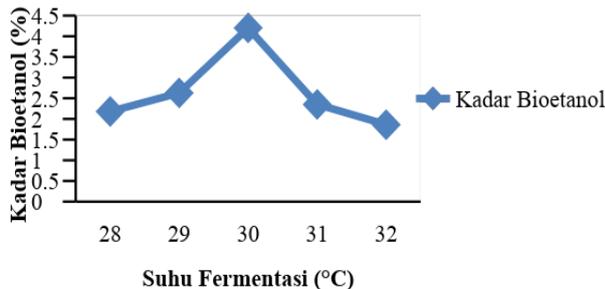


Fig. 13: Relationship between Fermentation Temperature and Bioethanol Content.

From the graph above it can be seen that the best density is at 30 ° C ie 0.9917 gr / ml, which is the closest to the density of ethanol which is 0.789 gr / ml. The best refractive index is at 30 ° C, which is 1.33310 which is the closest to the refractive index of

ethanol, which is 1.361. Whereas the best bioethanol content is also at a temperature of 30 ° C, reaching 4.20%. So it can be concluded that the best bioethanol content is at room temperature [11].

4.4. Effect of addition to urea nutrition on density, bias index and bioethanol level

The results of testing the effect of fermentation temperature on density, refractive index and bioethanol content in the process of making bioethanol from coconut fiber waste with a concentration of H₂SO₄ 1N, the weight of fermented coconut husk 9.8 gr, the fermentation time is 3 days, the amount of yeast is 0.98 gr and the fermentation temperature is 30 ° C can be seen in figure 8 to 10.

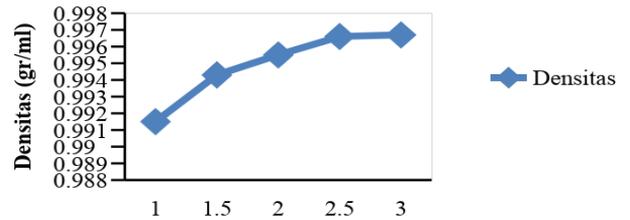


Fig. 14: Relationship between Addition of Nutrition and Density.

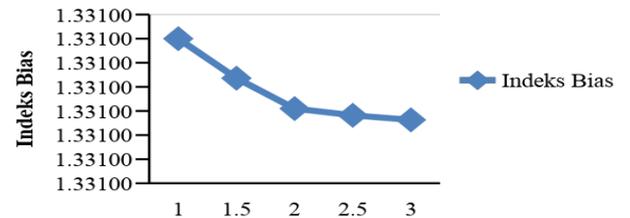


Fig. 15: The Relationship between Addition of Nutrition and Bias Index.

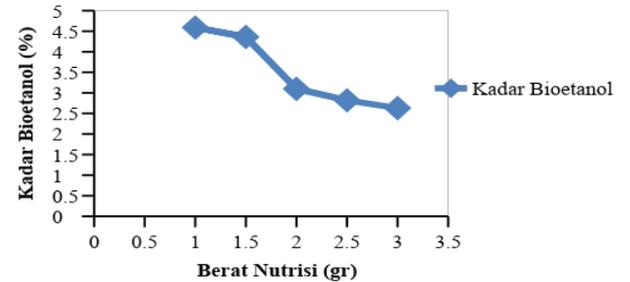


Fig. 16: Relationship between Addition of Nutrients and Bioethanol Levels.

From the graph above, it can be seen that the best density is in the addition of 1 gram of nutrition, which is 0.9915 gr / ml, which is the closest to the density of ethanol which is 0.789 gr / ml. The best refractive index is the addition of 1 g of nutrients, which is 1.33103 which is the closest to the refractive index of ethanol, which is 1.361. Whereas the best bioethanol content is also in the addition of 1 gram of nutrients which is 4.59%. So that it can be concluded that the higher the amount of nutrients added, the less the level of bioethanol produced because the higher the urea will inhibit the growth of cells Saccharomyces Cerevisiae, and if urea is consumed in large quantities will form NH₃-N which is toxic and inhibits the growth of microorganisms [13].

4.5. The effect of the addition to diammonium phosphate and urea nutrients on density, bias index and ethanol levels

The results of the analysis of the addition of Diammonium Phosphate and Urea nutrients in the process of making bioethanol from coconut fiber waste with a concentration of H₂SO₄ 1N, the weight of fermented coconut husk 9.5 grams, 3 days fermentation time,

the number of yeast 0.95 gr can be seen in Figures 4.5, and 6 below.

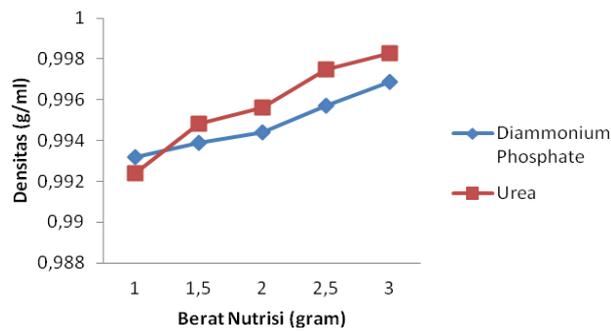


Fig. 17: Relationship between Addition of Diammonium Phosphate and Urea Nutrients to Density.

Based on the picture above shows the relationship between the addition of Diammonium Phosphate and Urea nutrients in the density test. In addition to the two nutrients as much as 1 gram, the lowest density value is 0.9932 gr / ml and 0.9924 gr / ml. On the addition of both nutrients as much as 3 grams, the highest density values were 0.9969 gr / ml and 0.9983 gr / ml. In addition to the second nutrient 1 gram obtained the lowest density because the amount is equivalent to the needs of *Saccharomyces cereviceae* and the value of ethanol content is high. The increasing nutrition of Diammonium Phosphate and Urea, the higher the density produced and the lower the value of ethanol content, this is due to yeast *Saccharomyces Cerevisiae* converts glucose to ethanol, where if the yeast given is excessive then the ethanol produced will also be lower and vice versa, so that the density is higher [14].

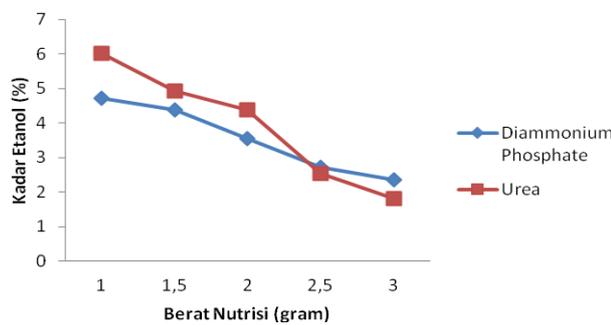


Fig. 18: The Relationship between Addition of Diammonium Phosphate and Urea Nutrients to Ethanol Levels.

Based on the graph above shows that the addition of 3 grams of Diammonium Phosphate and Urea nutrients obtained the lowest ethanol content of 2.35% and 1.82%. Urea as much as 1 gram obtained the highest ethanol content of 4.71% and 6.01%. On the addition of 3 grams of Diammonium Phosphate and Urea nutrients, the lowest ethanol content was obtained, this was due to insufficient bacteria in both nutrients for the growth of bacteria. *Saccharomyces cereviceae* Meanwhile, the addition of 1 gram of Diammonium Phosphate and Urea nutrients was the highest ethanol content due to this amount. equivalent to the needs of *Saccharomyces cereviceae* for fermentation for 3 days. The function of *Saccharomyces cereviceae* as converting glucose to ethanol. Meanwhile, in addition to more than 1 gram of nutrients obtained ethanol levels decreased as this was due to the addition of excess nutrients can inhibit the growth of *Saccharomyces cereviceae* [11].

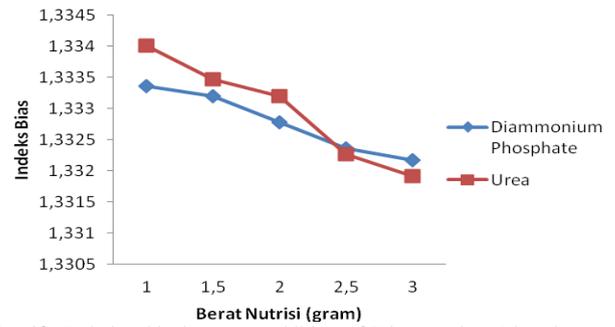


Fig. 19: Relationship between Addition of Diammonium Phosphate and Urea Nutrients to Bias Index.

Based on the graph above, it shows that the addition of 1 gram of Diammonium Phosphate and Urea nutrients is the highest refractive index is 1.3336 and 1.33405 where this value is close to 1.361 (refractive index bioethanol), so that the higher the refractive index, the higher the bioethanol content. In addition to the 3 grams of nutrients, the lowest refractive index is 1.332175 and 1.33191. On the addition of both nutrients as much as 3 grams obtained the lowest refractive index value this is due to the addition of nutrients both are not equivalent to yeast and can inhibit the growth of *Saccharomyces cereviceae* [11].

2.5. Results of Cromatographic Gas Analysis

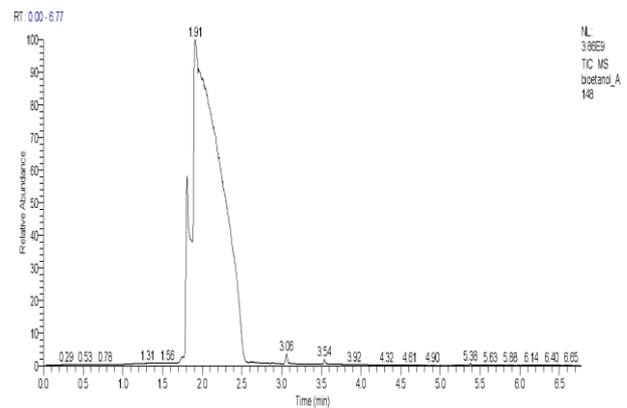


Fig. 20: Graph of Cromatographic Gas between Concentration and Fermentation Time.

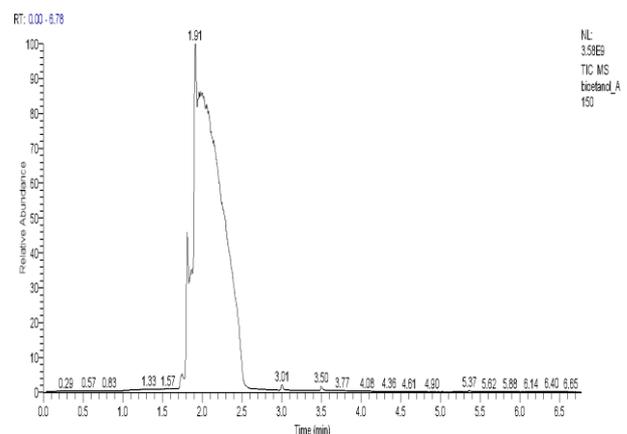


Fig. 21: Graph of Gas Chromatography between Fermentation Temperature and Urea Nutrition.

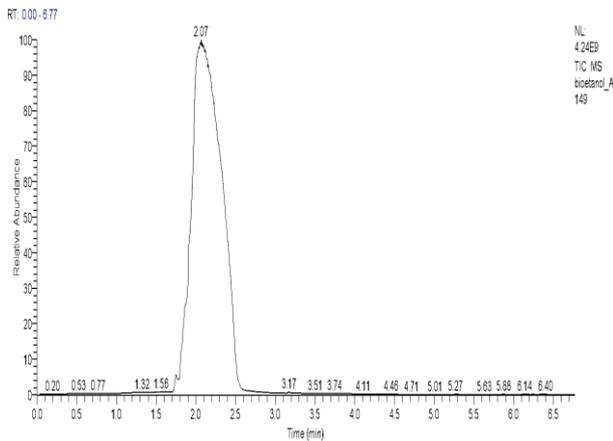


Fig. 22: Graph of Chromatographic Gas with Delignification of KOH and Variations of Diammonium Phosphate and Urea Nutrients.

In Figures 20, 21 and 22, it can be seen that there are several chart peaks which state that there are other components besides bioethanol. But the sample is said to be ethanol if it is at runtime between 1.91 to 2.65 min. In Figure 20 the bioethanol content with GC-MS was 8.845% different from the calculated levels of 4.01%, while in Figure 21 the bioethanol levels of GC-MS were 8.337% different from the calculated levels of 4.59% and in figure 22 bioethanol GC levels -MS 9.845% is different from the level of the calculation, which is 6.01%. GC-MS analysis results are greater because the analyst using GC-MS lasts for 6.74 minutes with high separation sharpness and uses a longer column to produce high separation efficiency [15].

5. Conclusion

Based on the research that has been carried out, it can be concluded as follows, The treatment of fermentation affects the fermentation process that takes place. Fermentation treatment affects the level of bioethanol produced. Delignification affects the level of bioethanol produced. The optimum yield of bioethanol content was 9.845%, with delignification using KOH, Hconcentration₂SO₄ 1N, 3 days fermentation time, 30°C fermentation temperature and 1gr urea nutrition addition.

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