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Bioethanol Production from Waste of the Cassava Peel (*Manihot esculenta*) by Acid Hydrolysis and Fermentation Process

Zainal Abidin[1]*, Ellena Saraswati[1], Tadjuddin Naid[2],

[1]Muslim University of Indonesia, Makassar, Indonesia.
[2]Hasanuddin University, Makassar, Indonesia.

***Corres.author: zainal243@yahoo.co.id**
Tlp. 082338805287

Abstract: Bioethanol is ethanol from biomass through fermentation process by microorganism. Raw material of bioethanol are sugary, starch, and fibrous material. Waste of cassava peel are one of many source fibrous material, so this study would utilise it as raw material to produce bioethanol. The aim of this study were determine the optimum concentration of H_2SO_4 and optimum time in the hydrolysis process and determine the optimum time in the fermentation process. Waste of the cassava peel (*Manihot esculenta*) was hydrolysed by using sulphuric acid solution with concentration 0.1 ; 0.5 ; and 1.0 M, at temperature 100 ^oC for two time 30 and 60 minute, respectively, and then the optimum result of the concentration of sulphuric acid of hydrolysis process was fermented by yeast for 1 ; 2 ; 3 ; 4 ; 5 ; and 6 days, respectively. The result of this study obtained that 0.5 M sulphuric acid solution was the optimum concentration of the hydrolysis process at temperatur 100 ^oC for 60 minute and 4 days was the optimum time of the fermentation process, where from that obtained 3.58 % v/v bioethanol.
Keyword : Bioethanol, Waste Cassava Peel (*Manihot esculenta*), Hydrolysis Acid.

Introduction

Cassava is the old stuff plant in Indonesia. This plant can grow in the less fertile land and harvest time can be done every year, cassava has high enough carbohidrate contens is 32 – 35 %, and it is the most important carbohydrate source after rice, but advance in preparation technology of cassava make it not only for food [9].

Cassava peels is main waste food in the developing nation, where the high number harvest of cassava so more and more high waste of cassava peels. Every one kilogram cassava can result 15 % - 20 % cassava peel, and every weight of roof is peeled contain 8 – 15 % cassava peel, where cassava peel contain carbohydrate about 50 % from content carbohydrate of roots. Carbohydrate content of cassava peel is high enough [3,7].

Bioethanol is ethanol from biomass through hydrolysis and fermentation process, where can be produced from foodstuff that contain starch, such as cassava, sweet potato, corn, and sago, beside that the raw material to produce bioethanol can be obtain from sugary, starch, and fibrous material[8]. In manufacturing industry, ethanol is commonly used as raw material to produce ethanol derivative, mixture of alcoholic liquor, and pharmaceutical and cosmetic raw material[9].

By virtue of that, this study will utilize cassava peels to produce bioethanol in a way acid hydrolysis and fermentation process. From this study is hoped to know how much the optimum concentration of sulphuric acid of the hydrolysis process and how long duration time of the hydrolysis and fermentation process. The result of this study could be used as consideration that cassava peels can be used as bioethanol source to fulfill requirement raw material of fuel industry, pharmaceutical and cosmetic.

Materials. Waste of cassava peels (*Manihot esculenta*) were obtained from Makassar and determined in the laboratory of Phytochemistry, Muslim University of Indonesia.

Chemicals. Sulfuric acid (pa, Merck), Distilled water, Ethanol 96 % (pa. Merck), Luff Schoorl solution, Sodium thiosulphate (pa. merck), Sodium carbonate (pa. Merck), Plumbi acetate (pa. Merck), Yeast (Technis, Fermipan), and Cassava peels (*Manihot esculenta*).

Experimental Procedure

Preparation Sample

Waste of cassava peels were washed and cut to be small, after that dryed and milled to powder. it was sieved to get homogeneous powder. It was done in Laboratorium Pharmacy chemistry, Muslim University of Indonesia.

Optimization Time and Concentration Acid of Hydrolysis Process

Waste of cassava peel powders, 15 gram, were hydrolyzed in 100 ml by using sulphuric acid with various concentration (0.1 ; 0,5 ; and 1.0 M) at temperature 100 ^{o}C for 30 minute, after that the optimum concentration of sulphiric acid was used to hydrolyze waste of cassava peel powder at temperature 100 ^{o}C for 30 and 60 minute, to get the optimum time of hydrolysis process[8]. The concentration of glucose from hydrolysis process was determined by using Luff Schoorl Mathod[10].

Optimization Time of Fermentation Process

The Result of the optimum time and concentration acid of hydrolysis process was added NaOH until pH 4 – 6, after that added yeast (*Saccharomyces* cerevisiae) as many as 10 % of volume sample. Fermentation process was done at room temperature for 1 ; 2 ; 3 ; 4 ; 5 ; and 6 days, respectively. Ethanol would be collected every day for 6 day by destilation process at temperature 70 – 80 ^{o}C [8], after that determination of ethanol by using Gas Chromatography (GC-2010 Chromatograph Shimadzu) Mathod, with injector and column temperature were 100 and 70 ^{o}C, respectively.

Result and Discussion

Bioethanol is ethanol from biomass through fermentation process by microorganism. Raw material of bioethanol are sugary, starch, and fibrous material. Waste of cassava peels are one of many source fibrous material, so this study would utilise it as raw material to produce bioethanol. Cassava peel was cut and milled to increase the surface area, so increase too contact between the hydrolysis solution with surface of cassava peel powder. It made carbohydrate content of cassava peel more and more a lot of to be hydrolyzed.

Hydrolysis process used acid solution to hydrolyze carbohydrate (polysaccharide) content of cassava peel to monosaccharide (glucose). The acid solution was sulphuric acid with various concentration (0.1 ; 0.5 ; and 1.0 M). Hydrolysis process were done twice, the first to get the optimum concentration of sulphuric acid and the second to get the optimum time of hydrolysis process. The result of the optimization concentration sulphuric acid solution of hydrolysis process could be seen in the following table :

Table 1. The result of optimization concentration of sulphuric acid of hydrolysis process

Sample	Concentration of Sulphuric acid (M)	Weight of sample (gram)	Volume titran (ml)	Concentration of glucose (%)
1	0.1	15.0013	10.43	1.63
2	0.5	15.0018	4.90	5.24
3	1.0	15.0012	6.81	2.85

Table 1 showed that concentration sulphuric acid 0.5 M resulted the highest concentration of glucose. It was 5.24 % glucose. After that, the optimum concentration sulphuric acid 0.5 M was used to do the optimization time of hydrolysis process was done at temperature 100 ^{o}C for 30 and 60 minute. The result of this process could be seen in the following table :

Table 2. The result of optimization time of acid hydrolysis process

Sample	Hydrolysis process time (minute)	Weight of sample (gram)	Volume titran (ml)	Concentration of glucose (%)
1	30	15.0012	4.00	5.55
2	60	15.0010	3.93	5.75

Table 2 showed that the optimum time of hydrolysis process was 60 minute, where the result of concentration of glucose was 5.75 %. By virtue of that, hydrolysis process to polysaccharide to monosaccharide was influenced by concentration sulphuric acid and duration of heating.

Determination of monosaccharide (glucose) from hydrolysis process was done by Luff Schoorl Mathod. Principle of this method was oxydation reduction reaction between reagent of Luff Schoorl with glucose. This method is indirect titration, where the result of that reaction would released iodine from potassium iodide, and it was titrated immediately by sodium thiosulfate. This method used starch indicator to detect the end point of titration[10].

Fermentation process by using yeast (*Saccharomyces cereviceae* source). It could convert glucose to ethanol (bioethanol) [2]. The result of the optimum hydrolysis process of cassava peel was prepared as many as 6 sampel to be fermented at room temperature and pH 5 for 6 days. Bioethanol from each sample in the fermentation process was collected by using destilation process, it was done for 1 ; 2 ; 3 ; 4 ; 5 ; and 6 day to each sample. The destilation process is one of many process to separate bioethanol from water at 70 – 80 ^{o}C, where bioethanol will prior evaporate and will condense to get it [11]. The result of the fermentation process was obtained volume of bioethanol solution that could be seen in the following table :

Table 3. The result of optimization time of fermentation process

Time of Fermentation process (day)	Volume of Destilation process (ml)
1	16,5
2	18,0
3	32,6
4	41,0
5	15,0
6	15,0

Table 3 showed that the fourth day was the highest volume of the result of destilation process, but to know quantities of bioethanol (the difference concentration of bioethanol) in each volume every time the result of fermentation process was determinated by using index of refraction method. According Mellissa Castillo *et al*. (2006) that the index of refraction is directly proportional to the solution concentration[1], it is caused that the more molecules added into a solution, the more of a chance the light has of hitting the molecules. This slows down the light as it goes through absorbtion and reemission, causing an increase in the index of refraction [5]. The result of index of refraction each sample every day could be seen in the followed table :

Table 4. The result of index of refraction each sample every day

Time of Fermentation process (day)	Index of refraction
1	1,3326
2	1,3326
3	1,3329
4	1,3331
5	1,3328
6	1,3323

Table 4 showed that the highest index of refraction of the result of fermentation process was the fourth day, it was mean that the fourth day resulted the highest consentration of bioethanol. The difference concentration of bioethanol in every day of the fermentation precess were caused by its duration time and quantities of *Saccharomyces cereviceae*, where the first time, quantities of bioethanol increase in line with the changing of day, but after the fourth day, quantities of bioethanol decrease, it was caused that *Saccharomyces cereviceae*, the microorganism produced enzyme as biocatalisis of glucose to convert it to be bioethanol, had been worked out death phase.

Determination of concentration of bioethanol from the highest index of refraction was done by using chromatography gas method. It could be used to determinate every gas sample or volatile sampel, and it doesn't work out dissociation[4], but this method has lack that was only used to the volatile sample[6]. The area of bioethanol solution of the fermentation process could be seen in the followed table :

Table 5. The area of bioethanol of the fermentation process

Object	Area (μm^2)
Sample solution of the fourth day (The highest of the index of refraction)	7569993
Standart solution (Ethanol 96%)	203017383

Table 5 showed the area of bioethanol of the sample solution of the fourth day and the area of ethanol standart, where determination concentration of bioethanol sampel was obtained by comparison the area of sample solution of the fourth day (The highest of the index of refraction) with the area of standart solution (Ethanol 96%), and obtained the concentration of bioethanol sampel was 3,58 % v/v.

Conclusion

From this study could be concluded that the optimum concentration of sulphuric acid of the hydrolysis process was 0,5 M and its duration was 60 minute, and duration time of fermentation process was 4 days, while the concentration of bioethanol for the optimum of hydrolysis and fermentation process was 3,58 % v/v. From this conclusion, then cassava peels can be used as bioethanol source to fulfill requirement raw material of fuel industry, pharmaceutical and cosmetic.

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