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To cite this article: Adulsman Sukkaew 2021 *J. Phys.: Conf. Ser.* **1835** 012113

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The Optimal Conditions of Saccharification and Fermentation Processes for Ethanol Production from Bagasse and Economic Feasibility Analysis

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Abstract. Thailand is one of the agricultural countries with an excellent natural resource like rice, sugarcane. The residue of sugarcane is a Bagasse as the main agricultural product of the sugar industry. Since Thailand is one of the significant sugar exporters, it is reported that Thailand produces sugarcane bagasse over 50 million metric tons every year. This research aimed to study the optimal condition of saccharification and fermentation Bagasse converted to ethanol production. The research found that seven days after saccharification and fermentation from *Aspergillus*, yeast has the highest ethanol content at 12.45±0.43%. While reducing sugar contents 285.65±0.74 g/L. And the results of ethanol concentration from the first and the second distillation were shown as 65.29±0.57% and 92.05±1.24%, respectively. When a study of economic feasibility analysis from the selected ethanol condition process has a sensitivity analysis and the payback period is approximately five months to seven months. In conclusion, the proposed project is attractive for the investors because the current price of ethanol is 18.21 THB (\$ 0.59) per liter, and its trend is expected to keep rising. However, if sugarcane bagasse were managed correctly, it would support the community's robustness and sustainability. It can also reduce agricultural waste and weeds. It can solve the energy crisis as an alternative avenue in the future.

1. Introduction

Nowadays, countries are supporting and developing more renewable energy. The reduced proportion of dependence on energy imports, especially Biofuel, can be produced from various agricultural products such as Soybean, Water Hyacinth, Sugarcane, Cassava, Palm oil, and Corn oil.[1] Energy is vital to the national economy. Especially in the industrial sector, transportation sector, agriculture and business sectors, etc. [2, 4, 7] The allocation of energy is sufficient and has a reasonable price per the demand. It is something that energy policymakers should act on. At present, the amount of energy usage increases gradually while Thailand still imports a lot of energy from foreign countries.[3, 5] Therefore, to ensure that Thailand will have enough energy to meet demand in the future. It results in the concept of alternative energy development and domestic energy sources to promote the country's energy security. The energy source should be cheap and large enough. For this reason, biomass renewable energy is considered an energy source. It has excellent potential and potential to be used because Thailand has many leftovers from the agricultural sector, which are sources of energy that are easy to find and cheap [6,8]. Bagasse is the part of the sugar cane stalk that has been taken out of sugar



cane juice or sugar. Nowadays, Bagasse can be used for many uses, such as developing autoclaved aerated bricks, biogas, ethanol, and others. [9] Ethanol is considered the energy chosen to replace the oil on the market, whether diesel or benzene. Because ethanol possesses clean fuel, complete combustion process without lead and can help reduce environmental pollution.[10] The utilization of ethanol can be used in agriculture and industry. The ethanol production process occurs from the fermentation process using sugar source degraded from waste materials from Bagasse. Bagasse is classified as a raw material with a lower production cost than other raw materials. In general, the application of Bagasse is usually preferable by combustion. The combustion will directly affect the atmosphere. [3,11] the Causing of the greenhouse effect and also the affecting environment as well. This research was to study the optimal condition of saccharification and fermentation Bagasse converted to ethanol production. And a study of economic feasibility analysis from selected ethanol condition process has a sensitivity analysis and the payback period. To solve the energy crisis as an alternative avenue in the future.

2. Theoretical Background

Sugarcane has the scientific name *Saccharum officinarum* L. It belongs to the same family POACEAE with bamboo, grass, and grains such as wheat, rice, corn, and barley.[11] Bagasse is the sugar cane trunk remnants that have taken sugar cane juice or sugar from the logs. When the sugar cane passes through the first set of chests, there may still be water left over. But when passing through the foreskin set 3 to 4, minimal residue means only pure fibers. [12-14] Subsequent by-products are Filter Mud or Filter Cake or Filter Muck, which are filtered or purified by bagasse juice by any means. [15] The final by-product cake filter, molasses, thick and sticky Dark brown, can be extracted sugar by standard methods. In the past, Bagasse was used as a fuel for boiling water in a steam boiler and then using steam power for operating steam engines and for electricity generation. However, many Bagasse remains due to not being used and exhausted, causing problems in removal and destruction, Although some places are used as distilled liquor or alcohol. But many remaining Bagasse. [14, 16] Bagasse consists of 18% cell texture and 82% cell wall, with 40% cellulose, 29% hemicellulose, 13% lignin, and 13% silica. 2 components of the cell wall of rice and sugarcane by-products.[17]

Lignocellulose Is a complex of Lignin, hemicellulose, and Cellulose, a component of plant cell walls. It is classified as dietary fiber insoluble in water in different ratios depending on the type of lignocellulose material. Generally, 40-60 percent cellulose is found, 20-30 percent hemicellulose, and 15-30 percent lignin.[18] Cellulose is a component found in lignocellulose materials, found in plants' cell walls, combined with hemicellulose and Lignin. [18] The amount of the substance varies depending on the type and part of the plant, for example. Wood is found in 40-50 percent, and cotton fibers are around 98 percent. Cellulose is a homopolymer, characterized by a straight line without branches, consisting of sub-units, beta-D-glucopyranol (β -D -Glucopyranose). The β -1,4-glycosidic bond is formed as a glucan polymer with a natural length of approximately 10,000 units bonded with hydrogen bonds.[17, 19] In general, two types of Cellulose are crystalline. Cellulose and amorphous Cellulose. The crystalline Cellulose is degraded by enzymes more difficult than amorphous Cellulose, as shown in Fig. 1[18, 20]

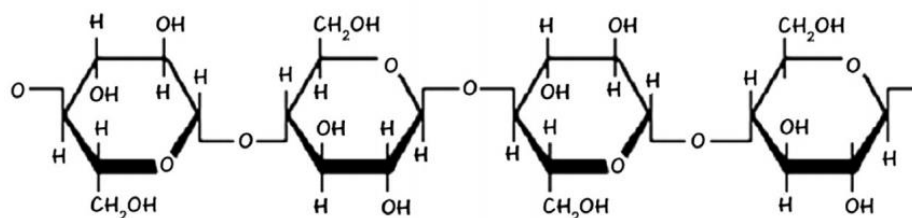


Figure. 1 Cellulose Structure

Hemicellulose, one of the components in lignocellulose materials, is a heteropolymer of many different types of sugars, such as glucose, mannose, xylose, and arabinose. In the form of polymers, xylan, man-galactan, and Arabinan, the average length is about 200 units, with the highest Xylan-Xylose polymer being 85-93 percent. As glucose, glucose acid Galacturonic acid is found in small amounts. The xylose is bonded with beta 1,4-glycosidic bonds for xylan hemicellulose's chemical structure in plants found in plant tissues. It is combined with other substances such as Lignin. Cellulose is the structure of the cell wall. The chemical formula is $(C_6H_{12}O_5)_n$. Hemicellulose is the polymer of pentose sugar. [18, 19] Most D-xylan, which consists of many sugar molecules connected by bonds 1-4-glycosidic Hemicellulose polymer cables, have heterogeneous characteristics. It consists of many types of polysaccharides, which are mainly pentosans, silansans, and arabs. When digested, the sugar was obtained. And arabinoxylan is a substance that is contained in hemicellulose than other substances, as shown in Fig. 2 [20]

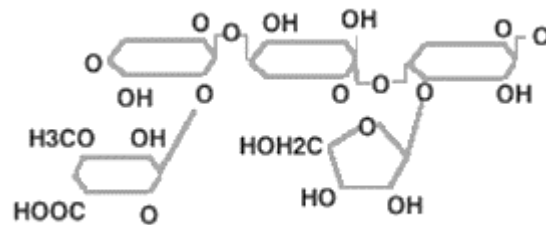


Figure 2 Hemicellulose Structure

Lignin is an aromatic compound known as Phenolic polymers. The -Group can bond with the aldehyde group as hemiacetal and the ketone group as ketal, randomly connected phenyl propane units. The phenol unit may be guaiacyl or sparingly at the lignin molecule's alpha and beta positions. There may be a link between molecules or carbon in the phenolic unit. Bonds may be formed in another unit within the polymer chain that composes the lignin molecules. Lignin has a strong structure, insoluble in water, but can dissolve in some organic solvents such as ethanol or hot methanol. And sodium hydroxide solution Lignin is usually found in the structure of plant cells around Cellulose. [21] As a cellulose protector from digestion, Lignin is resistant to microbes. Anaerobic process in which microbes cannot react with the aromatic ring of Lignin or react will be prolonged for a time. Several days Lignin is found in nature as a binder between Cellulose and hemicellulose. Most of them are found in the cell walls of plants with different amounts according to the type of plants in nature. Lignin protects Cellulose from being easily digested by enzymes of micro-lignin. [22] It is a heteropolymer with a 3-dimensional crystallization structure consisting of 3 aromatic compounds consisting of trans-p-coumaryl Alcohol, trans-coniferyl Alcohol, and trans-p-sinapyl Alcohol. Besides, lignin molecules are connected to many other aromatic compounds such as vanillin and syringaldehyde. The structural formula of trans-p-coumaryl alcohol, trans-coniferyl alcohol, and trans-p-sinapyl alcohol as shown in Fig. 3 [20, 23]

The Pretreatment of lignocellulose into raw materials consisting of natural fibers, Cellulose, hemicellulose-cellulose, Lignin, and other substances. Raw material preparation must be done in two processes. Is the Pretreatment and hydrolysis, which will reduce hemicellulose and Lignin to the maximum Cellulose. It also reduces the crystallization of cells—increased porosity in the raw material Resulting in a better effect on the hydrolysis process for enzymatic digestion. Besides, Pretreatment requires the improvement of sugar structure, helping avoid the deterioration of carbohydrates in raw materials. And reducing the production of inhibitors that will interfere with the enzyme activity. as shown in Fig. 4 [20, 24]

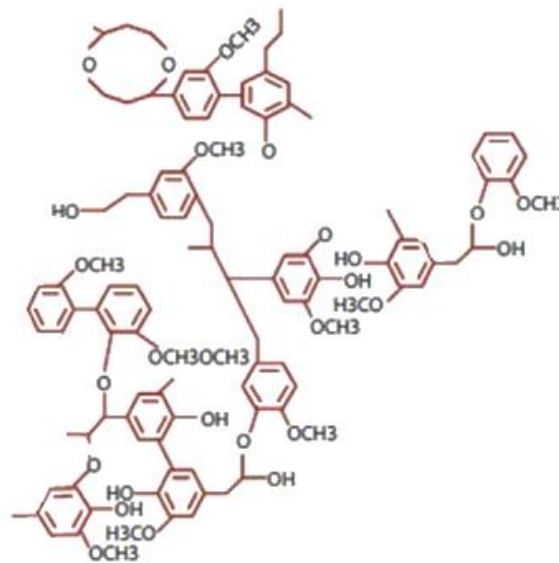


Figure 3 lignin Structure

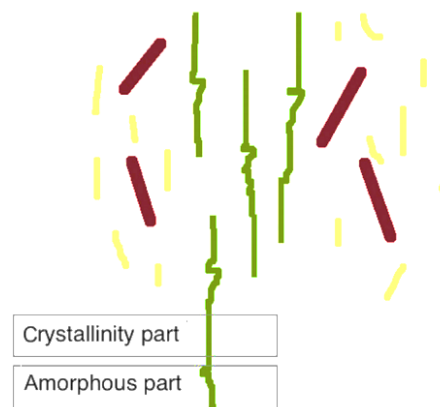


Figure 4 Pretreatment of lignocellulose Structure

Ethanol fermentation, In most cases, yeast is used to change glucose. Or fructose sugar Into alcohol and with carbon dioxide by-products. As the equation below, In theory, alcohol is about 50% of the amount of sugar used. But in practice, it is often not due to the production of many different flavoring substances. As shown in Fig. 4 [25]. Therefore, if fermenting sugar with a sugar content of 20 ° Briggs or 200 grams per liter, we should get about 100 grams of alcohol per liter or 10 percent or less a bit. Because waste is a by-product, but when calculated as a percentage by volume, we need to use the Alcohol specific gravity to calculate 0.7893, resulting in an increased alcohol content of more than 10 percent. For example, if having 96.3 grams of alcohol per liter, it will get 12.2 percent by volume. (9.63 divided 0.7893) in the yeast, fermentation will increase during the first 2-3 days, after which the growth will slow down until the number does not increase, but in this period Still increasing the amount of alcohol And the amount of sugar decreased And creating various flavoring substances During this period, so must continue to ferment Even though the yeast has stopped growing [23, 26-28]

3. Methods

3.1 Bagasse Preparation

The Bagasse is cut to 1-2 centimeters in size. It reduces the humidity by drying in the sun for two days, then drying at 80 degrees Celsius for 8 hours, then grinded with an ultra centrifugal mill and sorted. Particle size by sieve test until the bagasse size is obtained 500 micrometers by placing in a closed container at room temperature. [22, 29] The components were analyzed using the Detergent method, Neutral detergent fiber (NDF), Acid detergent fiber (ADF), and Acid detergent lignin (ADL) analysis. [28, 30]

3.2 Optimal Condition of Saccharification and Fermentation Bagasse

Bagasse from 3.1 was treated and fermented with *Aspergillus* and *saccharomyces* in different conditions, as shown in Table 1, and then analyzed for Reducing sugar content And The ethanol content [5, 12, 20, 28]

Table 1 The Condition of Saccharification and Fermentation Bagasse

| Sampling | Bagasse (g) | Time (day) | % of <i>Aspergillus</i> | % Yeast | % Water |
|----------|-------------|------------|-------------------------|---------|---------|
| 1 | 60 | 1 | 5 | 15 | 80 |
| 2 | 60 | 3 | 5 | 15 | |
| 3 | 60 | 7 | 5 | 15 | |
| 4 | 60 | 1 | 10 | 10 | |
| 5 | 60 | 3 | 10 | 10 | |
| 6 | 60 | 7 | 10 | 10 | |
| 7 | 60 | 1 | 15 | 5 | |
| 8 | 60 | 3 | 15 | 5 | |
| 9 | 60 | 7 | 15 | 5 | |
| 10 | 120 | 1 | 5 | 15 | |
| 11 | 120 | 3 | 5 | 15 | |
| 12 | 120 | 7 | 5 | 15 | |
| 13 | 120 | 1 | 10 | 10 | |
| 14 | 120 | 3 | 10 | 10 | |
| 15 | 120 | 7 | 10 | 10 | |
| 16 | 120 | 1 | 15 | 5 | |
| 17 | 120 | 3 | 15 | 5 | |
| 18 | 120 | 7 | 15 | 5 | |

3.3 Economic Feasibility Analysis from Ethanol Production was selected

The best condition of Bagasse Ethanol Production was to selected for economic calculation as Net present value (NPV), Return On Investment(ROI) and Payback Period, It was used from the 1st, 2nd and 3th equations. [27, 31-32]

$$NPV = \sum_{t=0}^N \frac{CF_t}{(1+r)^t} \dots\dots\dots[1]$$

CF = Cash flow
 r = Required rate of return
 t = year of cash flow
 N = the nth year

Return On Investment= ((Discounted Benefits Discounted Costs))/(DiscountedCosts).....[2]

Payback period = the number of years before full recovery+((Unrecovered cost at the start of the year)/(Cash flow during full recovery year)).....[3]

4. Results and Discussion

The study of the optimum conditions for ethanol production from Bagasse found that the physical characteristics of Bagasse after being exposed to the sun to expel moisture from the light yellow appearance, as shown in Figure 5. When studying the composition of Bagasse, the highest amount of Cellulose was 45.35, followed by hemicellulose 32.54 and Lignin equal to 5.96 percent, respectively, as shown in Figure 6.



Figure 5 Bagasse from Yala Province, Thailand

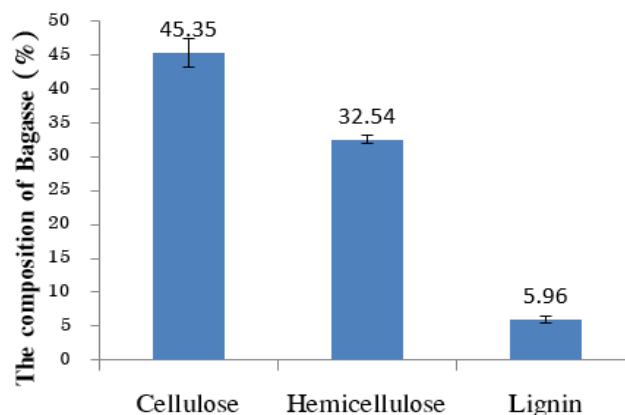


Figure 6 The lignocellulosic composition of Bagasse

When the samples were taken to study the optimum conditions according to Table 1, it was found that the amount of ethanol which using 60 grams of Bagasse mixed with *Aspergillus* sp. at 15 percent and yeast at 5 percent at a seven day fermentation time of 12.45 percent and with reducing sugar content of 285.65 grams per liter as follows shown in pictures 7 and 8

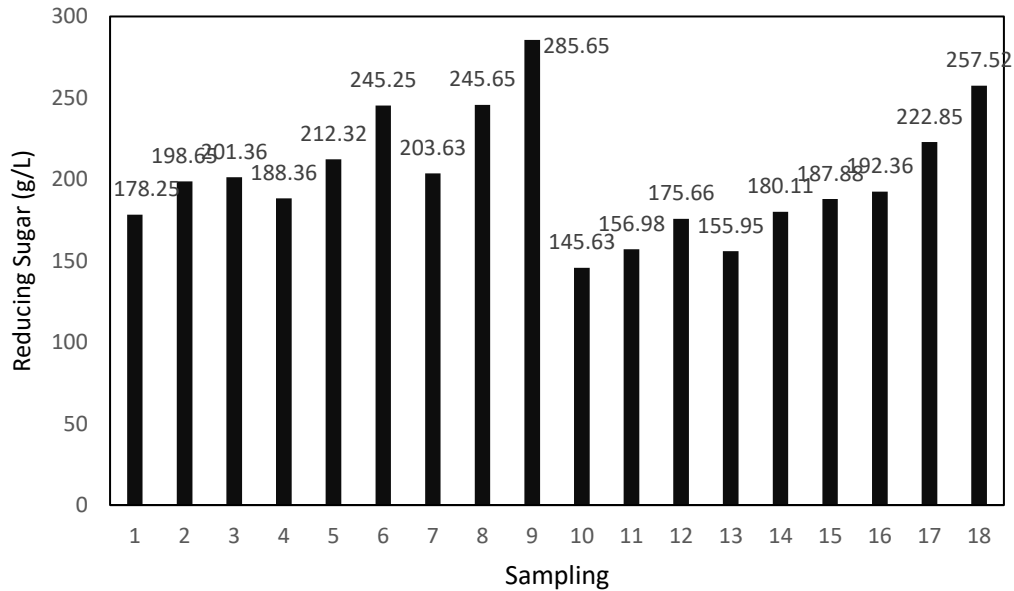


Figure 7 Reducing sugar content of Condition of Saccharification and Fermentation Bagasse

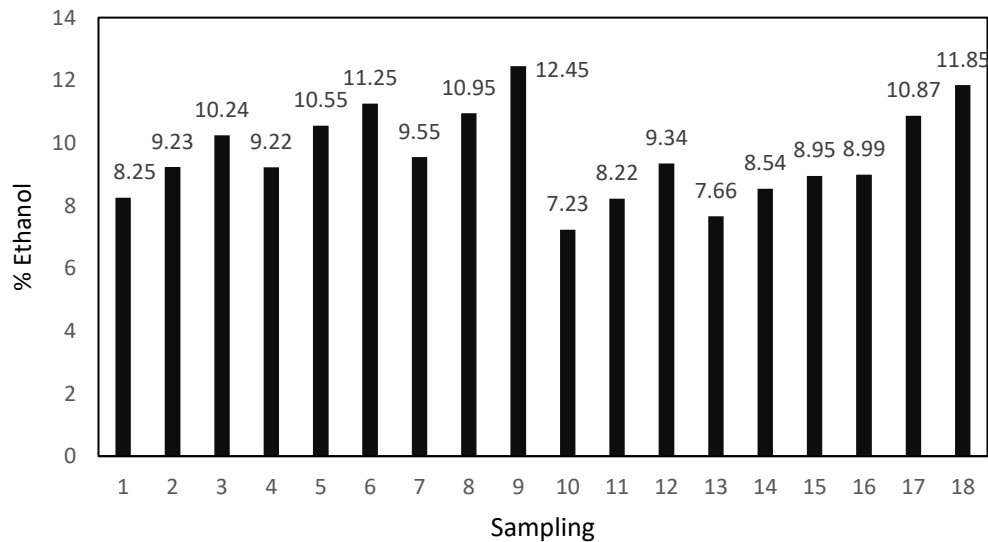


Figure 8 The percent ethanol content of Condition of Saccharification and Fermentation Bagasse

When the optimum conditions were used to distillate two rounds, then analyzed with Gas Chromatography Graph found that The first ethanol distillation cycle was 65.29. The second distillation was 92.05 percent, respectively. As shown in picture 9

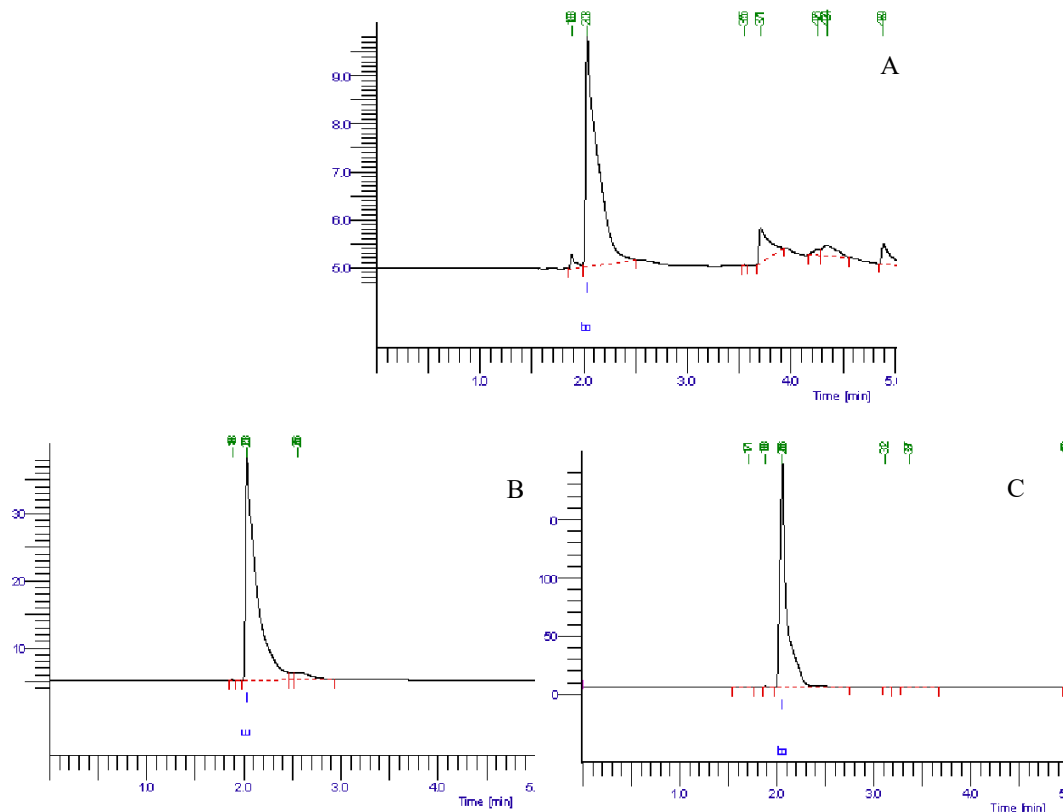


Figure 9 The amount of ethanol that has been analyzed by gas chromatography, where A is 12.45 percent of the amount of ethanol obtained from fermentation gave, B is 65.29 percent of the amount of the first distilled ethanol, and C is 95.05 percent of the amount 2nd ethanol distilled

When a study of economic feasibility analysis from the selected ethanol condition process has a sensitivity analysis and the payback period is approximately five months to seven months. In conclusion, the proposed project is attractive for the investors because the current price of ethanol is 18.21 THB (\$0.59) per liter, and its trend is expected to keep rising. By using the initial cost of an investment equal to 99,050 baht (\$ 3,225.97), divided into expenses such as *Bagasse*, *Aspergillus* sp, *S. cerevisiae* yeast, plastic bucket, water filter, Distiller, Sodium meta-bisulfite ($\text{Na}_2\text{S}_2\text{O}_5$), etc., As shown in Table 2

Table 2 The cost of ethanol production from Bagasse with *Aspergillus* and *S. Cerevisiae* yeast compared per year.

| Item | Unit | Cost(Bath) |
|-------------------------------------------------------------|--------------|------------|
| Bagasse | 100 kilogram | 200 |
| <i>Aspergillus</i> sp | 1 strain | 400 |
| <i>S. cerevisiae</i> yeast | 1 Strain | 400 |
| Plastic bucket | 30 Bottle | 900 |
| water filter | 1 | 12,000 |
| Distiller | 1 | 80,000 |
| Sodium meta-bisulfite ($\text{Na}_2\text{S}_2\text{O}_5$) | 1 kilogram | 150 |
| Other | - | 5,000 |
| Total | | 99,050 |

5. Conclusion

The optimal condition of saccharification and fermentation Bagasse converted to ethanol production. It was found that seven days after saccharification and fermentation from *Aspergillus* and yeast has the highest ethanol content gave $12.45 \pm 0.43\%$. While reducing sugar contents gave 285.65 ± 0.74 g/L. And the results of ethanol concentration from the first and the second distillation were shown as $65.29 \pm 0.57\%$ and $92.05 \pm 1.24\%$, respectively. When a study of economic feasibility analysis from the selected ethanol condition process has a sensitivity analysis and the payback period is approximately five months to seven months. In conclusion, the proposed project is attractive for the investors because the current price of ethanol is 18.21 THB (\$0.59) per liter, and its trend is expected to keep rising.

Acknowledgments

The authors would like to thank for supporting chemical and instrument in this research by Renewable Research Unit, Program of Renewable Energy Technology, Faculty of Science Technology and Agriculture, Rajabhat Yala University, Thailand; and Energy Laboratory, Program of Biotechnology, Faculty of Agro-industry, Rajamangala University of Technology Srivijaya, Thailand.

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