

Production of Biofuel (Bio-Ethanol) From Fruitwaste: Banana Peels

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Abstract: Bio-ethanol, a type of biofuel, is known as renewable energy source as it is derived from biomass as its raw material. Biomass can be found in abundance and sustainable i.e. sources are available continuously, unlike the currently used conventional fossil fuels where these sources are limited and depleting. In this study, biomass from fruit waste, banana peels, were utilized to produce bio-ethanol via hydrolysis and fermentation process. Banana peels, a lignocellulosic biomass, possesses compositions which favour these processes, where the banana peels are rich in cellulose content and low in lignin content. Mechanical pre-treatment of the banana peels was conducted to further ease the hydrolysis process by reducing the particle size of the biomass. Hydrolysis was carried out for 24 hours at 50°C at different pH using sulfuric acid H_2SO_4 acid and sodium hydroxide NaOH as the base, to study the effect of pH on the hydrolysis process and hence the final bio-ethanol production, in terms of concentration. Fermentation of the hydrolysis products were carried out using glucose-yeast broth for 4 days at temperature of 35°C. Water content in the bio-ethanol product from fermentation process was separated using rotary evaporator, prior to ethanol analysis using Gas Chromatography (GC-MS). Concentration of ethanol was found to be the highest at acidic pH conditions; pH 4 to 6. Lowest ethanol concentration was recorded at higher pH values, indicating alkaline conditions do not favour the hydrolysis process.

Index Terms: Banana Peels, Bioethanol, Fermentation, Fruit waste, Hydrolysis.

I. INTRODUCTION

Fossil fuels have been a major source of energy worldwide for ages and both natural gas as well as petroleum are going to deplete in the future. Furthermore, the use of fossil fuels for energy generation via combustion process also affects the climate change and is a major factor contributing to global warming issues. Biofuel or bio-ethanol is one of the various alternative sources of energy besides fossil fuel-derived energy; where the energy generation process using biofuels are much cleaner and environmental-friendly as compared to fossil fuels, natural gas and petroleum processes.

Biofuels are known as the alternative source of energy that can replace the need of petroleum and natural gas in the future since both of these sources are going to be consumed up sooner or later. Currently, the world has been relying on

fossil fuels as the primary source of energy, while other renewable energy sources are secondary source of energy, such as hydroelectric, wind, solar, and biomass-source related energy. Biofuels, which is derived from biomass sources, are known as the cleaner type of energy since biofuels are low in carbon intensity where it can't directly affect the global warming, hence reducing the impact on climate change. In the year 2010, the worldwide production of biofuel has reached an estimated value of 105 billion litres, in which biodiesel, another form of biofuel, contributed to 2.7% of the total fuel consumption for road transportation sector [1].

Fruit waste, which is part of food waste; the largest composition in the overall municipal solid waste (MSW) content at approximately 37% [2], possesses a great potential to be used as feedstock in the production of bioethanol. Moreover, fruit and vegetables are consumed in large scales globally every single day. After consumption of the fruits, around 50% of the initial weight of the fruits are recovered as food waste, hence making fruit waste a suitable raw material in bioethanol production [3]. Furthermore, the utilization of this abundantly available biomass to produce bioethanol can also help in solving both energy security and also waste management problem. Fruit wastes is a type of lignocellulosic biomass; a type of biomass whose major compositions are cellulose, hemicellulose, and lignin, in which the compositions of these three materials play an important role in determining the efficiency of the process where the biomass is being utilized [4].

Different types of fruit wastes possess different lignocellulosic compositions; some are rich in cellulose while some others are high in lignin content [5]. Table- I shows the lignocellulosic compositions for some common fruit waste, where the wastes are in form of fruit peels.

From Table- I, it can be seen that fruit wastes possess high moisture content and low ash content. In terms of lignocellulosic compositions, out of the four common fruit waste types found, banana peels are the most promising source, as banana peels have a high cellulose content as well as low lignin content.

Revised Manuscript Received on 20 October, 2019.

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Table- I: Lignocellulosic Compositions of Common Fruit Wastes (Source: Jahid.M, Gupta.A, & Sharma.DK, 2018)

Fruit waste	Banana peels	Pineapple peels	Papaya peels	Mango peels
Cellulose (% dry basis)	34.8	22.4	20.4	38.4
Hemicellulose (% dry basis)	9.4	11.1	24.6	13.9
Lignin (% dry basis)	4.5	6.5	2.7	27.9
Moisture content (%)	85.7	75.1	91.3	77.8
Ash (% dry basis)	12.9	11.8	5.8	4.0

In the structure of lignocellulosic biomass, as shown in Fig 1, the cellulose; major component in biomass, is constrained by the lignin structure, which acts as a shield for the cellulose and hemicellulose as well. The lignin structure in lignocellulosic biomass possesses a high resistance towards microbial attacks, in which the latter is necessary in hydrolysis process for the decomposition of cellulose into glucose; the major step in bio-ethanol production. Hence, high lignin content in the biomass will restrict access to the cellulose. Cellulose is the main component in biomasses that will undergo degradation steps to yield high glucose during hydrolysis. Therefore, higher lignin leads to lower glucose conversion from cellulose, and will also affect the yield of bio-ethanol at the end of the process; whether in terms of concentration or volume.

Banana biomass is one of the second generation of biomass where production of bioethanol from second generation interests many researchers. [6] The optimal temperature for banana to grow is at 26.67 °C where banana plant can grow easily within the Malaysia's geographical location since Malaysia climate is being hot and humid throughout the year [6]. Banana peel residue was exerted about 117,250 tonnes in year 2012 [6]. Ethanol is also known as alcohol which possess colorless, flammable, and volatile properties. Ethanol has a low density of 0.789 g/cm³, melting point of -114°C and the boiling point of 78.37°C. [7] Ethanol can be produced from several processes, such as ethylene hydration process; addition of water to ethylene, as well as fermentation process; conversion of sugars, starch, and lignocellulosic materials into alcohols in the presence of bacteria [7] From banana peel wastes, bio-ethanol can be extracted via several processes as shown in Fig. 2.

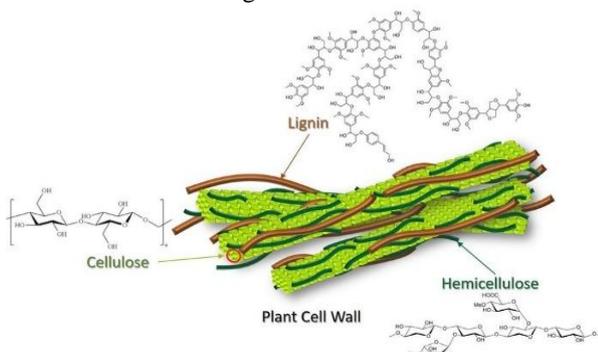


Fig. 1. Structure of Lignocellulosic Biomass

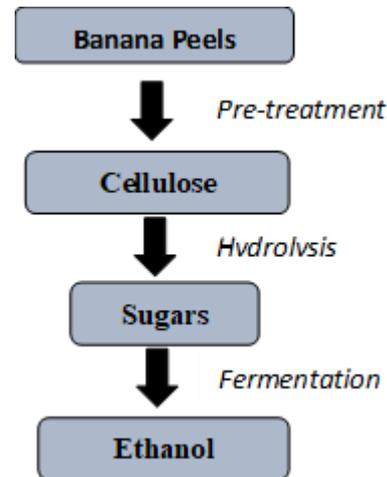


Fig. 2. Steps involved in conversion of banana peels to bio-ethanol

Pre-treatment process is the first step that are required in order to produce bioethanol where the lignocellulosic biomass will be pre-treated with either enzymes or acid in order to reduce the feedstock in terms of size. During pre-treatment process, the structure of the plant will be altered where the lignin seal shielding the cellulose and hemicellulose will be destroyed, and these two components will be reduced or converted from carbohydrates polymers to sugars monomers and fermentable sugars [8]. The production process of bio-ethanol will be proceeded with hydrolysis process where a chemical reaction occurs; releasing sugars or glucose. In early conversion of lignocellulosic biomass to bio-ethanol, acid was used in the hydrolysis process, but recent researches have also focused on the use of enzymes or also called as bio-enzymes. Enzymes action will attack the chains of sugars release during the hydrolysis process affectively which would lead to a higher yield of fermentable sugars [8]. Fermentation is the last step required in order to produce bioethanol where microorganisms are used to ferments the sugars that have been released during hydrolysis process. During this process, yeasts and bacteria are the most common types of microorganisms used in fermentation process. *Saccharomyces cerevisiae* is one type of yeast that usually used in the fermentation process where it also known as a baker yeast.

Banana peel fruit waste possesses high cellulose content and low lignin content, hence rendering it suitable for bio-ethanol production. The conversion of cellulose polymer to bio-ethanol involves different processes. The first step, pre-treatment is applied to separate the mixed polymers of hemicellulose, lignin and also the cellulose in order to provide the sugars needed for the hydrolysis and also fermentation process.

Pretreatment process is necessary in order to enhance the digestibility of solid content so that the hydrolysis process can be improved. Hydrolysis process is highly affected by particle size of the feedstock, structure and also the composition of the substrate [9]. Therefore, for every process an appropriate choice of pre-treatment is needed since the pre-treatment method used, whether physical,

chemical or physicochemical method, brings an impact on the subsequent steps in the overall conversion; significant or not. Physical pretreatment involves the disruption of the biomass structure where it aimed to maximize the accessibility of the surface area for the chemical and also enzymatic reaction in order to increase rate of the hydrolysis [9]. Physical pretreatment can be achieved by reducing the particle size through chopping, grinding or milling technique.

Biomass with fine particle size may generate clumps during the pre-treatments and also enzymatic hydrolysis which may lead to negative effects of total sugars produced [10]. Chemical pre-treatment is one of the mostly used type of pretreatment process, in order to enhance the biodegradability of organic substrate where the effect of this type of pretreatment process is highly dependent on the type of method or chemical used, as well as the characteristics of the biomass substrates. Chemical pretreatment is widely applied on the lignocellulosic substrates in order to remove lignin and also hemicelluloses where this would increase the pore size and the surface area of the substrate [9]. Acid hydrolysis is one of the commonly used chemical pre-treatment process, due to its ability to adapt with wide a variety of feedstocks [11]. The concentration of sugars produced from acid hydrolysis is dependent on the type of biomass used, the composition of substrates, temperature, time, acid concentration, and pH of hydrolysis. The pH effect gives an impact on the reactions which occur during the hydrolysis step, thus affecting the final result of the glucose or sugars produced, which in return, affect the bio-ethanol production during fermentation process which follows after the hydrolysis step. Previous studies on bio-ethanol production via hydrolysis and fermentation focused on various parameters, such as type of pre-treatment [12], and method of operation; whether separate hydrolysis and fermentation (SHF) method or simultaneous saccharification and fermentation (SSF) method [13]. However, limited researches have been conducted on effect of pH on the bio-ethanol production from banana peels. Similar research of pH effect has been conducted by [14] and [15], where these researches have focused on optimizing pH value for maximum bio-ethanol production from hydrolysis of microalgae. Hence, this study was carried out to determine the effect of pH on the banana peel-derived bio-ethanol production via hydrolysis and fermentation.

II. MATERIALS AND METHODS

A. Banana Peels

Bananas were collected from a local supermarket in Section 7, Shah Alam, Selangor, whereas the location, time, and type of banana collected was consistent throughout the research period. Chopping and grinding steps were carried out as mechanical pre-treatment to reduce the overall particle size of the banana peels. Dilute (5%) sulfuric acid H_2SO_4 was mixed with the grinded banana peels as second pre-treatment step; acid pre-treatment, to further alter the lignocellulosic structure of the banana peel biomass. The banana peels were weighed around 40 g and added into 200 ml of 5% H_2SO_4 . The mixture of banana peels and 5% of H_2SO_4 solution was

then placed inside autoclave for sterilization process for 15 minutes at $120^\circ C$. After 15 minutes of autoclaving process, the mixture was then cooled down at room temperature for 3 hours. After the mixture has cooled down, the solid residue and the solution were separated via filtration. The solid residues were then washed by using distilled water and dried by using microwave for 30 minutes at $40^\circ C$. Fig. 3 shows the condition of the banana peels after pre-treatment processes.

B. Hydrolysis Step

The dried banana peels from pre-treatment process were weighted and divided into six samples of equal weight. 10% of H_2SO_4 was added to the dried sample of banana peels with ratio of acid to dried sample (1.5:1) inside 250 ml of conical flask. The six samples for acid hydrolysis were prepared with different pH value within the range of 4 to 12. The pH value was adjusted via the addition of 1M of NaOH at different volumes. All of the samples were then incubated by using incubator shaker for 24 hours at $50^\circ C$.

C. Fermentation Step

In the fermentation step, glucose-yeast broth was used to ferment the sugars produced from hydrolysis step. The broth was prepared by mixing 10g of peptone, 5g of NaCl of sodium chloride and 3g of yeast extract in distilled water. The solution was then well-mixed by using glass rod stirrer. After the mixing process, the broth was then placed in incubator shaker for 24 hours before being used in the fermentation step.

To initiate the fermentation process, 25ml of the prepared glucose-yeast broth was added into the sample which has undergone complete hydrolysis process mentioned in part A. The samples were then placed again into incubator shaker at $35^\circ C$ and 200 rpm, as shown in Fig. 4. The fermentation process was carried out for 4 days. After the fermentation process, all of the samples were then centrifuged by using centrifuger at 10,000 rpm for 15 minutes, to separate the bio-ethanol layer from other contents, which was observed as in Fig. 5.

The water content in each sample were removed by using rotary evaporator, as shown in Fig. 6. where the water was separated from the bio-ethanol product. The process of the removal of water was done for 30 minutes for each sample.



Fig. 3. Banana peels after pre-treatment processes

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Fig. 4. Fermentation process in incubator shaker



Fig. 5. Fermentation products after centrifuge step



Fig. 6. Rotary Evaporator

III. RESULTS AND DISCUSSIONS

The analysis for each sample was done by using Gas Chromatography GC-MS, where the inlet temperature was maintained at 250°C and the injection volume was at 2µl. The GC was operated with a 1:50 ratio by using helium as the carrier gas. The concentration and percentage of ethanol was calculated by using Equations (1) and (2), respectively, as shown below:

Concentration of Ethanol :

$$= \frac{\text{Vol. of standard ethanol} \times \text{Area of unknown sample}}{\text{Area of Standard Ethanol}} \quad (1)$$

Percentage of Ethanol :

$$= \left(\frac{\text{Vol. of standard ethanol} \times \text{Conc. Ethanol}}{\text{Area of Standard Ethanol}} \right) \times 100 \quad (2)$$

The volume of standard ethanol used was 2µl meanwhile the area of standard ethanol was found to be around 1500. Both samples were run at room temperature since [7] reported that the optimum temperature in order to produce ethanol is around 30°C to 35°C. Therefore, all of the samples have been conducted under room temperature where it was approximately around 32°C. Table- II and Fig. 7 shows the data of percentage of ethanol that was obtained from the experiment.

From the results above, sample with pH value 6 possessed the highest percentage of ethanol, followed by the sample with pH value 4 where the percentage for both samples were 20.9433 and 19.6076 respectively. Meanwhile, sample at pH value 12 recorded the lowest value of percentage of ethanol at 8.50277 and at pH value 8 the percentage of ethanol at 18.3879. Hence, hydrolysis step performance was more efficient in the acidic region, as compared to the alkaline region. As observed from the data, the production of ethanol from banana peels favoured acidic condition in order to obtain a higher yield of ethanol. Similar trends in the results from this study were also observed in other bio-ethanol studies, such as [14] which recorded highest glucose yield from hydrolysis at acidic pH of 4.8, also in [15], which recorded 57% of glucose yield at pH of 5, and [7], in which this study observed highest glucose concentration when using acid pre-treatment. High glucose concentration and yield from hydrolysis will also give high concentration of bio-ethanol during the fermentation process. Furthermore, acidic conditions are also favoured by yeasts during the fermentation process where it is to ensure the yeast function at the minimal stress. The optimum pH value in the production of ethanol by using banana peels is highly dependent on the type of yeast that has been used for the fermentation process.

Table- II: Concentration of Ethanol at different pH values

pH	Concentration of ethanol (mg/ml)
4	147.0573
6	157.0747
8	137.9093
12	63.77067

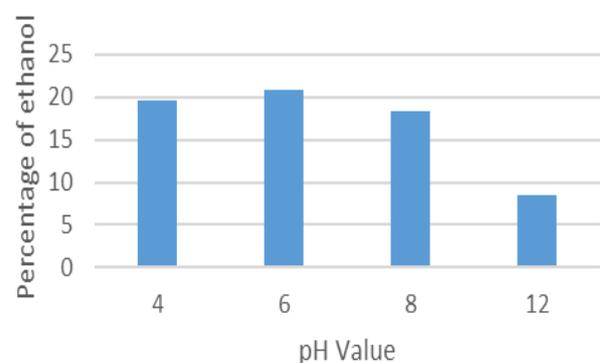


Fig. 7. Percentage of ethanol for pH values of 4, 6, 8 and 12

IV. CONCLUSION

In conclusion, hydrolysis of banana peels at different pH resulted in different ethanol concentrations at the end of the fermentation process. Highest ethanol concentration was recorded at pH of 6, 157.0747 mg/ml, followed by pH of 4, 147.0573 mg/ml, both acidic pH values, hence indicating that optimum ethanol production from banana peels is at acidic conditions for hydrolysis step. On the other hand, lowest ethanol concentration was recorded at pH of 12, 63.77067 mg/ml, indicating that alkaline conditions do not favour hydrolysis step to produce high glucose yields and high ethanol yields.

ACKNOWLEDGMENT

The authors would like to thank the Universiti Teknologi MARA (UiTM) and Ministry of Education Malaysia for the financial supports. The research is conducted at the Faculty of Chemical Engineering, UiTM under the support of FRGS grant (600-IRMI/FRGS 5/3 (189/2019)).

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