Jour. Myan. Acad. Arts & Sc. 2016 Vol. XIV. No. 1

OBSERVATION ON THE YIELD OF BIOETHANOL FROM REJECTED PAPAYA AND PINEAPPLE

Aye Aye Aung[1], Soe Soe Than[2], Pansy Kyaw Hla[3]

Abstract

Rejected Papaya (*Carica papaya* L.) and Pineapple (*Ananas comosus* L.) were considered as sources of sugary material for the preparation of bioethanol. Direct fermentation of rejected and sterilized pulp was carried out by using baker's yeast (*Saccharomyce cerevisiae*) under anaerobic conditions. Different fermentation conditions such as amount of substrate such as (100, 200, 300, 400, 500 and 600)g, amount of yeast such as (0.2, 0.4, 0.6, 0.8, 1.0 and 1.2) % (w/w), temperature of fermentation such as (28, 30, 32, 34, 36 and 38) °C and different pH such as (3.5, 4, 4.5, 5, 5.5 and 6), period of fermentation (1, 2, 3, 4, 5 and 6) days were employed during fermentation. The strength of the resulting bioethanol was analyzed by using gas chromatography (GC). It was pointed out that the maximum strength of bioethanol 20.154 % (v/v) was obtained from sterilized pulp of papaya and 85.276 % (v/v) from sterilized pulp of pineapple respectively.

Key words: Bioethanol, rejected fruits, sugary material, fermentation

Introduction

Bioethanol is being widely explored as a renewable liquid fuel because it is superior to gasoline fuel under many considerations.There is a great demand for ethanol as it is widely accepted for clean burning. In many countries, ethanol is used as either an alternate fuel or blended with gasoline.

Selection of suitable and economical source is considered as an important cost component for industrial ethanol production.Technically, ethanol can be produced from a wide variety of renewable feedstock, which can be roughly classified into three main groups: (1) those containing considerable amounts of readily fermentable sugars (sugar cane, sugar beets, and sweet sorghum), (2) starches (corn, potatoes, rice, wheat) and (3) celluloses (cornstover, grasses, corn cobs, wood, sugar cane bagasse). Sugary materials such as sugar cane, sugar beet and sweet sorghum maintain the simple sugars, as sucrose, glucose and fructose that can be readily fermented by yeast. Carbohydrates such as starch from cereal and tuber crops are chemically or enzymatically converted into simple sugars and sugars are then

1. PhD candidate, Department of Industrial Chemistry, Dagon University.
2. Dr.,Associate Professor, Department of Industrial Chemistry, University of Yangon.
3. Dr.,Professor (Retired), Department of Industrial Chemistry, University of Yangon.

fermented using yeast to produce a mash containing ethanol and water. Ethanol is separated from the fermented mash by distillation (www.gea.wiegand).The conversion of celluloses to monosaccharide usually involves three steps: pretreatment, acid or enzymatic hydrolysis, and fermentation (Yu et al., 2010).

Many investigators have studied the production of ethanol using various raw materials. Fruit wastes are also included as the cheapest and easily available source of sugary material for the production of bioethanol and considered as a potential energy source (IJIRSET, 2013).The fruit wastes such as papaya, mangoes, banana peels, pineapple and grapes were used as sources of sugary material. Yeast and fungus are commonly used for fermentation of sugary materials. *Saccharomyces cerevisiae* strains are being usually used in fermentation.

In this study, ethanol was prepared from different rejected fruits such as papaya (*Carica papaya* L.) and pineapple (*Ananas comosus* L.) by fermentation. Fermentation of pulp was carried out using different amount of substrate, amount of yeast, at various temperature of fermentation, pH and period of fermentation.

Materials and Methods

Materials

Rejected fruits such as pineapple (*Ananas comosus* L.), produce of Thipaw Township, Northen Shan State and papaya (*Carica papaya* L.) produce of Kyonpyaw Township, Ayeyarwady Region were collected from Thiri Mingalar Market, Hlaing Township, Yangon Region. Potassium sodium tartrate (analar grade), anhydrous Copper II sulphate (analar grade), sodium hydroxide (analar grade) and *Saccharomyces cerevisiae* (baker's yeast) were purchased from Kemiko,(Chemical Dealers), 28th Street, Pabedan Township, Yangon.

Methods

Fermentation Process

Rejected fruits were peeled and other spoiled parts were removed. Then, flesh of fruits were washed, sliced and ground into pulp using a blender. Some pulp were sterilized in an autoclave at 121°C for 15 minutes. Some pulp were fermented with baker's yeast (*Saccharomyces cerevisiae*) under anaerobic conditions. Fermentation of 500 g of sterilized pulp was conducted with 0.6% (w/w) yeast for three days. Its pH was maintained at pH 4 and the temperature was kept at 32°C. The same procedure was carried out for rejected pineapple pulp. For papaya, 500 g of sterilized pulp was fermented with 0.4% (w/w) yeast for five days, keeping the pH of 4.5 and temperature of 32°C. The same producer was carried out for rejected papaya pulp. The highest strength of ethanol was observed under controlled conditions of fermentation process. Fermentation was maintained at different amount of substrate such as 100, 200,300, 400, 500 and 600 g, amount of yeast such as 0.2, 0.4, 0.6, 0.8, 1.0 and 1.2 % (w/w), temperature of fermentation such as 28, 30, 32, 34, 36 and 38 °C, at different pH such as 3.5, 4, 4.5, 5, 5.5 and 6 and , period of fermentation such as 1, 2, 3, 4, 5 and 6 days were employed during fermentation. Ethanol was then separated from the fermented broth by distillation at 78 ± 1°C. The first distillate was further purified by fractional distillation at 78 ± 1°C using fractionating column (50cm in length, 2cm in outside diameter, 1.75 in inside diameter) packed with glass tubes (length = 1.5 cm, diameter = 0.25cm). After 4 hr fractionating time, the resulted second distillate was analyzed by gas chromatography (GC).

Determination of Ethanol Content

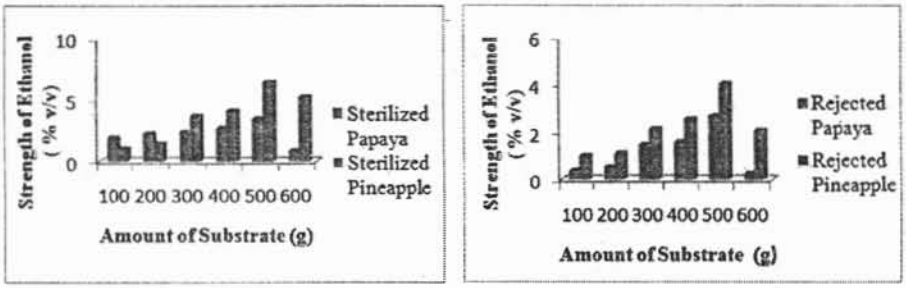
The strength of ethanol for first distillate was measured by specific gravity method. Gas Chromatography (GC) (2010, SHIMADZU) was then used for identification of second distillate at the Laboratory of Australasia Marketing Trade & Technology Co. Ltd (AmttCo. Ltd), Mayangone Township,Yangon Region.

Results and Discussion

In this research work, pulp of rejected papaya and pineapple were used as sources of sugary material and converted into ethanol and carbon dioxide through fermentation by inoculating the baker's yeast, *Saccharomyces cerevisiae.*

Effect of Amount of Substrate

Figure (1) (a) and (1) (b) pointed out that 500 g of substrate was the suitable amount for sterilized rejected pineapple and rejected pineapple to obtain the maximum strength of ethanol 6.45% (v/v) and 4.07 % (v/v) respectively. For sterilized rejected papaya and rejected papaya, 500 g of substrate was the suitable amount for the maximum strength of ethanol 3.5 % (v/v) and 2.71 % (v/v) respectively. When the amount of substrate was increased, the strength of ethanol was decreased. In the fermentation process, yeast consumed sugary materials as source of carbon and microorganism to food ratio may influence on its metabolic action to produce ethanol.



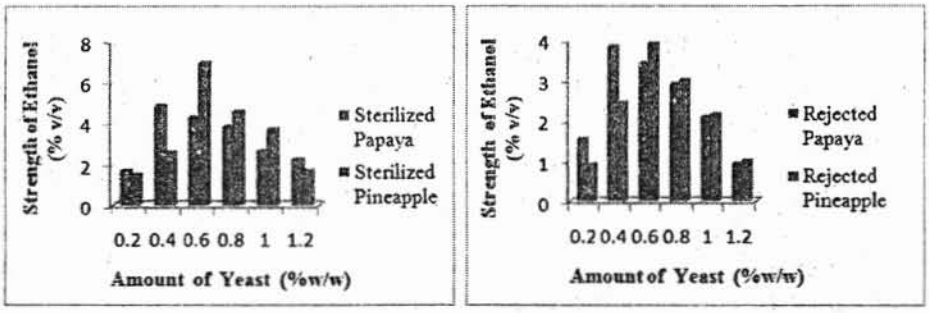
(a) (b)

Figure (1) Effect of Amount of Substrate on the Strength of Ethanol for (a) Sterilized and (b) Rejected Pulp of Papaya and Pineapple

Effect of Amount of Yeast

From the results of Figure (2) (a) and (2) (b), the maximum strength of ethanol 6.92% (v/v) and 3.92 % (v/v) for sterilized rejected pineapple and rejected pineapple were obtained by using the same amount of yeast 0.6 %(w/w).The maximum strength of ethanol 4.87 % (v/v)and 3.85 % (v/v) for sterilized rejected papaya and rejected papaya were obtained by using the same amount of yeast 0.4 % (w/w). When the amount of yeast was increased,

the strength of ethanol gradually decreased.This fact showed that the amount of yeast inoculated affect the yield of ethanol. It was also observed that higher ethanol content has resulted for both sterilized pulp of pineapple and papaya in which fermentation was based on substrate amount and carried out by using inoculated baker's yeast only. On the other hand, fermentation process of rejected pulp may contain backbone microorganisms apart from inoculated baker's yeast.



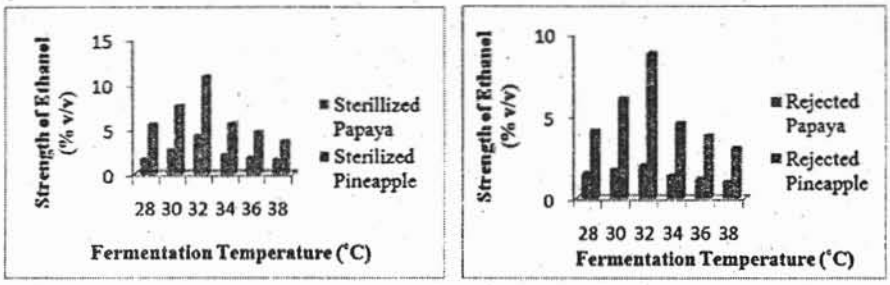
(a) (b)

Figure (2) Effect of Amount of Yeast on the Strength of Ethanol for (a) Sterilized and (b) Rejected Pulp of Papaya and Pineapple

Effect of Fermentation Temperature

Fermentation Temperature is one of the most important factors that affect the ethanol production by yeast using sugary material as a carbon source. The fermentation process is always accompanied with the evolution of heat that raises the temperature of the fermenter and fermentation. Ethanol fermentation at enhanced temperature is a main necessity for effective ethanol production. It is, therefore, necessary to select the optimum temperature at which yeast strains can ferment the sugars. The rate of alcoholic fermentation increases with the increase in temperature (William, 2008). Figure (3) (a) and (3) (b) show the effect of fermentation temperature on the strength of ethanol. The maximum strength of ethanol 10.92% (v/v) and 8.845 % (v/v) for sterilized rejected and rejected pineapple pulp and the maximum strength of ethanol 4.285 % (v/v) and 2.03 % (v/v) for sterilized rejected papaya and rejected papaya pulp have resulted at the fermentation temperature of 32°C. The maximum yield of alcohol for both samples was obtained at that temperature. It was found that increasing the temperature, above 32 °C, could cause reduction in the yield of ethanol. It may be mainly due to the denaturing

of the yeast cells at elevated temperature. It was also observed that when temperature goes below the optimal range, their ability to catalyze the intended reaction slows down. On the other hand, when the temperature increases, the cells begin to denature and thus become inactive.

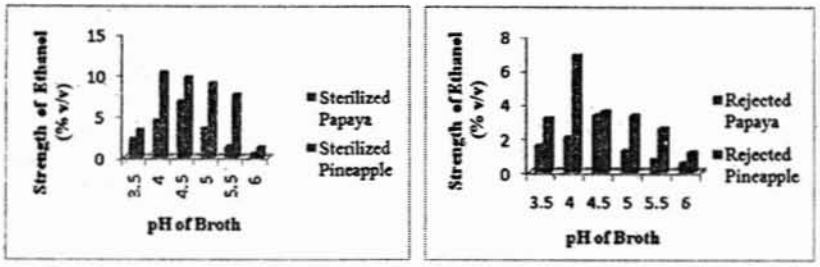


(a) (b)

Figure (3) Effect of Fermentation Temperature on the Strength of Ethanol for (a) Sterilized and (b) Rejected Pulp of Papaya and Pineapple

Effect of pH

The pH is an important environmental factor affecting cell growth and metabolite production. Therefore, several studies investigating the influence of pH on microorganisms have been published. With increase in pH, yeast produces acid rather than alcohol (IJIRSET, 2013). pH during fermentation should be controlled at low pH for two reasons: (1) the growth of other harmful microorganisms is retarded by acidic solution: (2) yeast grows well in acidic condition. Yeast survives in a slightly acidic environment with the pH between 4 to 6. As shown in Figure. (4)(a) and (4)(b), the maximum strength of ethanol 10.42 % (v/v)and 6.84 % (v/v) have resulted for both the sterilized rejected pineapple and rejected pineapple pulp at the optimum pH 4. The maximum strength of ethanol 6.85 % (v/v) and 3.35 % (v/v) had resulted for both the sterilized rejected papaya and rejected papaya pulp at the optimum pH 4.5.

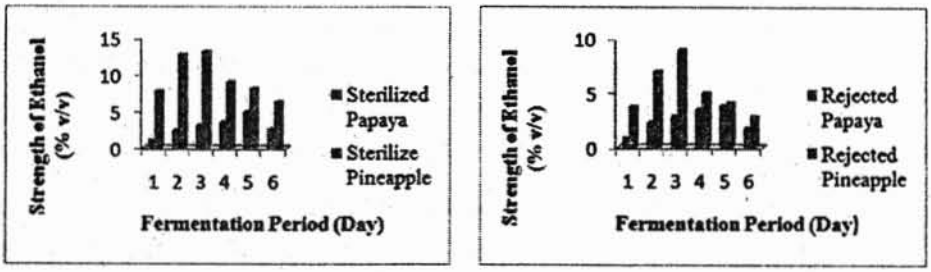


(a) (b)

Figure (4) Effect of pH of Broth on the Strength of Ethanol for (a) Sterilized and (b)Rejected Pulp of Papaya and Pineapple

Effect of Fermentation Period

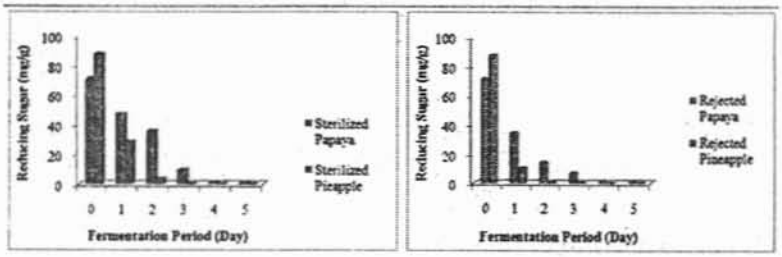
Longer fermentation period may offer some infection and unwanted microorganisms such as acetobacter and lactobacillus that could produce vinegar and lactic acid. In addition, a small amount of glycerol and lactic acid may be formed during fermentation process (William, 2008). From the results shown in Figure (5)(a) and (5) (b), the maximum strength of alcohol of 13.23% (v/v) and 9.01% (v/v) were obtained at the fermentation period of 3 days for sterilized rejected pineapple and rejected pineapple pulp respectively. The maximum strength of alcohol of 4.99 % (v/v) and 3.89 % (v/v) were also obtained at the fermentation period of 5 days for sterilized rejected papaya and rejected papaya pulp respectively. It has been observed that the strength of ethanol did not increase even though its longer fermentation period was subjected to. In addition, fermentation period of papaya pulp was longer than that of pineapple pulp. It may depend on the structure and composition of fruits.



(a) (b)

Figure (5) Effect of Fermentation Period on the Strength of Ethanol for (a) Sterilized and (b) Rejected Pulp of Papaya and Pineapple

From the results as shown in Figure (6)(a) and (6)(b), the reducing sugar of sterilized and rejected pulp of pineapple during fermentation decreased from 87.916 mg/g to 0.130 mg/g and to 0.032 mg/g on the 3rd day, whereas the reducing sugar of sterilized and rejected pulp of papaya decreased from 71.851 mg/g to 0.871 mg/g and to 0.598 mg/g on the 5th day respectively. It showed that baker's yeast used was well adapted in the chosen conditions by consumption of sugar as sole carbon source. During fermentation, the inoculated yeast consumed the maximum amount of reducing sugar.



(a) (b)

Figure (6) Amount of Reducing Sugar under Controlled Conditions of Fermentation (a) Sterilized and (b) Rejected Pulp of Papaya and Pineapple

The strength of ethanol identified by GC analysis is shown in Table1. Based on the GC analysis, both sterilized and rejected pineapple pulp had the highest ethanol content when compared to the ethanol content of sterilized and rejected papaya pulp. The maximum strength of ethanol 85.276 % (v/v) and 72.172 % (v/v) were obtained for sterilized rejected pineapple and rejected pineapple meanwhile 20.154 % (v/v) and 16.418 % (v/v) had resulted for sterilized and rejected papaya respectively.

Table 1. The Strength of Ethanol by GC Analysis

Sr.No	Sample	Strength of ethanol (by GC)(%v/v)
1	Sterilized papaya pulp	20.154
2	Sterilized pineapple pulp	85.276
3	Rejected papaya pulp	16.418
4	Rejected pineapple pulp	72.172

Conclusion

Since the rejected fruits have not been animal feed, they should be considered for bioethanol production. In this study, the satisfactory strength of ethanol specifically 85.276 % (v/v) has been resulted from sterilized pineapple pulp.

Acknowledgements

We would like to express our gratitude to Professor DrYin Shwe, Head of Department of Industrial Chemistry, Dagon University, for permission to use laboratory facilities at the Department of Industrial Chemistry. We are also grateful to Dr Khin Thet Ni, Head of Department of Industrial Chemistry, University of Yangon, for giving permission to perform this research, and advice throughout this research work.

References

Byarugaba, G.W., *The Effect of Enzymatic Processing on Banana Juice and Wine*, Institute For Wine Biotechnology at Stellenbosh University, Faculty of AgriScience, Doctor of Philosophy Science, (Unpublished).

International Journal of Innovative Research in Science, (2013),"*Comparative Studies of Ethanol Production from Different Fruit Wastes Using Saccharomyces cerevisiae*", Journal Engineering and Technology (IJIRSET) , vol 2,India.

Kumar, H.D., (1998), Mordern Concept of Biotechnology, Banaras Hindu University, Varanasi.

Kumar Soni.N, (2014),*Deviation in Production of Bioethanol Applying Stress Tolerant Yeast: A Current Approach,* International Journal of Pharmaceutical Science Invention Volume 3 Issue 5,PP.10-19