

Comparative Studies of Bioethanol Production from Different Fruit Wastes Using *Saccharomyces cerevisiae*

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Abstract: - Biologically produced alcohol, most commonly bioethanol are produced by the action of microorganisms and enzymes through the fermentation of sugars. Bioethanol is another biofuel capable of providing enough energy when burnt to be used as a fuel for transport. The production of bioethanol must be increased using cheaper and ecofriendly raw materials. Based on these characteristics, fruit wastes can be considered as cheaper and ecofriendly. In this study different fruit wastes were used as a raw material for the production of bioethanol by using *Saccharomyces cerevisiae* and the result were compared. The results of this work shows that the rate of ethanol production through fermentation fruit waste by *Saccharomyces cerevisiae* yield is very high at temperature 33° C than other fruit wastes. This study suggest that wastes from fruits that contain fermentable sugar should not be discarded into our surroundings or environment, but should be converted to useful products like bioethanol that can serve as an alternative energy sources.

Keyword: *Saccharomyces cerevisiae*, microorganisms, biofuel, bioethanol, fermentation, ecofriendly, cheaper, alternative energy sources.

1. INTRODUCTION

Ethanol has been described as one of the most exotic synthetic oxygen – containing organic chemicals because of its unique combination of properties as a solvent, a fuel. Ethanol fuel is the most common biofuel worldwide. Ethanol can be used in petrol engines as a replacement for gasoline. Ethanol has a smaller energy density than that of gasoline; this means it takes more fuel (volume and mass) to produce the same amount of work. The main sources of sugar required to produce ethanol come from fuel or energy crops. These crops are grown specifically for energy use and include corn, maize, and wheat straw. Ethanol or ethyl alcohol (C₂H₅OH) is a clear colourless liquid, it is biodegradable, low in toxicity and causes little and causes little environmental pollution if split. Ethanol burns to produce carbon dioxide and water. The first generation of ethanol production used corn as a substrate, later corn was considered, as a feedstock lead to the second generation of production of ethanol which used microorganisms and different wastes as substrate.

The cheapest and easily available source for the production of bioethanol is fruit wastes. It is a potential energy sources, from which ethanol can be obtained. Fruit waste is thrown outside or away has very good antimicrobial and antioxidant potential. The ethanol produced from fermentation process by different fruits wastes such as sweet lime, papaya, pineapple, water melon, banana were compared in this study. The ethanol concentration was estimated by alcohol estimate ion method (potassium dichromate K₂CR₂O₇ method). *Saccharomyces cerevisiae* is used in the fermentation process since it convert sugars with oxygen to give carbon dioxide.

(*Carica papaya*) is one the most commonly used fruit available with highest energy sources and invert sugar. Bananas (*Musaceae*) are readily available agricultural waste that is under utilized as potential growth medium for yeast strain. (*Citrus limetta*), is commonly known as sweet lime, and they are cheap and suitable substances for growth. Water melon (*Citrullus lanatus*) is cheaply available fruit and its belongs to the family *Cucurbitaceae*. Pineapple (*Ananas comosus*) is a tropical plant with edible multiple fruit consisting of coalesced berries and the most economically significant plant in the *Bromeliaceae* family. On the basis of these characteristics, the fruits were chosen for the production of ethanol. The main objective of this study is the comparison of the ethanol efficiency obtained from the different fruit wastes.

2. MATERIALS AND METHODS

2.1 Materials and Equipments

In this research the chemicals used are 5% potassium permanganate (KMnO₄), 25g of sucrose, 0.5g of urea, *Saccharomyces cerevisiae* strains 5ml, five different fruit wastes such as papaya, banana, sweet lime, watermelon, pineapple. The equipments used are Erlenmeyer flasks, incubator, distillation unit.

2.2 Preparation of the substrate for the fermentation process

About 100g of each fruit wastes such as papaya, banana, sweet lime, watermelon, pineapple (fig 1) were weighed separately and were taken in five different beakers, which was washed with 5% potassium permanganate solution and then washed with distilled water. The five wastes were crushed separately in a mixer and collected in five different beakers.



Fig 1: Different fruit wastes

0.5g of urea, 25g of sucrose and 5ml of *saccharomyces cerevisiae* was mixed in warm water in a separate beaker. This mixture was taken as an inoculum for fermentation process for each fruit mash. The fruit mash and inoculum were transferred into conical flask (1 liter) and made up to the final volume 500ml with distilled water. The samples were subjected to fermentation process for 7 days, kept at an incubator at 36° C. During fermentation process, the enzyme zymase from yeast changes the simple sugars into ethanol and releases the carbon dioxide.

After the process the sample was distilled using simple distillation unit to collect the ethanol from the different fruit wastes. For distillation batch distillation method was adopted (fig 2). The components of distillation units are: a reboiler, condenser pipe and a distillate or receiving flask. The filtered samples were transferred into the reboiler and the samples were boiled for each fruit waste. The vapor started to rise into the still head and passed through the condenser pipe. The continuous circulation of cold water around the condenser pipe assisted in cooling the alcohol rich vapors back to the liquid state. The condensed liquid enters the still receiver and is then collected in the distilled. To find out the presence of ethanol iodine test was made. Then the final tests such as alcohol estimation test, reducing sugar analysis test to find the concentration of bioethanol were determined. The amount of ethanol obtained from different fruit wastes was recorded and the solution was subjected to iodine test to confirm the presence of ethanol.



Fig 2: Recovery of the product

3. RESULTS AND DISCUSSION

In this study, it has been shown that ethanol was produced from different waste. Thus the comparative study has been carried out to check the efficiency of ethanol produced from these fruit wastes by determining various parameters. The influence of various parameters on the production of ethanol is presented as follows

3.1 Effect of temperature

Temperature plays a major role in the production of ethanol, since the rate of alcoholic fermentation increases with the increase in temperature. The optimum temperature of ethanol ranges between 25° C to 40° C which depends on room temperature. Bioethanol produced from pineapple has a temperature of about 33° C, and ethanol from watermelon has 25° C temperature, and papaya has 27° C and banana has 30° C temperature, whereas sweet lime has a temperature of about 29° C. When temperature goes below the optimal range, their ability to catalyze the intended reaction shows down (table 1; fig 3). On the other hand, when the temperature increases, enzymes begin to denature or unfold and thus become inactive. Each enzyme will have a different temperature range where it becomes inactive. Even if one essential enzyme stops working, the organisms fail to grow. Hence, the first essential enzyme that gets deactivated defines the maximal temperature at which that organisms can grow. At the lower end, it gets more complicated. Usually, the enzymes are not inactivated but rather just shown below.

Table 1: comparison of production of bioethanol at different temperature

S.NO	SAMPLE	TEMPERATURE
1	Watermelon	25° C
2	Banana	30° C
3	Pineapple	33° C
4	Papaya	27° C
5	Sweet lime	29° C

3.2 Method of analysis

The analysis of fruit wastes were carried out to assess the contents of the various components in them.

3.3 Reducing Sugar Analysis

Reducing sugar analysis was estimated by using DNS (dinitrosalicylic acid) method (miller, 1959). It involves preparation of the standard reference curves the method for which is given below

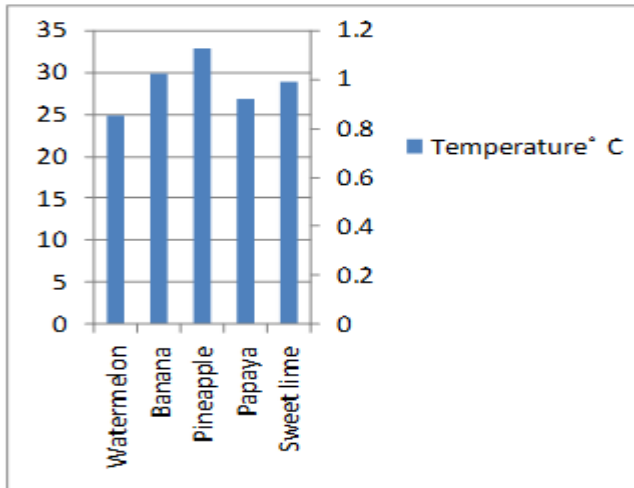


Fig 3: Comparison of bioethanol production of different temperature obtained from different fruit sample.

3.4 Preparation of standard curves

10 mg of D- sucrose was dissolved in 50 ml distilled water. Aliquots of triplicates of 0.2, 0.4, 0.6, 0.8, 1ml sucrose solution were taken and volume adjusted to 1ml using distilled water. To each tube 1ml of DNS reagent was added and boiled for 15 min. The absorbance was measured at 540 nm by using a digital photo calorimeter. See in fig 4.

3.5 Alcohol Estimation by Potassium Dichromate Method

Alcohol content of the extract was estimated by the Potassium Dichromate Method.

The standard reference curve was prepared for this To each tube add 3ml of dichromate reagent and it was followed by adding 1ml of conc. Sulphuric acid. Absorbance was measured at 620 nm by using spectrophotometer and it was shown below

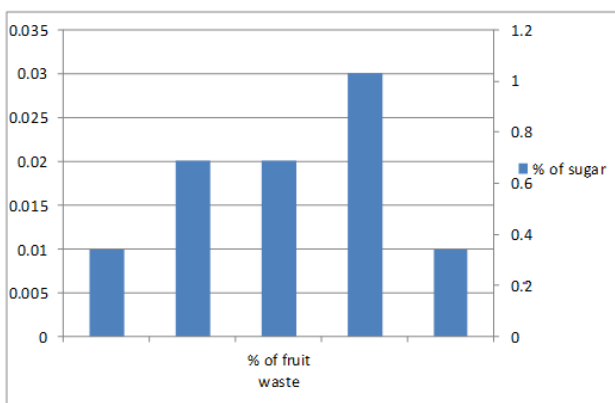


Fig 4: sugar estimation for % of fruit wastes

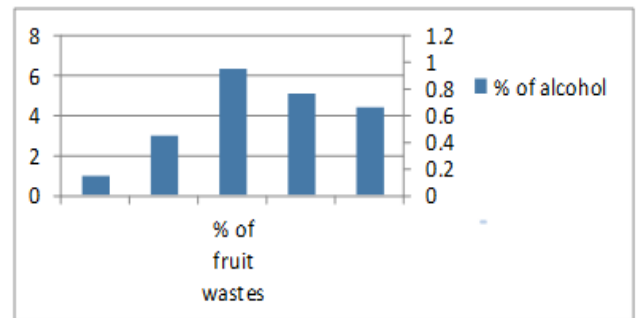


Fig 5: Alcohol estimation for various % of fruit wastes

The result of this work has shown that different fruit waste can serve as raw material for the ethanol was obtained from pineapple wastes at temperature 33 °c, and concentration of ethanol 6.3 % which is appropriately close to constant value of ethanol. Finally pine apple fruit waste shows the higher efficiency than the other fruit wastes.

4. CONCLUSION

Bio ethanol is a good alternative source compared to other fossil fuel. It can used to run the vehicles, because of these characteristics it can be utilized for future prospective.

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