

THE FERMENTATION OF BANANA (*Musa acuminata*), MANGO (*Mangifera indica L.*) AND PINEAPPLE (*Ananas comosus*) MASH IN THE ABSENCE AND PRESENCE OF ADDITIVES

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ABSTRACT

There has been increased interest in the use of renewable energy such as Bio-ethanol, with a view to decrease dependence on fossil fuel as the world population increases and to curb global warming. Bio-ethanol can be used for a variety of purposes, of which blending with gasoline to produce gas alcohol to power automobiles is on the increase. Ethanol can be obtained via the fermentation of glucose or sucrose under the influence of *Saccharomyces cerevisiae* at room temperature or acid hydrolysis of lignocellulose material followed by subsequent fermentation. Good sugar rich sources include ripe fruits etc. In this research, the fermentation of fruit mash: Banana, *Musa acuminata* (500g), pineapple, *Ananas comosus* (500g), and mango, *Mangifera indica* (500g) with an initial total soluble solid, TSS of 18.0, 12.2 and 13.4 °Brix was conducted at room temperature (30.8°C) at a pH of 4-5 over a period of three days in triplicates. Experiments were also subsequently conducted in the presence of additives such as 1%, 5% and 10% Urea, Ammonium Tartrate and Zinc Sulphate under the same conditions. In the absence of additives, the alcohol strength was found to be in the range 1.42 ± 0.43 %, v/v to 5.41 ± 1.92 , v/v using cultured yeast and in the range 5.51 ± 0.61 , v/v to 6.81 ± 1.75 , v/v using uncultured yeast. In the presence of additives, the alcohol strength was (6.19, 4.88, 5.1) %, v/v for the ZnSO₄ solution at the specified concentration. Urea at 1%, 5% and 10% induced alcoholic strength of (4.25, 4.67 and 4.39) %, v/v, whereas Ammonium tartrate induced alcoholic strength of (5.08, 5.44 and 4.88) %, v/v respectively. This research should be of interest to the Agroindustry.

Keywords: Fossil fuel, global warming, renewable energy, fermentation, banana, pineapple, mango, *Saccharomyces cerevisiae*, additives, alcoholic strength.

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INTRODUCTION

With a view to decrease dependence on fossil fuel as a result of depletion, increasing global fuel price, increasing population and increasing global warming, there has been increased interest in the use of renewable energy sources of which bioethanol is one ^{1,2,3}. Bioethanol (b.p: 78.5°C) can be used for a variety of purposes, of which blending with gasoline to produce gas alcohol to power automobiles is of current utilisation ^{1,2,3}. It's a clean burning renewable energy source⁴. Ethanol is also an important component of alcoholic beverages such as wine, beer, cider, vodka, gin. whisky, brandy etc. It is also an important starting materials for aldehydes, ketones, carboxylic acid, carboxylic acid derivatives and the hydroxyl group is a component of many pharmaceutical drugs ⁵.

Ethanol doesn't have significant environmental impact as fossil fuel combustion ³. It has low air polluting effect and low atmospheric photochemical reactivity, further reducing impact on the ozone layer⁶. It contributes little net CO₂ accumulation to the atmosphere and thus should curb global warming⁶. To solve the above problem, one alternative is to produce bioethanol from fruits, other grown organic matter or waste^{3,4,6-8,13}.

Bioethanol can be obtained via the fermentation of glucose, fructose or sucrose under the influence of *Saccharomyces cerevisiae* at room temperature, ^{4,6-25}. Also, acid hydrolysis of lignocellulose material followed by subsequent fermentation ^{3,10}. Sugar rich sources include ripe fruits ⁸⁻¹⁸ etc. Other sources include biodegradable fraction of products, waste and residues from agriculture like vegetables and animal origin ^{10-12, 17, 20-21} etc. The percentage yield of ethanol ranging from 6-10% have been reported ³⁻¹².

Fermentation is the process of energy production in a cell in an anerobic environment with the lack of an external electron acceptor ²². Sugars are the common substrate of fermentation and the products include ethanol, lactic acid and hydrogen. In some instances compounds such as butyric acid and acetone are produced ²². During Fermentation, starch is first hydrolysed to maltose by the action of the enzyme diastase. This enzyme is obtained from germinating barley seeds or malt. Maltose is converted to glucose by the enzyme maltase. Glucose is then fermented to ethanol via the enzyme zymase ²², Fig. 1.0.

This paper reports the use of fruits (mango, pineapple and banana) mash as fermentation substrates to produce ethanol in higher yield in the presence and absence of additives (ZnSO₄, Urea and ammonium tartrate) and to promote the Agro economy. It is anticipated that a higher

% yield of ethanol than that which is usually achieved using *Saccharomyces cerevisiae* under the temperatures that are normally used (35°C, 40°C and 45°C) is achieved.

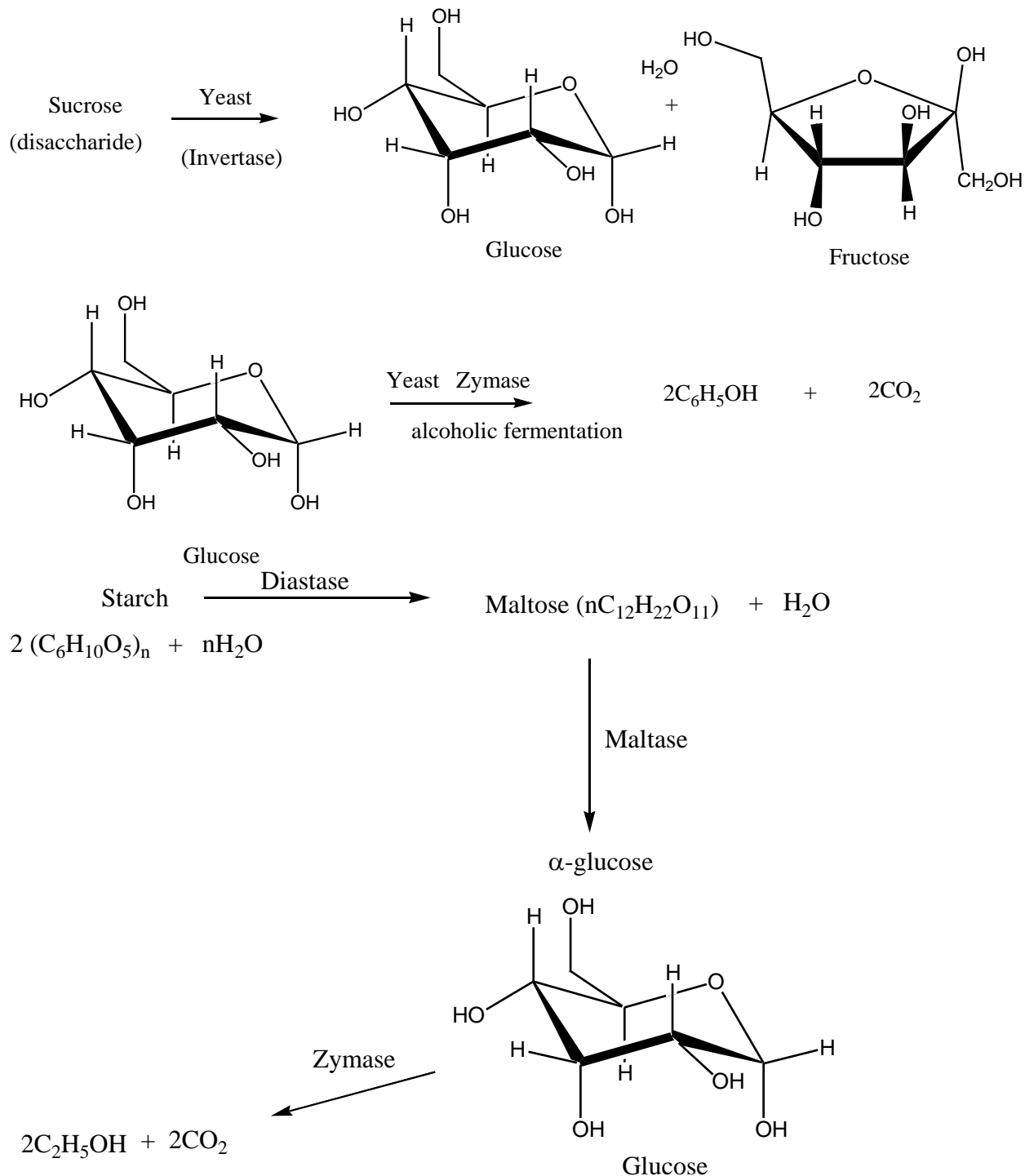


Fig. 1.0

Procedure

Each fruit was subjected to the same experimental conditions. 200 ml of distilled water was added to blended fruits (banana, mango and pineapple) (500g) in a sterilised fermentation jar. The pH was kept between 4-5 using citric acid at room temperature (25-31°C). 3g of Yeast/10 loopfuls of uncultured/ cultured yeast was embedded into the pulp/juice matrix. Fermentation was done in triplicates over a three days period. The mixture was filtered after the specified time and Fehling's solution was used to test for the presence of any reducing sugar. The filtrate was distilled and the alcohol strength tested using a Pictometer. The experiment above was repeated on pineapple mash using additives such as zinc sulphate solution, $ZnSO_4$, Urea, $CO(NH_2)_2$ and Ammonium Tartrate.

RESULTS

TABLE 1: CHARACTERISTICS OF FRUITS THAT WERE SELECTED FOR FERMENTATION WITHOUT ADDITIVES

FRUIT	TSS (°BRIX) INITIAL	TSS (°BRIX) FINAL	PH (INITIAL, FINAL)	TEMP. (INITIAL, FINAL)
Banana, <i>Musa acuminata</i>	18.0	6.8	4.65, 4.69	31.7°C, 29.5°C
Pineapple, <i>Ananas comosus</i>	12.2	4.40	4.20, 4.30	30.0°C, 29.0°C
Mango, <i>Mangifera indica</i>	13.4	6.2	4.11, 4.61	30.6°C, 28.5°C

TABLE 2: CHARACTERISTICS OF PINEAPPLE THAT WAS SELECTED FOR FERMENTATION WITH ADDITIVES

FRUIT	TSS (°BRIX) INITIAL	TSS (°BRIX) FINAL	PH (INITIAL, FINAL)	TEMP (INITIAL, FINAL)
Pineapple with $ZnSO_4$	11.8	4.5	4.16, 4.2	25°C, 25°C
Pineapple With Urea, $CO(NH_2)_2$	10.4	3.6	4.18, 4.89	25.0°C, 28.6°C
Pineapple with ammonium Tartrate	11.3	4.3	4.37, 4.40	29.7°C, 30.2°C

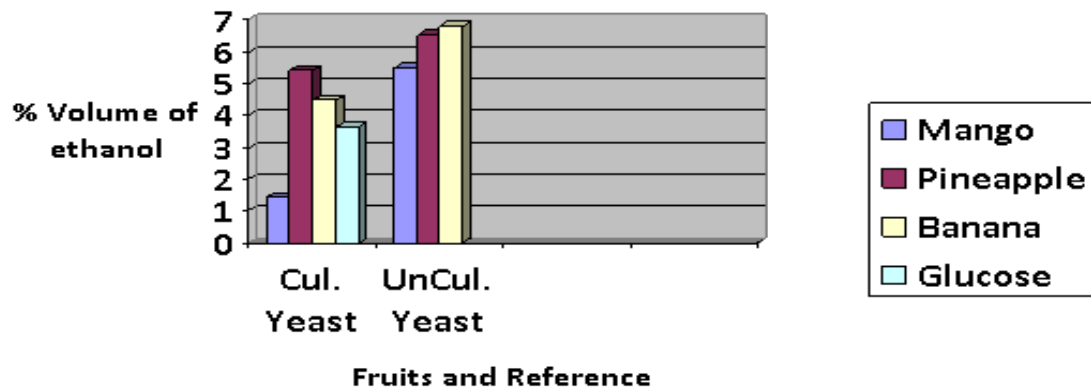
TABLE 3: AVERAGE % YIELD OF ETHANOL WITH STANDARD DEVIATION FOR THE SAMPLES WITHOUT ADDITIVES FOR WHICH CULTURED AND UNCULTURED YEAST WERE USED

Fruit mash in water	Yeast Type	Average % Yield of Ethanol \pm SD
Mango	Cultured Yeast	1.42 \pm 0.43
	Uncultured Yeast	5.51 \pm 0.61
Pineapple	Cultured Yeast	5.41 \pm 1.92
	Uncultured Yeast	6.55 \pm 0.32
Banana	Cultured Yeast	4.48 \pm 2.63
	Uncultured Yeast	6.81 \pm 1.75
Glucose (Reference)	Uncultured Yeast	3.62 \pm 0.21
Glucose (Reference)	Cultured Yeast	4.12 \pm 0.41

TABLE 4: THE % YIELD OF ETHANOL FOR THE PINEAPPLE SAMPLES WHEN DIFFERENT ADDITIVES WERE EMPLOYED

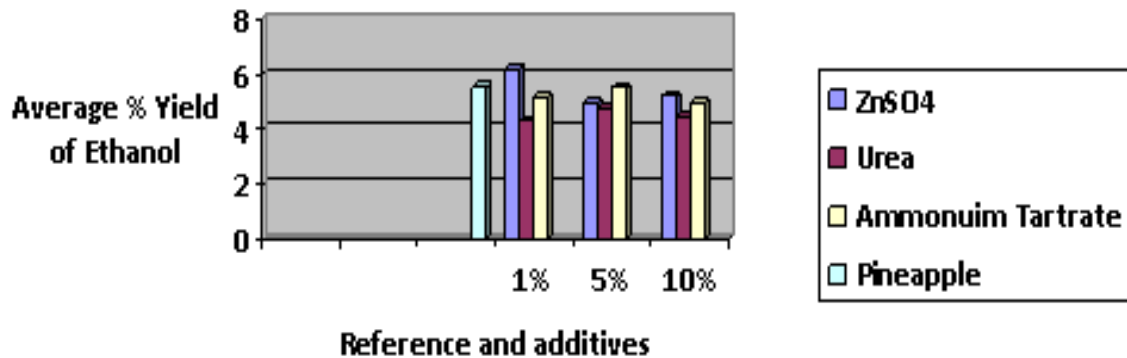
Fruit + % additives	Yeast Type used	% Yield of Ethanol		
Pineapple	Uncultured Yeast	6.55 \pm 0.32, v/v		
Pineapple + 1%, 5%, 10% ZnSO ₄ solution	Uncultured Yeast	6.19	4.88	5.13
Pineapple + 1%, 5%, 10% Urea solution	Uncultured Yeast	4.25	4.67	4.39
Pineapple + 1%, 5%, 10% Ammonium Tartrate solution	Uncultured Yeast	5.08	5.44	4.88

Graph Showing % volume of ethanol obtained for the different fermented fruits



GRAPH 1: % VOLUME OF ETHANOL OBTAINED FOR THE DIFFERENT FERMENTED FRUITS USING CULTURED AND UNCULTURED YEAST.

% Yield of ethanol for the pineapple samples at different concentrations of additives



GRAPH 2: % YIELD OF ETHANOL FOR THE PINEAPPLE SAMPLE AT DIFFERENT CONCENTRATIONS OF ADDITIVES.

DISCUSSION

Mango, banana and pineapples were selected because of their commercial abundance in Guyana. Their usage for the production of ethanol via fermentation will promote the Agro industry. *Saccharomyces cerevisiae* was used as the source of biocatalyst for fermentation processes. The pulp rather than the skin peel was preferred for more significant yield. After three days, Fehling's tests on the filtrate was negative, indicating that all the reducing sugars were utilised in the fermentation process. This further indicates that there were no inhibitory substances in the mash of three fruits, a prerequisite for ethanol production.

Using cultured yeast, the % yield of ethanol ranged from (1.42 ± 0.43 , v/v) to (5.41 ± 1.92 , v/v). Pineapple produced the highest yield of ethanol, (5.41 ± 1.92 , v/v) whereas mango produced the lowest yield (1.42 ± 0.43 , v/v). Thus, using cultured yeast, the % of ethanol produced, v/v follows the sequence: pineapple > banana > mango. These values are in accordance with those obtained in the literature¹⁻²⁸. All samples, with the exception of mangoes, subjected to cultured yeast produced a higher % yield of ethanol, v/v. than the reference sample, glucose (4.12 ± 0.41 , v/v).

Using uncultured yeast, the % yield of ethanol ranged from (5.51 ± 0.61 %, v/v) to (6.81 ± 1.75 %, v/v). Banana produced the highest yield of ethanol (6.81 ± 1.75 %, v/v) using uncultured yeast on fruit mash without additives, whereas mangoes produced the lowest (5.51 ± 0.61 %, v/v). The order being: banana > pineapple > mango. These values are in accordance with those obtained in literature¹⁻²⁸. These values are higher than those for the reference sample glucose (3.62 ± 0.21 , v/v), Table 3.0.

As the concentration of the additives increase, there seems to be a decrease in the % v/v of ethanol produced. An exception to the above is urea. Thus, in the presence of 1%, 5% and 10% ZnSO₄, the % yield of ethanol was 6.19, 4.88 and 5.13 %, v/v respectively. Thus, in the presence of 1% additives, the % yield of ethanol, v/v follows the sequence: ZnSO₄ > Ammonium Tartrate > Urea, Table 4.0.

It was found in all cases the Total Soluble Solid (TSS), expressed in °Brix was less after the fermentation process for both processes with and without additives. This indicates that the substrate was being acted upon by the enzyme *Saccharomyces cerevisiae*. As an example, the TSS for Pineapple without additives, before and after fermentation was 12.2 ° brix and 4.4 ° brix respectively. With an additive such as ZnSO₄, pineapple registered an initial and final brix value of 11.8 and 4.5 respectively. The pH values showed variation. It ranges from 4.20 to 4.30 without additives and 4.16 to 4.2 with ZnSO₄. There was a general increase in the pH after fermentation process, with and without additives. The temperature showed variation. For example, with banana, *Musa acuminata*, the temperature registered was 31.7°C and 29.5°C before and after fermentation respectively.

As the concentration of the additives increase from 1 to 10%, there seem to be an increase in the % yield of ethanol produced at the 5% mark, followed by a decrease at the 10% level. As an example, with ammonium tartrate, values of 5.08, 5.44 and 4.88, v/v ethanol were recorded at the 1%, 5% and 10% level respectively. However, with the addition of the additive $ZnSO_4$, there is a decrease at the 5% concentration followed by an increase at the 10% level. Values of 6.19, 4.88 and 5.13 v/v were obtained at the 1%, 5% and 10% level respectively. It should be noted that without the additives, % yield of ethanol for pineapple was 6.55 ± 0.32 , v/v). Thus, the addition of the additives seem to decrease the volume of ethanol produced via fermentation.

CONCLUSION

Banana mash in the presence of uncultured yeast produced the highest % of ethanol of (6.81 ± 1.75 , v/v.). This was followed by pineapple, registering a value of (6.55 ± 0.32 , v/v). Mango mash the lowest, registering a value of (5.51 ± 0.61 , v/v). The presence of the additives seem to decrease the % yield of ethanol. $ZnSO_4$ solution at a concentration of 1% proved to be the best additive, with the ethanol yield being 6.19, v/v. Ammonium Tartrate is a better nitrogen source than Urea for Pineapple. Thus, the pulp of bananas, pineapples and mangoes can be used to produce ethanol for commercial and industrial applications. However, research should continue towards improving the % yield of ethanol.

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