Wine Production from Banana *(Musa sapientum)* Using Yeast (*Saccharomyces cerevisiae*) Isolated from Grape *(Vitis vinifera*).

Abstract

This research was carried out to produce wine from banana *(Musa sapientum)* using yeast (*Saccharomyces cerevisiae*) isolated from grape *(Vitis vinifera*). The fermentation of the banana wine lasted for 21days. During fermentation, liquor of the fermenting "must" were removed every 48hours from the fermentor for analysis of pH, titratable acidity, specific gravity and reducing sugar using standard procedures.. The results from the experiment showed that Specific gravity of the wine was observed to reduce drastically as the fermentation progresses. The pH of the Banana wine during fermentation increased from 4.16- 4.22 while the titrable acidity of the Banana wine produced increased from 1.05-1.77. The alcohol content of the wine increased from 0.0 to 9%. The higher the fermentation temperature, the faster the conversion of sugars into alcohol and carbon dioxide by the yeast. The flavor and taste was appreciable. This study showed that acceptable wine can be produced from banana with the yeast *Saccharomyces cerevisiae* isolated from grape.

Keywords: Banana, Fermentation, Grape, Yeast, Fermentor, Carbondioxide

1. Introduction

Wine is a product of alcoholic fermentation by yeast of the juice of ripe grapes or any fruit with a good proportion of sugar [1]. Wine is one of the most recognizable high value added products from fruits. It can also be used as a substrate for the manufacture of vinegar, a by-product of wine manufacture. Fruit juices are fermented to produce wine, an alcoholic beverage. Grapes are usually preferred because of the natural chemical balance of the grape juice which aids their fermentation process without addition of sugars, acids, enzymes, or other nutrients. However, fruits such as banana, cucumber, pineapple and other fruits are used in wine production [2].

Wine manufacture is challenging in which marketable product can be obtained, but the processes involved in its production are relatively straight forward [3]. Highly acceptable wines can be made from practically all fruits. Wine can be fermented with yeast that occurs naturally in grape which is the main organism responsible for alcoholic fermentation which belongs to the genus *Saccharomyces*. Although, many genera and species of yeast are found in must, *Saccharomyces cerevisiae* is the main yeast strain that is commonly reported to be responsible for alcoholic fermentation [1] and in other countries where grape is not produced, emphasis is usually placed on other fruits for wine making. There are some soft fruits from both temperate and tropical regions whose pigment stability and flavor profiles match those of any wine from grapes, but suffer from the lack of intensive research and development given to grape wine.

Reports on tropical fruit wines have been mainly on exotic species such as banana, pineapple, citrus, mango, pawpaw, apple, strawberries etc.[4]. Wine represents a safe and healthful beverage; it also provides calories and vitamins. During period when life was often strenuous, it offered relaxation and relief from pains.

Bananas *(Musa sapientum)* are an important staple starchy food in Nigeria. It is a seasonal and highly perishable fruit, which can be available all year round. The large quantity of bananas and plantains provides the potential for industrial use [5]. In addition, any application to produce a marketable, value-added product will improve banana farming economies and eliminate the large environmental problem presented by banana waste. Banana could then compete in the market, either as banana juice or as mixtures with other juices because of its flavor and aroma [6].

Bananas has a lot of nutritional benefits, thus demands in the market are high. They are highly recommended by doctors for patients whose potassium is low, because of its impressive potassium content. Potassium is an important component of cell and body fluids that helps control heart beat and blood pressure, countering bad effects of sodium. Banana is considered as an important food to boost the health of malnourished children, it contains good amount of soluble dietary fiber that helps normal bowel movements; thereby reducing constipation problems. Medicinal uses of banana have positive contribution towards successful treatment of anemia, heartburn, temperature control, ulcer, overweight etc.

Banana juice can also be applied to wine production; however, banana juice is turbid, gray in color, very viscous, tends to settle during storage and, therefore must be clarified prior to commercialization [6]. The turbidity and viscosity of banana wine are caused mainly by the polysaccharides in banana juice such as pectin and starch and therefore make the clarification process harder. Application of pectinase and α-amylases that affect the quality of wine is important for improving the process of banana wine production.

2. Material and Methods

2.1. Sample Collection

Ripe banana fruits used for this experiment were bought from Eke Market, Awka, in Anambra State.

2.2. Preparation of Must/ Fermentation

The ripe bananas were washed with distilled water. The bananas were thoroughly disinfected with cotton wool soaked in ethanol. The banana slices were blended with water in a Super Mark blender. The slurry was filtered through a "muslin cloth" to obtain the juice (must) and autoclaved after which it was allowed to cool and poured into the fermenting vessel and the

250ml inoculum developed was inoculated into the fermenting vessel. 0.14 Sodium metabisulphite was added to inhibit the growth of micro-organism and about 100g of Sucrose was added to fortify the must and kept for fermentation to commence. The must was analysed for reducing sugar, titratable acidity, specific gravity and alcoholic content before fermentation.

2.3. Inoculum Development

The banana fruits were washed thoroughly with 0.1% sodium metabisulphite in water. The fruits were cut, manually deseeded, blended and filtered to obtain the juice. About 250ml of the juice was introduced into a clean sterile 500 ml conical flask and sterilized by autoclaving. Upon cooling, three (3) loopful of the yeast culture isolated from the palm-wine was used to inoculate the juice and incubated in a rotary shaker for 48 hours.

All procedures were done under aseptic condition.

2.4 *pH* Determination

The pH was determined using a standard pH meter. The pH meter was standardized with buffer solution, the buffer solution was prepared with pH buffer powder of pH 4.00 at $25^{o}C$ dissolved in 250ml distilled water. Ten ml of the "must" was put into a sterile beaker, The electrode of the pH meter was immersed in the beaker containing the "must". The pH of the "must" was determined using a digital *pH* meter (Model No: pH S-25)

2.5 Determination of Reducing Sugar

The quantitative estimation of reducing sugar of the wine was determined using the method described by Amerine and Ough (2000). 1ml of 3,5-Dinitro Salicyclic acid (DNS) is added to 1ml of supernatant of wort (sample) in a test tube and the mixture heated in boiling water for 10mins. The test tube is cooled rapidly in tap water and the volume adjusted to 12ml with distilled water. A blank containing 1ml of distilled water and 1ml of DNS is also prepared. The Optical Density (OD) of sample is read against the blank in a Spectrophotometer at 540nm absorbance. The concentration of reducing sugar is estimated from a glucose standard curve.

2.6 Determination of Specific Gravity

Fifty ml specific gravity bottle was thoroughly cleaned with distilled water, dried in an oven for $50^{o}C$ and allowed to cool. The weight of the cooled dried bottle (W1) was recorded. The dried bottle was filled with deionized water and surface of the bottle was cleaned with a cotton wool and weighed as (W2).

The bottle was empty and cleaned twice with 10ml of the "must" thereafter the bottle was filled to the brim with the "must" and the bottle cleaned with cotton wool and weighed as (W3). The specific gravity (S.G) was calculated.

$$S.G = \frac{W3 - W1}{W2 - W1} = \frac{S}{W}$$

Where
S= weight of volume of must (W_3 - W_1)
W= weight of volume of water (W_2 - W_1)

2.7 Estimation of Titrable Acidity

This was determined by the methods described by [7]. 1% of aqueous alcoholic phenolphthalein as indicator was added to 200ml of distilled water. It was titrated with 0.1m of Na0H. Titration was stopped when a faint but definite pink colour appeared. The titre was taken, this served as the initial titre. 5ml of the must was added to the neutralized solution. The same 0.1m NaOH was used to titrate it. The titration was stopped at the appearance of faint, but definite pink colour. The titre was taken. This served as the final titre. The titratable acidity was calculated with reference to tartaric acid.

2.8 Alcohol Content Determination

The distillation method According to the Association of Official Analytical Chemists Methods of Analysis [8]. Fractional distillation of the wine was carried out and 100ml of the wine was measured into a round- bottom flask with side arms. The flask was connected to the fractioning column and an anti-bumping chip was added to the wine. This ensured adequate dispersal of heat in the wine. It was then heated and the distillate collected. The distillate was poured into a clean dry conical flask and made up to 100ml with distilled water. It was left to stand overnight on the bench. The specific gravity of the distillate was determined and the result was taken to determine the alcoholic strength.

3. Results

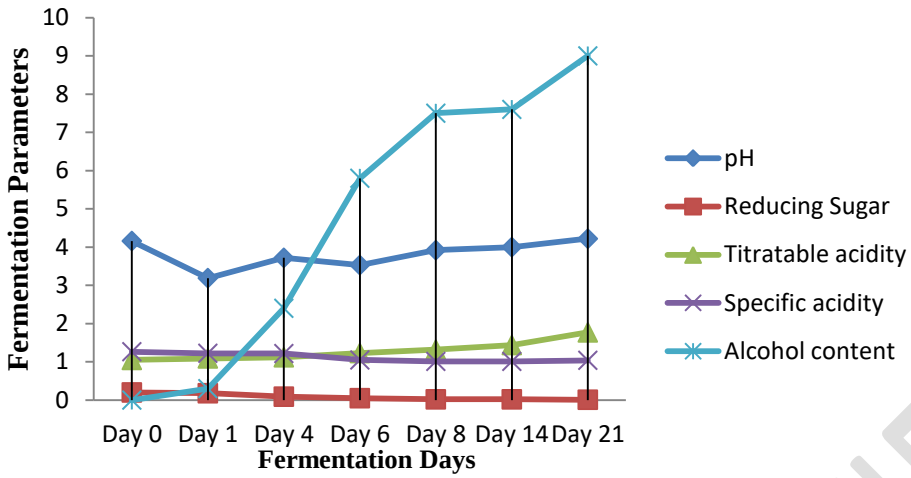
Table 1: Analysis of the fermenting wine

Number of Days	pH	Reducing sugar(mg/ml)	Titratable acidity	Specific Gravity	Alcohol content
DAY 0	4.16	0.20	1.05	1.266	0.0
DAY 2	3.91	0.18	1.09	1.224	0.3
DAY 4	3.72	0.09	1.12	1.218	2.4

DAY 6	3.53	0.05	1.23	1.049	5.8
DAY 8	3.92	0.02	1.32	1.012	7.5
DAY 14	4.00	0.02	1.44	1.012	7.6
DAY 21	4.22	0.01	1.77	1.04	9.0

Throughout the period of fermentation, *pH* of the must was within the acidic range. The pH of the fermenting wine as shown in table 1 and Fig. 1 indicate a gradual decrease in the pH as the fermentation time increases and increased at the end of fermentation. At day 0, the pH is 4.16. While at day 21 the pH is 4.22. The result of titrable acidity carried out on the sample as shown in table indicates that the titrable acidity ranges from 1.05 at 0 day to 1.77 at day 21 in the fermenter. Therefore, the titrable acidity increases as fermentation time increases. The result shows an an increase in the alcohol content of the fermenting sample as shown in the table.

Figure 1. Wine analysis



The alcohol content ranges from 0.0 to 9.0% at day 21. In the case of specific gravity of the fruit wine gradual decrease in valves were observed throughout the period of fermentation. These decreases were observed to be irrespective of the yeast strain and the fruits used in the wine production. Between 0 day to 21 days of the fermentation, specific gravity values were observed to range from 1.266 to 1.04kgm-3.

4.0 Discussion

In this research, it is observed that the fermentation period agrees with the result of [9] who observed that *Saccharomyces. cerevisiae* could strive under low pH. The result of this experiment shows that pH values of banana wine decrease progressively as the fermentation period increases and finally increased at the end of the fermentation. The pH value at Day 0 is 4.16 which decreases progressively as fermentation continues and increased at the end of the fermentation. The pH is 4.22 at Day 21 (Table 1). This shows that the wine became more acidic with the period of fermentation. The drop in pH also records the utilization of the sugar present in the must for growth. This observation is similar to that reported by [10].

Result of this study also revealed consistent increase in titratable acidity of the wine throughout the perod of fermentation. The titratable acidity is recorded as 1.05 at Day 0 which increased to 1.77 at Day 21. It is observed that the titratable acidity increases with the period of fermentation. Studies have shown that during fermentation of fruits, low pH is inhibitory to the growth of spoilage organisms but creates conducive environment for the growth of desirable organisms. Also, low pH and high acidity are known to give fermenting yeast competitive advantage in natural environment [7]. The alcohol content of the wine increased from 0.00% to 9% on the last day of fermentation (Fig 1). This result is found to conform to that of [11] who observed gradual increase in the titratable acidity and the alcohol content in the fermentation of plantain.

The specific gravity of the fruit wine produced in this study reduces as the fermentation days of the wine increases. After the 21 days fermentation the specific gravity of the wine reduced drastically to 1.04kgm-3 this was due to the type of yeast used in the wine production. *Saccharomyces cerevisiae* isolated from palm wine has been reported to reduce specific quality of fruit wines during fermentation9. The short shelf-life of beverages are however a major problem faced by their producers and consumers in Africa [12].

In texture, the wine produced was completely watery at the end of fermentation. This result is similar to that recorded by [7] that as fermentation rate proceeded, gas was formed and this rose through the liquid then during active fermentation, forth or foam is formed on the surface. The gas carries the cells through the fermenting must cause it to be cloudy and as a result, a strong odor of alcoholic fermentation developed.

The result of the banana wine produced by the fermenting with yeast strains isolated from grape (*Saccharomyces cerevisiae*) shows high rate of alcoholic production at the end of day 21. This result is similar to the observation reported by [13, 14] who recorded that pure culture of *Saccharomyces cerevisiae* produces more ethanol and give a faster fermentation than the native yeast.

5.0 Conclusion

Banana wine is very nutritious and easy to produce and could then compete in the market with other wines because of its flavor, aroma and the successful production of using indigenous fruits as substrates for wine production and the efficiency of locally isolated yeast *Saccharomyces cerevisiae* strains from grape (*Vitis vinifera*) for fruit wine production. The pH of the fermenting must was shown to increase at the last day of fermentation ranging from 4.16 to 4.22. the alcohol content also gradually increased ranging from 0.00% to 9% showing the strength of the fermenting must. Banana wine has a lot of nutritional benefits, Vitamins including B5, B6, C, A are all present in banana wine and this makes it one of the high ranking beverage over other alcoholic ones. Vitamin A helps in restrain of eye sight. Banana wine is infused with high taste and health. Commercially produced banana wine is a clear, slightly sparkling alcoholic beverage with a long shelf life than banana beer, which is spoiled easily and therefore not stored for long periods. However, there is the need for further research to ascertain the shelf life for the wines.

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