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Production of wine from zobo (*Hibiscus sabdariffa*) flower juice

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Abstract

Wine was produced from zobo flower juice using a yeast (*Saccharomyces cerevisiae*) extracted from palm wine. The maximum alcohol productivity of 2.6 %(v/v) was achieved by the fermentation of zobo flower juice. The pH decreased from 4.20 to 3.48 and there was an increase in viable count from an $9.10x10^5$CFU per ml to a maximum $5.92x10^8$CFU per ml on the 5th day. Decrease in concentration of reducing sugar from an initial concentration of 0.43 mg ml^{-1} to a final concentration of 0.176 mg ml^{-1} on the eleventh day was observed. The growth kinetic studies were performed and the specific growth rate of yeast on "Zobo" juice was found to be 0.0667 s^{-1} and doubling time was 10.4 s^{-1}.

Keywords: Zobo flower juice, wine, monod kinetics, yeast

INTRODUCTION

Zobo (*Hibiscus sabdariffa*) is a tropical plant found mainly in the northern part of Nigeria. It grows even in the wild, producing a dark red flower which is used for the production of the very common Zobo juice. The matured flowers were harvested and dried and the juice was extracted with hot water.

Nigeria had spent about ₦ 171.56 million for importing wine. The temperature restriction of grape to the temperate regions predisposes this trend. However, the very high duty on imported wines has stimulated interest in producing wines from tropical fruits (Okoro et al. 2007). Many investigations were conducted for production of wine using Zobo (*Hibiscus sabdariffa)*. Okara et al. (2007) produced red wine from roselle (Hibiscus sabdariffa) and pawpaw (Carica papaya) using palm-wine yeast (Saccharomyces cerevisiae) and Arubi et al, (2009) produced colored wine from *Hibiscus sabdariffa* calyx extract.

An investigation was conducted for producing wine using Zobo flower juice. The fermentation was conducted by local yeast (*Saccharomyces cerevisiae*) isolated from palm wine. Process parameters like pH, Colony Formation Unit (CFU), initial concentration were evalutated. The monod growth kinetics was applied and specific growth rate of *Saccharomyces cerevisiae* was determined for enhancing the production of wine from Zobo flower juice.

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Materials and methods

Preparation of Zobo juice

The matured Zobo flowers (dried) were bought from mile-1 Market, Portharcourt, Nigeria. The juice is extracted from Zobo Flowers with hot water. The fermentation was carried out using local yeast (saccharomyces cereisiae) isolated from fresh palm wine. The fresh Palm wine was obtained from a local palm tapper in Ozuoba, portharcourt. 1000ml of Zobo juice was prepared by washing 250g of zobo flowers with 1000ml of boiling water for a period of 20 minutes.

Six Petri-dishes, each containing 10ml of potato dextrose agar were inoculated with the flesh palm wine. All Petri dishes were incubated for 24hrs at 37^{o}C. Yeast was isolated from the colonies using serial dilution technique.

The Zobo juice was autoclaved at 121^{o}C for 15 minutes at 15 psi. The juice was fermented for eleven days at 37^{o}C. 3ml of the fermentation broth was taken using a sterile pipette and centrifuged at 6000 rev. per minutes for 30 minutes in a bench-top centrifuge. The supernatant was carefully decanted and the optical density was determined. The amount of reducing sugar was determined using a calibration graph. The PH was measured at every 24hrs. The viable count was done on zero day and at intervals of 24hrs.

Results and discussion

Zobo wine production sounds common but the novelty of this work is the use of zobo flower and zobo juice which is fermented using a locally made yeast (*Saccharomyces cerevisiae*) isolated from fresh palm wine for wine production. Below are the findings.

The Figure 1 shows the results obtained for reducing sugar during fermentation. It shows that the reducing sugar concentrating decreased as fermentation progressed. The initial value of 0.43 ml/ml was obtained on the zero day and 0.170 mg ml at the end of the fermentation. This gives a yield on sugar of 22.7%. The decrease

in reducing sugar values may be attributed to utilization of the sugars for growth and other metabolic activities by the organism.

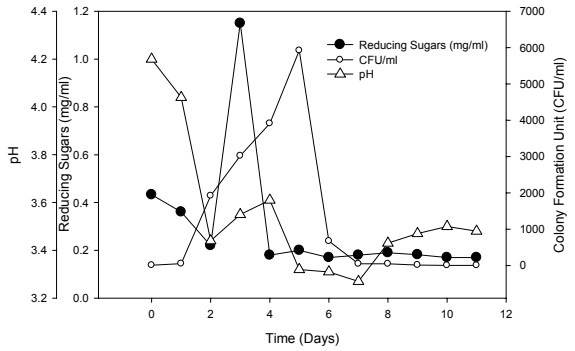

Figure 1: Reducing Sugars, viable count and pH with respects to time (Days).

The Figure-1 also shows the results obtained for viable count during must fermentation. There was a steady increase in viable count. From an initial value of $9.1x10^5$ C.F.U /mL to a maximum value of $5.92x10^8$ colony forming units (C.F.U) per ml on the 5th day. From the 6th day the population density started to decrease. The decrease continued until the 11th day.

The decrease in population density could be as a result of depletion of some nutrients especially nitrogen and phosphate which have been reported to enhance growth of yeast during wine fermentation. These elements were not added to the culture medium. Another reason may be due to the accumulation of toxic metabolites. It is known that glucose by yeast results in acidic metabolites.

This is evident from the results obtained for the changes in pH during fermentation which lended towards the acidic range. The Figure 1 equally shows that the pH decreased from an initial value of 4.20 on zero day to pH of 3.48. Theoretically, sugars are converted to alcohols, then alcohols to aldehydes, aldehydes to ketones and ketones are finally converted to acids during fermentation

The Table-1 show the results obtained for total count during must fermentation. There was a steady increase in total count from a initial value of 1.64×10^6 at day zero to a value of 2.46×10^8 at 2nd day. After which the total count became approximately constant. The first day represents the exponential growth phase. The last days represents both the stationary phase and the death phase. The population density at the stationary and death phases represent both life and dead yeast cells. The death may have been caused by the

Table 2: Total count in must fermentation

Time (Days)	Total unit (C. FU/ML)
0	1.64×10^6
1	5.26×10^6
2	2.46×10^8
3	3.30×10^8
4	5.49×10^8
5	5.24×10^8
6	7.20×10^8
7	7.76×10^8
8	8.70×10^8
9	8.93×10^8
10	8.90×10^8
11	8.80×10^8

lack of some essential nutrients such as nitrogen and phosphate which have been reported to be essential for yeast growth in wine fermentation. Death might also have been caused by the accumulations of some toxic metabolites.

Table-2 shows the results obtained for percentage alcohol produced during fermentation. There was a steady increase in percentage alcohol from an initial value which was not detectable at day zero to a value of 2.6% at the 11th day. The specific gravity decreased steadily from an initial value of 1.00 on the first day to 0.9952 on the 11th day. This is due to the increasing percentage of alcohol which is less dense than water. The increasing alcohol in the wine is due to the conversion of sugar, which has been shown to reduce as fermentation proceeded.

Table 2: Percentage of alcohol by volume during fermentation

Time (Days)	Specific Gravity	Percentage Alcohol by volume
0	1.0.000	Not *Detected*
1	1.9995	0.28
3	1.9991	0.61
5	1.9986	0.92
7	1.9983	1.15
9	1.9974	1.72
11	1.9961	2.60

Specific growth rate = $\mu = 1/X * dX/dt \Rightarrow \mu = \ln (X_0/X_t)/t$

X_0 = Initial cell concentration at time 0; X_t = final cell concentration after time t.

Based on the exponential growth phase (Table 1), the specific growth rate is calculated to be 0.0667/hours (μ) and doubling time td ($\ln 2/\mu$) is calculated to be 10.4 h.

Conclusion

Saccharomyces cerevisiae isolated from palm wine has been found to be suitable for fermenting "zobo" to produce wine. The main parameter that varied with time which were monitored were PH, viable count, concentration of reducing sugar and alcoholic Content. zobo juice is a new drink which has just been introduced into Nigeria market. Zobo flowers is grown locally in Nigeria, even in the wild. Therefore being easily obtained in Nigeria it becomes a very good and economical source of wine.

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