

WINE PRODUCTION FROM APPLE (*Maluspumila*) USING YEAST ISOLATED FROM PALMWINE

ABSTRACT

The study was aimed at the production of apple (*Maluspumila*) fruit wine with the use of yeast *Saccharomyces cerevisiae* isolated from palmwine. Primary and secondary fermentation of the fruits lasted 28 days. During fermentation, aliquot samples were removed daily from the fermentation tank for analysis of alcohol content, specific gravity, *pH*, titratable acidity, and reducing sugar using standard procedures. *pH* of the fruit must during the period of fermentation ranged from 5.0 to 3.2. During the fermentation period, consistent increases in alcohol content were observed with time. At the end of the 28th days fermentation, the concentration of alcohol in the fruit wine was observed to be 3.2%. Also titratable acidity concentration of the wine was observed to show steady increase with time throughout the period of fermentation. This study has revealed that acceptable wine could be produced from apple with *Saccharomyces cerevisiae* isolated from palm wine. Sensory evaluation results showed there were no significant differences ($p > 0.05$) in flavor, taste, clarity and overall acceptability between apple wine and a reference wine. The apple wine was generally accepted.

INTRODUCTION

Wine is an alcoholic beverage typically made of fermented fruit juice (Okafor, 2007). Any fruit with good proportion of sugar may be used in producing wine and the resultant wine is normally named after the fruit.

In the European Union, wine is legally defined as the fermented juice of grapes [Harding, 2005]. In the United Kingdom, wine is commonly called country wine. Wine can be made from virtually any plant matter which can be fermented [Harding, 2005]. Most fruits and berries have the potential to produce wine. Few fruits other than grapes have the balanced quantities of sugar, acid, tannin, nutritive salts for yeast feeding and water to naturally produce a stable, drinkable wine. During the past few decades, grapes are the main fruits that used for wine production. Despite that, several studies have investigated the suitability of other fruits as substrates for the purpose of wine production (Joshi and Bhutani, 1991, Ndipet *al* . 2001). The non-availability of grapes, which is usually the fruit of choice for wine production in the tropics has necessitated the search for alternative fruits source in Nigeria and other tropical countries (Alobo and Offonry, 2009).

In Nigeria, there are abundances of tropical fruit which includes; plum, watermelon, apple, pineapple, passion fruit etc, these fruits are highly perishable, and susceptible to bacterial and fungal contamination as a result they fail to reach the market due to spoilage, mechanical damage and over ripeness (Ihekoroye and Ngoddy, 1985). Besides, these fruit are difficult to keep for considerable length of time; hence the ripe fruits are utilized either as fresh or processed into juice and specially products (Oyeleke and Olaniyan, 2007). High rate wastage of these fruits especially at their peak of production during their season necessitates the need for alternative method of preservation of food and enhanced utilization of these fruit. The production of wines from common fruits could help reduce the level of post harvest losses and increase variety of wines (Okoro, 2007; Aloba and Offonry 2009).

The type of wine to be produced dictates the fruit and strain of yeast to be involved (Amerine and Kunkee, 2005). Preservatives used in wine making include sulphur dioxide, potassium sorbate, sorbic acid and metabisulphites (Idise and Izuagbe, 1988). High concentration of these preservatives in wine, aside causing off odors, can induce lots of systemic disorderliness such as breathing problems in asthmatic patients and gastrointestinal disturbances in allergic persons. The effects of bioaccumulation of these chemicals could further compound these situations (Okafor, 2007).

Wine making involves the use of wine yeast to ferment the 'must' of fruits. Yeast which is the main organism responsible for alcoholic fermentation usually belongs to the genus *Saccharomyces*. Although, many genera and species of yeast are found in musts, *Saccharomyces cerevisiae* is the main yeast strain that is commonly reported to be responsible for alcoholic fermentation (Querol, 2003).

The objective of this study is to produce wine from apple using yeast isolated from palm wine, to study fermentation of the wine and utilization of agricultural produce from perishing. *pH*, temperature, titratable acidity and reducing sugar tests were assayed quantitatively.

MATERIALS AND METHODS

Sample collection

Apple fruit was obtained from local Eke Awka Market in Anambra State, (South east), Nigeria.

Identified as *Maluspumila* at Botany Department Nnamdi Azikiwe University, Awka. Other materials used were Sucrose, Ethanol, Sodium metabisulphite, Dinitrosalicylic acid (DNS) and distilled water.

Palm wine was purchased from Umuawulu, Awka in Anambra State and stored for fermentation for 24 hours to isolate the yeast. The yeast was isolated from palm wine, sub-cultured, characterized and kept for use.

Inoculum Development

Development was done to obtain large quantities of yeast cell for pitching. To build up the inoculums, 200mls of apple juice each were put into a 250mls conical flask separately and autoclaved at 121⁰C for 15 minutes. The mixture was allowed to cool then three loopful of stock culture from SDA plate was transferred into the 200ml standard 'must' in a conical flask and kept in the refrigerator.

Preparation of "must" juice:

The apple fruit (*Maluspumila*) was plucked and rinsed with distilled water, they were peeled for easy blending. The grated edible portions (mention the amount of pulp taken) were blended in the electric blender with constant addition of water respectively. The overall water added during the blending was 2000mls distilled water to avoid friction in the blender. 2000mls of distilled water was added extract the "must" by filtering the juice with sterile muslin cloth. 4000mls of the whole "must" was poured into the fermenting jar for fermentation and then, 0.28g of sodium metabisulphite were added, transferred into the fermenting vessel of 5 liters volume, and corked then allowed to stay for 24hours.

Fermentation

The 200mls of developed inoculum was poured into the fermenter jar containing the "must" making it a total of 4.2 liters. 358.9g of sucrose was then added to the fermenter to fortify the must, then the mouth of the jar stuffed plugged tightly with cotton wool and kept on the bench for fermentation (Archibonget *al.*, 2015).

Determination of physico-chemical Test

The pH, titratable acidity, and alcohol percentage were determined. These were carried out in

accordance with standard methods reported by (A.O.A.C., 1990). While reducing sugar(brix) was determined using Miller (1959).

Organoleptic evaluation

This was carried out in accordance with the procedure reported by Maragatham and Panneerselvam (2011). The sensory evaluation was done using 8 judge panels after aging for 28 days. Observations recorded for color, clarity, body and taste on a 5 point scale with 5 points for excellent quality and 1 point for bad quality.

Statistical analyses

These were carried out using comparative analysis at 95% confidence level.

RESULTS

Table 1a: PROXIMATE ANALYSIS OF APPLE JUICE BEFORE FORTIFICATION

Parameters	Values
<i>pH</i>	4.01
Reducing sugar(⁰ Brix)	2.803
Specific gravity	1.013

Table 1a nd 1b, maintain the same row order

TABLE 1b: PROXIMATE ANALYSIS OF APPLE JUICE AFTER FORTIFICATION

PARAMETER	% QUANTITY
PH pH	5.6
Specific Gravity	0.081
Reducing sugar (⁰ Brix)	14.0

TABLE 2: PHYSICAL AND ORANOLEPTIC PROPERTIES OF APPLE JUICE

Days	SWEETNESS	COLOUR
1	+	CREAM
2	+	CREAM
3	-	DIRTY CREAM
4	-	DIRTY CREAM
5	-	DIRTY CREAM
6	-	DIRTY CREAM
7	-	DIRTY CREAM
14	-	PALE CREAM
21	- PALE CREAM	
28	- PALE CREAM	

TABLE 3 : WINE ANALYSIS OF APPLE JUICE

Days of fermentation	Specific gravity (kg/m ³)	pH	Alcohol content (%)	Titrateable acidity	Reducing sugar (g)	Temp
1	1.0801	5.0	0	0.50	10.803	32

2	1.0564	4.9	0.2	0.55	6.738	31
3	1.0355	4.6	0.4	0.63	1.722	31
4	1.0215	3.9	0.8	0.65	0.671	31
5	1.0179	3.6	1.0	0.70	0.644	30
6	0.9140	3.5	1.4	0.76	0.563	30
7	0.8134	3.3	1.8	0.82	0.513	30
14	0.4105	3.3	2.4	0.88	0.474	29
21	0.4041	3.2	2.8	0.96	0.421	27
28	0.4002	3.2	3.2	1.00	0.384	27
Standard Wine	0.9722	3.5	6.0	0.63	0.546	32

RESULTS AND DISCUSSIONS

In this research, the choice of the fruits, apples(*Maluspumila*)was as a result of its high yield of sugar. The proximate composition of the fruits used was in agreement with the general case for fruits as reported by Pearson (1976), Okaka(1997) and Archibonget *al.*, (2015).

The produced wine was characterized by measurements of parameters such as organoleptic attributes, physico-chemical properties, and physical screening. The physically observable and taste changes in the wines with period of fermentation are presented in Table 2. It was observed that there were appreciable changes in the apple wine during the fermentation period.

The present study showed low pH values, pH in the wines were within the acidic range. The pH of the wine ranged from 5.0 to 3.2 (table 3) **Sentence does not read properly.** The observed changes in the pH of the wines could be due to production of acids with period of fermentation probably arising from microbial succession. This result agrees with the reports of previous workers (Amerine and Kunkee, 2005; Okafor, 2007). A similar observation has been reported by Reddy and Reddy (2005) in their study on mango fruit wine production and Noah *et al.* (2013) and Idise (2012), who noted that optimum pH value for quality wine production is within the range of 4.0.

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.

Low pH and high acidity are known to give fermenting yeasts competitive advantage in natural environments (Reddy and Reddy, 2005).

Trend in titratable acidity was observed to show a steady increase with time throughout the fermentation. At the 28th day of fermentation, the acid concentration of the wine was observed to increase from the initial range of 0.5% (on the 1st day) to 1.00% (on the 28th day). The titratable acidity of the final wine was 1.00. This was in agreement with the report of Idise (2012). Nell and Ettre, (1974), also observed that titratable acidity on fruit wine production is expected to be between the range of 0.5 to 1.0%.

In order to supplement the sugar content of the “musts” granulated sugar which is a source of sucrose was added during the production of the wine. Reports have shown that the major problem associated with the use of tropical fruits in wine production is their low sugar content (Alobo and Offonry, 2009).

Table 3 also indicated a steady increase in alcohol content of the wine during the period of fermentation with the fermenting yeast strain. At the end of the 28th day of the fermentation, the concentration of alcohol content of the wine was observed to range from 0% to 3.0%. The observed changes in specific gravity and % alcohol (v/v) of the wines with period of fermentation support the occurrence of microbe with varying tolerance for metabolic end

products. These results agree with reports of Idise and Izuagbe (1988), Robinson, (2006), Okafor (2007) and Archibong *et al.*, (2015).

Remarkable amount of alcohol was produced from the fruit with the yeast strain used during fermentation and was consistent during the course of fermentation. Generally, the percentage alcohol produced from the apples fruits at the end of the fermentation by the yeast strain was above 2% which is comparable with moderate grape wines (Querol, 2003; Okunowo, 2005). In the present study, the amount of alcohol produced by the isolate from the palm wine did not show any difference from wines produced from yeast from other sources apart from palm wine.

Throughout the period of fermentation, the specific gravity of the wine gradually decreases in values. Between 21 day to 28 days of fermentation, specific gravity values of the wine was observed to decrease from 1.004 to 0.922 kg/m³ as shown in table 3. This was due to the yeast activity in the degradation of sugars producing alcohol during the wine production. The specific gravity value of the wine was observed to diminish by significant value at $p \leq 0.006$.

There was observed changes in the temperature of the wines, These results agree with reports of previous workers (Idise and Izuagbe, 1985, 1988; Amerine and Kunkee, 2005; Okafor, 2007; and Archibong *et al.*, 2015).

In the case of reducing sugar of the wine, the values of the reducing sugar were observed to be decreasing. It was high on the first day (0.803g) but started to diminish from the second day till the 28th day. This was as a result of fermentation by the fermenting yeast and the production of alcohol. The type and aroma produced during wine production is reported to depend on duration and physico-chemical characteristics of the “musts” and the strain of yeast used.

The present study revealed the effectiveness of other yeast strains apart from the commercial yeast in wine produced from tropical fruits (apple), Also, an appreciable quality wine can also be produced using apple fruit, this is in support with the work of Agbor *et al.* (2011).

CONCLUSION

This successful production of wine from apple (*Malus pumila*) using yeast *Saccharomyces cerevisiae* isolated from palm wine was found to have a good quality, analytically acceptable to potential consumers. This shows that another profitable utilization of apple fruits apart from consumption could be undertaken. Apple wine production in small-scale level should be encouraged instead of importing wines from other countries or relying on the traditional grape.

This will be economical and also generate employment as well as to provide value addition to the agricultural produce.

This study has also given an insight into the efficiency and role of local yeast strains during alcohol fermentation of fruits.

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