IJIRR

International Journal of Information Research and Review
Vol. 03, Issue, 10, pp. 2834-2840, October, 2016

Research Article

PRODUCTION OF WINE FROM FERMENTATION OF *VITIS VINIFERA* (GRAPE) JUICE USING *SACCHAROMYCES CEREVISIAE* STRAIN ISOLATED FROM PALM WINE

Yabaya, A., *Bobai, M. and Adebayo, L. R.

Department of Microbiology, Faculty of Science, Kaduna State University, Kaduna, Nigeria

ARTICLE INFO

Article History:

Received 25th July 2016
Received in revised form
17th August 2016
Accepted 26th September 2016
Published online 31st October 2016

Keywords:

Wine, *Vitis Vinifera*,
Saccharomyces Cerevisiae,
Fermentation,
Alcohol,
Acidity.

ABSTRACT

This study aimed at evaluating the fermentative performance of palm wine *Saccharomyces cerevisiae* isolate on *Vitis vinifera* (grape) wine production.Sterilegrape fruit must was inoculated with standard inoculum of the *Saccharomyces cerevisiae* strain isolated from palm wine. Variationsin Physicochemical parameters were determined during five days' fermentation using standard procedures. Proximate analysis of the test fruit revealed low protein content (0.53%), but high moisture content (82.43%). During the fermentation, the pH values ranged from 3.50 to 3.27, percentage titratable acidity ranged from 0.020 to 0.051, while percentage volatile acidity from 0.085 to 0.125. The temperature of the fermentation ranged from 20 to 23^0C and the specific gravity values decreased steadily from 1.100 to1.010. Consistent increases in alcohol content was observed and at the end of five days' fermentation period, the percentage alcohol produced was14.6%. The fermentation recorded total viable yeast count from $2.5x10^6$ to 13.0 x 10^6 cfu/ml. Sensory evaluation of the wine revealed acceptable aroma/flavor and taste. This study indicates that *Saccharomyces cerevisiae* strain from the palm wine have good fermentative performance which suggested that it could be used for fruit wine production and other industrial applications preceded by further experiments.

INTRODUCTION

Wine is by common usage defined as a product of the normal alcoholic fermentation of the juice of sound ripe grapes.Grape wine is perhaps the most economically important fruit juice alcohol (Kelebek, Selli, & Canbas, 2013), because of the commercialization of the product for industry, the process has received most research attention. There is tremendous scope and potential for the use of microorganisms towards meeting the growing world demand for food, through efficient utilization of available natural food, feed stocks and the transformation of waste materials.Moreover, the role of yeast in wine making is the most important element that distinguishes wine from fruit juice, and most industrial production of wine utilizes commercial *Saccharomycescerevisiae* wine yeast strain. Most studies on palm wine have reported its potentials as sources of yeast isolates for the fermentation industries because it is a nutritionally rich medium for the growth of microorganism, among which is the yeast species (Nwachukwu *et al*., 2006, 2008; Nanknean, 2010).

Corresponding author: Bobai, M.,
Department of Microbiology, Faculty of Science, Kaduna State University, Kaduna, Nigeria.

Purified yeasts from palm wine showed highly viable cells and good metabolic activity during grape must fermentation (Ukwuru and Awah, 2013). *Saccharomyces cerevisiae* isolated from palm wine were tested and reported by Ukwuru and Awah (2013) to have high ethanol tolerance - a unique properties of the yeast that makes it exploitable for industrial application.
Non-availability and relatively high cost of obtaining effective commercially alcoholic fermentative *Saccharomyces cerevisiae* strain is a major constrain in development and sustaining local industrial fermentation process. Hence, there is need for search of indigenous strains that could be used as an alternative. Utilizing the palm wine yeast for industrial process requires a comprehensive knowledge of their technological and alcoholic fermentative performance, which however prompted this research. Therefore, this study aimed at evaluating the fermentative performance of *Saccharomyces cerevisiae* strain isolated from palm wine on *Vitis vinifera*wine production.

MATERIALS AND METHODS

Samples Collection and Transportation

Fresh palm wine samples were collected from tapped sources into sterile stomacher bags from Kagoro in Kaura Local Government Area, Kaduna State, Nigeria and transported in ice pack thermo-flask to Kaduna State University Microbiology Laboratory for the isolation of yeast.

Isolation and Identification of Yeast from Palm Wine

Sabouraud Dextrose Agar (SDA) was prepared according to manufacturer's instruction and supplemented with 50mg/L Chloramphenicol for selective enumeration of yeast. Serial dilution of the wine was carried out and inoculated using pour plate techniques. Pure culture was made on yeast glucose agar plates. Microscopic examination of the isolate was carried out using wet mount method according to Thais and Danilo (2006).

Carbohydrates Utilization Test on Yeast Isolated from Palm Wine

1% each of glucose, fructose, sucrose, lactose, xylose, mannitol, raffinose, galactose and maltose sugar were prepared using Yeast fermentation broth and dispensed 10ml volume into clean test tubes. Clean Durham tubes were introduced into the tubes, displaced all bubbles and then autoclave at 120^0C for 15minutes and allowed to cool. The sterile broth was inoculated with 0.2ml yeast culture broth and incubated at room temperature for 24-72hours and observed evidence of fermentation. Presumptive *Saccharomyces cerevisiae* isolate was confirmed using Analytical Profile Index (API 20 CAUX, BIOMAX).

Percentage Ethanol Tolerance Test

The test was carried out according to Alobo and Offonry (2009), where 2%, 5%, 8%, 11%, 14%, 16% and 19% of ethanol in molten yeast glucose agar medium were prepared and pour in triplicates plates. The plates were inoculated with the pure culture yeast isolates and incubated at room temperature for 24-72hours.

Proximate Analysis of the Fruit

Determination of Percentage Moisture Content

5g of the sample was weighed into Petri dish and placed in air draught oven at 100°C for 1 hour. The Petri dish was then weighed after cooling. The process was repeated thrice until a constant weight was obtained. Loss in weight was calculated as the percentage moisture content (Moronkola *et al*., 2011) and this was expressed by the following formula:

$$\% \text{ moisture} = \frac{loss\ in\ weight\ due\ to\ dryness\ x\ 100}{Weight\ of\ sample\ taken} = \frac{W2 - W3\ x\ 100}{W2 - W1}$$

Where; W_1 = weight of empty crucible, W_2 = weight of crucible + sample before drying,

W_3 = weight of crucible + sample after attaining constant weight on drying

Determination of Percentage Ash Content

This was carried out as describe by Moronkola *et al.* (2011), where porcelain crucible with lid was ignited in a hot Bunsen burner flame and transferred into desiccator to cool and the crucible was weighed. 5g of the sample was accurately weighed into the crucible and gently placed in the muffle furnace set at 600^0C for 4 hours. The crucible was place in desiccator to cool. The ashedsample in the crucible was weighed after cooling without the lid and the process repeated thrice for the sample. The result was calculated using the following formula:

$$\% \text{ Ash content} = \frac{W_3 - W_1}{W_2 - W_1} \times 100$$

Where, W_1 = weight of empty crucible,
W_2 = weight of crucible + sample before ashing
W_3 = weight of crucible + sample after ashing

Determination of Percentage Crude Fat

2g of the sample was transferred into a beaker and weighed as W, 10ml of water was added, and the solid was dispersed by agitating it. 10ml of concentrated hydrochloric acid was added and immersed in a boiling water bath until the solid particle dissolved and the mixture become brown in colour. This was allowed to cool and 10ml of alcohol added and agitated vigorously. A dried clean flask was weighed and recorded as W_1 and the ether layer was transferred into the flask and placed in a boiling water bath to evaporate the ether. The extraction was repeated by adding 50ml diethyl ether in order to evaporate the ether living the fat behind. The fat and the flask was weighed and recorded as W_2, then the fat content was calculated as follows:

$$\% \text{ fat} = \frac{W2 - W1}{W} \times 100$$

Where, W = weight of the sample
W_1 = weight of dried flask
W_2 = weight of dried flask fat residue.

Determination of Percentage Crude Protein Content

5g of dried finely sample was weighed on ash less filter paper. The paper with sample was folded and dropped into the digestion flask. 20ml of sulphuric acid (H_2SO_4) and 4 pieces of granulated zinc were added and shook, then heated gently inside a fume cupboard for 6 hours. The content in the flask was allowed to cooled. The solution was diluted with distilled water and transferred into 800ml Kjehldah flask. 100ml of 40% NaOH was added and distilled. This followed by titration against 0.05% of boric acid solution using methyl red as indicator. The protein content was estimated from the amount of nitrogen present in the sample.

$$\% \text{ nitrogen content } (N_2\%) = \frac{0.014 \times M \times V \times 100 \times D.F}{\text{Weight of sample}}$$

Where, M=is the actual molarity of acid
V = is the volume of acid used
D.F= is the volume ratio of solution

% protein content = Nitrogen content x conversion factor based on the sample

Determination of Total Carbohydrate Content

The Total carbohydrate content of the sample was obtained as described by Moronkola *et al*. (2011), where the results from fat, protein, moisture and ash content analyses were sum-up and the carbohydrate content was calculated as follows:
100% (% moisture + % protein + % fat + % ash).

Fermentation of the Fruit Must

The fermentation tank was wash and 1litre pasteurized fruit must was transferred aseptically to the fermentation tank. 0.2ml 0f 10% Sodium metabisulphite mixed well follow by addition of 2ml of 1.0 x10^6cells/ml of the standard yeast inoculum to the tank and shook to dispersed evenly in the tank.

This was kept at room temperature in a sterile laminar flow unit for fermentation up to 5days (Butz, 2007).

Determination of pH

The pH was determined directly during fermentation using a digital pH meter (model pHS -25) as described by Ochai and Kolhatkar (2008).

Determination of Titratable Acidity

The titratable acidity was determined during the fermentation based on Association of Analytical Communities International (2000) method 962.12. The sample was degassed by agitation. The pH of the water was adjusted by adding 1 ml phenolphthalein indicator for each 200ml of water. 0.1 N NaOH was used to neutralize the water to a distinct but faint pink colour (desired endpoint). 5ml of the degassed sample was pipette into a 250ml conical flask, 100ml of distilled water was added to the flask. The flask was swirled to release any remaining CO_2. 0.1 N NaOH was titrated against the content of the flask until the pale pink colour endpoint which persist for 30 seconds was achieved. Titratable acidity was calculated as follows:

$$\% \text{ Tartaric Acid} = \frac{(ml\ alkali\ )x(normality\ of\ alkali)x\ 7.5}{weight\ of\ sample\ (mls\ of\ sample)}$$

Determination of Volatile Acidity

Volatile Acidity was determined during the fermentation based on Association of Analytical Communities International (2000) method 962.12. The sample was degassed by agitation. The pH of the water was adjusted by adding 1ml drop of phenolphthalein indicator for each 200ml of water. 0.05 N NaOH was used to neutralize the water to a distinct but faint pink colour (desired endpoint). 5ml of the degassed sample was pipette into a 250ml conical flask, 100ml of distilled water was added to the flask. The flask was swirled to release any remaining CO_2. 0.05 N NaOH was titrated against the content of the flask until the pale pink colour endpoint which persist for 30 seconds was achieved and calculated as follows:

$$\% \text{ Acetic acid} = \frac{(ml\ alkali\ )x(normality\ of\ alkali)x\ 6.0}{weight\ of\ sample\ (mls\ of\ sample)}$$

Determination of Specific Gravity (S.G)

This was carried out using relative density bottle, where the bottle was wash with tap water and dried. The surface of bottle was further clean with ethanol and allowed to dried again. Using an electronic balance, the empty weight of the bottle was determined (M_0). The weight of the bottle plus 5ml of the wine sample (M1) was determined, follow by the weight of the empty bottle plus

5ml of distilled water (M2), and the specific gravity then calculated from these values as follows:

$$Specific\ gravity = \frac{wt\ of\ volume\ of\ sample\ (M1 - Mo)}{wt\ of\ an\ equal\ \ volume\ of\ water\ (M2 - Mo)}$$

Measurement of Temperature

120^0C mercury bulb thermometer was inserted to the side arm of the fermentation tank through a sterile rubber cork. The periodic temperature changes during fermentation was recorded.

Determination of Alcohol Content

This was determined using the %Sugar/Specific gravity/Brix/ PA equivalent table according to AOAC (2000).

Determination ofTotal Colony Yeast Count

Sabouraud dextrose agar supplemented with 50mg/L Chloramphenicol was used for selective enumeration of yeast according to Steger and Lambrechts (2010) and Mir and Mohammed (2014). Serial dilution of the wine was carried out and inoculated using pour plate techniques. Pure culture was made on yeast glucose agar plates. Colonies from the plates of 10^{-6} dilution were count using colony counter.

Sensory Evaluation of the Fermented Fruit Wine

The sensory evaluation of the fermented fruit wine was made by ten panelist comprising of staff and students of Microbiology Department, Kaduna state University, Kaduna. The samples were evaluated using "Hedonic Scale" Test method (Hodgson, 2008) following the instructions provided in the sensory evaluation questionnaires.

RESULTS AND DISCUSSION

Results

***Saccharomyces cerevisiae* strain Isolated from Palm Wine**

The *Saccharomyces cerevisiae* isolate revealed a high ethanol tolerance up to 16%.

Proximate composition of grape fruit

Table 1 showed the proximate composition of grape fruit with low protein content, but high moisture and total carbohydrate content.

Variations in the Physicochemical and Microbiological Parameters during Fermentation

As revealed in Figure 1, the pH of the fruit must was 3.64, but during fermentation, the pH ranged was 3.50 to 3.27.The total titratable acidity of the must before fermentation was 0.023 (Figure 2) and during fermentation, the titratable acidity was recorded as 0.020 to 0.051 with fluctuation showing an initial increased from the 0.023 of the must at zero hour to 0.040 at 36hours, then a decrease to 0.020 at 72hours and a further increased to 0.051 at 120hours.Figure 3 shows that the volatile acidity of the must before fermentation was 0.075 and during fermentation, the volatile acidity was recorded as 0.087 to 0.121, then fluctuate with an initial increased from the 0.075 of the must at zero hour to 0.092 at 36hours, then a decrease to 0.087 at 48hours and a further increased to 0.120 at 120hours.

Table 1. Proximate composition of grape fruit

Parameter	Moisture	Ash	Fat	Protein	Total Carbohydrate	Crude Fibre	Energy (Kcal/G)
Composition(%)	82.43	2.27	0.535	0.53	14.23	2.43	63.90

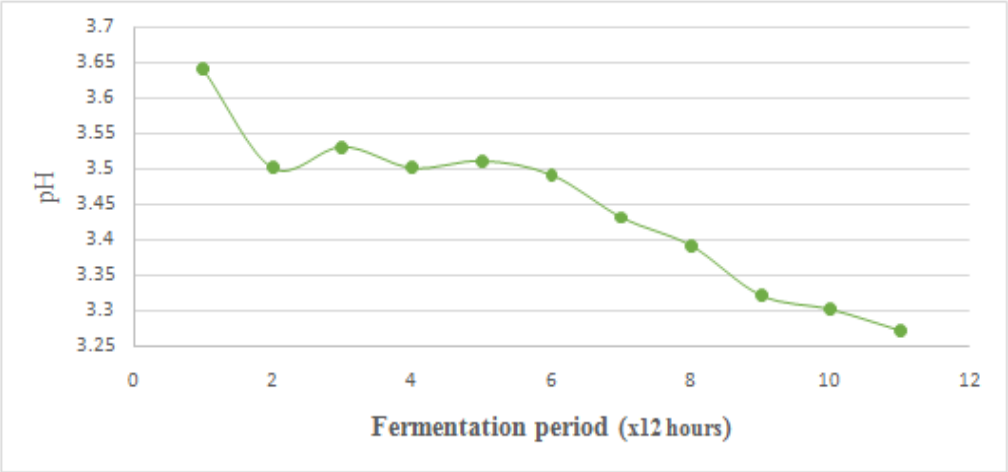

Figure 1. Variation in pH during Fermentation

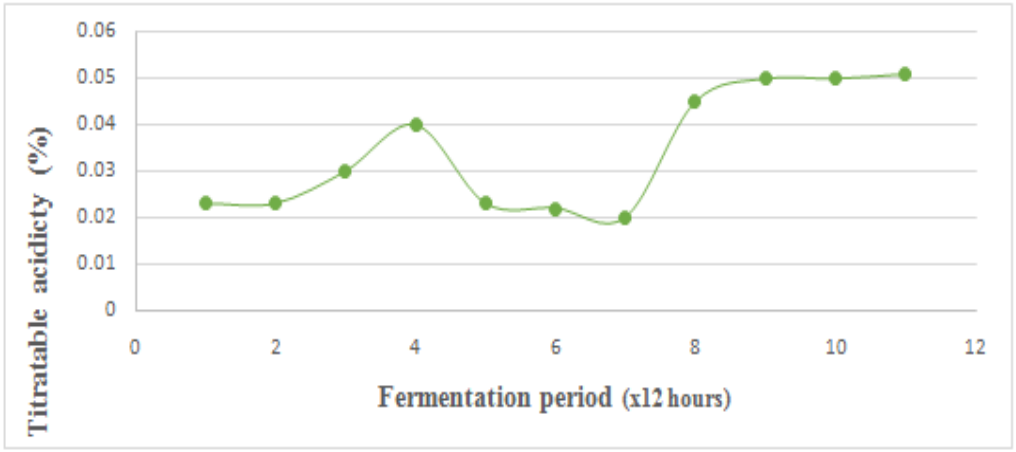

Figure 2. Variation in Titratable Acidity during Fermentation

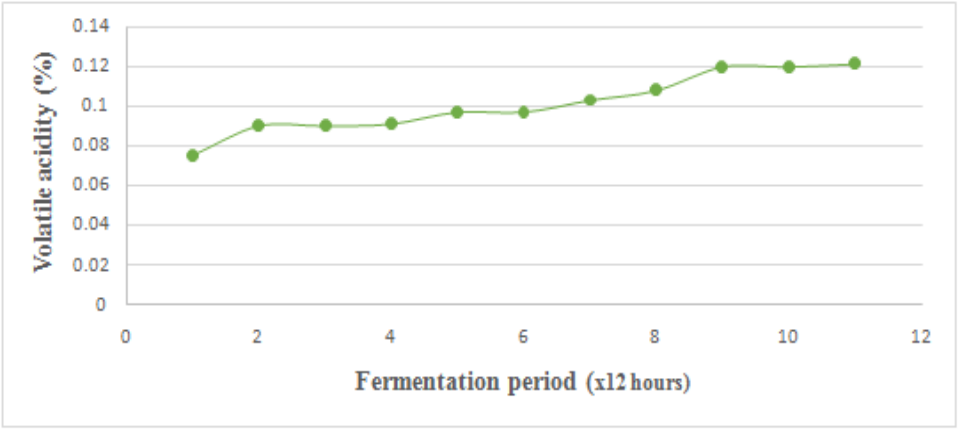

Figure 3. Variation in Volatile Acidity during Fermentation

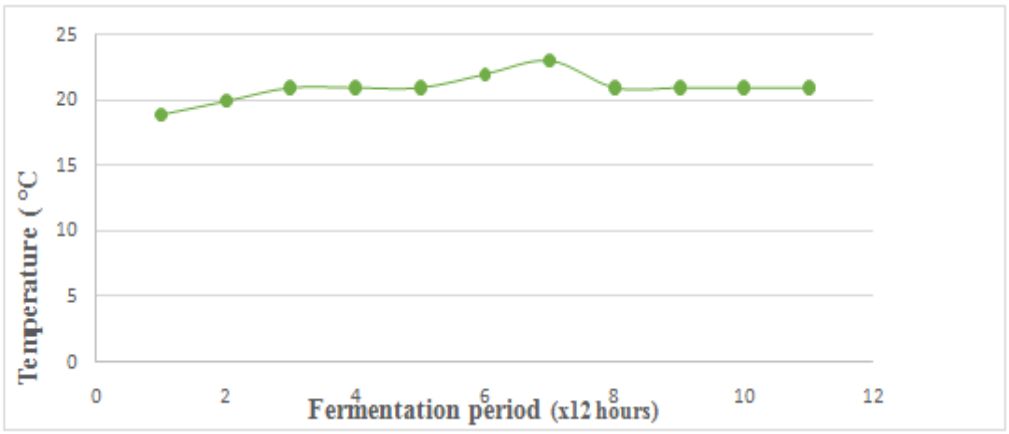

Figure 4. Variation in Temperature during Fermentation

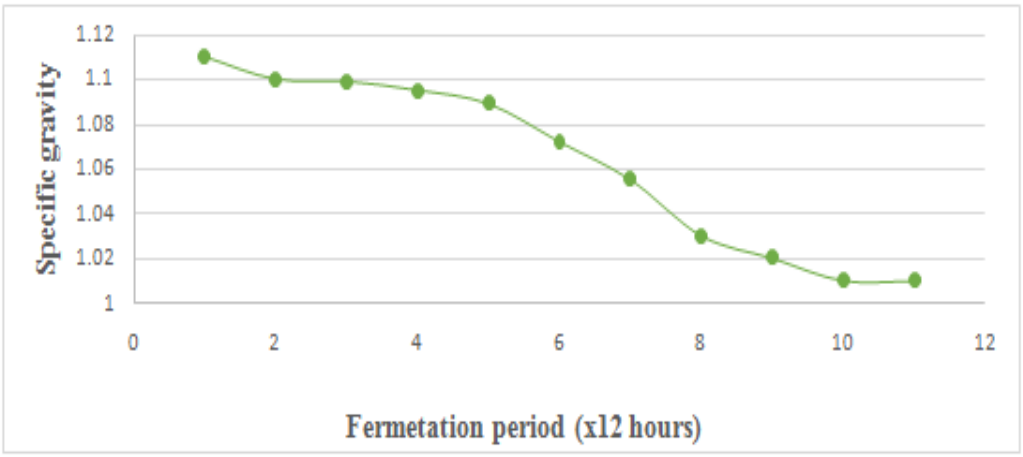

Figure 5. Variation in Specific Gravity during Fermentation

According to figure 4, the temperature of the fruit must before fermentation were recorded as 19^0C and it rises during fermentation to 23^0C at 72hours, then decreases to 21^0C at 120hours.As revealed in figure 5, the specific gravity determined before fermentation was 1.11, while during fermentation the values determined decreasesteadily from 1.100 to 1.010 for 12hours to 120hours. The percentage alcohol determined was 14.6% (Figure 6).During fermentation, the percentage alcohol increases steadily from 12hours to the 120hours. Figure 7 revealed no growth on the pasteurized fruit must before the fermentation, but after inoculation of sterile fruit must and the onset of the fermentation, total colony yeast count (cfu/ml) ranged from 2.5 x 10^6 at 12hours to 13.0 x10^6 at 84hours and then decline to 8 x 10^6at 120hours.Sensory evaluation of the wine revealedacceptable aroma/flavor and taste.

DISCUSSION

In this study, *Saccharomyces cerevisiae* strain isolated from the palm wine samples revealed a high ethanol tolerance up to 16%. This implies that this yeast strain can remain metabolically active in the fermentation medium and tolerate up to 16 % alcohol produce during the fermentation period. Bechem *et al. (*2007) stated that high percentage ethanol tolerance is a property that can be exploited for industrial applications.

The results of the proximate composition (Table 1) revealed high percentage moisture content (82.43%) and this according to Okaka (2010) accounts for their high perishable nature and their short shelf life under normal storage condition. The fruit also contained reasonable amount of total carbohydrate (14.23%) which invariably account for their high caloric values suggesting the presence energy source for metabolic activity of the yeast.The protein content was 0.53%, and according to Okegbabile and Taiwo (2009), the low protein and mineral contents of the fruit as reported in this study is a probable indication that fear of over accumulation due to consumption of the fruits do not arise. The proximate composition in this investigationwas in agreement with the general case for fruits as reported by Pearson (2007). The present study revealed low pH values (3.50 to 3.27) in the fruit wine fermentation period. The trend of the changes in pH revealed consistent increases in acidity of the fruit wine during the fermentation. Chilaka *et al*. (2010) also recorded a similar pH ranged of 3.0 to 4.8 during fermentation of passion fruit, water melon and pineapple fruits must using commercial *Saccharomyces cerevvisiae*. Studies have shown that during fermentation of fruits, low pH is inhibitory to the growth of spoilage organisms but creates conducive and competitive advantage environment for the growth of desirable organisms. (Reddy & Reddy, 2009). Temperature of the fermentation of fruit wine in this current investigation ranged from 20 to 23°C.

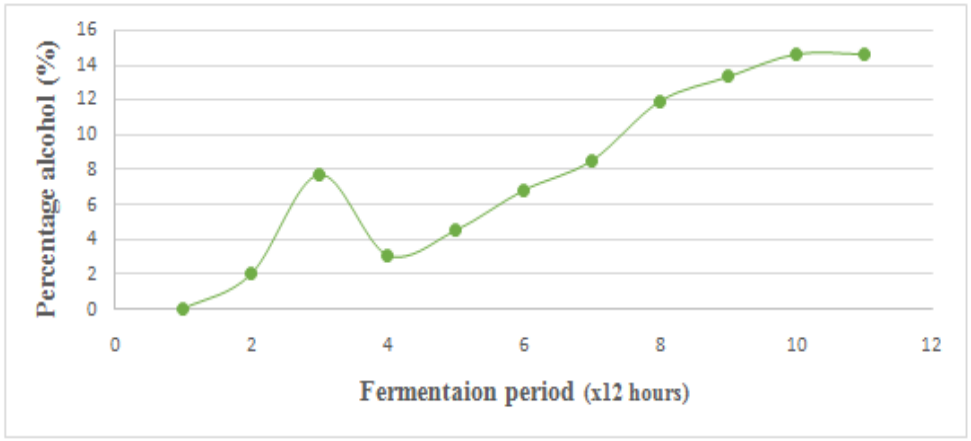

Figure 6. Variation in Percentage Alcohol during Fermentation

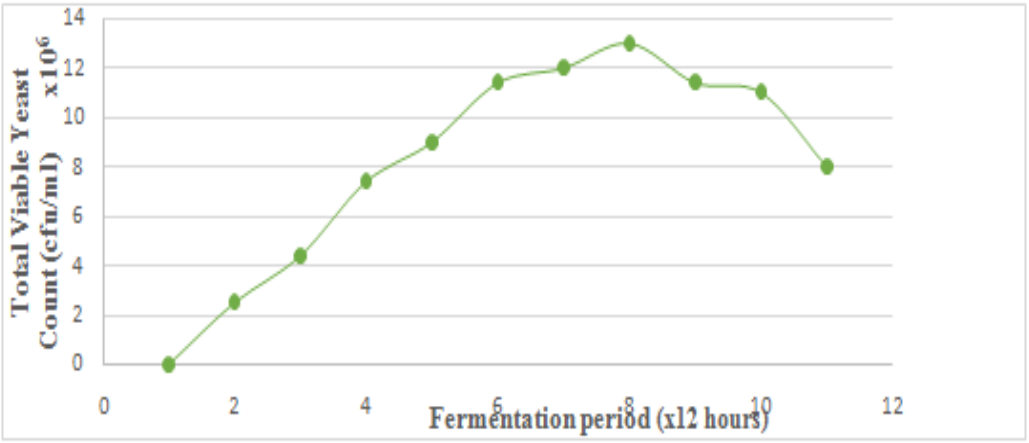

Figure 7. Variation in Total Viable Yeast Count during Fermentation

A similarobservation has been reported by (Reddy & Reddy, 2009), where the temperature values for quality mango fruit wine production was 5to 30°C. The rises in temperature recorded may be due to the catabolic processes of sugars by yeast cells resulting in metabolic heat that ultimately increased the temperature. The production of heat during fermentation as an exothermic process means that the temperature of the fermentation in the vessel rises. Malic acid and tartaric acid are the primary acids in wine grapes, and these acids have direct influence on growth and vitality of yeast during fermentation (Bellman and Gallander, 1979). The measure of the amount of acidity in wine is known as the "titratable acidity or total acidity" which refers to the test that yields the total of all acids present, while the strength of the acidity is measure according to pH, with most wines having pH values between 2.9 and 3.9. The result of this investigation recorded ranged of pH values between 3.27 and 3.50, % titratable acidity between 0.020 and 0.051, and %volatile acidity between 0.087and 0.120 during the fermentation. Generally, the lower the pH the higher the acidity in the wine (Bellman and Gallander, 1979). Also, acetic acid is a two-carbon organic acid produced in wine during or after the fermentation period. In this study, the specific gravity values were observed to decreases steadily with ranged from 1.100 to1.010 during fermentation. Decreasedin specific gravity values was observed to inversely related to increases in alcohol content, as remarkable amount of alcohol was produced from the fruit wine during fermentation with the test yeast.

The percentage alcohol produced at the end of fermentation was recorded as 14.6%. Similar findings by Chilaka *et al*. (2010) during fermentation of passion, water melon and pineapple fruit must have revealed percentage alcohol content ranged from 10.14 to 12.8%. This however, according to Okunowo and Okotore (2005) is comparable with moderate grape wine. The fermentation recorded highest total viable yeast count of $2.5x10^6$to 13.0 x 10^6 cfu/ml. The yeast viable count (figure 7) of this study showed rapid increases during the first three days (72hours) fermentation and thereafter, a shaped decline. This is probably because in the first three days, there was much nutrients (fermentable sugars) available to the yeast, active enzymes, viable yeast cells, etc. which might be responsible to the increase in the yeast count. In the contrary, nutrient depletion in the fermenter, high cells density, and possible present of toxic metabolic by- products could be responsible to the decline in the yeast count after the third day of the fermentation. More so, consistent increase in the alcohol concentration may result to the yeast cell membranes becoming susceptible to rapture by the ethanol, the possible contributing factor for declined in growth recorded on the third days of fermentation. High alcohols are known to be important precursors for the formation of esters, which are associated with pleasant aromas (Clemente *et al.,* 2007). Report have shown that the fermentation of fruits juices using yeast from different sources creates variety in flavor and varying levels of alcoholic contents in wines. More so, that alcoholic fermentation leads to a series of by-products in addition to

ethanol; and that some of the byproducts include carbonyl compounds, alcohols, esters and acids, all of them influencing the quality of the finished product.The type and aroma produced during wine making is reported to depend on yeast, environmental factors and physio-chemical characteristics of the musts. In this investigation, the sensory evaluation of the fruit winerevealed acceptable aroma and taste. This study revealed the effectiveness of palm wine *Saccharomyces cerevisiae* strain on *Vitis vinifera*fruit wine production with characteristic ability to complete fermentation; probably due to its high ethanol tolerance; hence, resulting in production of generally acceptable flavor, taste, and colour fruit wine.

Conclusion

In this study, grape wine was produced from the palm wine *Saccharomycescerevisiae* isolate. High percentage alcohol was produced by the test yeast strain during fermentation of the grape wine and the pH level of the fruit wine fall within acceptable limits. This study therefore indicates that *Saccharomycescerevisiae* strain isolated from the locally tapped palm wine can be used to make grape fruit wine. However,process optimization and scale up will be required; and hence starter culture obtain to augment the more expensive and non-available commercial wine *Saccharomyces cerevicae* strain for better applications of this study.

REFERENCES

Alobo , A. P., and Offonry, S. U. 2009. Characteristics of coloured wine produced from roselle (Hibiscus sabdariffa) calyx extract. *Journal of Institute Brewing*, 91-94.

Association of Analytical Communities International. 2000. Official Methods of Analysis of wine . *AOAC International* (pp. 25-30). USA: AOAC International press.

Bechem EET., Omoloko, C., Nwaga, D., and Titanji, V.P.K. 2007. Characterization of palm wineyeasts using osmotic, ethanol tolerance and the isozyme polymorphism of alcohol dehydrogenase. *African Journal of Biotechnology*, 6(14):1715-1719.

Bellman, R. B. and Gallander, J.F. 1979. Wine deacidification in Chichester. *Advance in Food Research* Vol.25, Academic press Pp. 3.

Berry, C. J. 2007. First Step in Wine Making. *Nexus Special Interests*, Berry, C.J.J., 1996. First Step in Wine Making. Nexus Special Interests Ltd., Hertf.

Berti, L. A. 2009. Sparkling Wine Production in California. *American Chemical Society, 145*, Berti, L.A., 1981. Sparkling Wine Production in California. In: Wine Production Technology in the United 85-121.

Butz, E. 2007. Practical considerations for managing wine acidity. *Proceedings of the Spring.*

Clemente, J. M., Mingorance, C. L., Martíne, R., Heras, F. J., & Rodríguez, V. S. 2007. Influence of sequential yeast mixtures wine fermentation. *International Journal of Microbiology*, 301-308.

Chilaka C. A., Uchechukwu N., Obidiegwu J. E. and Akpor O. B. 2010. Evaluation of the efficiency of yeast isolates from palm wine in diverse fruit wine production. *African Journal of Food Science* Vol.4(12):764-774

Hodgson, R. T. 2008. An examination of judge reliability at a major US wine competition. *Journal of Wine Eccon*. 3(2): 105-113

Kelebek, H., Selli, S. and Canbas, A. 2013. HPLC determination of organic acids, sugars, phenolic compositions and antioxidant capacity of orange juice and orange wine. *Microchemical Journal 91*, 20-24.

Mir , N. A. and Mohammed, M. K. 2014. Screening, identification and characterization ofalcohol tolerance potential bioethanol producing yeast. *Current Research in Microbiology and Biotechnology*, 2-4.

Moronkola, B. A., Olowu, R. A. Tovide, O.O. and Ayejuyo, O. O. 2011. Determination of proximate and mineral contents. *Sc. Revs. Chem.Commum.* 1(1): 1 - 6

Naknean, P., Meenune, M. and Roudaut, G. 2010. Characterization of palm sap harvested in Songkhla province, Southern Thailand. *International journal of Food Research* 17 (4):977-986.

Nwachukwu, I. N., Ibekwe, V. I., Nwabueze, R. N. and Anyanwu, B. N. 2006. Characterization of palm wine yeast isolates for industrial utilization. *African Journal of Biotechnology* 5 (19): 1725-1728.

Ochai, J. O. and Kolhatkar, A. 2008. Medical Laboratory Science and Practice. Tata McGrewHill Publishing Limited New Delhi, New York. Pp 50-53, 803.

Okaka, J. C. 2010. Tropical Plant Perishable Processing, Storage and handling. *New Heaven*, 55-60.

Okegbile, E. O., and Taiwo, E. A. 2009. The Nutritional potential of black velvet,. *Nigerian Journal of Nutrition Science*, 115-121.

Okunowo , W. O., and Okotore, R. S. 2005. The alcoholic fermentative efficiency of indigenous yeast strains of different origin. *African Journal of Biotechnology*, 1290-1296.

Pearson, D. 2007. The Chemical Analysis of Foods. *Churchill Living*, 56-89.

Reddy, L. V., and Reddy, O. V. 2009. Production and optimization and characterization of wine from mango. *Natural Product journal*, 426-435.

Samson, J. A. 1992. *Tropical Fruits.* New York.: Longman Publishers.

Shrikant, B., and Thakor, A. D. 2006. Fruit Wine Production: A Review. *Journal of Food Research and Technology*, 33-34.

Steger, C., and Lambrechts, M. G. 2010. The selection of yeast strains for fermention of fruit wine. *Journal of Industrial Microbiology and Biotechnology*, 431-440.

Thais, M. G., and Danilo, G. M. 2006. Isolation and characterization of Saccharomyces cerevisiae strains. *Brazilian Journal of Pharmaceutical Sciences*, 4-6.

Ukuru, M. U. and Awah J.I. 2013. Properties of palm wine yeasts and its performance in wine making. *African Journal of Biotechnology* 12(19): 2670-2677

****