
ABSTRACT

Wine is an alcoholic beverage made from fermented grapes or other fruits. Many tropical fruits such as mango, jackfruit, litchi, banana and cashew apple have been shown to be suitable for fermentation, mainly because of their appropriate taste, flavour, availability, high sugar and water content and overall chemical composition (Muniz et al. 2008). The natural chemical balance of these fruits lets them ferment without the addition of sugars, acids, enzymes, water, or other nutrients. Yeast consumes the sugars in the fruits and converts them into alcohol. Study was conducted to produce red wine without using any sugar and making use of the kitchen yeast strain i.e. *Saccharomyces cerevisiae* and was tested for the different physical and chemical characteristics of the wine such as acidity, sugar content and other quantitative and qualitative tests. The wine was produced in simple lab conditions and using simple lab utensils and instruments so this technique may very well reduce the overall cost of wine production. The production and tests were conducted in small scale but this technique can be converted to large scale with few changes.

KEYWORDS: Fruit Wine, Wine Production.

INTRODUCTION

Wine is an alcoholic beverage made from fermented grapes or other fruits. The natural chemical balance of grapes lets them ferment without the addition of sugars, acids, enzymes, water, or other nutrients.^[1] Yeast consumes the sugars in the grapes and converts them into alcohol and carbon dioxide. Different varieties of grapes and strains of yeasts produce different styles of wine. The well-known variations result from the very complex interactions between the biochemical development of the fruit, reactions involved in fermentation, terroir and subsequent appellation, along with human intervention in the overall process. The final product may contain tens of thousands of chemical compounds in amounts varying from a few percent to a few parts per billion.

Wines made from produce besides grapes are usually named after the product from which they are produced (for example, rice wine, pomegranate wine, apple wine and elderberry wine) and are generically called fruit wine. The term "wine" can also refer to starch-fermented or fortified beverages having higher alcohol content, such as barley wine, huangjiu, or sake.

Wine has a rich history dating back thousands of years, with the earliest production so far discovered having occurred c. 6000 BC in Georgia.^{[2][3][4]} It had reached the Balkans by c. 4500 BC and was consumed and celebrated in ancient Greece and Rome

Red wine – Red Wine is made from the must (pulp) of red or black grapes that undergo fermentation together with the grape skins.

White wine – White wine is made by fermenting juice which is made by pressing crushed grapes to extract juice; the skins are removed and play no further role. Occasionally white wine is made from red grapes; this is done by extracting grapes' juice with minimal contact with their skins.

Sparkling wine – Sparkling wine is a wine with significant levels of carbon dioxide. A classic example of a sparkling wine is Champagne. There are two levels of fermentation involved in making sparkling wine. It is the initiation of a secondary fermentation that distinguishes sparkling wine production and gives the wine bubbles.

Fortified wine - It can be red or white wine that has been flavoured by the addition of herbs and barks like cardamom, cinnamon, marjoram and chamomile. It is used primarily as pre-meal appetiser.

Rose wine – Rose wine is made from red grapes where the juice is allowed to stay in contact with the dark skins long enough to pick up a pink colour.

REVIEW OF LITERATURE

Litchi wine fermentation process

One kilogram of litchi fruits were peeled by hand; skin and seeds were separated from the pulp. Then litchi pulp was crushed in a mixture/grinder (TTK Prestige Ltd., Bangalore, India) and the juice was extracted by using a juice squeezer. The juice (must) filtered through cheese cotton cloth had 17° Brix and was treated with sodium metabisulphite (SMS) (100 µg/ml) to inhibit the growth of undesirable microorganisms such as acetic acid bacteria, wild yeasts and moulds. The pH of the juice was adjusted to 3.8 with 1 N tartaric acid and then inoculated with 2% (v/v) starter culture (prepared with grape juice) of *S. cerevisiae* var. *bayanus*.^[5] Fermentation was carried out at a temperature of 30 ± 2°C for 6 days. Racking of wine was carried out when total soluble solids (TSS) reached 4-6° Brix. Two or three more rackings were done at 15 days intervals to remove any sediment deposited in wine. After racking, the wine was clarified with the addition of 0.04% bentonite. Another dose of SMS (100 µg/ml) was added as preservative before bottling^[6].

Preparation of mango wine

S. cerevisiae var. *ellipsoideus* CFTRI 101 (wine yeast) was received from Central Food Technological Research Institute (CFTRI), Mysuru (India). Culture maintenance and inoculum preparation was done as mentioned in Reddy and Reddy¹⁰. The culture was maintained on MPYD (Malt extract 0.3%, Peptone 0.5%, Yeast extract 0.3%, Dextrose 2% and Agar 1.5%) slants at 4°C. The inoculum was prepared by inoculating the slant culture into 25 ml of the sterile MPYD liquid medium taken in 100 ml Erlen Mayer flask and allowed to grow it on a rotary shaker (100 rpm) for 48 h at 37°C. This inoculum (3×10⁶ cells/ml) was transferred to 250 ml conical flasks having 100 ml mango juice.

Batch fermentation of the inoculated must was carried out in conical flasks by incubating at pH 4.5 and at temperature 20°C for 15 days at 22 ± 2°C. The samples were collected by separation of the cells by centrifugation at 5,000 × g for 10 minutes. The fermented samples were kept at -20°C for a few weeks for chemical and sensory analyses.

Banana wine preparation: One and half kg of banana was taken it was completely peeled off. This yielded 300ml of Banana pulp. The pulp was macerated in blender and pasteurized at 85°C-90°C for 05 minutes. Similar procedure was carried out as like papaya wine preparation.^[7]

Citrus wine preparation: One and half kg of orange and one and half kg lime was taken it was completely peeled off. This yielded 300ml of orange and lime pulp. The pulp was macerated in blender and pasteurized at 85°C- 90°C for 05 minutes. Similar procedure was carried out as like papaya wine preparation.^[8]

Papaya wine preparation:

One and half kg of papaya was taken it was completely peeled off. This yielded 300ml of Papaya pulp. The pulp was macerated in mixer /blender and pasteurized at 85°C-90°C to 5 minutes. After cooling required amount of cane^[9]

MATERIALS AND METHODS

Grapes: Grapes were purchased from market followed by its washing and then dried in air for half an hour.

Yeast: Dry yeast powder was purchased from market and then cultured in YEPD media for further use.

Lactic acid bacteria: We purchased Nutrolin B plus capsules from market and then cultured Lactic acid bacteria in luria broth.

Wine preparation:

- 1.5 kg grapes were weighed for the extraction of juice.
- Grapes were crushed in TLC tank to separate the skin of grapes and juice.
- Yeast paste was prepared and then added to the must (Grape skin & juice).
- This mixture is then kept in order to achieve primary fermentation for 15 days.
- After 15 days the must is then strained through muslin cloth to separate the juice from the skin of grapes.
- To the samples (grape juice), lactobacillus is then added for the secondary fermentation (malo-lactic fermentation) and kept for 20 days.

Produced wine is then followed by many tests (qualitative and quantitative) in order to check for its alcohol and sugar content.

ACIDITY ESTIMATION:-

Material required : 4 pH tablet, 7 pH tablet, 9.2 pH tablet, Water, Buffer solutions, Juice sample (primary and secondary fermented)

- 1) Three solutions were prepared by adding the following:
 - i) 100 ml water + 4 pH buffer tablet
 - ii) 100 ml water + 7 pH buffer tablet
 - iii) 100 ml water + 9.2 pH tablet
- 2) Standardized the pH meter with the above three solutions.
- 3) Checked the pH of juice (primary)
- 4) Checked the pH of wine (secondary)

SUGAR ESTIMATION:-

Material required : Juice sample (primary and secondary fermented)
Anthrone , Glucose, Conc. H₂SO₄, Test tubes

ANTHRONE METHOD:

For juice sample after primary fermentation

- 1) Glucose stock solution preparation (1%): 1 gm glucose is added to 100 ml water.
- 2) Anthrone stock preparation (0.2%): 0.2 gm of anthrone is added to conc. H₂SO₄.
- 3) 1 ml juice (wine) is then added to 100 ml water.
- 4) Three test tubes were prepared for glucose stock solution with following composition:
 - i) 9 part water + 1 part glucose stock
 - ii) 8 part water + 2 part glucose stock
 - iii) 7 part water + 3 part glucose stock
- 5) Similarly three test tubes were also prepared for our juice sample with following composition:
 - i) 9 part water + 1 part juice
 - ii) 8 part water + 2 part juice
 - iii) 7 part water + 3 part juice
- 6) In all of these six test tubes we added 4ml of anthrone stock solution.
- 7) OD was taken at 450 nm for each of the 3 samples by using the 3 samples of glucose stock as the blank. The same procedure was repeated for the samples of juice (wine) after the secondary fermentation.

ALCOHOL ESTIMATION (QUALITATIVE):-

Material required: Juice sample (primary and secondary fermented), Iodine, Potassium iodide, Sodium hydroxide, Test tubes, Water bath, Beaker.

- 10 ml of both the samples were taken in two test tubes, one from primary fermented and second one from secondary fermented juice (wine).
- 100 µl of iodine solution is added to the samples.
- 1 mg potassium iodide is then added.
- An addition of a small crystal of sodium hydroxide is then followed by the above step.
- This mixture is then kept in a water bath for 30 minutes.

- Yellow precipitate then confirms the presence of alcohol in the sample.

ALCOHOL ESTIMATION (QUANTITATIVE)

Evaporation method:

- 1) 2 empty beaker was taken and weighed using weighing balance.
- 2) 50 ml juice is then transferred to the one beaker and normal water to the other and again weighed.
- 3) The beakers were then kept on digestion unit at 35 °C for 20 minutes.
- 4) Beaker was then again weighed using weighing balance.
- 5) Using above data alcohol percentage (v/v) present in the wine was calculated.

RESULT AND DISCUSSION

Many tropical fruits such as mango, jackfruit, litchi, banana and cashew apple have been shown to be suitable for fermentation, mainly because of their appropriate taste, flavour, availability, high sugar and water content and overall chemical composition (Muniz *et al.* 2008).

In this study we were able to prepare a 750 ml i.e a bottle of grape wine at a very economic cost which was rupees 200 including all the resources used .the different characteristics of the prepared wine is discussed as follows:

Cost of different constituents used in preparation of wine:

1. 1.5 kg of black grapes : 80-100 rupees
2. Dry yeast packet : 40 rupees.
3. Nutrolin B capsule: 5 rupees
4. Cost of other components: 30-50 rupees.

1) pH ESTIMATION: The pH of the wine was calculated to be 3.7 which is consumable for us.

2) SUGAR ESTIMATION: The test results show that, the amount of sugar present after the primary fermentation of wine is 86.67% and after secondary fermentation 62.68%.

Total amount of sugar converted to alcohol in pr. Fermentation =13.33%

Total amount of sugar converted to alcohol in sec. fermentation = 37.32%

3) ALCOHOL ESTIMATION (QUALITATIVE): After addition of iodine, potassium iodide and sodium hydroxide to both the samples i.e- juice sample and wine sample, a yellow colored precipitate was formed which confirmed the presence of alcohol in both the samples.

4) QUANTITATIVE ESTIMATION OF ALCOHOL CONTENT IN WINE: Amount of alcohol calculated came out to be 9.4% v/v

COMPARISION OF DIFFERENT WINE

Alcohol content of different wines:

In the study conducted the alcohol content of the prepared red wine came out to be 9.4% . when the result of the other wine were studied , the wine prepared from the banana had the highest alcohol content i.e 15.49% . this is due to the higher sugar level present in banana . pH of different wine is shown in following^[10] table and graph

Sr. No.	FRUITS USED	ALCOHOL CONTENT (%)
1	Papaya wine	8.73%
2	Banana wine	15.49%
3	Orange wine	8.65%
4	Lime wine	0.93%
5	Grape wine	9.4%
6	Litchi wine	11.4%
7	Mango wine	7.4%

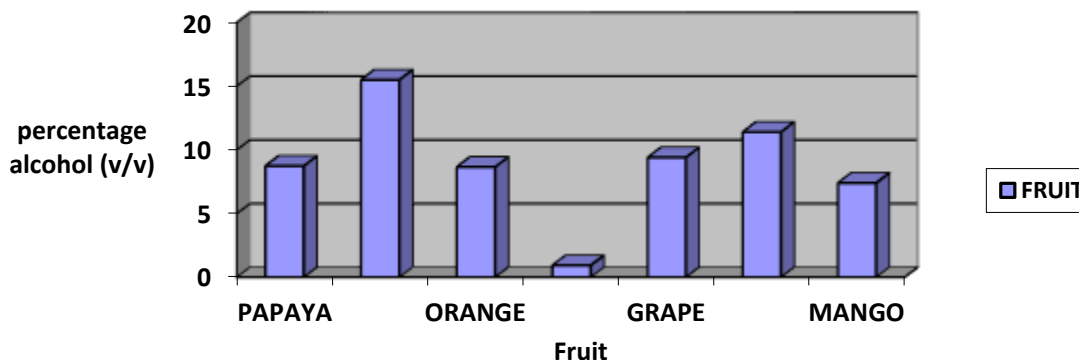


Figure 1: graph showing alcoho pcentage of different wine

pH of different wines

In the study the pH of the prepared grape wine came out to be 3.7 and the pH of other wine was in the range of 3-3.7 which is of the required range.

Sr. No.	FRUITS USED	Ph
1	Papaya wine	3.3
2	Banana wine	3.3
3	Orange wine	3.1
4	Lime wine	3
5	Grape wine	3.7
6	Litchi wine	3.05
7	Mango wine	4.1

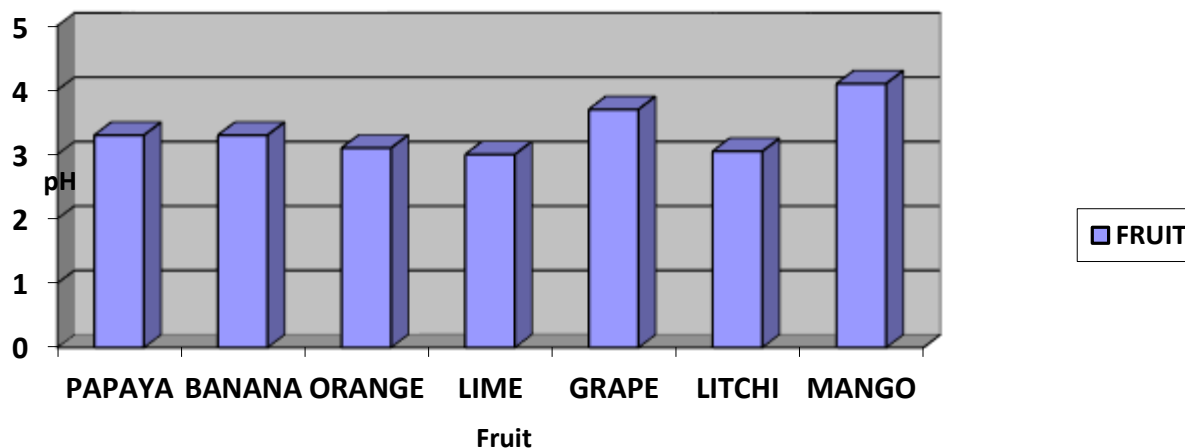
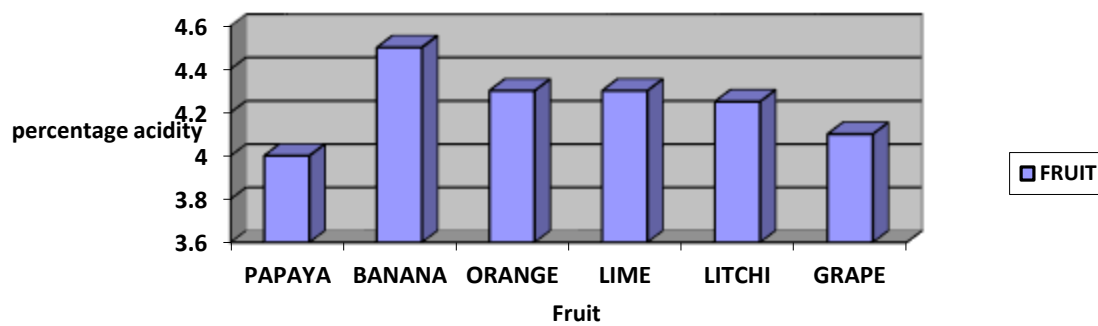


Figure 2: Graph showing pH of different wine

Percentage acidity of different wines

The percentage acidity of different wines were in the range of 4-4.5. the acidity of different wines are given in following table

Sr. No.	FRUITS USED	Percentage acidity (after 30 days)
1	Papaya wine	4
2	Banana wine	4.5
3	Orange wine	4.3
4	Lime wine	4.3
5	Litchi wine	4.25
6	Grape	4.1



CONCLUSION

In the study conducted it was found that it is possible to prepare a wine at low cost as low as rupees 200. The wine was prepared without adding any extra sugar but to enhance the flavor sugarcane juice can be added which will also be economical and also improves tastes. Also different fruits can be used to prepare wines like mango, banana, litchi, etc. of which banana gives the highest alcohol output due to high sugar content.

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