



Production of Wine from Guava (*Psidium guajava*L.) By Using *Saccharomyces cerevisiae* Isolate

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ABSTRACT

Wine is one of the functional fermented foods having many health benefits like anti-aging effects, improvement of lung function (from antioxidants in white wine), reduction in coronary heart disease, and development of healthier blood vessels. Many wines are made from fruits having medicinal value. The production of Guava wine in India was found to be of great benefit. A samples of Guava fruit was collected from the local vegetable market of Karad, Maharashtra, India. The wine was prepared from Guava fruit juice by using the yeast *Saccharomyces cerevisiae*. The fermentation process was optimized by adjusting the sugar level and pH. It was seen that wine prepared from juices has a sugar level of 0.42 mg/mL and a pH of 5. The alcohol content in Guava fermented juice was found to be 5.43%. From the results, it was also concluded that *Saccharomyces cerevisiae* is suitable for Guava wine production.

Key words: Wine, Guava fruit juice, *Saccharomyces cerevisiae*

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INTRODUCTION

Guava (*Psidium guajava* L.), a good source of ascorbic acid, pectin, sugars and certain minerals, is an important natural fruit and its juice consumptions serve as an alternative to the traditional caffeine-containing beverages such as coffee, tea or carbonated soft drinks. Guava, with its widely appreciated flavor and aroma, is able to compete in the market, either as guava juice or as mixtures with other juices or guava wine. However, raw guava juice is turbid, gray in color, very viscous and tends to settle during storage, and therefore, it must be clarified prior to commercialization. It is easy to extract the maximum juice and then directly going for storage or wine production [1]. Yeast, during the fermentation process, can utilize the sugars in quantities present in the fruits. During fruit wine production, the initial sugar concentrations, fermentation temperatures, SO₂ concentrations and specific yeast strains are key factors along with pH values, sugar contents and nitrogen contents in determining successful fermentative processes of fruit wine [2]. The use of *Saccharomyces cerevisiae* yeast starters has been extensively applied in wine production processes. It allows rapid and reliable fermentations, reduce the risk of sluggish or stuck fermentations and prevent microbial contaminations [3]. For optimization of the raw material quality and individual characteristics of the wine, yeast starter cultures are specifically selected on the basis of scientifically verified characteristics, creating a more desirable product [4]. Higher quality wine can be produced with selected yeasts than by spontaneous fermentation [5]. By anaerobic fermentation by yeast, the sugars are converted into alcohol & carbon dioxide during guava (*Psidium guajava* L.) wine production. There is a report on ethanol production from guava pulp [6] but there is no study found in literature for guava must fermentation for guava fruit wine production. Therefore, the aim of the present study was to produce guava fruit wine and check its quality and characteristics.

MATERIALS AND METHODS [1, 5, 7]

Collection of Fruit Sample

Over ripened (Guava) fruits, disease free were collected from local market in Karad, Maharashtra, India and were selected for the preparation of wine.

Enrichment and Isolation of Yeast and Inoculum Preparation [8]

The yeast was isolated by using enrichment culture technique. The (Guava) fruit cut into small pieces and put in 100mL sterile Yeast extract potato Dextrose broth (YEPDB) in sterile conical flask and kept in room temperature for 48 h. After incubation the growth was cultured on yeast extract peptone dextrose agar plate for the isolation of yeast. The typical yeast colony (white, convex curd like smelling) was selected

and preserved in triplicates at refrigeration on medium slant for further use. The yeast growth from media plates was scrapped and suspended in 30mL fruit juice with 12% sugar adjusted with sucrose and hand refractometer and incubated on shaker at 150 rpm for 24-h. This was used as inoculum.

Pretreatment on Fruit

Before the production of wine, the pretreatment of fruits (10 in number) was carried out. The fruits were washed with warm water to remove the toxicity and pesticides as well as contaminants on the skin of the fruit. During the washing care was taken to avoid the crushing of fruit. After the repeated washing with sterile water, the fruits were dried at room temperature.

Preparation of Must Fruits

After the pretreatment, the fruits were weighed, sliced, and rewashed with the removal of seed. The fruit were added with 300mL of sterile distilled water and then blended with a sterile blender into homogeneous solution.

Filtration

The filtration was carried out by using muslin cloth. The filtration removes skin and other impurities from fruit juice. Filtration was repeated once. About 800mL juice was obtained. The 12% sugar adjusted with sucrose using hand refractometer. This was used as must for fermentation.

Sterilization of Must

To remove the bacterial contaminants, the must was heated at 68°C for 25 min. After this, must was cooled at freezing temperature. Before starting of the fermentation pH of must was adjusted at 6.5.

Production of Wine: Three sets each with 250mL of must were added with 25mL of inocula in each flask. The flasks were kept for incubation at 25°C for 7 days.

Determination of pH and Alcohol Content

1) Determination of pH: It was determined by taking small amount crude wine into a beaker and the pH was checked with hand pH meter.

2) Determination of Alcohol Content: It was determined by specific gravity method using the distillate. The aroma and flavor of wine was tasted organoleptically.

Distillation: After the fermentation distillation was carried out. The 100mL of fermented musts from three fermentation flasks were taken in clean distillation flasks, which were attached to plane condenser having water inlet and outlet and then distillation flasks were cork sealed using plaster of Paris and the distillation flasks were subjected to distillation with the help heating mantle. Distillation was allowed for 30 minutes or until about 50ml distilled was collected and subjected to determine of alcohol content by specific gravity method as described below.

Specific Gravity Method for Alcohol Estimation: Specific gravity bottle was cleaned with distilled water, and was allowed drying completely. After drying weight of empty specific gravity bottle was taken and noted down as W_1 . Then specific gravity bottle was filled with distilled water and stopper was placed and then weight of specific gravity bottle with water was measured on electric weighing balance and noted down W_2 . Then distilled water from specific gravity bottle was removed. Then specific gravity bottle was rinsed with some amount of distillate and filled with same and again weight of specific gravity bottle was measured and noted down as W_3 .

After noting the values W_1 , W_2 and W_3 the calculation for specific gravity of distillate was done as below::

$$A = (W_2 - W_1) / (W_3 - W_1)$$

Where,

A = Value of specific gravity which was taken and used for determination of alcohol % from table

W_1 = weight of empty specific gravity bottle

W_2 = weight of specific gravity bottle with distilled water

W_3 = Weight of specific gravity bottle with distillate

After calculations of A value, the percentage of alcohol was determined from the Table showing percentage by volume at 30°C (82.4°F) of alcohol corresponding to apparent specific gravity at various temperatures [7].

RESULTS AND DISCUSSION

Isolation of Yeast

The yeast was isolated from Guava fruit solid medium YEPD agar medium. The isolate was subjected to characterization. The result is presented in the Table-1. The yeast was from fruit.

Physico-Chemical Parameters of Wine

The average physicochemical properties of three sets indicate (Table-2) that the pH of wine was acidic (5.0), with pinkish white colour and 4% was residual sugar. The aroma and flavor of wine was fruity and pleasant. Thus the quality of wine was desirable [1, 9].

The result of determination of sugar concentration in the must after fermentation was less because sugar was converted into alcohol.

Calculation:

$$\begin{aligned} \text{\% of alcohol} &= (W_2 - W_1) / (W_3 - W_1) \\ &= 47.61 - 26.70 / 50.95 - 26.70 \\ &= 20.91 / 24.25 \\ \text{\% of alcohol} &= 0.8622 \end{aligned}$$

As per standard table used for determination of % of alcohol by volume at 30°C is 7.43%. Results of characterization of wine produced from guava pulp are present in table 4.4.

Table 1 Results of Morphological Characteristics and Identity of The Isolate.

Tentative Identity	Size	Shape	Colour	Margin	Elevation	Opacity	Consistency	Gram nature
<i>Saccharomyces cerevisiae</i>	1mm	Circular	Yellow	Regular	Flat	Smooth	Opaque	Gram positive budding yeast cells

Table 2 Results of Physico-Chemical Parameters of Wine

Sr. No.	Parameters	Average result of three sets
1	pH	5.0
2	Colour	pinkish white
3	Sugar content of wine	4 %
4	Aroma and flavor	Pleasant fruity
5	Alcohol content %	5.43

Table 3: Results of Determination of Alcohol Content

W₁	Weight of empty specific gravity bottle	26.70 gm
W₂	Weight of empty specific gravity bottle with distilled water	47.61 gm
W₃	Weight of specific gravity bottle with water	50.95 gm

CONCLUSION

Wine is one of the functional fermented foods having many health benefits like anti-ageing effects, improvement of lung function (from antioxidants in white wine), reduction in coronary heart disease, development of healthier blood vessel. Many wines are made from fruits having medicinal value. The production of Guava wine in India can be found to be of great benefit.

Samples of Guava fruit were collected from the local vegetable market, Karad and were subjected to enrichment of *Saccharomyces cerevisiae* by enrichment culture technique. Wine was prepared from Guava fruit juice by using yeast *Saccharomyces cerevisiae*. The fermentation process was optimized by adjusting sugar level and pH. It was seen that wine prepared from juices having sugar level rich (4%) and pH 5. The maximum alcohol yield in Guava fermented juice was 5.43%. The results indicated that *Saccharomyces cerevisiae* isolate was suitable for Guava wine production.

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