

Optimizing Wine Production: A Comparative Study of Yeast Fermentation in Seasonal Fruit Juices

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1 Introduction

Fruit juices are naturally enriched with a diverse range of bioactive compounds, including essential vitamins, antioxidants, organic acids, minerals, and varying levels of fermentable sugars. This biochemical richness not only contributes to their nutritional value but also makes fruit juices ideal substrate media for microbial fermentation processes. In particular, yeast-based fermentations commonly employed in wine making and ethanol production rely heavily on such nutrient-dense substrates to support microbial growth, enzymatic activity, and efficient metabolic conversion. The complex composition of fruit juices can also enhance the organoleptic properties, aroma profiles, and functional characteristics of the resulting fermented beverages, adding further value to their utilization in fermentation industries.

Despite their potential, the tropical fruit sector faces significant challenges related to seasonality and perishability. Many fruits have short post-harvest shelf lives due to high moisture content and the absence of advanced storage, preservation, and transportation facilities in many developing regions. As a result, a substantial portion of harvested fruits becomes unsuitable for fresh consumption or commercial sale. This leads to considerable post-harvest losses, which not only reduce farmer profitability but also contribute to resource wastage and food insecurity. One practical and sustainable approach to mitigating these losses is the conversion of excess or surplus fruits into fermented products. Fruit wines and ethanol represent value-added commodities that can be produced economically from such raw materials while providing an avenue for waste reduction and rural-level industrial development.

According to [1], fermentation is a fundamental anaerobic metabolic process in which yeast converts carbohydrates, primarily sugars, into ethanol and carbon dioxide through the catalytic action of the enzyme complex **zymase**. In this process, complex carbohydrates or starches present in fruit substrates are hydrolysed by yeast enzymes into fermentable monosaccharides such as glucose and fructose, and

disaccharides like sucrose. These simple sugars undergo glycolysis via the **Embden–Meyerhof–Parnas (EMP) pathway**, yielding pyruvic acid as an intermediate metabolite. The pyruvate is then enzymatically decarboxylated to form acetaldehyde, which is subsequently reduced to ethanol through a dehydrogenation reaction. The efficiency of ethanol formation is strongly influenced by key parameters such as **temperature, incubation time, pH, and substrate composition**, which collectively determine yeast metabolic activity and overall fermentation yield.

The present study investigates the fermentation potential of *Saccharomyces cerevisiae* in six locally available fruit juices: king coconut, Kurumba (tender coconut), papaya, star fruit, rambutan, and tomato. These fruits were selected based on their availability throughout Sri Lanka, diverse sugar compositions, and their varying nutritional profiles, representing a broad range of tropical substrates with practical relevance for small-scale and artisanal fermentation. The objective of this work was to evaluate and compare the fermentability, yeast growth behavior, and ethanol yield across these different fruit juices, ultimately identifying which substrates are most promising for the development of locally sourced fruit-based wines or ethanol products. Additionally, by exploring the use of these fruits for fermentation, the study aims to highlight their potential role in reducing post-harvest losses and promoting value-added processing within local agricultural systems.

2 Objectives

The main objective of this study was to compare the ethanol yield from different fruit juices fermented by the microorganism *Saccharomyces cerevisiae*. Specific aims included:

- Evaluating the sugar consumption of each juice sample upon *Saccharomyces cerevisiae* (yeast) cell growth.
- Monitoring the amount of ethanol produced per 1 L of fruit juice sample.
- Determining the most suitable fruit substrate for wine and ethanol production.

3 Experiments

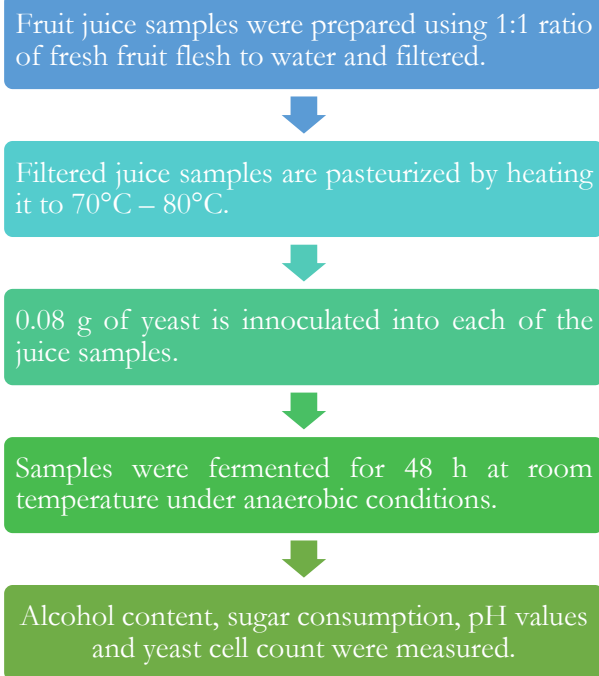
Initially fresh juice samples were prepared using a 1:1 ratio of fresh fruit flesh to water and filtered to remove coarse particles. Each juice sample is pasteurized by heating it to 70 °C – 80 °C in a stainless-steel cooking pot to prevent any microbial contamination. From each juice sample, four sets of 100 ml samples were prepared by adding 11 g, 22 g, and 33 g of glucose and without adding any glucose to obtain 0%, 10%, 20% and 30% glucose containing samples respectively. Then into each of the samples 0.08 g of activated yeast was inoculated under sterile conditions to prevent any unwanted microbial contamination in the fruit juice samples. Then the samples were kept for fermentation for 48 h at room temperature (~ 30 °C) ensuring the anaerobic environment to encourage anaerobic respiration in the inoculated yeast cells. pH values, glucose concentration, and yeast cell count were measured by using a pH meter, a refractometer (HINOK WZS 32), and a spectrophotometer respectively at the moment of yeast inoculation and after 48 h. The yeast cell concentration was estimated indirectly by measuring the turbidity of the fermentation samples using a spectrophotometer at a wavelength of 600 nm, where the absorbance values corresponded to the cell density in each fruit juice medium. Alcohol content was calculated by performing iodoform test to the each of the fruit juice samples after 48 hours of fermentation [2].

4.1 Materials

Pasteurized fruit juices king coconut water, Kurumba water, papaya, star fruit, rambutan, and tomato were used as fermentation substrates. Commercially available dry yeast (*Saccharomyces cerevisiae*) served as the fermenting microorganism. Analytical-grade glucose was added to selected samples to enhance the fermentable sugar content in the prepared fruit juice samples. Concentrated iodine solution and 1 M sodium hydroxide (NaOH) were used for qualitative determination of alcohol content in the fermented juice samples via iodoform test. Sterilized tap water was used for dilution where required. All glassware, including conical and flat-bottom flasks, beakers, and pipettes, were sterilized prior to use. Ethanol or isopropyl alcohol (IPA) was used for disinfection. Fermentation was carried out in flat-bottom flasks equipped with stoppers and gas-release tubes to maintain anaerobic conditions (Figure 4) for the fermentation process. A refractometer was used to determine initial and final sugar concentrations,

spectrophotometer was used to obtain the yeast cell count on Day 0 and Day 2, capped bottles were employed for temporary storage of fermented samples, and a pH meter was used to check the pH values of the fruit juice samples on Day 0 and Day 2.

4.2 Methods



5 Results and Discussion

All fruit juices supported yeast cell growth and ethanol production via fermentation, though with varying substrate consumption upon cell growth. Pasteurization allows to preserve heat labile compounds that could add both nutrient and flavour to final product. Figure 1 shows that the 30% glucose-added Kurumba water juice sample has resulted in the highest ethanol amount per 1 L of juice sample, while the no sugar-added star fruit juice sample has resulted lowest alcohol amount per 1 L of juice sample. However, Figure 1 indicates that by adding additional amount of glucose to the media, amount of alcohol produced per 1 L of fruit juice sample can be increased. All in all, Figure 1 shows the amount of alcohol produced in the 1 L of fermented juice sample which was calculated by performing the iodoform test for each of the juice samples.

Figure 2 shows the yeast cell growth after 48 h of fermentation at room temperature under given anaerobic conditions. According to Figure 2, 20% glucose-added tomato juice sample and the no glucose-added rambutan juice sample resulted the highest yeast cell growth (6.704×10^7 cells/ml), while the no glucose-added

kurumba juice sample resulted in the lowest yeast cell growth. Yeast cell growth can be determined by obtaining the cell count at 0 h of yeast inoculation to the media (Day 0) and 48 h of yeast inoculation to the media (Day 2). Total yeast cell growth can be calculated as indicated in Eq. (1) where $N_{Day 0}$ indicates the number of yeast cells on Day 0, $N_{Day 2}$ indicates the number of yeast cells on Day 2 and N_{Growth} indicates the total yeast cell growth after 48 h.

$$N_{Day 2} - N_{Day 0} = N_{Growth} \quad \text{Eq. (1)}$$

Figure 3 shows the glucose consumption in each of the juice samples upon yeast cell growth where glucose is provided to the media as a substrate. Moreover, Figure 3 illustrates that papaya juice has resulted the highest substrate (glucose) consumption as the substrate in the 20% glucose-added juice sample upon yeast cell growth in the media, while no glucose-added tomato juice sample resulted the lowest substrate consumption upon yeast cell growth. Glucose consumption of each of the fruit juice samples were calculated by after obtaining the brix values of all the fruit juice samples at Day 0 and Day 2, according to Eq. (2) where $S_{Day 0}$ indicates the glucose concentration in the samples on Day 0, $S_{Day 2}$ indicates the glucose concentration in the samples on Day 2 and $S_{Consumption}$ indicates the total glucose consumption upon yeast cell growth after 48 h.

$$S_{Day 0} - S_{Day 2} = S_{Consumption} \quad \text{Eq. (2)}$$

Figure 4 shows that highest ethanol yield is obtained from 20% glucose added Kurumba water while lowest ethanol yield has resulted from no glucose added rambutan juice sample. Ethanol yield can be calculated according to Eq. (3) where $Y_{P/S}$ is ethanol yield, P is the amount of ethanol produced (g/L), and S is the amount of glucose consumed (g/L) during the entire fermentation process.

$$Y_{P/S} = \frac{P}{S} \quad \text{Eq. (3)}$$

The pH values measured on Day 0 and Day 02 are presented in Table 01. On the initial day pH value remains at a same range for a specific fruit juice sample even the glucose concentrations are different. A decrease in pH was observed across all fruit juice samples after 48 hours of fermentation, indicating increased acidity in the medium. This reduction in pH is consistent with metabolic by-products generated during yeast fermentation, including organic acids and dissolved carbon dioxide, accompanying ethanol production.

Table 2. pH variation in fruit juice samples after 48 h of fermentation

Fruit Juice	Glucose Concentration	pH Value	
		Day 01	Day 02
King Coconut	0%	4.27	3.61
	10%	4.23	3.54
	20%	4.22	3.27
	30%	4.20	3.29
Papaya	0%	4.88	4.15
	10%	4.81	3.67
	20%	4.79	3.55
	30%	4.78	3.55
Kurumba	0%	4.63	3.97
	10%	4.67	3.86
	20%	4.64	3.82
	30%	4.59	3.77
Star Fruit	0%	3.46	3.16
	10%	3.47	3.03
	20%	3.46	2.98
	30%	3.40	2.95
Rambutan	0%	3.85	3.33
	10%	3.91	3.17
	20%	3.85	3.11
	30%	3.85	3.27
Tomato	0%	4.04	3.81
	10%	4.00	3.44
	20%	3.99	3.32
	30%	3.97	3.21

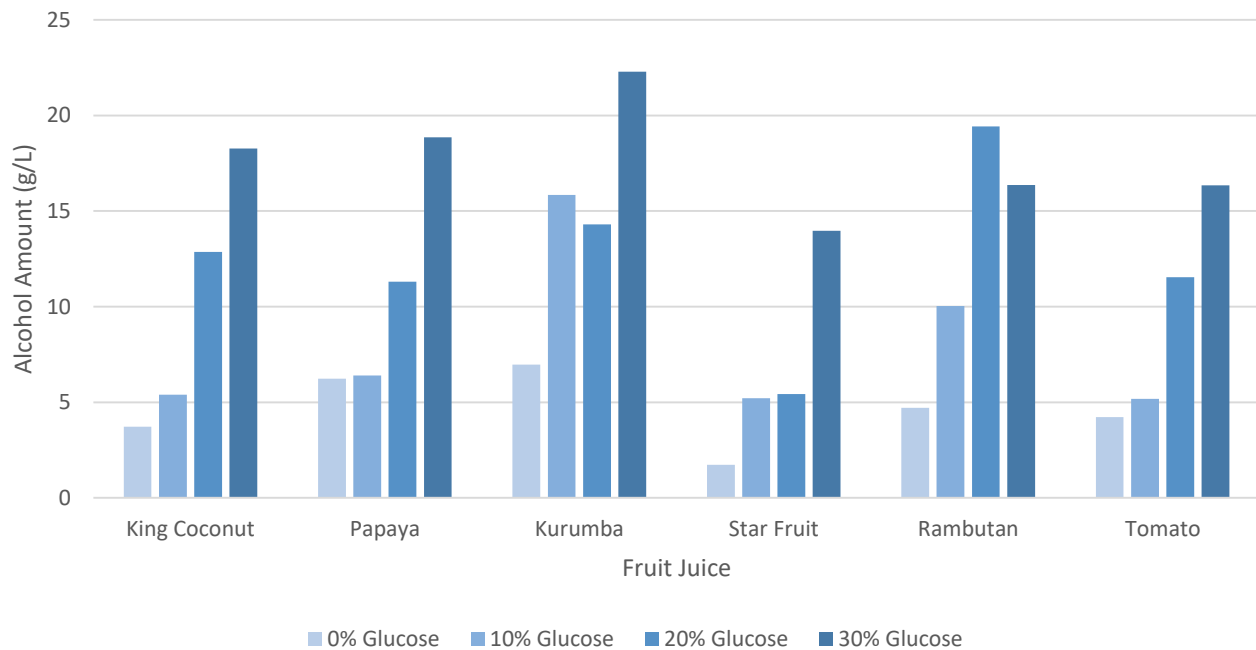


Figure 1. Alcohol amount per 1 L of fruit juice sample obtained after 48 hours of fermentation.

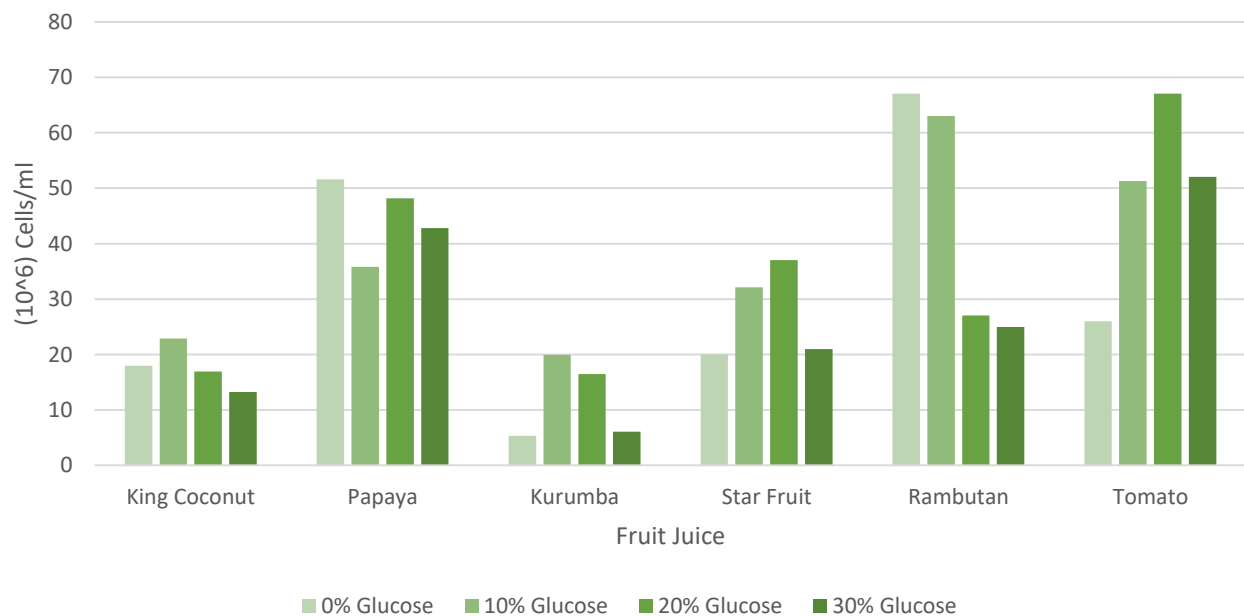


Figure 2. Yeast cell growth amount after 48 hours of fermentation.

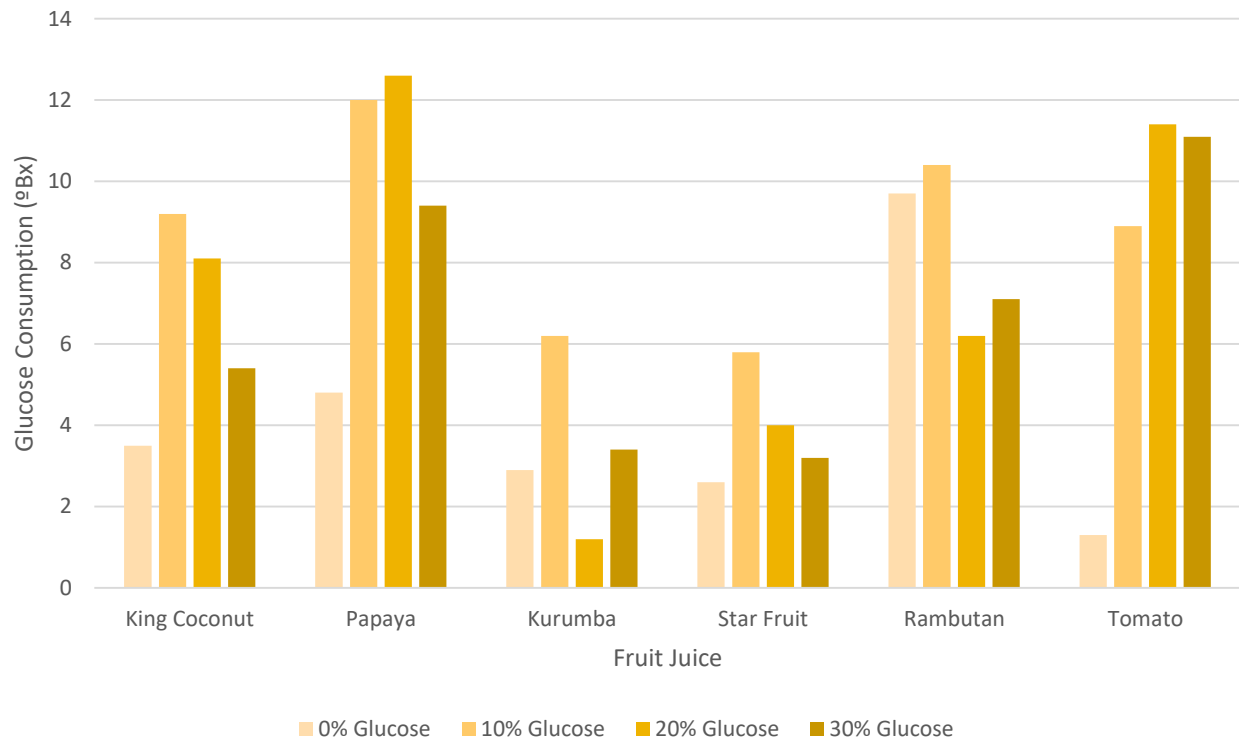


Figure 3. Glucose consumption of each fruit juice samples after 48 hours of fermentation.

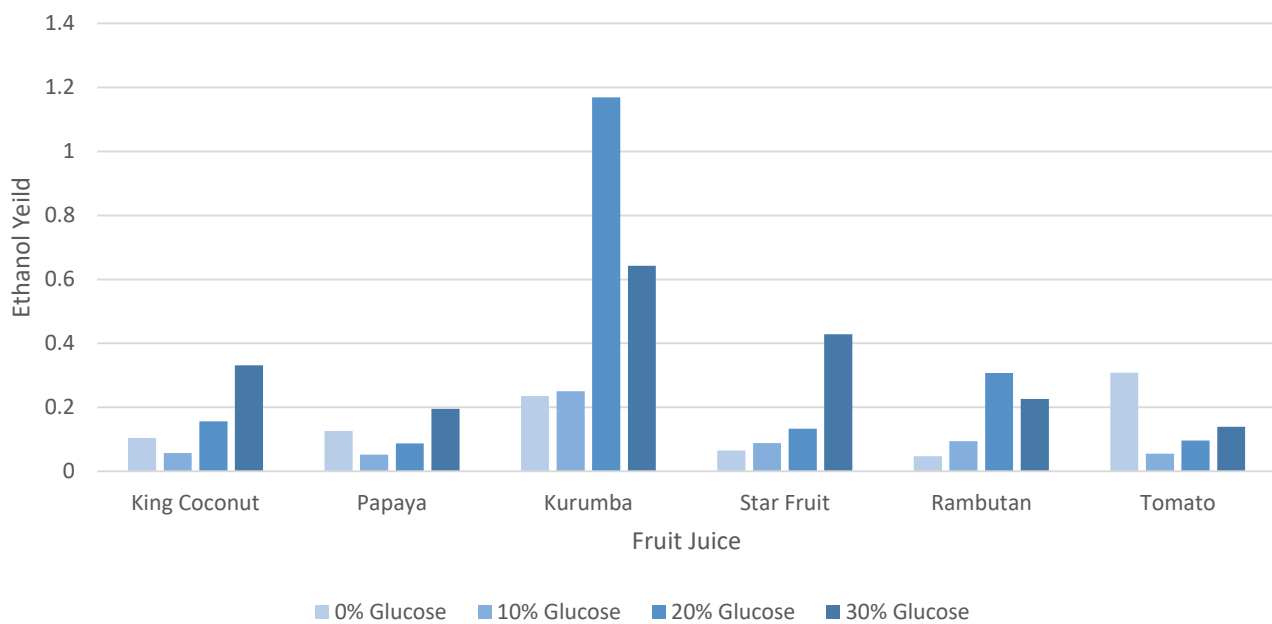


Figure 4. Ethanol yield of each fruit juice samples with varying glucose concentration.



Figure 5. Fruit juice samples fermentation experimental set up.



Figure 6. Obtained precipitations after conducting the iodoform test.

6 Conclusions

The present study demonstrated that all six fruit juices (king coconut, papaya, Kurumba, rambutan, star fruit, and tomato) serve as viable substrates for ethanol fermentation by *Saccharomyces cerevisiae*. However, their ethanol yields, and fermentation behaviours varied considerably depending on the biochemical composition of the fruit matrix and the level of glucose enrichment. Among the tested substrates, **Kurumba (tender coconut water) supplemented with 30% glucose** exhibited the **highest ethanol yield**, indicating that its balanced nutrient composition and suitable pH provided favourable conditions for yeast growth and metabolic activity.

Beyond the fermentation efficiency, this study highlights the potential of using surplus or seasonally abundant fruits as fermentation substrates to **minimize post-harvest losses** and promote **value addition** through the production of fruit-based ethanol or wine. The

findings suggest that locally available fruits, when optimized for sugar content and fermentation parameters, can contribute to sustainable small-scale wine production while addressing issues of food waste management and agricultural sustainability.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this study.

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Credit Authorship Statement

Tashini Inoli; Conceptualization, Data analysis, Visualization, Writing – original draft. **Kalindu S. Fernando**; Supervision, Conceptualization, Data analysis, Writing – review & editing.

Keywords

Wine fermentation, Fruit juice, Ethanol, *Saccharomyces cerevisiae*

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