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RESEARCH NOTE

Process Optimization of Low Alcoholic Beverage from Guava Using Different Yeast and Temperature Combination of Fermentation

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ABSTRACT

The aims of this study was to determine the effects of temperature and yeast count on the fermentation kinetics and chemical properties of guava fruits were determined for its suitability to produce low alcoholic beverage. The fermentation conditions were optimized by varying temperature at 22,27 and 32°C and the yeast concentration of 1,2 and 3%. The increase in temperature and yeast count, sugar level was deceased. However, at low temperature (22°C±2°C) and yeast concentration (1%) the sugars were not completely utilized during fermentation At high temperature of 32°C ±2°C, the alcohol yield was highest (6.02%) relative to the low temperature of 22°C±2°C that gave the lowest yield (2.60%), the yeast count of 3% and fermentation temperature of 32°C±2°C gave the optimal characteristics for guava low alcoholic beverage.

Keywords: Guava low alcoholic beverage, yeast count, fermentation temperature

Guava (*Psidium guajava* L.) is one of the most important commercial fruit crops in (India) consumed locally. It is a good source of ascorbic acid, pectin, sugars and certain minerals, its skin and flesh colours vary from variety to variety depending on the amount and type of pigments. Guava is often marketed as "super-fruits" which has a considerable nutritional importance in terms of vitamins A and C with seeds that are rich in omega-3, omega-6, poly unsaturated fatty acids and especially dietary fiber, riboflavin, as well as in proteins and mineral salts. The high content of vitamins-C (ascorbic acid) in guava makes it a powerhouse in combating free radicals and oxidation that are key enemies that couse many degenerative diseases. The anti-oxidant virtue in guavas is believed to help reduce the risk of cancers of the stomach, oesophagus, larynx, oral cavity and pancreas. The fibres in guavas promote digestion and ease bowel movements. The high content of vitamin A in guava plays an important role in maintaining the quality and health of eye sight, skin, teeth, bones and the mucus membranes (Kocher and Pooja, 2011).

With the changing consumer attitudes, demands and emergence of new market products, it has become imperative for the producers to develop products, which have nutritional as well as health benefits. In this context, guava has excellent digestive and

nutritive value, pleasant flavour, high palatability and availability in abundance at moderate price. The fresh fruit has limited shelf-life therefore it is necessary to utilize the fruit for making different products to increase its availability over an extended Period to stabilize the price during the glut season. Guava, with its widely appreciated flavour and aroma, is able to complete in the market, either as guava juice or as mixtures with other juices or fermented guava beverage (Nguyen Phouc Minh, 2014).

Guava (*Psidium guajava* L.) alcoholic beverages is the product of anaerobic fermentation by yeast in which the sugars are converted into alcohol and carbon dioxide. Ethanol production from guava pulp is reported (Kocher and Pooja, 2011; Sevda 2011; Kaiser Younis, 2014; Nguyen Phuocminh, 2014) still there is no study found in literature for guava must fermentation for guava fruit low alcoholic beverage production. Therefore, the aim of our research is to optimization of the fermentation parameter for production of low alcoholic beverage from guava.

MATERIALS AND METHODS

Raw material: Guava fruits were purchased from local market.

Chemicals: Chemicals of analytical grade like Indigo carmine, 2,6.-Dichlorophenol Indophenol, sodium hydroxide (NaOH), copper sulphate, alkali, Rochelle salt, starch solution, di-ammonium phosphate were used.

Preparation of low alcoholic beverage: Healthy, ripened and even sized guava fruits (Sadabahar variety white guava, procured from Daulatabad, Aurangabad, Maharashtra, India were selected and their calyx portions were removed. The initial analysis of various parameters like length and diameter, of fruit, weight of fruit, initial brix, final volume of juice. After washing, the fruit were cut into small pieces, dipped in water (1:1, w/v) and heated to just boiling. The softened fruit pieces were crushed and the "pulp" so obtained was filtered through a muslin cloth and juice obtained was used for further study. The details of treatments used are shown in Table 1. the details of low alcoholic beverage is depicted in Fig. 1. The fermentation was carried out for 10 days. During the fermentation I was monitored the TSS, pH, acidity and ethanol percent.

Analytical Methods

Chemical Analysis

The physico-chemical analysis of guava pulp included the estimation of TSS (using hand refractometer) (Ranganna, 1979), pH (laboratory pH meter) (Ranganna S. 1979) and Titrable acidity (Ranganna S. 1979), reducing sugars (Ranganna 1979), moisture and protein (A.O.A.C. 1990), tannins (Ranganna 1986), ascorbic acid (Ranganna 1979), specific gravity (BIS, 1989) and ethanol content (BIS, 1989).

Sensory Analysis

The sensory evaluation of low alcoholic beverage was conducted by a panel of 10 semi-trained judges. The evaluated parameters consisted of colour, appearance, flavour, taste, Mouthfeel and over all acceptability.

Statistical analyses

Microsoft excel 2007 was used for statistical analysis

Table 1: Samples code and combination of yeast percent and fermentation temperature

Parameter	Treatment								
	T_1^A	T_2^A	T_3^A	T_1^B	T_2^B	T_3^B	T_1^C	T_2^C	T_3^C
Yeast	1%	2%	3%	1%	2%	3%	1%	2%	3%
Temperature	22°C±2°C			27°C±2°C			32°C±2°C		
Total soluble solid	15°Bx			15°Bx			15°Bx		

at 95% confidence level or ANOVA used for sensory evaluation. Analyses were done in triplicate and data of sensory assessed using single way analysis of variance. Significance was determined at $P<0.05$ and the values have been displayed with standard deviation of the means.

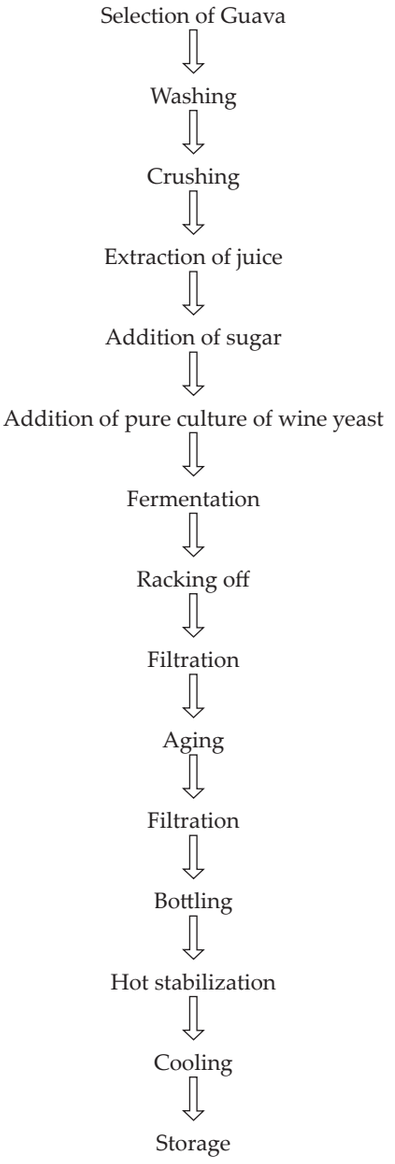

Fig. 1: Preparation of low alcoholic beverage

Flow sheet 1. Flow chart for the production of low alcoholic beverage from guava.

RESULTS AND DISCUSSION

Physico-chemical characteristics of guava pulp

The results was revealed that the moisture content of fruits are 83.67 per cent on fresh weight basis. The total sugar content in the guava fruit were found as 10.52%. The ascorbic acid content of guava fruit was is found (230.33mg/100g). While the protein content of fruits were in the range of 3.55 to 5.63 per cent. The reducing sugar, pH and per cent acidity or tannin percent in fruit pulp were 5.58, 4.0, 0.39 and 0.16 respectively.

Changes occurring during the alcoholic fermentation

The TSS was the highest during the first 2 days of fermentation (Fig. 2). This may be due to the higher concentration of substrate available for the activity of yeast during the initial period of fermentation during which maximum amount of sugars present in the must was utilized to produce alcohol and CO_2. TSS was monitored till the constant reading with refractometer was achieved. Sample T_2^B, T_3^B, T_2^C and T_3^C had the least TSS (4.0°Bx) which indicated that it was best fermented as compared to the other five samples.

Effect of fermentation on pH

Variations in pH was observed during the period of fermentation. Among all the nine samples, the sample containing 100% guava extract showed a decrease in pH from 4.5 to 3.6, whereas the sample T_3^C showed a decrease in pH from initial value of 4.2 to 3.6 mentioned in (Fig. 3).

During the fermentation process there was a decrease pH in all low alcoholic beverage samples and may be due to metabolic activity of microorganisms. Lower pH indicates long stability of low alcoholic beverage. Most stable low alcoholic beverage have been reported to have a pH in the range 2.5 – 4.0 (Zhang *et al*., 2006).

Effect of alcoholic fermentation on titrable acidity of guava juice

The titrable acidity was found to increase as the

Table 2: Composition of low alcoholic beverage after fermentation

Constituents	T_1^A	T_2^A	T_3^A	T_1^B	T_2^B	T_3^B	T_1^C	T_2^C	T_3^C
TSS (°B)	8.0 ± 0.00	4.0 ± 0.00	5.03 ± 0.06	4.50 ± 0.00	4.00 ± 0.00	4.00 ± 0.00	4.17 ± 0.29	4.00 ± 0.0	4.00 ± 0.00
pH	4.10 ± 0.02	4.23 ± 0.06	4.20 ± 0.00	4.33 ± 0.06	3.90 ± 0.01	4.13 ± 0.02	3.87 ± 0.15	3.60 ± 0.01	3.57 ± 0.04
Titrable acidity (%)	0.43 ± 0.01	0.42 ± 0.01	0.40 ± 0.01	0.41 ± 0.00	0.44 ± 0.01	0.46 ± 0.01	0.46 ± 0.01	0.45 ± 0.02	0.50 ± 0.03
Specific gravity	0.9962	0.9942	0.9934	0.9948	0.9942	0.9928	0.9942	0.9923	0.9915
Tannin (%)	0.07 ± 0.01	0.09 ± 0.01	0.10 ± 0.01	0.08 ± 0.00	0.06 ± 0.04	0.08 ± 0.01	0.09 ± 0.02	0.10 ± 0.01	0.08 ± 0.01
Ascorbic acid (mg/100g)	140.33 ± 0.58	142.33 ± 0.58	139.67 ± 0.58	132.66 ± 1.53	134.33 ± 0.58	132.00 ± 0.00	121.66 ± 1.53	120.67 ± 0.58	120.33 ± 0.58
Ethanol (%)	2.60 ± 0.00	4.00 ± 0.00	4.60 ± 0.00	3.60 ± 0.00	4.00 ± 0.00	5.00 ± 0.00	4.00 ± 0.00	5.50 ± 0.00	6.02 ± 0.03
Reducing sugar (%)	0.82 ± 0.00	0.75 ± 0.00	0.72 ± 0.00	0.78 ± 0.00	0.73 ± 0.00	0.68 ± 0.01	0.72 ±0.00	0.67 ± 0.00	0.62 ± 0.00

TSS= Total Soluble Solid, TA= Titrable Acidity (%), SG= Specific Gravity, Ascorbic acid mg/100gm RS= Reducing Sugar %.

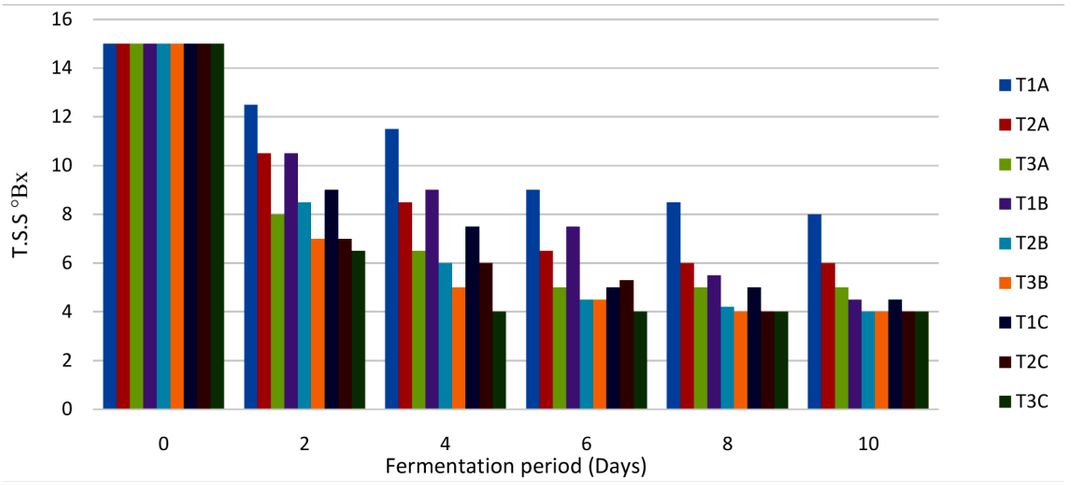

Fig. 2: Changes in TSS (°Bx) during fermentation

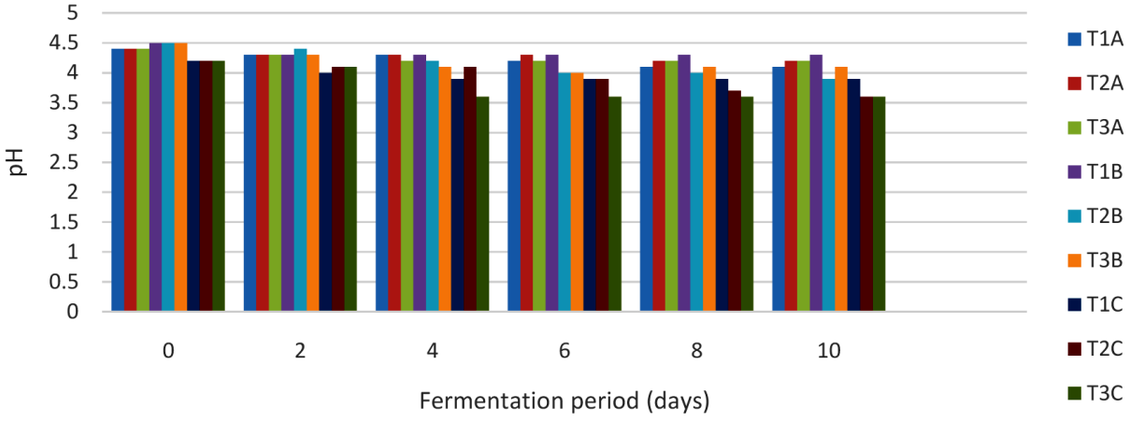

Fig. 3: Changes in pH during fermentation

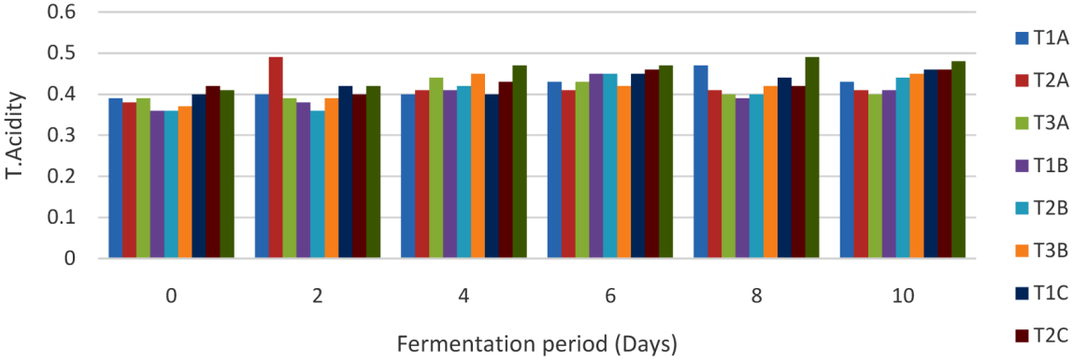

Fig. 4: Changes of titrable acidity during fermentation

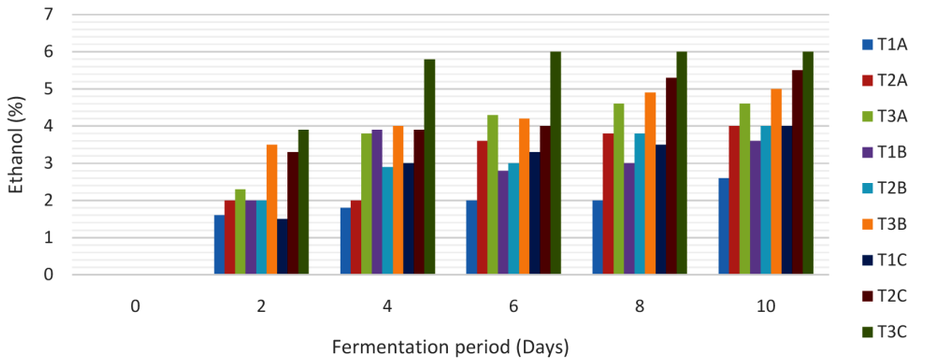

Fig. 5: Changes in Ethanol during fermentation

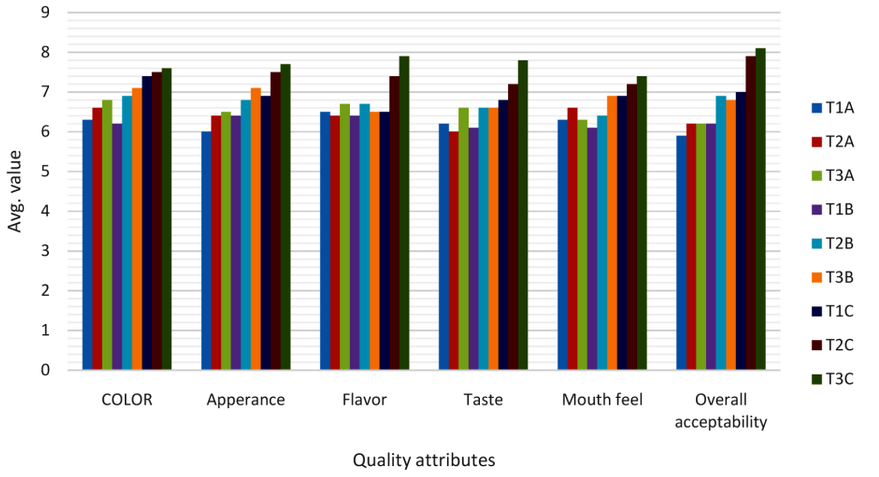

Fig 6: Organoleptic quality of low alcoholic beverage

pH decreases showing that more organic acids were produced as substrates were utilized by the microorganisms (Fig 4). The titrable acidity of alcoholic beverages prepared from fruit is an important characteristics varying between 0.39 to 0.48 percent. During the fermentation process there was an increase in acidity. Similar values are reported by the (Parram *et al*., 2010, Pooja *et al*., 2015 and Tanwar *et al*., 2014).

Ethanol concentration during the fermentation

The data presented in above Fig. 5 show that alcohol content of sample was higher in sample T_3^C as compared to other sample. Alcohol content of T_3^C sample at the first day was nil and as fermentation went on the alcohol content was found increased.

The variations in the alcohol content may be due to utilization of sugar by yeast during fermentation process. Pooja *et al*. (2015) reported that ethanol content of wine after fermentation of 10th days the similar to low alcoholic beverage sample.

Composition of low alcoholic beverage after fermentation

All the nine sample of low alcoholic beverage where subjected to chemical analysis after the fermentation of 10th days At different temperature i.e. 22°C±2°C, 27°C±2°C and 32°C±2°C by using baker's yeast

saccharomyces cerevisiae. The composition of low alcoholic beverage after fermentation is shown in Table 2.

The results show that alcohol content of sample T_3^C was higher than other samples of low alcoholic beverage. The variation in alcohol content may be due to utilization of sugar by yeast during the fermentation process. After fermentation of 10th days, the sugar content of sample T_1^A to T_3^C were decreased.

The TA of different low alcoholic beverage sample was increased after the fermentation of 10th days. The sample T_3^C it had the highest TA. The increase in TA was concomitant with a decrease in pH. The reducing sugar content decreased after the fermentation. In sample T_3^C the reducing sugar were more decreased as compared to other 8 samples of low alcoholic beverage.

The specific gravity was found to be least (0.9915) in sample T_3^C and it was found to be the highest (0.9962) in sample T_1^A. Sugar is converted into alcohol and alcohol content had lower activity specific thus, lowered the gravity of low alcoholic beverage. The tannin and ascorbic acid of low alcoholic beverage sample is decreased after the fermentation.

Sensory Evaluation

After completion of fermentation for the period of 10th days, the sensory properties of prepared low alcoholic beverage was evaluated and the average values are presented in Fig. 6.

Colour is an important parameter for consumer acceptability. The score for colour ranged from 6.3 to 7.6. The low alcoholic beverage prepared from guava by using treatment T_3^C obtained the highest score (7.6) while treatment T_1^B was having minimum score of 6.2. The colour of low alcoholic beverage was white. The score obtained for appearance ranged from 6.0 to 7.7. It was observed that colour and appearance changes with treatment (Table 1). The low alcoholic beverage prepared using treatment T_2^C obtained 7.5 score compared to other treatments. The score for taste ranged from 6.0 to 7.8. There was a significant difference in the score among the treatment under the investigation. Taste was acceptable for the treatment T_3^C after found decrease. The low alcoholic beverage prepared using T_3^C(7.8) the highest score whereas treatment T_2^A obtained the lowest score (6.0).

Score for flavour was found to be decreasing as levels of temp and yeast decreased. The score for flavour ranged from 6.4 to 7.9. The score for mouthfeel ranged from 6.1 to 7.4. The score obtained for treatment T_3^C (7.4) was more than compared to other treatment. The overall acceptability of the prepared low alcoholic beverage by T_3^C had greater acceptability followed the other low alcoholic beverage.

CONCLUSION

It may be concluded from the present study that the increase in fermentation temperature and yeast count, sugar could be completely utilized or fermentation yielding more alcohol content. Fermentation temperature of 32°C±2°C and yeast count of 3% gave the optimal characteristics for the production of low alcoholic beverage from guava.

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