Indian J. Hort. 71(4), December 2014: 554-559

Evaluation of local guava varieties for quality wine production

Pooja, G.S. Kocher*, M.I.S. Gill and N.K. Arora****

Department of Microbiology, Punjab Agricultural University, Ludhiana 141004

ABSTRACT

Guava (*Psidium guajava* L.) varieties (Punjab Pink, Arka Amulya and Lucknow-49) were evaluated for wine production. Pre-fermentation treatment using pectinase (3.5 units/ mg) was performed under optimized conditions to enhance the juice yield and clarification. The effect of different fermentation parameters, *viz.* sugar, temperature, inoculum size and DAHP supplementation revealed 25°Brix, 25°C, 9% v/v and 300 mg/100 ml as optimum levels in all the three varieties, with an ethanol production of 13.8, 13.6 and 13.6% v/v in 6, 8 and 8 days for Punjab Pink, Arka Amulya and Lucknow-49, respectively. Post fermentative storage of wine (at 15°C) for 90 days freed the wine off viable yeast cells and led to reduction in ascorbic acid, total phenolics content along with percent decrease in ethanol levels. The prepared wine was subjected to sensory analysis (at 15 and 90 days of storage). The wines prepared from Punjab Pink and Arka Amulya varieties were found to be of standard quality. After storage of 90 days, wine from Punjab Pink scored a superior quality score (68.8 ± 3.27), whereas wines from Arka Amulya and Lucknow-49 scored the same, *i.e.* of standard and below standard quality (54.2 ± 3.11 and 47.2 ± 2.38, respectively).

Key words: Fermentation, guava, optimization, *Saccharomyces cerevisiae*, wine.

INTRODUCTION

Guava (*Psidium guajava* L.) (Family Myrtaceae) known as 'Apple of Tropics' is one of the exotic fruits prized for its very pleasant, sub-acid and aromatic nature. India ranks fourth in its production after mango, banana and citrus (Shankar *et al*., 14). In Punjab, it ranks second after Kinnow with an annual production of 1.5 lakh MT. Guava is rich in vitamin A (200-400 IU), ascorbic acid (88.2-250.8 mg/100 g), lycopene (45.3 µg/ g FW), total sugars (10-15.3%), reducing sugars (2.05-6.08%), acids (10-15.3%), pectins (0.62%) and phenols (170- 345 GAE/ g FW) (Kaur *et al*., 8). Besides, it is also a good source of calcium, magnesium, thiamine and niacin (Hiwale and Singh, 4). However, guava has a very low shelf-life of about two days at room temperature and thus reflects 10-15% post-harvest losses, which make it an ideal candidate for value-addition. Therefore, to utilize the produce at the time of glut and to save it from spoilage, development of low cost processing technologies for guava is much required. Though guava nectars/ juices are available in market, very little work has been carried out towards guava-wine production (Kocher and Pooja, 10).

The easy availability, comparatively low cost, high nutritive value and good sugar content of guava, together make it a suitable alternative substrate for wine production. Further, wines are health friendly alcoholic beverages, which contain antioxidant components in the form of anthocyanins, flavanoids, vitamins and minerals. The quality of wine depends upon a number of factors like cultivars and their characteristics such as adequate sugar level, acidity, colour and aroma, besides different fermentation parameters like type and size of inoculum, substrate concentration, temperature, pH etc. for obtaining optimal alcohol production. Further, this primary fermentation period of alcohol production is followed by a slow secondary fermentation that produces aromas due to esters, higher alcohols like isobutyl, isoamyl and acetaldehyde. These volatile compounds improve the aroma and bouquet of wine. It is therefore very important to evaluate the local guava varieties for both alcohol production and sensory properties so that guava-wine may be accepted commercially. Among the different varieties available in Punjab, Punjab Pink, Lucknow-49 and Arka Amulya possess good sugar levels (~10% TSS) and appropriate acidity of 0.25-0.34% (Kocher and Pooja, 10). Hence, these varieties can act as a suitable alternative substrate (to grape) for wine production.

MATERIALS AND METHODS

Three varieties of guava var. Arka Amulya, Punjab Pink and Lucknow-49 procured from Department of Fruit Science, PAU, Ludhiana and Fruit Research Sub-Station, Bahadurgarh were used in the study. Yeast culture, *Saccharomyces cerevisiae* (MTCC 11815) an own isolate used in the study was maintained

*Corresponding author's E-mail: gskocher@pau.edu

**Department of Fruit Science, Punjab Agricultural University, Ludhiana 141004

on Glucose Yeast Extract agar and stored at 4°C. Healthy ripened and even sized fruits were selected after manual sorting, washed in hot water (containing 0.1% potassium meta bisulphite), cut into pieces, which were dipped in water and boiled for 30-40 min. Thereafter, softened fruit pieces were crushed and filtered through muslin cloth. The juice so obtained was stored in flasks under refrigerated conditions (4°C) till further use. The physico-chemical analysis of guava pulp was performed included the estimation of TSS (digital refractometer, ATAGO), pH (Hanna HI96107), titrable acidity (Amerine and Roessler, 1), reducing sugars (Miller, 12), ascorbic acid (AOVC, 2) and total phenols (Malik and Singh, 11).

Pre-fermentation treatment of guava juice was performed with pectinase enzyme (3.5 units /mg; SRL, Mumbai) using the pre-optimized conditions for guava pulp enzymatic treatment (temperature-45°C, enzyme concentration- 0.50 mg/100 ml for Punjab Pink; 0.84 mg/100 ml for Arka Amulya and Lucknow-49 and treatment time of 6 h) (Pooja and Kocher, 13). The inoculum of *S. cerevisiae* (MTCC 11815) for carrying out alcoholic fermentation of pretreated guava juice was prepared in glucose yeast extract (GYE) broth (general culture media for growing yeasts) where a loopful of slant culture was inoculated in 250 ml Erlenmeyer flasks containing 100 ml GYE broth. It was incubated at 100 rpm and at 28 ± 2°C for 24 h to raise seed inoculum. From the seed inoculum, starter culture was prepared by inoculating 2% of seed inoculum in pasteurized guava juice and incubated at 28°C for 24 h under shaking (100 rpm) conditions. Pre-treated guava juice was optimized for fermentation parameters, *i.e.* Brix (15-30°B) and temperature (15-35°C) by Factorial Complete Randomized Design. For this the pre-treated guava juice (100 ml) was taken in 250 ml Erlenmeyer flasks and chaptalized with sugar solution to different brix levels (15-30°B), inoculated with (5% v/v) inoculum and incubated at different temperatures (15-35°C). While the effect of inoculum size and DAHP supplementation was studied individually by taking different combinations under the optimized brix and temperature conditions. The periodic samples from fermentation treatments were taken, spun at 6000 rpm for 5 min. and analyzed for TSS, pH and ethanol content till no further decrease in TSS was noted.

The fermentation efficiency was calculated as:

$$= \frac{\text{Actual ethanol produced}}{\text{Theoretical ethanol produced}} \times 100$$

Theoretical ethanol (%v/v) = sugar utilized × 0.64

Sugar utilized = Initial sugar - Residual sugar left after fermentation

The flasks / bottles containing prepared young wine were stored at 4°C and lees and suspended particles were allowed to settle. No settling or fining agent was added. The racking was repeated after every 15 days till there was no further settling. The cleared mature wine was stored in glass bottles (washed earlier with boiling water and cotton plugged) for upto three months (with racking every four weeks) under refrigerated conditions. The shelf-life of guava-wine (var. Arka Amulya, Lucknow-49 and Punjab Pink) stored at refrigerated temperature (4°C) was studied for 3 months**.** The refrigerated stored wine was analysed for total microbial count using plate count method on GYE medium at different periods of time. The clarified wine was subjected to sensory analysis on 80 point modified Davis's score card by ten semi-trained panellists.

RESULTS AND DISCUSSION

The parameters like TSS, titratable acidity and brix: acid ratio determine the final sensory quality attributes like appearance, colour, aroma, taste, bouquet, body, flavour, astringency and overall acceptability of the wine. The comparative physicochemical characteristics of guava pulp evaluated on the basis of chemical analysis are presented in Table 1. The data showed that parameters, *viz*., TSS, brix: acid ratio, total sugars, reducing sugars, ascorbic acid and total phenols vary significantly amongst three varieties, while pH and acidity did not vary significantly. Elsewhere, different parameters studies have revealed TSS of 9.6 to 11.0%, acidity ranging from 0.26 to 0.38% and vitamin C in the range of 167 to 210 mg 100 g^{-1} in different cultivars of guava (Jain and Neema, 6). Among the three guava varieties (Punjab Pink, Arka Amulya and Lucknow-49), though the TSS°Brix of pulp ranged between 9.0-10.2, post water supplementation, *i.e.* 'must' preparation decreased the value by more than half in all the varieties. The pulp yield was found to be 28.8, 22.3 and 19.0% in Punjab Pink, Arka Amulya and Lucknow-49, respectively. Elsewhere, Jain and Neema (6) have reported that in five different guava cultivars studied by them, the pulp yield ranged between 54.0-54.8%. Our pulp yield was less as we added water to prepare 'must' for fermentation. The pre-fermentation treatment of guava 'must' with pectinase enzyme is known to enhance the recovery of juice from fruits (Kaur *et al*., 8). In our study, pectinase treatment for three guava varieties, was performed under the optimized conditions (Pooja and Kocher, 13). The effect of total soluble solids (15-30°Brix), temperature (15-35°C), inoculum size (3-15% v/v) and DAHP supplementation (0.1-0.5%, w/v) on ethanolic fermentation of guava juice by

Table 1. Physico-chemical analysis of guava varieties.

Parameter	Variety			CD at 5%
	Arka Amulya	Lucknow-49	Punjab Pink	
TSS (°Brix)	9.2 ± 0.28	9.0 ± 0.28	10.1 ± 0.14	0.139
Acidity (%)	0.33 ± 0.03	0.30 ± 0.02	0.38 ± 0.02	NS
pH	4.3 ± 0.07	4.4 ± 0.07	4.2 ± 0.0	NS
Brix: acid ratio	28.4 ± 2.26	31.3 ± 1.13	28.0 ± 0.42	0.956
Total sugars (%)	5.36 ± 0.04	5.04 ± 0.09	6.26 ± 0.02	0.548
Reducing sugars (%)	3.68 ± 0.04	3.29 ± 0.03	3.40 ± 0.12	0.252
Ascorbic acid (mg/100 g)	195.0 ± 3.60	169.7 ± 3.46	229.6 ± 2.26	7.25
Ascorbic acid (mg/100 ml) of 'must'	84.05 ± 2.97	73.14 ± 3.05	98.97 ± 3.25	8.36
Total phenols (mg/100 g)	297 ± 2.12	262 ± 3.53	235 ± 1.41	8.14
Total phenols (mg/100 ml) of 'must'	128.0 ± 2.54	112.9 ± 3.61	101.9 ± 2.35	6.84

NS = non significant at 5% level of significance; ± = Standard deviation

S. cerevisiae was studied. The effect of sugar and temperature were studied using factorial CRD revealed that the guava juice having a Brix level of 25°Brix fermented at 25°C produced better levels of ethanol - 11.1, 11.0 and 11.0% in Punjab Pink, Arka Amulya and Lucknow-49, respectively (Table 2). The significant changes in parameters, *viz.* decreasing Brix and increasing per cent ethanol levels were observed for upto 10 days of fermentation in all varieties with no further change. Hence, in all the three varieties, the combination of 25°B and 25°C was found to be optimum for the ethanolic fermentation, which was completed in 8, 10 and 8 days of fermentation in Punjab Pink, Arka Amulya and Lucknow-49, respectively. Similarly, maximum ethanol production was recorded at the temperature range of 25-30°C by several workers. Overall, there was non-significant decrease in pH from 4.2 to 3.7, 4.3 to 3.8 and 4.2 to 3.7 in case of Punjab Pink, Arka Amulya and Lucknow-49, respectively.

The effect of inoculum size was studied individually by varying the initial inoculum level (3 to 15% v/v) of 24 h old culture of *S. cerevisiae* in the fermentation flasks (having previously optimized conditions of 25°Brix and 25°C). An inoculum size of 9% (v/v) led to maximum percent ethanol production for all the three varieties with fermentation efficiencies of 83.1, 83.8 and 83.0% in Punjab Pink, Arka Amulya and Lucknow-49, respectively (Table 3). Ethanol production was enhanced with increase in inoculum concentration upto 9% (v/v) for a fermentation period of 8 days. There was a statistically significant decrease in ethanol production beyond inoculum level of 9% (v/v). Earlier, Srivastava *et al*. (15) reported that 10% inoculum size added in guava pulp led to the production of 5.8% ethanol (w/v) by *S. cerevisiae*. A combined source of nitrogen and phosphorus, DAHP was supplemented (100-500 mg/100 ml) in the 'must' of all three varieties optimized at 25°B, 25°C and fermented by inoculum size of (9% v/v) *S. cerevisiae*. Results revealed 300 mg/100 ml as the optimum concentration of DAHP in guava juice for maximum ethanol production (in the range of 13.6-13.8% v/v). However, there were varietal as well as DAHP concentration differences with respect to ethanol production profile (Table 4). The supplementation of nitrogen and phosphorus sources in the fermenting 'must' has been found to increase yeast growth and sugar catabolic rate. Nitrogen and phosphorus supplementation also increased the sensory characteristics of wines prepared from different substrates (Ghosh *et al*., 3). It was observed that higher concentrations of DAHP, *i.e.* 400 and 500 mg/ 100 ml juice produced lower ethanol. Ghosh *et al*. (3) also found that higher DAHP concentrations produces less ethanol as compared to less concentrations as with higher concentration of nitrogen, cellular activity of yeast get inhibited and ethanol production is affected. Overall, ethanol production from guava juice was increased from 11.0% v/v (fermentation efficiency-86.3%) prior to optimization to 13.7% (fermentation efficiency- 97.8%) after the optimization of fermentation parameters, *viz.* Brix, temperature, inoculum size and DAHP supplementation.

The young guava-wines (var. Punjab Pink, Arka Amulya and Lucknow-49) so prepared under the optimized fermentation conditions of Brix (25°B), temperature (25°C), 9% (v/v) inoculum size and 0.3% DAHP supplementation was subjected to settling for 7 days at refrigerated conditions (4°C) in glass bottles. Thereafter, *i.e.* every 15 days upto 3 months, the process of racking was repeated. During this course various physio-chemical and microbiological

Table 2. Effect of Brix (°B) and temperature (°C) on ethanolic fermentation of guava juice.

Treatment*	Fermentation time (days)														
	2			4			6			8			10		
	PP	AA	L-49	PP	AA	L-49	PP	AA	L-49	PP	AA	L-49	PP	AA	L-49
15.0°B, 15.0°C	1.2	1.2	1.2	2.6	3.1	2.4	4.5	4.5	5.1	6.5	6.5	5.9	6.6	6.5	5.7
15.0°B, 20.0°C	1.7	1.7	2.0	2.3	3.9	3.9	5.1	5.1	6.5	7.0	7.1	6.6	7.2	7.1	6.5
15.0°B, 25.0°C	2.0	2.0	3.8	2.9	4.9	6.8	7.8	7.9	6.7	8.0	8.0	6.6	7.8	7.9	6.5
15.0°B, 30.0°C	1.9	1.9	4.7	2.7	5.0	6.4	7.3	7.4	6.3	7.3	7.3	6.5	7.3	7.3	6.4
15.0°B, 35.0°C	3.6	3.6	2.2	5.1	6.4	5.2	6.4	6.4	5.1	6.5	6.5	5.1	6.5	6.4	5.2
20.0°B, 15.0°C	0.8	0.8	1.4	1.4	2.1	3.1	3.2	3.2	6.0	6.3	5.6	6.6	6.3	6.1	7.8
20.0°B, 20.0°C	1.9	1.9	1.9	3.0	4.1	6.1	5.8	5.8	6.5	9.9	9.9	6.6	9.9	9.8	6.7
20.0°B, 25.0°C	3.9	3.9	2.3	6.9	8.8	6.7	10.5	10.3	9.9	10.4	10.4	9.8	10.0	10.0	9.7
20.0°B, 30.0°C	4.3	4.3	6.4	7.2	9.0	8.6	10.5	10.8	8.6	10.9	10.8	8.7	10.1	10.2	8.7
20.0°B, 35.0°C	4.6	4.6	4.9	5.3	6.5	5.7	6.5	6.5	5.7	6.6	6.5	5.8	6.5	6.4	5.8
25.0°B, 15.0°C	1.3	1.3	1.7	2.7	4.0	3.1	5.7	5.7	7.1	8.0	7.8	7.8	8.1	8.3	9.0
25.0°B, 20.0°C	1.7	1.7	2.0	2.9	3.9	4.6	5.0	5.0	7.1	9.0	8.3	8.0	9.9	9.8	9.3
25.0°B, 25.0°C	2.9	2.9	2.7	4.0	5.3	5.9	6.9	6.9	11.0	11.1	10.0	11.0	11.1	11.0	11.0
25.0°B, 30.0°C	3.9	3.9	3.3	5.7	6.0	6.0	7.5	7.5	10.3	10.6	10.8	10.4			
25.0°B, 35.0°C	3.1	3.1	3.3	5.2	5.9	8.0	6.9	6.9	8.0	7.2	7.1	8.1			
30.0°B, 15.0°C	1.0	1.0	1.8	1.6	2.1	4.3	3.0	3.0	5.0	5.3	5.0	5.4			
30.0°B, 20.0°C	1.0	1.0	1.6	1.3	2.9	4.6	3.6	3.6	6.4	6.7	5.4	7.9			
30.0°B, 25.0°C	2.7	2.7	2.3	4.0	4.7	4.8	5.3	5.3	6.7	7.0	7.0	7.8			
30.0°B, 30.0°C	3.6	3.6	3.2	3.9	5.2	5.4	7.0	7.0	6.9	8.0	8.1	8.3			
30.0°B, 35.0°C	4.0	4.0	4.6	4.9	5.7	5.6	6.0	6.0	7.0	7.1	7.1	8.1			
CD at 5%	Fermentation time = 0.140 Treatment = 0.209														

*Treatments as mentioned in materials and methods; %Eth = Ethanol (%) v/v Cultural conditions:
All values are mean of triplicates. Scale of fermentation : 100 ml
PP = Punjab Pink; AA = Arka Amulya; L-49 = Lucknow 49 Inoculum : 5% (v/v)

Table 3. Effect of inoculum size on ethanolic fermentation of guava juice.

Time (days)	*Inoculum size % (v/v)																	
	Control			3			6			9			12			15		
	PP	AA	L-49	PP	AA	L-49	PP	AA	L-49	PP	AA	L-49	PP	AA	L-49	PP	AA	L-49
0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.3	0.0	0.0	0.2	0.0	0.0	0.4
2	3.0	2.3	2.3	2.4	2.0	1.9	3.0	2.5	2.7	3.9	3.7	3.8	4.9	4.4	7.1	5.8	5.1	8.6
4	6.9	5.2	5.1	5.5	3.7	4.4	7.5	5.3	5.0	8.9	6.2	5.5	9.0	7.3	8.2	9.7	7.5	9.2
6	8.7	6.9	9.0	6.9	6.0	7.9	9.0	7.0	9.7	10.4	9.1	9.9	10.9	11.0	10.0	10.2	10.1	10.1
8	10.6	9.6	10.5	8.0	7.6	8.5	10.7	9.9	10.8	11.8	11.7	11.7	11.0	11.1	10.5	10.3	10.1	10.2
10	10.7	10.4	10.7	8.7	8.4	8.6	10.7	10.4	10.8	11.8	11.9	11.8	11.0	11.2	10.5	10.3	10.1	10.3
CD at - (5%)	Fermentation time = 0.126 Inoculum size = 0.941																	

*Initial inoculum size - 6.3×10^8 cfu/ ml ; R.S. = Reducing sugars %; %Eth = Ethanol (%) v/v; PP = Punjab Pink; AA = Arka Amulya; L-49 = Lucknow 49
Cultural conditions
Scale of fermentation : 100 ml Brix : 25°B Temperature : 25°C

Table 4. Effect of di-ammonium hydrogen ortho phosphate (DAHP) concentration on ethanolic fermentation of guava juice.

Time (days)	DAHP conc. (mg/100 ml)																	
	Control (0)			100			200			300			400			500		
	PP	AA	L-49	PP	AA	L-49	PP	AA	L-49	PP	AA	L-49	PP	AA	L-49	PP	AA	L-49
0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.2
2	4.0	3.0	1.6	6.8	4.4	2.5	9.1	6.0	2.7	9.5	8.6	3.1	9.0	8.1	3.6	7.7	7.3	3.8
4	7.6	7.3	3.6	9.7	7.9	5.3	12.0	9.0	7.9	12.4	12.4	9.7	12.8	12.5	10.1	10.6	10.6	10.5
6	11.2	9.9	6.6	11.9	10.4	8.15	12.9	11.3	10.3	13.8	13.4	12.3	12.8	12.4	11.2	10.5	10.4	10.8
8	11.7	11.6	10.0	11.9	11.7	10.9	12.9	13.3	12.8	13.8	13.6	13.6	12.7	12.6	12.1	10.6	10.6	10.9
CD at (5%)	Fermentation time = 0.189 DAHP conc. = 0.164																	

*Initial inoculum size - 6.3 × 108 cfu/ml; RS = Reducing sugars %; %Eth = Ethanol (%) v/v; PP = Punjab Pink; AA = Arka Amulya; L-49 = Lucknow 49

Cultural conditions

Scale of fermentation : 100 ml Brix : 25°B Temperature : 9%

parameters were studied. The results revealed that the yeast was undetectable after 45 days of storage in all three varieties. This was attributed to the absence of sugars and removal of settled yeast during racking from wines. There was insignificant change in pH during the storage. However, ethanol decreased by around 1% v/v over the storage period of 90 days. Inspite of differences in initial ethanol levels, the final ethanol (at 90 days) was constant at 12.6-12.8%, which is reasonably good for wine. Though total phenols decreased significantly during storage in all the three varieties, they were still higher than the 'must'. The decrease in phenols during storage in white wines (Kallithraka *et al*., 7) and Merlot wines have been reported (Ivanova *et al*., 5). While, Kallithraka *et al.* (7) observed a decrease in total phenols upto 6 months of storage and an increase thereafter, Ivanova *et al.* (5) observed a continuous decrease upto 16 months of storage that they studied. Guava being rich in ascorbic acid is a good source of vitamin C. Results presented previously revealed that initial ascorbic acid content (76-91.2 mg/100 ml in all the three varieties) decreased gradually and significantly during storage and was 63.0, 64.2 and 76.0 mg/100 ml in wines prepared from Punjab Pink, Arka Amulya and Lucknow-49, respectively. Literature also reveals that guava rapidly losses ascorbic acid (21-83%) in 4-8 weeks. However, it is retained to a large extent in wines. This retention of ascorbic acid in white wines is a good sign for guava-wine as it prevents browning during storage and also decreases in phenolic compounds. Further, ascorbic acid is a known antioxidant as it quenches free radicals otherwise harmful to health. The aged wine (at 15 and 90 days) was subjected to evaluation by a panel of 10 judges on a 80 point Modified Davis's Score Card. The wine prepared from Punjab Pink and Arka Amulya varieties under the optimized conditions having 13.8 and 13.5% ethanol (v/v) was found to be of standard quality with a mean score of 60.0 ± 3.91 and 58.6 ± 5.02, respectively, whereas wine prepared from variety Lucknow-49 having 13.5% ethanol (v/v) was found to be of below standard quality with mean score of 45.6 ± 9.63 at 15 days of wine age. However, after a period of 90 days, wine from Punjab Pink variety improved to superior quality with the mean sensory score of 68.8 ± 3.27, whereas wine from Arka Amulya was of same standard quality having 54.2 ± 3.11 score. Lucknow-49 wine was of below standard quality with mean score of 47.2 ± 2.38 even at 90 days of storage. The results revealed that with the aging of wine the sensory quality of wine becomes better in terms of taste, aroma, total acidity and overall feel in Punjab Pink variety. Only the wine from Punjab Pink was of superior quality (at 90 days of storage) among three guava varieties studied. Wine prepared from pink fleshed guava variety by Pooja and Kocher (13) was also of superior quality with respect to taste and aroma. Earlier, Kocher *et al.* (9) studied the sensory characteristics of grape wines (white and red) prepared from five different grape varieties/ hybrids and reported species specific variation in sensory characteristics of grape wines.

REFERENCES

1. Amerine, M.A. and Roessler, E.B. 1976. *Wines: Their Sensory Evaluation* (2nd Edn.), W.H. Freeman & Co., New York, pp. 36-37.

2. AOVC. 1996. *Methods of Vitamin Assay*. Association of Vitamin Chemists Inc. (Ed.), Interscience Publishers, New York, pp. 79-81.

3. Ghosh, S., Chakraborty, R. and Raychaudhri, U. 2010. Modulation of palm wine fermentation by the control of carbon and nitrogen source on metabolism of *Saccharomyces cerevisiae*. *J. Fd. Tech.* **8**: 204-10.

4. Hiwale, S.S. and Sharma, S.P. 2003. Prolonging shelf life of guava (*Psidium guajava* L.). *Indian J. Hort.* **60**: 140-46.

5. Ivanova, V., Stefova, M. and Vojnosk, B. 2009. Assay of the phenolic profile of Merlot wines from Macedonia: Effect of maceration time, storage, SO_2 and temperature of storage. *Macedonian J. Chem. Chem. Engg.* **28**: 141-49.

6. Jain, P.K. and Nema, P.K. 2007. Processing of pulp of various cultivars of guava (*Psidium guajava* L.) for leather production. *Agri. Engg. Int.: CIGRE J.* **9**: 1-9.

7. Kallithraka, S., Salacha, M.I. and Tzourou, I. 2009. Changes in phenolic composition and antioxidant activity of white wine during bottle storage: Accelerated browning test versus bottle storage. *J. Fd. Chem.* **113**: 500-05.

8. Kaur, S., Sarkar, B.C., Sharma, H.K. and Singh, C. 2009. Optimization of enzymatic hydrolysis pretreatment conditions for enhanced juice recovery from guava fruit using response surface methodology. *Fd. Bioprocess. Tech.* **2**: 96-100.

9. Kocher, G.S., Phutela, R.P. and Gill, M.I.S. 2009. Evaluation of grape varieties for wine production. *Indian J. Hort.* **66**: 410-12.

10. Kocher, G.S. and Pooja. 2011. Status of wine production from guava (*Psidium guajava* L.): A traditional fruit of India. *African J. Fd. Sci.* **5**: 851-60.

11. Malik, C.B. and Singh, M.B. 1991. *Plant Enzymology and Histoenzymology* (1st Edn.), Kalyani Publishers, New Delhi, pp. 231-33.

12. Miller, G.L. 1959. Use of dinitrosalicylic acid reagent for determination of reducing sugar. *Anal. Chem.* **31**: 426-28.

13. Pooja and Kocher, G.S. 2014. Fermentative production of guava-wine (*Psidium guajava* L.) using *S. cerevisiae* MTCC 11815. *Curr. Nut. Fd. Sci.* **10**: 1.

14. Shankar, S., Dilip, J. and Narayana, R.Y. 2006. Fermentation of guava pulp with grape grown yeast (*S.cerevisiae* var. *ellipsoideus*) for wine production. *Indian J. Hort.* **60**: 171-73.

15. Srivastava, S., Modi, D.R. and Garg, S.K. 1997. Production of ethanol from guava pulp by yeast strains. *Biores. Tech.* **60**: 263-65.

Received : January, 2014; Revised : September, 2014;
Accepted : October, 2014