Value Addition of Tropical Sugarbeet through Microbiological Process: An Innovative Approach

Harish H. Deshpande1*, P. Jones Nirmalnath2 and C.S. Hunshal2

1Water and Land Management Institute (WALMI), Aurangabad – 431 005, Maharashtra, India
2University of Agricultural Sciences, Dharwad - 580 005, Karnataka, India

*Corresponding author

A B S T R A C T

A laboratory experiment was conducted at Microbiology Lab (AICRP-WC), University of Agricultural Sciences, Dharwad during 2012-13 in order to prepare sugarbeet wine from different varieties of Tropical Sugarbeet (TSB). Four TSB varieties were selected viz Magnolia, PAC 60008, Calixta and SZ 35 for wine preparation. In the first four treatments, the sugarbeet juice was obtained and directly assembled for fermentation without any TSS adjustment (T1 to T4). The obtained juice TSS was adjusted to 23ºbrix in the next four set of treatments (T5 to T8). Totally wine was prepared in eight treatment combinations. The wine was subjected to chemical analysis such as alcohol content, pH and TSS. Colour and brightness at different stages and its organoleptic evaluation was also done after proper aging. The experimental results revealed that, wine prepared out of variety calixta without adjustment in its TSS was found superior to other treatment combinations. Either calixta or Magnolia varieties were found superior with respect to wine quality and alcohol production point of view.

Keywords: Sugarbeet wine, Value addition, PAC 60008, Magnolia and Tropical sugarbeet.

Introduction

Tropical sugarbeet (Beta vulgaris L. sp. vulgaris var. altissima Doll.) is an important commercial biennial root crop of the world, extensively grown for sugar and ethanol production. It is second important sugar crop after sugarcane, producing annually about 40% of sugar all over the world (Leilah et al., 2005). In India, sugarbeet can supplement the sugar industries with respect to sugar production point of view due to its higher sucrose content than sugarcane. This crop can give wider scope for development of wineries in India, due to non availability of beet processing mills in potential beet growing regions. Beet is a major economic part which is utilized for the production of white sugar, alcohol, ethanol and pharmaceutical value. The beet molasses is used as a raw material for special fermentations, rich source of lactic acid and vitamin B (Anonymous, 2011).

Wine is a healthful beverage. It has been consumed through ages as food and also as food adjunct. Since antiquity, the virtue of wine as a panacea has been widely exploited in folklore and in the medical arts and sciences (Lucia, 1954, 1963). Wine is unique among beverages in that it contains both alcohol and antioxidants. This coexistence has profound health benefits on consumers. Wine making as a form of food preservation is as old as civilization. Wine has been an integral
component of people’s daily diet since its discovery and had also played an important role in the development of society, religion and culture. Modern wine makers now rely on basic science and the systematic application of their art to produce product pleasing to the increasingly knowledgeable consumer base that enjoys wine as part of its civilized society.

Sugarbeet is commercial crop and having lower value in the sugarcane processing mills. The same raw material can be well utilized in wineries as a high value crop. Through this study effort has been made to prepare wine from sugarbeet as a part of value addition. The wine prepared from beet root has been well proved (Raghavendra Kumar, 2006), but there is no literature on sugarbeet wine. Being a source of alcohol attempt has been made to prepare wine out of this. Fruit wines are undistilled alcoholic beverages which are nutritive, more tasty and mild (Darby, 1979).

Every major civilization since ancient times has drunk wines and Poets, Painter’s and Writer’s have praised it. Wine is fermented juice of fresh fruits (Patankar, 2005). The sugarbeet is modified tap root and classified as a berry type of fruit.

Wines made from fruits are often named after the fruits. The present finding deals with the wine production from different cultivars of sugarbeet with varied TSS levels. The procedure followed and methodologies adopted are presented under the heading Material and Methods.

Materials and Methods

The present investigation on preparation of sugarbeet wine was carried out at the Microbiology Lab (AICRP-Weed Control), MARS, UAS, Dharwad during the period of 2011-12.

Selection of tubers and yeast culture

The fully matured (180 days old) healthy and disease free sugarbeet tubers were obtained from the experimental field at ARS, Mudhol. Four different varieties were selected for the investigation viz., SZ 35, PAC 60008, Magnolia and Calixta. A pure culture of Saccharomyces cerevisiae (CFTRI) was obtained from Department of Agriculture Microbiology, UAS, Dharwad. It was kept in refrigerator at 0 to 5 °C for the further use.

Juice recovery

The sugarbeet tubers were washed thoroughly with tap water in order to remove the muddy particles, debris and adhering particles present in the side root grooves. After washing, the roots were peeled with a hand peeler. Peeled tubers were washed thoroughly with clean water at 50°C before chopping them into small pieces (1 cm³ size) with sterilized knife. The chopped tubers of 500 g were transferred to a clean glass beaker containing 500 ml of water (1:1 ratio) and ground in mixer grinder. Finally, the sugarbeet juice was filtered through muslin membrane cloth in order to get the extract for further fermentation process. Chemical analysis of each wine sample was done to know the parameters like pH, alcohol per cent and TSS. In addition to that colour and brightness, organoleptic evaluation of wine was also done.

Treatment details

T₁: Magnolia with TSS 23.0 brix
T₂: PAC 60008 with TSS 23.0 brix
T₃: Calixta with TSS 23.0 brix
T₄: SZ 35 with TSS 23.0 brix
T₅: Magnolia with TSS 13.2 brix
T₆: PAC 6008 with TSS 12.9 brix
T₇: Calixta with TSS 13.8 brix
T₇: SZ 35 with TSS 12.3 brix
Note: The external source of sugar was added to obtain TSS of 23 °brix

**Preparation of wine**

The fresh extracted juice was transferred into a fermentor (1000 ml). To this 75 g of sugar (T₁ to T₄) and 50 mg of potassium meta-bisulphite (KMS) was added and flask mouth was covered with polythene cover. After ½ an hour 5% of starter culture or inoculum (v/v basis) was added to the fermentor and kept for fermentation. This fermentation assembly was incubated at room temperature for 10 days. Raking was carried out after 5-6 days after incubation of the yeast. Clear wine was siphoned out into sterilized bottles after passing it through cheese cloth. Further, the wine was clarified with the help of bentonite clay. Finally, wines were stored in airtight bottles for further aging (Kim et al., 1998). The flow diagram illustrating wine preparation from sugarbeet is shown in Fig.1.

**Estimation of alcohol % (ethanol)**

The ethanol content of the fermented medium was estimated colorimetrically as per the method described by Caputi et al., (1968).

**pH**

The pH of the wine was measured using the pH meter of Analog model (Corion Research, USA) at two stages viz., immediately after the completion of fermentation and after the aging (4 months). Standard solutions of pH 4.0, 7.0 and 9.0 were used as reference to calibrate.

**Colour and brightness**

The colour of the wine was measured with the help of spectrophotometer (Onkarayya, 1986) at 420 nm and for brightness sum of absorbance of 420 and 520 nm after diluting the samples to 1:1 with water was measured.

**Brix**

Brix reading of the wine samples was determined with the help of ERMA Hand Refractometer immediately after the fermentation and after aging, having a range of 0-32 °brix at 20°C.

**Organoleptic evaluation**

Each sample was coded prior to testing and placed in a random manner. Different samples were placed along with glass of water (to rinse the mouth) in the laboratory and panelists were instructed to evaluate each sample by blind tasting as per the score card. The standard grape wine was kept for comparison. Twenty point scales (Amerine and Ough, 1980) was based mainly on the appearance, colour, aroma, taste and acceptability. All the wines were evaluated by 5 test panel members.

**Grading according to score**

1. 17-20 wines with outstanding characteristics and no marked defect
2. 13-16 standard wines with neither an outstanding character nor defect
3. 9-12 wines of commercial acceptability but with a noticeable defect
4. 5-8 wines of below commercial acceptability
5. 1-4 completely spoiled wines.

**Results and Discussion**

**Chemical analysis of the wine**

The data pertaining to chemical analysis of sugarbeet wine prepared by using different varieties at varied TSS levels of sugarbeet juice viz., modified and normal TSS levels are presented in table 1.
Alcohol content

Calixta with TSS level of 23 °brix recorded higher alcohol content (13.10%) followed by Magnolia with TSS level of 23 °brix (12.98%) and lowest in PAC 6008 with TSS level of 23 °brix (8.81%) among the modified TSS levels (T₁-T₄). However, in original TSS levels (T₅-T₈) the highest alcohol content was recorded in Calixta with TSS level of 13.8 °brix (8.81%). However, the lowest alcohol content was recorded in SZ 35 with TSS level of 12.3 °brix (5.95%).

In the present study, Calixta with TSS level of 23 °brix recorded highest per cent of alcohol (13.10%). This could be due to the fact that amount of alcohol produced depends upon fermentation efficiency of yeast strain and capacity of sugar uptake. These results are in conformity with data by Ayogu (1999) and Joshi et al., (1991). However, reduced TSS levels indicate lower sugar content present in it. The decline in sugars reduces the fermentation efficiency there by reducing the alcohol content. The low alcohol content in wine may be due to yeast growth suppression in the juice. Ethyl alcohol content of 6.6 % in pomegranate wine was reported by Adusule et al., (1992). Free Ethanol content of 12.6 % in banana wine and 10.4 % in tomato wine was reported by Mathapati (2005).

pH

The pH of the wine was recorded after fermentation and after the aging process. The pH values varied slightly. The highest pH was recorded with Calixta with TSS level of 13.8 °brix (4.76). The lowest value of pH was recorded with Magnolia with TSS level of 23 °brix (3.25) immediately after the fermentation.

The pH of the wine was also recorded after aging process. The pH values varied slightly. The highest pH values were recorded in Calixta with TSS level of 13.8 °brix (5.01) which were followed by SZ 35 with TSS level of 12.3 °brix (4.75). The lowest value of pH was recorded in Magnolia with TSS level of 23 °brix (3.98) immediately after the aging.

| Treatment | Alcohol content (%) | pH of the wine after fermentation | pH of the wine after aging (4 months) | TSS (°Brix) after fermentation |
|-----------|---------------------|-----------------------------------|--------------------------------------|-----------------------------|
| T₁        | 12.98               | 3.25                              | 3.98                                 | 4.3                         |
| T₂        | 8.81                | 3.90                              | 4.53                                 | 5.5                         |
| T₃        | 13.10               | 3.42                              | 4.13                                 | 5.6                         |
| T₄        | 10.60               | 3.76                              | 4.42                                 | 5.0                         |
| T₅        | 07.50               | 3.86                              | 4.51                                 | 2.9                         |
| T₆        | 06.19               | 4.12                              | 4.42                                 | 3.4                         |
| T₇        | 08.81               | 4.76                              | 5.01                                 | 3.0                         |
| T₈        | 05.95               | 4.12                              | 4.75                                 | 3.0                         |

T₁: Magnolia with TSS 23 °brix  
T₂: PAC 6008 with TSS 23 °brix  
T₃: Calixta with TSS 23 °brix  
T₄: SZ 35 with TSS 23 °brix  
T₅: Magnolia with TSS 13.2 °brix  
T₆: PAC 6008 with TSS 12.9 °brix  
T₇: Calixta with TSS 13.8 °brix  
T₈: SZ 35 with TSS 12.3 °brix
Table 2 Colour and brightness of sugarbeet wine prepared by using different varieties and TSS levels of sugarbeet juice

| Treatment | Optical Density (OD) values (Immediately after fermentation) | Optical Density (OD) values (After Aging/4 months later) |
|-----------|---------------------------------------------------------------|----------------------------------------------------------|
|           | Colour (420 nm) | Brightness (420nm+520nm) | Colour (420 nm) | Brightness (420nm+520nm) |
| T1        | 0.216           | 0.333                      | 0.004           | 0.563                      |
| T2        | 0.289           | 0.425                      | 1.677           | 2.940                      |
| T3        | 0.262           | 0.368                      | 1.025           | 1.541                      |
| T4        | 0.305           | 0.473                      | 1.148           | 2.014                      |
| T5        | 0.245           | 0.301                      | 0.987           | 1.578                      |
| T6        | 0.229           | 0.315                      | 1.276           | 2.160                      |
| T7        | 0.460           | 0.596                      | 1.315           | 1.933                      |
| T8        | 0.331           | 0.449                      | 1.290           | 2.357                      |

T1: Magnolia with TSS 23 °brix  
T2: PAC 6008 with TSS 23 °brix  
T3: Calixta with TSS 23 °brix  
T4: SZ 35 with TSS 23 °brix  
T5: Magnolia with TSS 13.2 °brix  
T6: PAC 6008 with TSS 12.9 °brix  
T7: Calixta with TSS 13.8 °brix  
T8: SZ 35 with TSS 12.3 °brix  

Table 3 Organoleptic evaluation of sugarbeet wine prepared by using different varieties and TSS levels of sugarbeet juice

| Sl. No | Quality character | T1 | T2 | T3 | T4 | T5 | T6 | T7 | T8 | C |
|--------|-------------------|----|----|----|----|----|----|----|----|---|
| 1      | Appearance        | 2  | 1.07 | 1.08 | 1.20 | 1.13 | 1.17 | 1.37 | 1.42 | 1.15 | 1.82 |
| 2      | Colour            | 2  | 1.20 | 1.25 | 1.61 | 0.98 | 1.23 | 1.59 | 1.52 | 1.14 | 1.89 |
| 3      | Aroma             | 2  | 1.08 | 0.95 | 1.20 | 1.06 | 1.48 | 1.43 | 1.20 | 1.24 | 1.50 |
| 4      | Bouquet           | 2  | 0.92 | 0.93 | 1.01 | 0.79 | 1.26 | 1.18 | 1.42 | 1.19 | 1.45 |
| 5      | Vinegar           | 2  | 0.62 | 0.65 | 0.77 | 0.77 | 0.57 | 0.75 | 0.72 | 0.55 | 0.88 |
| 6      | Total acidity     | 2  | 0.83 | 0.91 | 0.88 | 1.15 | 0.71 | 0.67 | 0.77 | 0.57 | 0.15 |
| 7      | Sweetness         | 1  | 0.70 | 0.83 | 0.77 | 0.69 | 0.83 | 0.94 | 1.00 | 1.00 | 1.00 |
| 8      | Body              | 1  | 0.93 | 0.90 | 1.00 | 0.97 | 1.00 | 0.87 | 0.96 | 0.98 | 1.00 |
| 9      | Flavour           | 2  | 1.10 | 0.92 | 1.23 | 1.18 | 1.45 | 1.28 | 1.36 | 1.09 | 1.73 |
| 10     | Astringency       | 2  | 0.58 | 0.60 | 0.63 | 0.77 | 0.67 | 0.60 | 0.80 | 0.67 | 0.53 |
| 11     | General quality   | 2  | 0.99 | 1.02 | 1.17 | 1.17 | 1.11 | 1.25 | 1.48 | 1.22 | 1.95 |
| 12     | Total score       | 20 | 10.02 | 10.04 | 11.47 | 10.66 | 11.48 | 11.93 | 12.65 | 10.80 | 13.90 |

T1: Magnolia with TSS 23 °brix  
T2: PAC 6008 with TSS 23 °brix  
T3: Calixta with TSS 23 °brix  
T4: SZ 35 with TSS 23 °brix  
T5: Magnolia with TSS 13.2 °brix  
T6: PAC 6008 with TSS 12.9 °brix  
T7: Calixta with TSS 13.8 °brix  
T8: SZ 35 with TSS 12.3 °brix  
C: Standard check (Grape wine)
Plate 1: Sugarbeet Wine Prepared from Different Treatment Combinations

**Treatment details**

T₁: Magnolia with TSS 23 °brix
T₂: PAC 60008 with TSS 23 °brix
T₃: Calixta with TSS 23 °brix
T₄: SZ 35 with TSS 23 °brix

T₅: Magnolia with TSS 13.2 °brix
T₆: PAC 60008 with TSS 12.9 °brix
T₇: Calixta with TSS 13.8 °brix
T₈: SZ 35 with TSS 12.3 °brix
Fig.1 Schematic illustration of wine preparation from sugarbeet

Sugarbeet roots

Washing peeling and chopping

Extraction of juice

Filtration

Brix adjustment

Addition of potassium meta-bisulphite (200 ppm)

Addition of starter culture (5% v/v basis)

Fermentation for 10 days

Filtration

Racking (3-4 times)

Addition of bentonite clay (400 mg/l)

Filtration

Pasteurization at 62°C for 30 min

Stored for maturation

The pH values after fermentation varied between 3.25 and 4.76 in general. The higher pH was observed in Calixta with TSS level of 13.8 °brix (4.76). The pH of the wine depends on the acid and sugar content of the wines according to Sanchez et al., (1987). Similar
work was done by Arun (2005) and reported that pH of wine prepared from different rice varieties ranged from 4.65 to 5.0. The pH of pineapple wine varied between 3.18 and 3.90 (Roodagi, 2010). The decrease in pH along with fermentation could be due to the efflux of H\(^+\) ions as a byproduct of the transport system and by organic acid production.

**Total soluble solids (TSS %)**

The highest TSS was recorded in Calixta with TSS level of 23 \(^{0}\)brix (5.6%) followed by PAC 6008 with TSS level of 23 \(^{0}\)brix (5.5%) among the modified TSS levels (T\(_1\)-T\(_4\)). The highest TSS was noticed in PAC 6008 with TSS 12.9 \(^{0}\)brix (3.4%) and was followed by SZ 35 with TSS 12.3 \(^{0}\)brix (3.0%).

The highest TSS was recorded in Calixta with TSS level of 23 \(^{0}\)brix (5.6%) among the modified TSS levels (T\(_1\)-T\(_4\)) and PAC 6008 with TSS 12.9 \(^{0}\)brix (3.4%) among the original TSS levels which is due to changes in the reducing sugar levels. The reducing sugar content in the sugarbeet varieties may vary from one genotype to another and reducing sugars constitute a major part of soluble solids present in the wine. The result showed that total sugar (%) content of wine was found to increase with increase in TSS levels. The variation in total sugar content of wine was due to the addition of sugars to maintain different TSS levels in must. This trend was similar to the study conducted by Lakshmana and Lingaiah (2006).

**Colour and brightness**

The data pertaining to colour and brightness are presented in table 2 and plate 1. The experimental results showed that, highest value of colour (0.460) and brightness (0.596) was recorded in wine prepared by using Calixta with TSS level of 13.8 \(^{0}\)brix followed by SZ 35 with TSS level of 12.3 \(^{0}\)brix (0.331 and 0.449 respectively) whereas, wine prepared by Magnolia with TSS level of 23 \(^{0}\)brix recorded the lowest value of colour (0.216) and brightness (0.333) immediately after the fermentation. The colour and brightness of the same treatments recorded after the aging and have showed much variation in colour and brightness.

The highest TSS was recorded in Calixta with TSS level of 23 \(^{0}\)brix (5.6%) followed by PAC 6008 with TSS level of 23 \(^{0}\)brix (5.5%) among the modified TSS levels (T\(_1\)-T\(_4\)). The highest TSS was noticed in PAC 6008 with TSS 12.9 \(^{0}\)brix (3.4%) and was followed by SZ 35 with TSS 12.3 \(^{0}\)brix (3.0%).

PAC 6008 with TSS level 23 \(^{0}\)brix recorded higher value of colour (1.677) followed by Calixta with TSS level of 13.8 \(^{0}\)brix (1.315). The lowest value of colour was obtained in Magnolia with TSS level of 23 \(^{0}\)brix (0.004). The brightness value of the wine was highest in PAC 6008 with TSS 23 \(^{0}\)brix (2.940) followed by SZ 35 with TSS level of 12.3 \(^{0}\)brix (2.357) and the lowest value of brightness was recorded with Magnolia with TSS level of 23 \(^{0}\)brix (0.563) after the aging process.

The highest value of colour (0.460) and brightness (0.596) was recorded in wine produced using Calixta with TSS level of 13.8 \(^{0}\)brix (Plate 1). However, Magnolia with TSS level of 23 \(^{0}\)brix recorded the lowest colour (0.216) and brightness (0.333) value. This variation in the colour intensity seems to be due to the inherent differences in composition of sugarbeet varieties. Arun (2005) recorded similar findings in the preparation of rice wine from different varieties. They reported that the highest colour and brightness was recorded in wine from Bharati variety (0.144 and 0.244) and lowest in wine from Intan variety (-0.042 and 0.056).

**Organoleptic evaluation of sugarbeet wine**

Wine quality evaluation scores (average of five members) for individual parameters of 20 point scale viz. appearance, colour, aroma, bouquet, vinegar, total acidity, sweetness, body, flavour, astringency, general quality, total score are presented in table 3.
The scores for the overall acceptability from organoleptic evaluation showed that wine produced from Calixta with TSS level of 13.8 °brix recorded highest score (12.65 out of 20.00) followed by PAC 6008 with TSS level of 12.9 °brix (11.93 out of 20.00 respectively). Whereas, the wine prepared by using PAC 60008 variety with TSS level of 23 °brix recorded the lowest score (10.04 out of 20.00).

Wine is made for human consumption, so, it cannot be evaluated only by chemical parameters. Therefore sensory evaluation is necessary to evaluate wine quality. Sensory evaluation is done by selected panel of members through organoleptic procedures. The score for overall acceptability of sugarbeet wine ranged from 10 to 13.0. The treatment Calixta with the TSS level of 13.8 °brix recorded maximum score for overall acceptability. The scores for sugarbeet wine for overall acceptability reported in the present study are in line with scores for rice wine (11 to 16) obtained by Arun (2005).

The organoleptic evaluation showed that Calixta with the TSS level of 13.8 °brix is supposed to be the best treatment combination for the production of good quality sugarbeet wine. This might be due to superiority in most of the characters like colour, appearance, body, taste, astringency, and overall acceptability. It can be considered as standard wine with neither an outstanding character nor defect because of its musty or muddy odour.

In conclusion the organoleptic evaluation showed that wine prepared out of Calixta with TSS level of 13.8 °brix recorded highest score (12.65 out of 20.00) for its commercial acceptability. For commercial alcohol production point of view either Calixta (13.10%) or Magnolia (12.98%) with TSS modification to 23 °brix may be preferred.

Further research in this area is required to improve its aroma by removing its musty or muddy odour, mainly for its commercial acceptability.

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