

STUDY ON RED WINE FERMENTATION FROM NEPALESE PLUM

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ABSTRACT

Red wine from indigenous Nepalese plum was prepared using suitable wine yeast isolated from different mucha samples. Two different yeast isolates namely M3, M8 and one commercial wine yeast were selected for plum wine production on the basis of CO₂ production and flavour in Yeast Malt extract broth. Wine was prepared using these strains separately from plum juice maintained at 20 °Brix. The various physico chemical analyses i.e. changes in °Brix, pH and acidity of wine during fermentation were performed. On completion of fermentation the ethanol content of these wines range from 5.5-6.5% by volume. Plum wine prepared by inoculating these strains at 20 °Brix were found to be comparable to market wine on the basis of sensory evaluation.

Key words: mucha, °Brix, fermentation, Yeast Malt extract broth

INTRODUCTION

Wine is an alcoholic beverage made from partial or complete fermentation of grape juice or various other fruit juices like apple, pear, cherry, plum etc. High demand of wine throughout the world indicates a wide scope of wine from different fruits such as plum (Joshi *et al.*, 2009).

Plum (*Prunus salicina*) is a type of stone fruit which is highly perishable with shelf life of 3 to 4 days at ambient temperature and up to 2 weeks in the cold storage (Joshi *et al.*, 1995). Plum fruit contains varying concentrations of sugars, anthocyanins, vitamins, acids etc. Average fruit weight of plum ranged between 5.86 g (red colored) and 7.83 g (dark purple colored). pHs were 3.13 (dark purple colored) and 3.70 (red colored); TSS were 11.98% (dark purple colored) and 14.98% (red colored). Wide variations in physico-chemical characteristic have been reported on plums. Vitamin C content of dark purple, red and yellow plum fruits ranged from a low of 3.8-12.1 mg/100 ml of fresh fruit. The highest amount of total anthocyanin was observed in dark purple fruits (41.3 mg/100 g). D-glucitol, sorbitol, galactose, mannose, arabinose, rhamnose, raffinose, and xylose are the main sugar detected in plum (Wills *et al.* 1993).

Wine is prepared from sugar containing raw material, yeast culture, sulphur dioxide etc. Wine fermentation comprises two different stages, the first stage is alcoholic fermentation where yeast converts sugar into ethanol, CO₂, and some esters (Joshi *et al.* 1991). While the second stage of fermentation is bacterial fermentation called malo-lactic fermentation where lactic acid bacteria

converts malic acid to lactic acid and CO₂ resulting in reduction of acidity of wine (Duenas *et al.*, 1994). The objective of the study was to select appropriate indigenous wine yeast and utilize it to produce plum wine with appealing colour and taste.

MATERIALS AND METHODS

Sample collection

Well ripened and undamaged indigenous plum fruits were collected from Gulmi district, Nepal. Wine yeast was isolated from 5 mucha samples collected from local market of Kathmandu valley.

Isolation of yeast

The mucha sample was powdered by clean mortar and pestle. One gram mucha sample was kept for ten fold serial dilutions. Isolation of yeast was done by pour plate technique using yeast malt extract agar (YMA) medium (Refai, 1992).

Screening of yeast:

The isolated yeasts were sub-cultured on YMA by streak plate technique and incubated at 28°C for 48 hours. The yeasts were confirmed by gram staining. The test for fermentative yeast was done by testing CO₂ production (Martini *et al.*, 1996). The yeast was grown in test tubes containing sterilized YM broth with inverted Durham's tube incubated at 28°C for 48-72 hours. After incubation the yeast with high CO₂ production in Durham's tube were considered as fermentative.

Preparation of must

Well ripened and undamaged fruits were washed

with clean water. Stones of plum fruits were removed mechanically. The juice was obtained by placing it in juicer. Fifty ppm (Parts Per Million) potassium meta bisulphate (w/v) was added to the extracted juice. The juice was then treated with pectinase enzyme (0.5%) and kept for 10 hours. The juice was strained through muslin cloth. The juice thus obtained was heated to 80°C for 15 seconds and cooled to 40°C. The initial parameters of juice were recorded as TSS (Total Soluble Solid) of must 11°Brix (°B), pH 3.60. Sugar was added to the juice to maintain 20°Brix.

Preparation of started culture

The yeast isolates M3, M8 and M1 were inoculated in the 5ml of sterile YM broth separately and incubated at 28°C for 24 hours. Then it was centrifuged at 5000 rpm for 30 seconds. The sediment (pellet) was then washed with normal saline and inoculated in 400 ml of the sterile plum juice. After 24 hours of incubation the load of the yeast in starter was measured and maintained at 10⁶ cfu/ml by the addition of sterile juice.

Inoculation and fermentation

The starter of each strain thus prepared was inoculated separately to 4000 ml of juice and incubated at room temperature (25-26°C) for 15 days. After fermentation siphoning was done and bottled. Pasteurization of wine was done at 70°C for 5 min.

Physico chemical analysis

The various physico chemical parameters (pH, acidity, °Brix) during fermentation of plum wine were measured at regular interval of 24 hours (AOAC, 2005). Alcohol content of wine was measured by specific gravity method. After ageing for three months sensory evaluation was carried out by a group of panelists.

RESULT AND DISCUSSION

Isolation and screening of fermentative yeast

From 5 different murcha samples, 20 different yeasts were isolated. Three yeast isolates showing high CO₂ production on Durham's tube were selected for fermentation. Code M3 and M8 were given for yeast strain isolated from murcha and B1 for commercial yeast strain. These three strains were used for wine production.

Fermentation

The selected yeast strains were inoculated separately in plum juice and kept at room temperature (25°C) for 15 days. Changes in pH, decrease in TSS (Total Soluble Solid or °B) and change in acidity (as malic) were determined (Satora and Tuszyński, 2009). The results for TSS (°B), pH and acidity were shown in figure.

Fig.2 and Fig.3 represent changes in pH and % titrat-

able acidity during the course of fermentation. The initial pH of 3.6 was gradually reduced to 2.55, 2.50 and 2.53 by yeast isolates M3, M8 and B1 respectively. There were no significant differences in pH change. The initial % titratable acidity of must was 0.87. The

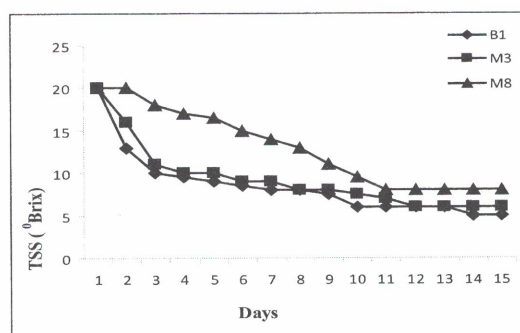


Fig. 1 shows the decrease in TSS of wine during course of fermentation. The rapid decrease in °B were shown up to 5 days by strain B1 and M3 where as strain M8 showed slow decrease up to 10 days. Then after 10 days no significant decrease in °B were observed by all strains. At the end of fermentation, the yeast strain M3 and M8 reduced 20 °B to 5 °B and 8 °B respectively whereas commercial strain reduced TSS to 6 °B.

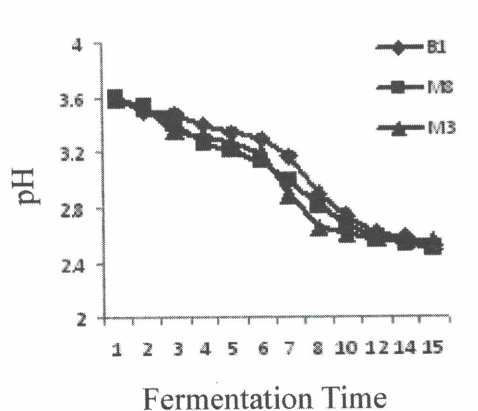


Fig 2. Change in pH during fermentation

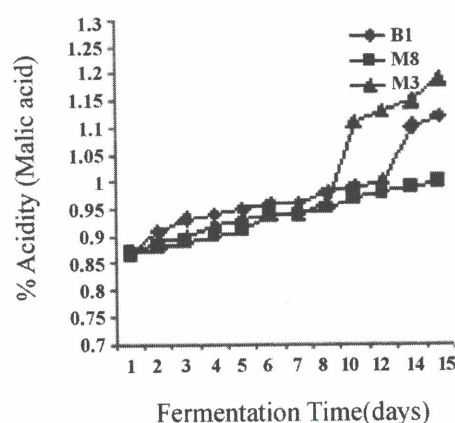


Fig 3. Change in acidity during fermentation

acidity of all the wine was found to be increase with progression of fermentation period. There was slight increase in titratable acidity during first week. Then onward it increases rapidly and remains stable at the end of fermentation. The final acidity of plum wine prepared by yeast strain M3, M8, and B1 were found to be 1.35%, 1.33% and 1.2% (as malic acid) respectively.

The alcohol content of plum wine was comparable with reduction in TSS. Fermentation of sugar is directly related to alcohol content. In case of plum wine fermented by yeast strain M3, M8 and B1, alcohol contents were found to be 6.039%, 5.760% and 6.315% respectively. The concentration of alcohol in wine was directly correlated by TSS present at the end of fermentation in each wine. The wine with comparatively high concentration of alcohol has reduced TSS than the other. The TSS and ethanol content obtained at the end of fermentation from the three yeast strains are comparable with TSS of 6.5-8.0 and alcohol content 5.5-7.0% of plum wine (Joshi and Attri, 2005).

Sensory evaluation of wine

Sensory evaluation of plum wine prepared was done on different attributes such as colour, aroma, taste, after taste and overall impression by the panelists of 10 judges. Out of 20, the overall mean score of wine fermented by M3, M8 and B1 were found to be 16.5, 16.3 and 16.8 respectively whereas commercial red wine has overall mean score of 17.9. There is a statistically significant difference ($P = <0.001$) between aroma of different wines, there is statistically significant difference ($P = 1.000$) in overall expression, there is a statistically significant difference ($P = <0.001$) among taste of wine, there is a statistically significant difference ($P = 0.006$) between colour whereas no statistically significant difference was observed on total score. Sensory evaluation of plum wine revealed that it was comparable with commercial red wine in terms of overall score.

CONCLUSION

Plum wine prepared using indigenous substrate and yeast strains isolated from murcha was found to be comparable to commercial red wine in terms of sensory quality. This study indicates the strains isolated from indigenous starter culture may be used for preparation of commercial wine from various substrates. The yeast isolated from the murcha need to be characterized and genetically identified to use in global market.

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