Original Research Article

Production, Optimization and Characterization of wine from Papaya using *Saccharomyces cerevisiae*

C. Maragatham Alagesan¹* and A. Panneerselvam²

¹Department of Biotechnology, Rajah Serfoji Government College (Autonomous), Thanjavur – 613 005, Tamil Nadu, India
²Department of Botany and Microbiology, A.V.V.M Sri Pushpam College, Thanjavur-613503, Tamil Nadu, India
*Corresponding author

A B S T R A C T

Carica papaya (pawpaw) is one of the fruits commonly used as food and medicine. It is eaten as fresh fruit or processed into desserts. It is a sugar crop with soluble saccharide in the form of glucose, fructose and sucrose. The fermentation of this juice to wine can be attractive alternative to explore its potential in alcohol beverage industry. The present investigation was carried to find out the optimal conditions for the efficient conversion of papaya juice into wine using *Saccharomyces cerevisiae*. The optimum process conditions for this fermentation process were 24ºBrix Total Soluble Solids, 26ºC temperature, 5ml of pectinase enzyme, 10% inoculum, and pH 4.5. Corresponding to these optimum conditions, the predicted value of ethanol production was found to be 11-12%, which was experimentally verified.

Keywords

Papaya fruits, *Saccharomyces Cerevisiae*, Fermentation, wine

Introduction

Wine, an alcoholic beverage is prepared by different fruit juices with appropriate processing and additions (Amerine and Singleton, 1968). The conventional process of wine making involves the fermentation of grape juice. However, there are numerous reports available on wine preparation from other fruits such as apple, plum, apricot, pomegranate, strawberry, kinnow, guava, jamun, sapota, litchi, etc (Sandhu and Joshi, 1995; Joshi et al., 1991; Joshi and Sharma, 1994; Adsule and Kadam, 1995; Joshi et al., 2005; Joshi et al., 1997; Bardiya et al., 1974; Shukla et al., 1991; Gautam and Chundawat, 1998; Zeng et al., 2008).

A large quantity of wines are produced and consumed all over the world (Amerine *et al.*, 1980). Traditionally, the fruit juice was fermented by wild yeast. However, with the developments in the field of fermentation, different strains of *Saccharomyces cerevisiae* and various fruits juices have been explored for wine production. The utilization of papaya juice for the preparation of wine can solve the problems of market surplus and related spoilage, apart from the development of a new variety of wine. Keeping this in view the present investigation was carried out for the optimization of process parameters for
papaya wine. Optimization of parameters by the conventional method involves changing one dependent variable while unchanging all others at a fixed level. Fermentation was performed under optimized condition. The papaya wine was characterized by various features such as TSS, Pectinase, inoculum level, temperature, pH.

Materials and Methods

Procurement of Papaya Fruit and Yeast Strain

The papaya fruits (Variety Co2) used for the preparation of papaya wine were procured from Tamil Nadu Agricultural University, Coimbatore. The yeast *Saccharomyces cerevisiae* culture isolated from rotten papaya fruits. The culture was maintained on malt agar medium and stored at refrigerator.

Inoculum Preparation

The inoculum was prepared by inoculating the slant culture into 10ml test tube incubated overnight then 3% inoculum transferred to 100ml of the sterile malt broth medium taken in 250ml flask and grown it on a rotary shaker for 48 h. After incubation period the culture was used for preparation of wine.

Fermentation Processes

Co 2 papaya fruits were taken and it was completely peeled off. The pulp was macerated in mixie/blender and pasteurized at 85-90°C for 5 minutes. A known volume of papaya juice was taken separately and potassium metabisulphite(KMS) at a rate of 200 ppm added to avoid growth of wild yeast and Diammonium orthophosphate at a rate of 0.2% was added as a source of nitrogen (N2) and phosphorus to yeast. In order to determine the optimum TSS, Pectinase, inoculum level, temperature, pH the experiments were carried out by incubating the appropriate number of inoculated flask at different TSS (18, 20, 22 24 & 26º Brix), Pectinase (1ml-5ml), inoculum level (2ml, 4ml, 6ml, 8ml and 10ml/1r), pH value (2,3,4,5 and 6), temperatures (15, 20, 25 and 35 º C). During the primary fermentation the must was aerated daily up to 9 days. All the treatment was kept for primary fermentation for 9 days with periodic aeration. After 9 days all the treatments were filtered through muslin cloth and filtrate was kept secondary fermentation in plastic carboys with air lock/water seal the carbon dioxide developed during fermentation. The secondary fermentation was carried out for a period of 2 weeks at same temperature. After two weeks the evolution of CO₂ ceased and the wine was clarified by centrifugation at 5000 rpm. The sediment was discarded and the clear wine was filled into sterile bottles of 200ml capacity and crown corked. The same bottles were pasteurized at 50°C for 15 minutes. The pasteurized bottles of wine were kept for aging at ambient temperature. The alcohol content was determined as per the standard procedures (AOAC, 2005).

Results and Discussion

Effect of Alcoholic Fermentation of Reconstituted Papaya Juice by *S.cerevisiae* with Total Soluble Solids as a Function

The total soluble solids of the reconstituted papaya juice were varied from 18-26 Brix, to study its influence on alcoholic fermentation. An increase in ethanol production was recorded with the increase in total soluble solids up to 24ºB and thereafter, no change was recorded for this parameter. Therefore, 24ºB was considered
optimal for further experimentation (Table 1, Figure 1). The optimization study the initial TSS 24ºB given maximum ethanol production. The maximum ethanol production has been reported in juice with 24ºB after 5 days of Kinnow juice fermentation Singh et al., 1998. Same has been the case for preparation of strawberry wine Joshi et al., (2005).

**Effect of Alcoholic Fermentation of Reconstituted Papaya Juice by S. cerevisiae with pH as a Function**

The maximum ethanol production (11.65%) was recorded at pH 5.0 and low alcohol content was recorded after 5 days of fermentation below and above this pH (Table 1, figure 4). The pH optimization study the initial pH 4.5 given maximum ethanol production. Earlier also pH 4.5 has been reported optimal for alcoholic fermentation of Kinnow juice Singh et al., (1998).

**Effect of Alcoholic Fermentation of Reconstituted Papaya Juice by S.cerevisiae with Temperature as a Function**

Alcoholic fermentation of papaya juice was carried out for 5 day at a temperature range of 15-30°C. The maximum ethanol production was recorded at a temperature range of 25-30°C. Comparatively, ethanol production was lesser at 20°C than at other temperature tried. Therefore, a temperature of 26°C been selected for further studies (Table 1, figure 5). The temperature optimization studies the initial temperature 25-30C given maximum ethanol production. The maximum ethanol production from kinnow juice has been reported at 30°C. However, technology of low temperature (13-16°C) and log fermentation time has been used for making of litchi wine Zeng et al (2008). It has been reported that when the fermentation temperature is between 10-15°C, most of the main aromatic compounds that endow the products with wine character can be preserved Wondra and Berovic, (2001).

**Effect of Alcoholic Fermentation of Reconstituted Papaya Juice by S.cerevisiae with Inoculum Level as a Function**

The results presented in table 1 depict the effect level of S. cerevisiae on ethanol production in papaya juice. Ethanol production was increased with the increase in inoculums concentration up to 10% for a fermentation period of 5 days. A slight decrease in ethanol production was recorded beyond inoculums level of 10%. Among the various concentration of inoculum level the 10% given maximum ethanol production (Table 1, figure 2). Similar result was observed by Singh et al., (1998). He reported an inoculums level of 10% has been reported optimal for the production of kinnow wine.

**Effect of Alcoholic Fermentation of Reconstituted Papaya Juice by S.cerevisiae with Pectinase Enzyme as a Function**

Pectinase treatment increased the yield of juice in all papaya cultivars. The optimized pectinase enzyme concentration was 5ml. It was considered very important because higher concentration may result in increased methanol accumulation due to the hydrolysis of methyl pectin esterases. Pectinase treatment significantly increased the ethanol production from papaya pulp. The increase in ethanol may be because of must clarity and increased sugar concentration in pectinase treated samples (Table 1, figure 2).
Sensory Evaluation

The sensory evaluation report reveals that the papaya wine was clean and golden brown colour. It was having a beautiful and plumy aroma of papaya fruit and a harmonious wine taste. In terms of the total score the wine has been evaluated as of superior quality.

*Saccharomyces cerevisiae* efficiently carried out the fermentation of papaya juice for wine production. Total soluble solids (TSS) concentration, process temperature, Pectinase, inoculum level, pH were most significant factors affecting the ethanol production and their optimal values obtained 24ºBrix, 26ºC temperature, 5ml of pectinase enzyme, 10% inoculum, and pH 4.5. The optimum process conditions were experimentally verified. The present studies have revealed that papaya fruit could be successfully utilized for the production of wine. The high quality wines should have a characteristic bouquet and taste which depends on the cultivar, maturity and phytosanitary conditions of the papaya fruit, pedoclimatic conditions and most importantly, on yeast fermentation physiology. All these factors cause the different aroma, fragrance, composition and quality among all kinds of fruits wines.

**Table.1 Effect of Various TSS, Pectinase Enzyme, Inoculum, pH, Temperature on pPapaya Wine Production by *Saccharomyces Cerevisiae***

<table>
<thead>
<tr>
<th>TSS (ºBrix)</th>
<th>Ethanol (%)</th>
<th>Pectinase (ml)</th>
<th>Ethanol (%)</th>
<th>Inoculum level(ml)</th>
<th>Ethanol (%)</th>
<th>pH</th>
<th>Ethanol (%)</th>
<th>Temp. (ºC)</th>
<th>Ethanol (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>18</td>
<td>7.26</td>
<td>1ml</td>
<td>6.46</td>
<td>2ml</td>
<td>5.43</td>
<td>2.0</td>
<td>3.00</td>
<td>15</td>
<td>07.21</td>
</tr>
<tr>
<td>20</td>
<td>8.25</td>
<td>2ml</td>
<td>7.33</td>
<td>4ml</td>
<td>6.41</td>
<td>3.5</td>
<td>7.89</td>
<td>20</td>
<td>08.45</td>
</tr>
<tr>
<td>22</td>
<td>10.25</td>
<td>3ml</td>
<td>7.89</td>
<td>6ml</td>
<td>7.00</td>
<td>4.5</td>
<td>10.44</td>
<td>25</td>
<td>10.89</td>
</tr>
<tr>
<td>24</td>
<td>11.05</td>
<td>4ml</td>
<td>8.44</td>
<td>8ml</td>
<td>8.21</td>
<td>5.5</td>
<td>10.98</td>
<td>30</td>
<td>11.46</td>
</tr>
<tr>
<td>26</td>
<td>11.55</td>
<td>5ml</td>
<td>11.63</td>
<td>10ml</td>
<td>11.69</td>
<td>6.0</td>
<td>8.42</td>
<td>35</td>
<td>06.00</td>
</tr>
</tbody>
</table>

**Figure.1 Alcoholic Fermentation of Reconstituted Papaya Wine by *S.cerevisiae* with Total Soluble Solids**

![Graph showing ethanol production with varying TSS](image-url)
Figure 2 Alcoholic Fermentation of Reconstituted Papaya Wine by S. cerevisiae with Total Soluble Solids

Figure 3 Alcoholic Fermentation of Reconstituted Papaya Wine by S. cerevisiae with Inoculum Level

Figure 4 Alcoholic Fermentation of Reconstituted Papaya Wine by S. cerevisiae with pH Level
Figure 5: Alcoholic Fermentation of Reconstituted Papaya Wine by S. cerevisiae with Temperature Level

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