



Physico-Chemical Analysis of Developed Rice Beer from *Oryza sativa*

Akansha Tiwari^{a++}, Neetu Singh^{a##} and Ayushi Singh^{a†}

^a Department of Food and Nutrition, BBAU, Lucknow, India.

Authors' contributions

This work was carried out in collaboration among all authors. Author AT designed the study, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Authors NS and AS managed the analyses of the study. All authors read and approved the final manuscript.

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ABSTRACT

Aim: Medicinal plants and their extracts represent a rich source of crude medications that possess therapeutic properties. Physico-chemical properties of two different rice varieties (Jandhan and Sugapankhi) of North India collected from Uttar Pradesh and a starter culture cake collected from Arunachal Pradesh used in the process of rice beer preparation were analyzed. This study evaluates the development of rice beer from *Oryza sativa* and physico-chemical analysis of developed rice beer.

Study Design: The present study was conducted in 3 phases-

1. Collection of raw materials
2. Production of rice beer
3. Physico-chemical analysis of developed rice beer

⁺⁺ Scholar;

[#] Associate Professor;

[†] Research Scholar;

*Corresponding author: Email: neetubbau@gmail.com;

Place and Duration of the Study: The present study was conducted in the Department of Food and Nutrition of Babasaheb Bhimrao Ambedkar University, Lucknow. The duration of the study is 3 months i.e. from Jan, 2023 - March, 2023.

Methodology: Rice beer was developed in the laboratory under optimum conditions through fermentation followed by filtration and distillation. Collection of the raw materials was done from the local markets of North India and North-east, India. In the present study we have compared physico-chemical properties between two rice varieties i.e. Jandhan and Sugapankhi.

Results: Various physico-chemical parameters (pH, TSS, total acidity, volatile acidity, turbidity, and alcohol content) of rice beer were determined. The pH decreased with storage time, and acidic pH was noted for the beer in ambient storage. An increase with time in the total acidity, volatile acidity, and alcohol % of rice beer was observed. The final recorded alcohol % value of rice beer was in the range of 8-10%. The alcohol content increased throughout the storage. Statistical analysis was performed for comparison between the two samples of rice. *Saccharomyces cerevisiae* isolated from the starter culture cake was found to be the major organism responsible for rice beer fermentation.

Conclusion: The preparation of rice beer needs scientific input for increasing its shelf-life and value addition for its marketing and improving its shelf-life for better quality and acceptability.

Keywords: *Oryza sativa*; rice beer; starter culture; North-East tribes; *Saccharomyces cerevisiae*.

1. INTRODUCTION

Rice beer is a traditional alcoholic beverage that plays a major role in the socio-cultural lives of people of North East India. Various ethnic tribes of North East India consume rice beer regularly in different forms. Many of them have been preparing it since time immemorial [1,2]. It plays an important role in the socio-cultural life of the tribal people as it is found to be associated with many occasions like merry-making, ritual ceremonies, festivals, and marriages (Saikia et al., 2007). These products are similar to shaosingiju and laochao of China, the sake of Japan, chongju and takju of Korea, brembali, tape-ketan and tapuy of Indonesia, khaomak of Thailand and tapaipulul of Malaysia.

The North-east India is inhabited by many ethnic tribes having diverse customs and life styles with difference in their preparation and consumption of fermented rice beer [3-5]. The occurrence of ethnic rice beer preparation and its varieties in consumption might have emerged due to varied climatic conditions and utilization of locally available natural resources. All of the tribes prepare their indigenous alcoholic beverages at home using round to flattened solid ball-like mixed dough inocula or starter [6,7] and “these contain amylolytic and alcohol producing yeasts, starch degrading moulds and lactic acid bacteria” (Dung et al.,2006). “The preparation and consumption of this type of liquor emerged mainly due to the climatic conditions and discovering the use of surrounding natural resources” [8,9]. “Plants are the primary source

for different pharmaceutical, perfumery, flavor, and cosmetics industries; the use of modern drugs dramatically resulted into resistant microorganisms toward different modern drugs; the researchers are now in search for alternate source of treatment of various disorders. For this purpose, the medicinal herbs are the best alternate to various drugs. Most of natural products possess interesting biological activities and medicinal potential. There are also reports of rice beer being used as a drug” [10,11]. “It works effective against insomnia, headache, body ache, inflammation of body parts, diarrhea and urinary problems, expelling worms and as treatment of cholera” [12,13]. These traditional rice beers that are prepared are reported to have medicinal and therapeutic properties and hence are considered as nutraceuticals.

The processes of preparation of rice beer begin with the making of a starter culture for fermentation. Starter cultures are traditionally made and preserved in a semi-sterilized medium. Usually, gluten-free rice of local variety is mixed with several plant parts having medicinal properties to make the starter cake [14]. For natural antioxidants, a larger number of medicinal herbs have been evaluated by applying laboratories’ developed procedures. Plants derived substances, collectively called phytonutrients or phytochemicals, been recognized as good source of natural antioxidants. The methodologies of fermentation followed by different tribes of these areas are not similar in many respects. Local rice varieties are used as the substrate for the preparation of

starter cakes along with some local plants and ingredients owes different appearance, taste, and aroma along with medicinal properties. The starter cakes are the source of amylolytic enzymes, starch-degrading moulds, lactic acid bacteria, and alcohol producing yeasts. Several microbes have been isolated like *Mucorcircinelloides*, *Rhuschinensis*, *Saccharomycopsis capsularis*, and *Pichia burtonii* having amylolytic properties.

The objective of this study is: to prepare developed rice beer from two varieties of *Oryza sativa* by *Saccharomyces* sps and to analyse the physico-chemical properties of developed rice beer.

2. METHODOLOGY

The methodology of starter culture cake preparation and time of incubation for a batch culture of rice beer is analysed. The present study is under taken to assess the major physico-chemical parameters (pH, TSS, total acidity, volatile acidity, turbidity, and alcohol content) for beer samples prepared in optimum laboratory conditions using two different rice varieties (Jandhan and Sugapankhi) in order to explore the possible potential of rice beer as a beneficial fermented beverage. *Saccharomyces cerevisiae* is responsible for rice beer fermentation.

2.1 Collection of Materials

Rice varieties were collected from the local market of Lucknow and starter culture cake was collected from the local market of Arunachal Pradesh. The collection was made from the location which was predominantly involved in the process of making rice beer, either for self-consumption or for commercial purposes. The starter culture cake was collected in replicates of 500mL sterile glass sample bottle (Borosil, India) marked according to the place of collection, brought to the laboratory under refrigerated condition.

2.2 Production of Rice Beer in the Laboratory

Collected Rice varieties (Jandhan and Sugapankhi) were used in the laboratory-scale production of rice beer, whereas only one type of starter culture was used during this process.

Rice beer was developed under optimum conditions, weighing the rice varieties and starter

cake by weighing machine, soaking both the varieties of rice, cooking them at room temperature. Fermentation of both the samples takes place in an incubator for 15 days followed by filtration and distillation process.

2.2.1 Weighing

1 kg of rice (Jandhan and Sugapankhi) and 5gm of starter cakes were weighed respectively.

2.2.2 Soaking

The rice varieties were soaked in water in two different utensils for about ten minutes at room temperature.

2.2.3 Cooking

Both varieties of rice were cooked for 20 minutes. This was followed by cooling of the rice to room temperature.

2.2.4 Mixing

The starter cakes were powdered in a clean mortar and pestle and then mixed with 1kg of boiled rice at a ratio of 5gm per kilogram of rice. This mixture was transferred to sterile glass containers. This was done for both the varieties of rice.

2.2.5 Fermentation

Fermentation was allowed to take place at 30 degrees C in an incubator for 15 days for both the samples.

2.2.6 Filtration and Distillation

After the completion of fermentation, the produce was strained using a muslin cloth, and the filtrate was further diluted with distilled water in a 1:1 ratio. After dilution with distilled water the rice beer was filtered using filter paper (Whatman Cat No: 1001125) provided by the department.

2.3 Physico-chemical Analysis of Rice Beer

The pH of the samples was measured by a digital pH meter (Eutech Instruments, Model LT-501) equipped with a glass electrode. The sample's Total Soluble Sugar (TSS) was measured by a digital refractometer (Milwaukee, MA871, 0-85% Brix). The alcohol percentage of both samples was determined by the specific

gravity. The acidity of the samples was measured in two forms. The first one was Total acidity i.e. expressed as % of tartaric acid in the given sample and second was Volatile acidity i.e. expressed as % of acetic acid present in the sample. These were determined by

titration tests of fermenting rice beer. Changes in total acidity, volatile acidity, and alcohol content of the produced ricebeer were analyzed for 5 days. The turbidity of the samples was measured by an instrument called a turbidity meter.

Conceptual framework of developed rice beer



Raw Ingredients



a) First Variety is Jandhan b) Second Variety is Sugapanki c) Starter Cakes

Step 1: Weighing



a) Weigh 1Kg Sample 1 b) Weigh 1Kg Sample 2



a) Weigh 5gm Sample 1 b) Weigh 5gm Sample 2

Step 2: Soaking



a) Soaking Sample 1 b) Soaking Sample 2

Step 3: Cooking



a) Boiling Rice For Sample 1 b) Boiling Rice For Sample 1

Step 4: Mixing



a) Starter Cake in Powder Form For Sample 1 b) Starter Cake in Powder Form For Sample 2



a) Mixing powdered starter cake in Sample 1 b) Mixing powdered starter cake in Sample 2

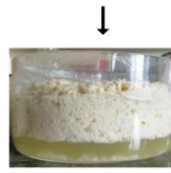
Step 5: Fermentation



a) Fermentation was allowed in an incubator for Sample 1 b) Fermentation was allowed in an incubator for Sample 2



a) Fermentation was allowed for 15 days



a) Fermentation was allowed for 15 days

Step 6: Filtration



a) Sample 1 was strained



b) Sample 1 was strained



a) Sample 1 before filtration



b) Sample 1 before filtration

Step 7: Distillation



a) Filtering Sample 1 using distilled water



b) Filtering Sample 2 using distilled water

Flow chart 1. Conceptual framework of developed rice beer

Table 1. Physico-chemical analysis of rice beer

Samples	pH	TSS	Total acidity (%)	Volatile acidity (%)	Turbidity	Alcohol content (%)
Sample1	3.46±0.012	4.6±0.081	1.25±0.012	2.26±0.012	5.1±0.081	8.49±0.012
Sample2	4.36±0.124	5.1±0.081	1.45±0.016	2.35±0.024	5.2±0.163	10.06±0.418
t-calculated	6.16*	0.04	0.08	0.68	0.72	08.38*

*denotes t- calculated value differ significantly at $P=0.05$ between two samples (sample1 and sample2) of rice beer.

(t- tabulated at $P=0.05=4.30$ at $df=2$)

3. RESULTS AND DISCUSSION

The changes in the various parameters in the laboratory-prepared rice beer were analyzed for 5 days under optimum laboratory conditions. Two varieties of rice (Jandhan and Sugapankhi) each of 1kg were used in the preparation of rice beer. Jandhan and Sugapankhi varieties were named Sample1 and Sample 2 respectively. The physico-chemical analysis of various parameters is shown in Table 1.

Sample 1 produced 600 ml of rice beer while sample2 produced 500 ml of rice beer.

Sample1 was found to have a pH value of (3.46±0.012) while sample2 showed a value of (4.36±0.124). "The pH values were in an acidic range of <3.0 which inhibits the growth of Coliforms and other members of the Enterobacteriaceae as previously reported (Mayer et.al, 2014) and (BHUYAN et.al, Biochemical and Nutritional analysis of Rice Beer of North East India)".

Sample 1 was found to have total soluble sugar (TSS) value (of 4.6±0.081) while sample2 showed a value (of 5.1±0.081) as previously reported (Narzi and Back, 2012b).

Sample 1 was found to have a total acidity value of (1.25±0.012) (% tartaric acid) while sample 2 showed value (1.45±0.016) (% tartaric acid).

Similarly, the volatile acidity of Sample 1 showed value (2.26±0.012) (% acetic acid) while sample 2 showed value (2.35±0.024) (%acetic acid).This indicates that the total acidity and volatile acidity increase with storage time as the fermentation proceeds.

The significant increase in alcohol content of sample 1 was observed (8.49±0.012) while sample 2 showed a value of (10.06±0.418). The alcohol content increased throughout the storage as previously reported (Mayer et.al, 2014) and

(BHUYAN et.al, Biochemical and Nutritional analysis of Rice Beer of North East India).

Sample 1 showed a turbidity value (of 5.1±0.081) while sample 2 showed (5.2±0.163).

4. CONCLUSION

Rice beer is a traditional alcoholic beverage prepared and consumed by almost all the ethnic tribes of North-east India. It is a common tradition to consume the beverage as a part of their socio-cultural development. The mode of preparation and formulation varies from tribe to tribe while the primary ingredient of the traditional beverage is rice (*Oryza sativa*) from which rice beer is prepared. The preparation of rice beer needs scientific input for increasing its shelf-life and value addition for its marketing and commercialization with increased acceptability by the common people. Rice beer was developed under optimum laboratory conditions using *Oryza sativa* as the primary ingredient. Physico-chemical analysis of rice beer produced by two varieties (Jandhan and Sugapankhi) was done to check the parameters (pH, TSS, total acidity, volatile acidity, turbidity, and alcohol content). These parameters play a significant role in the aging of rice beer. The findings show that there is an urgent need and sufficient scope for the enrichment and improvement of alcohol as well as the traditional varieties of rice beer with proper scientific input and research to improve shelf life, stability, and aging. There is ample of scope to validate the beneficial properties of rice beer and to standardize various parameters offer mentation for the commercialization of this staple drink.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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