# Assessment of Microbial Activity, Shelf Life, and Sensory Characteristics of Buttermilk Enhanced with Indian Prickly Ash (*Zanthoxylum rhetsa*) Extract

# Ashutosh Ghonge<sup>1</sup>, Dr. Sandip Ramod<sup>2</sup>, Dr. Mukund Patond<sup>3</sup>, and Dr. Vijay Kadav<sup>4</sup>

#### Abstract

The present investigation was carried out at the Dairy Science Laboratory of the Department of Animal Husbandry and Dairy Science, College of Agriculture, Dr. BSKKV., Dapoli (M.S.) to Assessment of Microbial Activity, Shelf Life, and Sensory Characteristics of Buttermilk enhanced with Indian Prickly Ash (Zanthoxylum rhetsa) extract.

This study aimed to evaluate the microbial activity, shelf life, and sensory attributes of Indian Prickly Ash extract fortified buttermilk during storage under refrigerated conditions. Microbial analysis was conducted on fresh and stored samples over 15 days, focusing on standard plate count, coliform count, yeast, and mold count. Results indicated that the initial standard plate count of fresh buttermilk increased marginally during storage. Shelf-life assessment revealed that the buttermilk remained suitable for consumption for up to 15 days under refrigerated conditions.

Sensory evaluation was performed concurrently with microbial analysis. Overall acceptability scores were highest for buttermilk fortified with 6% Indian Prickly Ash extract, followed by treatments with lower extract concentrations. Statistical analysis showed significant differences among treatments, with 6% extract incorporation yielding the most favorable sensory outcomes.

In conclusion, Indian Prickly Ash extract fortified buttermilk, particularly at 6% concentration, exhibited enhanced microbial stability and sensory appeal, suggesting its potential as a functional food product.

Keywords: Milk, Buttermilk, Indian Prickly Ash.

#### Introduction

Buttermilk, esteemed for its taste and nutritional value, is a staple in many cuisines due to its versatility and potential health benefits. Despite its popularity, maintaining its microbial quality and sensory appeal over time poses a challenge. To address this, we investigate the use of Indian Prickly Ash (*Zanthoxylum rhetsa*) extract to fortify buttermilk. Indian Prickly Ash is known for its antimicrobial properties and ability to enhance taste.

In recent years, there has been a growing interest in utilizing natural bioactive compounds to enhance the nutritional profile and sensory attributes of food products. Indian Prickly Ash extract, derived from the Zanthoxylum rhetsa plant, has garnered attention for its potential health benefits and culinary applications. Rich in phytochemicals such as alkaloids and flavonoids, this extract possesses antioxidant and antimicrobial properties, making it a promising candidate for fortifying dairy products like buttermilk.

Incorporating Indian Prickly Ash extract into buttermilk not only offers the potential to extend its shelf life by inhibiting microbial growth but also presents an opportunity to enhance its flavor profile and overall sensory experience. By tapping into the synergistic effects of bioactive compounds present in the extract, we aim to unlock new possibilities for creating healthier and more palatable dairy products that align with consumer preferences for natural, functional foods. Through rigorous scientific investigation, we seek to uncover the optimal formulation that balances microbial stability, sensory acceptance, and nutritional value, thus contributing to the advancement of both food science and consumer well-being.

# Material and Method

Flow chart of making Indian Prickly Ash extract:

Indian Prickly Ash 60g Washing and cleaning Soaking in water for 24 hours (1000ml water) Churning and Filtration Indian Prickly Ash (*Zanthoxylum rhetsa*) extract

Fully ripened Indian Prickly Ash were selected. The Indian Prickly Ash was washed with running tap water to remove dirt and dust. 60g of Indian Prickly Ash was added in 1000 ml of clean fresh water and kept for soaking for 24 hours. Next day it was grinded and filtered and made the extract. Extract obtained used to mix at different level during buttermilk preparation.



Flow chart of making Indian Prickly Ash extract Buttermilk: Receiving Milk Preheating  $(35-40^{\circ} \text{ C}/5 \text{ minutes})$ Filtration/Clarification Pasteurization (80-90<sup>°</sup> C/ 15 minutes) Cooling (35-37<sup>0</sup> C) Addition of starter culture@ 1.5% (w/v basis) Proper mixing Incubation 37<sup>°</sup>C for 8 hours Dahi Addition of water 1:1 proportion Churning Plain Buttermilk Addition of Salt @ 0.5 and Indian Prickly Ash (Zanthoxylum rhetsa) extract as per treatment Mixing Storage (5-10° C)

# Microbial and Shelf-Life Study:

The most effective concentration determined during the investigation underwent microbial analysis and shelf-life assessment over a refrigerated period of 15 days. The parameters studied included Standard Plate Count (SPC), Yeast and Mold count, and Coliform count. The product was evaluated at intervals of 0, 5, 10, and 15 days of storage.

# 1. Microbial Study:

## I) Standard Plate Count:

One milliliter of sample from 10-4 dilutions was aseptically transferred onto sterilized Petri dishes using sterilized pipettes. Subsequently, 10 milliliters of plate count agar medium were added to each dish at 45°C. After thorough mixing and uniform spreading, the dishes were allowed to solidify at room temperature for 10 to 15 minutes. Duplicates of each dish were then incubated at 37°C±10°C for 48 hours. Following incubation, colonies were enumerated using a colony counter, and microbial counts were expressed as colony-forming units per milliliter (cfu/ml).

## II) Yeast and Mold Count:

One milliliter of sample from 10-1 and 10-2 dilutions was aseptically dispensed onto sterilized Petri dishes using sterilized pipettes. Subsequently, 10 milliliters of potato dextrose agar at 45°C was added to each dish. After thorough mixing and uniform spreading, the dishes were allowed to solidify at room temperature for 10 to 15 minutes. Duplicates of each dish were then incubated at 37°C±10°C for 48 hours. Following incubation, colonies were enumerated using a colony counter, and yeast and Mold counts were expressed as colony-forming units per milliliter (cfu/ml) of milk.

# **III) Coliform Count:**

One milliliter of thoroughly mixed and macerated sample was aseptically dispensed onto sterilized Petri dishes using sterilized pipettes. Violet red bile agar (VRBA) was then added to each dish and mixed thoroughly before allowing the plates to solidify. The plates were then overlaid with the same agar and allowed to solidify. Subsequently, the plates were incubated at 37°C for 24 hours, and the number of coliform colonies formed was enumerated using a colony counter, expressed as colony-forming units per milliliter (cfu/ml) per gram.

#### 2. Sensory Evaluation:

Fresh samples of Indian Prickly Ash extract Buttermilk underwent organoleptic assessment using a nine-point hedonic scale. A panel consisting of approximately 8-10 semi-trained judges evaluated

various quality attributes, including general appearance, consistency, and flavour. Judges were provided with hedonic scorecards to rate the quality of the product, following the guidelines outlined in IS: 6273 (Part- II), 1971.

### Results

# 1. Microbial Analysis of Fresh and Stored Indian Prickly Ash Extract Buttermilk:

The most effective concentration discovered during the study was stored under refrigerated conditions for up to 15 days. Microbial characteristics of the product were assessed on days 0, 5, 10, and 15 of storage, concurrently with sensory evaluation. The parameters investigated included standard plate count, coliform count, yeast, and mold count. Table 1.1 presents the results of microbial evaluation for the most acceptable level of Indian Prickly Ash extract buttermilk at various time intervals.

# Table 1.1: Microbial evaluation of most acceptable level of Indian Prickly Ash extract buttermilk at various time intervals.

Microbial	Standard plate	Coliform count	Yeast and	Overall	
parameter	count	(10 <sup>1</sup> cfu/g)	Mold count	acceptability	
(Days)	(10 <sup>4</sup> cfu/g)		(10 <sup>1</sup> cfu/g)		
D <sub>0</sub>	4.03	ND	ND	8.3	
D5	4.12	ND	ND	7.8	
D <sub>10</sub>	4.21	ND	ND	6.7	
D <sub>15</sub>	4.27	ND	ND	5.9	

Upon careful examination of the data in Table 1.1, it was noted that the initial standard plate count of Indian Prickly Ash extract Buttermilk was  $4.03 \times 10^4$  cfu/g, which increased to  $4.27 \times 10^4$  cfu/g in the stored sample.

The microbial analysis and shelf-life study indicate that Indian Prickly Ash extract buttermilk remains in good edible condition for up to 15 days under refrigerated conditions.

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#### 2. Sensory Evaluation:

The overall acceptability of Indian Prickly Ash extract buttermilk was determined based on the average scores recorded for different sensory attributes, including colour and appearance, flavour, and consistency.

	R-I	R-II	R-III	R-IV	R-V	R-VI	Mean
T <sub>0</sub>	6.47	6.56	6.49	6.37	6.52	6.56	6.50°
T <sub>1</sub>	7.26	7.28	7.37	7.29	7.31	7.29	7.30 <sup>b</sup>
<b>T</b> <sub>2</sub>	7.63	7.74	7.39	7.68	7.71	7.74	7.65 <sup>a</sup>
<b>T</b> 3	7.20	7.42	7.22	7.33	7.37	7.32	7.31 <sup>b</sup>
T <sub>4</sub>	7.16	7.16	7.08	7.23	7.23	7.17	7.17 <sup>ab</sup>
Mean	7.144	7.232	7.11	7.18	7.228	7.216	7.185

#### Table 2.1: Overall acceptability of Indian Prickly Ash extract buttermilk (Nine-point hedonic scale)

The results, presented in a table 2.1, indicate that at a 6 percent addition of Indian Prickly Ash extract ( $T_2$ ), the product achieved the highest score of 7.65, followed by treatments  $T_3$ ,  $T_1$ , and  $T_4$ . Conversely, the control treatment ( $T_0$ ) received the lowest score of 6.50.

A consistent increase in overall acceptability scores was observed with the addition of Indian Prickly Ash extract at 4 percent and 6 percent levels. However, a declining trend in overall acceptability was noted with 8 percent and 10 percent additions of Indian Prickly Ash extract.

Statistical analysis revealed that the results were statistically significant at a 1 percent level of probability. Treatments  $T_1$  and  $T_3$  were found to be comparable, while  $T_0$ ,  $T_2$ , and  $T_4$  differed significantly from buttermilk fortified with Indian Prickly Ash extract.

The investigation demonstrated that the proportion of Indian Prickly Ash extract at 6 percent  $(T_2)$  was widely accepted by judges and obtained the highest score for overall acceptability. Therefore, treatment  $T_2$  was considered the most acceptable level.

#### **Conclusion:**

In conclusion, the microbial and sensory evaluation of Indian Prickly Ash extract buttermilk revealed valuable insights into its quality and shelf-life characteristics. The most effective concentration of Indian Prickly Ash extract demonstrated satisfactory microbial stability and sensory acceptance when stored under refrigerated conditions for up to 15 days. Microbial analysis, including standard plate count, coliform count, yeast, and mold count, indicated that the buttermilk remained within acceptable limits throughout the storage period, highlighting its suitability for consumption. Furthermore, sensory evaluation results emphasized the importance of Indian Prickly Ash extract concentration in influencing overall acceptability. Treatment with 6 percent addition of Indian Prickly Ash extract (T2) garnered the highest overall acceptability score, followed by treatments T3, T1, and T4. Conversely, the control treatment (T0) received the lowest score, indicating the beneficial impact of Indian Prickly Ash extract on sensory attributes.

Statistical analysis confirmed the significance of the observed differences in overall acceptability scores among treatments, with T2 being identified as the most acceptable level. These findings underscore the potential of Indian Prickly Ash extract as a functional ingredient for enhancing the microbial stability and sensory quality of buttermilk, offering opportunities for the development of innovative dairy products with improved nutritional and sensory profiles.

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Author Details-

# 1. Ashutosh Ghonge

M.Sc. Student, Department of Animal Husbandry and Dairy Science, College of Agriculture, Dr. B. S. K. K. V, Dapoli, Maharashtra, India.

Email id- ashutoshghonge84@gmail.com

## 2. Dr. Sandip Ramod

Assistant Professor, Department of Animal Husbandry and Dairy Science, College of Agriculture, Dr. B. S. K. K. V, Dapoli, Maharashtra, India.

Email id- sandipramod28081981@gmail.com

## 3. Dr. Mukund Patond

Assistant Professor, Department of Animal Husbandry and Dairy Science, Anand Niketan College of Agriculture, Warora, Maharashtra, India.

Email id- mukundpatond@gmail.com

## 4. Dr. Vijay Kadav

Assistant Professor, Department of Animal Husbandry and Dairy Science, College of Agriculture, Dr. B. S. K. K. V, Dapoli, Maharashtra, India.