Preparation and Comparative Evaluation of LP Treated Shelf-life Enhanced (SLE) Khoa and Fresh Milk Khoa

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ABSTRACT

The Konkan tract of Maharashtra is small narrow strip of about 700 km length with average breadth of 30-35 km. This tract is embodie with Arabian Sea towards west and ranges of Sahyadri Mountain towards east. This region is well known for typical hot- humid climate for about 8 to 9 months of the year. Dairy industry is setting its foot hold in this region. However, problems like accessibility to market, lack of cold storage facilities are poised as hurdles in maintaining milk quality. The hot humid climate coupled with frequent electricity failures and negligence for quality aggravates this problem further.

Although during this study Attempts were made to boost this natural system by addition of hydrogen peroxide (H2O2) and thiocyanate. This method got tremendous success and is recommended by SAARC for rural hilly areas. The lacto peroxidase (LP) system, when properly applied, is harmless to the mammalian cells.

Similarly, considerable work is carried out on utilization of such LP treated milk for preparation of various milk products. Present research work is therefore, aimed at exploring the possibility of LP treated shelf-life enhanced (SLE) milk for preparation of indigenous desiccated milk product like khoa with main objectives of exploring the possibility of preparing khoa from LP treated shelf-life enhanced (SLE) milk, to study the physicochemical, microbiological and sensory parameters of khoa prepared from LP treated SLE milk and to compare them with khoa prepared from fresh whole milk, to Study the shelf-life of khoa prepared from LP treated SLE milk and fresh whole milk at storage temperature of 24±1°C, to work out the cost of production.

Keywords: Lacto peroxidase, Shelf life enhanced milk, Dairy, Khoa.

INTRODUCTION

Milk plays an important role in the diet of most of the people in the world.¹ It is a well-accepted fact that milk is the most precious ideal liquid food in the nature because, it not only contains the various nutrients essential for human growth, but also contains them in proper proportions and in easily digestible and assailable form required by the body for maintaining the good health.²

Dairy industry in India is today poised for dynamic growth. The dairy farming plays a significant role in shaping the Indian economy. In the year of the independence, we started with a base line milk production of 17 million tonnes per year and according to Food and Agricultural Organization (FAO) in 2005-06, we have surpassed 90 million tonnes, which amount to almost more than one million tones increases every year. Today, we are largest milk producer in the world with 187.7 MT of milk production with present growth rate of 5.5 per cent per annum and per capita availability of 394 gm per day (2018-19); India is expected to produce 250 to 300 million tonnes of milk by the year 2022. At present, India produces half of the Asian milk output. The peculiarity about Indian...
The dairy industry is higher proportion of small milk producers. Thousands of milk producers spread over length and breadth of the country contribute to national milk grid in the range of 2 to 5 lit/day. [3]

Though, India has secured first position in milk production in the world, it is only in quantity and not in quality. The spoilage of milk because of bacterial action is estimated to be 10 per cent of the total milk production in India. [4] It is estimated that due to poor milk quality management, our country bears whopping annual loss of Rs 5,500 crore every year. [5] In terms of quality of milk and productivity of dairy animals, India ranks low among the major dairy nations. Dairy production in tropical countries is hindered by accelerated milk spoilage due to poor production techniques, handling, and storage facilities at farms. [6] High ambient temperature aggravates this problem. Over-night storage of milk on the farm has created problems like change in acidity and it makes the milk unfit for consumption as well as for product manufacturing. [7] Considerable attempts were made for enhancing the shelf life of milk so as to prevent financial losses being faced by dairy entrepreneurs. [8]

Food and Agriculture Organization (FAO), a statutory body of United Nations has also echoed serious concern over the quality of milk produced and financial setbacks, poor and marginal farmers received due to it. FAO categorically mentioned need to tackle this problem by developing easily adoptable method to enhance shelf-life of milk at farm. [9]

To increase the shelf life by lowering the bacterial load of raw milk various methods are in use. In the villages the milk is collected only once in a day i.e. in the morning for the processing at government dairy units. [10] The evening milk is mechanically sterilized by heating which deteriorates the chemical quality of milk and then sold to the dairies in the morning which either fetches lower price or not accepted by the dairy units because of lower and non-homogenous distribution of fat and SNF due to skim formation during mechanical sterilization of milk. [11]

Shelf-life of milk at farm or at milk collection centre can be enhanced by reducing rate of microbial multiplication in milk. In plant sciences a two way approaches is adopted for control of pest and diseases i.e. preventive and curative approach. [12] Similar approach is required to be adopted for producing milk with superior microbiological, chemical and sensory qualities. The raw fresh milk contains some anti-microbial components, which inhibits the microbial growth in milk, due to heat treatment some of these anti-microbial components get destroyed, similarly the heat treatment increases the operation cost and ultimately it affects the cost of milk besides altering its chemical nature. [13]

**MATERIAL AND METHODS**

**Material**

All the chemicals used were of analytical grade. Hydrogen peroxide and potassium thiocyanate were procured from Sd fine chemicals, Mumbai. Agar and other microbiological material was procured from Hi-Media.

**Selection of animals**

Healthy six milch animals free from mastitis or any other disease were selected from the milch herd. While selecting animals, cows in early or late lactation were avoided. The animals selected were crossbreds having 50 to 62.5 percent exotic inheritance with yielding capacity of 5 to 7 lit per milking, the animal selected was between 2nd to 5th stages of lactation.

**Collection of milk**

After taking all prerequisite precautions milk was collected in six different pre-disinfected milk cans of 1 lit capacity along with 7th can having control sample i.e. milk obtained from animal by adopting normal pre-milking cleaning procedures. The milk samples were then taken to laboratory and cans were kept in incubator at 22±1oC. After a period of two hours from milking, milk was poured in pre-sterilized milk cans.
sampling bottles. About 75 to 80 ml of milk was poured in each bottle by removing Aluminum foil cap and then again capped properly. These sample bottles were again placed in incubator at 22±1 °C. The incubator was switched on for one hour prior of keeping the cans, to reach the temperature at desired level.

**Preparation of stock solutions**

**Preparation of hydrogen peroxide (H2O2) stock solution**
The stock solution on v/v basis having strength equivalent to 10,000 ppm (1%) was prepared by dissolving 1 ml of AR grade hydrogen peroxide in 100 ml double glass distilled water. Fresh stock solution was prepared at the beginning of each trial in phase II and phase III.

**Preparation of potassium thiocyanate (KSCN) stock solution**
The stock solution having strength equivalent to 10,000 ppm (1%) KSCN on w/v basis was prepared by dissolving 1.7g of potassium thiocyanate in 100 ml double glass distilled water. Fresh stock solution was prepared at the beginning of each trial in phase II and phase III.

**LP System activation**
Milk was treated by using hydrogen peroxide and potassium thiocyanate in 40:25 ppm proportion to enhance shelf life. This treatment was carried out 2 hrs after milking. LP System activated milk was stored at 24±1°C.

**Treatments**
T0 – Preparation of khoa from fresh milk.
T1 – Preparation of khoa from SLE milk (10 hrs storage).
T2 – Preparation of khoa from SLE milk (12 hrs storage).

**Preparation of khoa:** Khoa was prepared by traditional open pan (Karahi) method from fresh as well as shelf-life enhanced (SLE) milk by concentration process. [12]

Packaging & storage of khoa: Khoa obtained was packed in parchment paper and stored in incubator at 24+1°C for different periods.

**Preparation of different media**

These dehydrated media were dissolved in prescribed amount of double distilled water in the prescribed proportion.

**Standard plate count:** The medium labeled as Plate Count Agar was used for this purpose. The dehydrated medium was dissolved in the proportions of 17.5g for 1000 ml distilled water, cooked, distributed in 10 ml quantity in test tubes and autoclaved at 121oC for 15 minutes.

**E. coli count:** The medium labelled as Violet Red Bile Agar was used for enumeration of E.coli in milk. The dehydrated medium was dissolved in the proportions of 41.53g in 1000 ml distilled water then cooked and filled in test tubes (10 ml) and autoclaved at 121oC for 15 minutes.

**Proteolytic count:** The medium labeled as Caseinate Agar was used for detecting proteolytic bacteria in milk. The dehydrated medium was dissolved in proportions of 40.13g in 1000 ml distilled water. It was heated to dissolve the medium completely. After the proper cooking it was sterilized by autoclaving at 15 lbs pressure (121oC) for 15 minutes.

**Lipolytic count:** The medium labeled as Spirit Blue Agar was used for estimation of lipolytic bacteria in milk. The dehydrated medium was dissolved in proportions of 32.15g in 1000 ml distilled water. It was heated to dissolve the medium completely. After the proper cooking it was transferred to the test tubes at 10 ml/tube and sterilized by autoclaving at 15 lbs pressure (121oC) for 15 minutes.

**Yeast and mould count:** The medium labeled as yeast malt extract agar was used. This dehydrated medium was dissolved in proportions of 45g in 1000ml distilled water. After the proper cooking it was sterilized by autoclaving at 15 lbs pressure (121oC) for 15 minutes.

**Analysis of khoa**
Either fresh or SLE milk was used for preparation of khoa. Khoa obtained was analyzed for following parameters immediately after production and 10 and 12 hrs of storage.
Physicochemical parameters like percent titrable acidity of milk expressed as per cent lactic acid was determined according to IS: 1479 (Part I) –1960. Percent fat was determined according to IS: 1479 (Part I) 1960. Percent total solids was determined. Microbiological parameters like standard plate count, coliform count, lipolytic count, proteolytic count and yeast and mould count was analysed as per IS: 1479 (Part III) 1962. Organoleptic evaluation such as colour, flavour and appearance were estimated by using 9 point Hedonic scale. [14]

Table 1: Hedonic ratings obtained

<table>
<thead>
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<th>Sr. No.</th>
<th>Remark</th>
<th>Score</th>
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<td>Like extremely</td>
<td>9</td>
</tr>
<tr>
<td>2</td>
<td>Like very much</td>
<td>8</td>
</tr>
<tr>
<td>3</td>
<td>Like moderately</td>
<td>7</td>
</tr>
<tr>
<td>4</td>
<td>Like slightly</td>
<td>6</td>
</tr>
<tr>
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<td>Neither like nor dislike</td>
<td>5</td>
</tr>
<tr>
<td>6</td>
<td>Dislike slightly</td>
<td>4</td>
</tr>
<tr>
<td>7</td>
<td>Dislike moderately</td>
<td>3</td>
</tr>
<tr>
<td>8</td>
<td>Dislike very much</td>
<td>2</td>
</tr>
<tr>
<td>9</td>
<td>Dislike extremely</td>
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</tr>
</tbody>
</table>

Statistical Analysis

The data were tabulated and analyzed by employing completely randomized design (CRD) using three treatments with eight replications.

RESULT AND DISCUSSION

The trial was conducted in duplicate with same procedure and with eight replications. T0, T1 and T2 treatments were subjected to physicochemical, microbiological and sensory parameters and the data obtained was analysed as per the prescribed statistical method. The cost of khoa production was calculated considering the prevailing market rates of the material.

The physicochemical parameters for T0, T1 and T2 treatments were found with percent titrable acidity of 0.155, 0.181 and 0.202 respectively, fat content of 4.427, 4.302 and 4.190 and percent total solids content of 13.40, 12.56 and 11.79 respectively.

The microbiological parameters such as standard plate count for T0, T1 and T2 treatments was found as 16.34 x 105, 26.39 x 105 and 44.01 x 105cfu/ml respectively.

The sensory parameters such as colour and appearance for T0, T1 and T2 treatments was observed as 8.59, 7.94 and 7.73 respectively by 9 point hedonic scale and flavour as 8.68, 8.26 and 7.74 respectively by 9 point hedonic scale. These parameters were repeated for second batch.

The physicochemical parameters T0, T1 and T2 treatments were found with percent titrable acidity of 0.402, 0.411 and 0.416, fat content of 22.26, 22.14 and 22.09 and percent total solids content of 62.17, 62.19 and 62.07 respectively.

The microbiological parameters such as standard plate count for T0, T1 and T2 treatments was found to be 13.53 x 105, 13.16 x 105 and 11.52 x 105 cfu/gm respectively, coliform count was found to be 2.908 x 102, 2.854 x 102 and 2.824 x 102 cfu/gm, lipolytic count 2.16 x 102, 2.04 x 102 and 1.88 x 102 cfu/gm, proteolytic count 3.16 x 102, 3.00 x 102 and 2.64 x 102 cfu/gm and yeast and mould count 3.01 x 103, 3.00 x 103 and 2.84 x 103 cfu/gm respectively.

The sensory parameters such as colour and appearance for T0, T1 and T2 treatments was found to be 7.22, 7.28, 6.52 estimated by 9 point hedonic scale.

From these parameters it is clear that as the storage period increases, bacterial flora also increases.

From the results it is evident that, physicochemical parameters are concerned, acidity goes on increasing whereas fat and total solids content goes on decreasing as the storage period progresses.

The cost of production of khoa prepared from fresh as well as from SLE milk of 10 and 12 hr storage was calculated.

The cost of khoa production was estimated considering the cost of ingredients and preservatives used in preparing khoaviz., milk, hydrogen peroxide, potassium thiocyanate and the yield of khoa, Average cost of khoain T0 (khoa prepared from fresh milk), T1 (khoa prepared from
SLE milk of 10 hr storage, T2 (khoa prepared from SLE milk of 12 hr storage) was worked out to Rs 144.44/-, Rs 161.11/- and Rs 170.58/- per kg respectively.

**CONCLUSION**

From the present study, it can be concluded that good and acceptable quality khoa can be prepared of LP treated SLE milk stored up to 12 hr of storage. Khoa prepared by using SLE milk was at par, in almost all physicochemical, sensory and microbiological parameters as compared to khoa prepared from fresh milk. The cost of khoa so obtained is slightly higher than khoa prepared from fresh milk.

The results affirms the fact that LP system activation helps in enhancing shelf life of milk and khoa prepared from such milk is by no means inferior in quality.

**REFERENCES**

2. www.nddb.coop.com


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