



IJFANS

International Journal of Food
And Nutritional Sciences

Volume 2, Issue 4, Oct-Dec-2013,

www.ijfans.com

e-ISSN: 2320-7876



Official Journal of IIFANS

SHRIKHAND - VALUE ADDED TRADITIONAL DAIRY PRODUCT

G. Swapna* and Suvarna V. Chavannavar

Department of Agricultural Microbiology, University of Agricultural Sciences, GKVK, Bangalore. Karnataka, India

*Corresponding Author: sapnamicro@rediffmail.com

ABSTRACT

The investigation was conducted to study the characterization of lactic acid bacterial isolates, physicochemical properties and sensory evaluation of shrikhand, a fermented dairy product prepared using cattle, buffalo and dairy milk by adding curd as well as lactic acid bacterial isolates and probiotics as a starter culture. The isolates obtained from shrikhand were tested for Gram's reaction, colony morphology, and biochemical tests like catalase, fermentation of carbohydrates, gelatin hydrolysis, spore production, growth on neutral red chalk agar and dextran production and were analysed for pH, residual sugars, total sugars and for titrable acidity. Result revealed that, all the lactic acid bacterial isolates were Gram +ve, rods and cocci, catalase -ve, could not hydrolyse gelatin and they showed acid and gas production and isolates from dairy milk (D₂ and D₃) produced dextran and the shrikhand prepared using probiotics in combination and with lactic acid bacterial isolates showed highest pH, residual sugars, total sugars and for titrable acidity. the highest score was recorded with combinations of probiotics *i.e.* *Lactobacillus acidophilus* + *Lactobacillus sporogens*, and the lowest score was recorded in *Lactobacillus rhamnosus* in terms of colour, appearance, aroma, texture, taste and overall acceptability of the product and lactic acid bacterial isolates using cattle milk isolates recorded highest score followed by dairy milk and buffalo milk.

Key words: Characterization, chemical composition, Lactic acid bacteria, probiotics, organoleptic evaluation.

INTRODUCTION

India's market potential and current growth rate of traditional dairy products is unparalleled and all set to boom further under the technology of mass production. An estimated 50 to 55 % of the milk produced in India is converted into a variety of traditional milk products, using processes such as coagulation, desiccation and fermentation. Indian fermented milk products utilize 7% of total milk produced and mainly includes three product dahi, shrikhand (sweetened concentrated curd) and lassi which may be considered the western equivalent to yogurt, quarg and stirred yogurt, respectively. Fermented milk products constitute a vital component of the human diet in many regions of the world. In the Indian sub-continent such products are also classified as "indigenous milk products" like dahi (curd), lassi, shrikhand etc. which are prominent in people's diet. Shrikhand is the indigenous fermented milk product prepared by the fermentation of milk by using known strain of lactic acid bacteria. Shrikhand is extensively used as a sweet dish after meals. It is also used as a festive sweet in India. Sugar is added as additive to the Shrikhand to enhance taste and does not have any preservative effect. Other natural additives like dried fruits are added to the shrikhand to enhance flavour. Shrikhand is traditionally made at home in western India. The name shrikhand is derived from the Sanskrit work "Shikharini". A lactic acid bacterium refers to a large group of beneficial bacteria that have similar properties

and all produce lactic acid as an end product of the fermentation process. They are widespread in nature and are also found in our digestive systems. Although they are best known for their role in the preparation of fermented dairy products. Lactic acid bacteria are therefore excellent ambassadors for an often maligned microbial world. They are not only of major economic significance, but are also of value in maintaining and promoting human health. Probiotic fermented milks, is one major segment amongst fermented milks that has tremendous potential for growth and development. Milk is an excellent medium to carry or generate live and active cultured dairy products. The technology of application of probiotic organisms in fermented dairy products aims to combine the potential health benefits of the bacteria with their ability to grow in milk, resulting in a nutritionally healthy and desirable product for the consumers. Therefore, the present study was undertaken with the objective to study the development of traditional dairy products with an addition of probiotics and lactic acid bacterial isolates as starter cultures on the quality of shrikhand by their physico-chemical characteristics.

MATERIAL AND METHODS

ISOLATION OF LACTIC ACID BACTERIA FROM SHRIKHAND

Lactic acid bacteria (LAB) were initially isolated

from shrikhand prepared using three different types of milk *i.e.* Cattle milk, buffalo milk and commercially available dairy milk by standard plate count technique using Mann, Rogosa and Sharpe's (MRS) agar medium (11). Lactic acid bacterial colonies *i.e.* cattle milk (C₁, C₃ & C₄), buffalo milk (B₁) and dairy milk (D₁, D₂ & D₄) thus obtained were further purified and were maintained in MRS broth for further preparation of shrikhand instead of using curd as starter culture. Then, the lactic acid bacterial isolates were further characterized for their biochemical changes. In similar fashion, the three different commercially available probiotics *i.e.* *Lactobacillus acidophilus*, *Lactobacillus sporogenes* and *Lactobacillus rhamnosus* are also used as starter culture alone or in combination of probiotics. (Fig. 1).

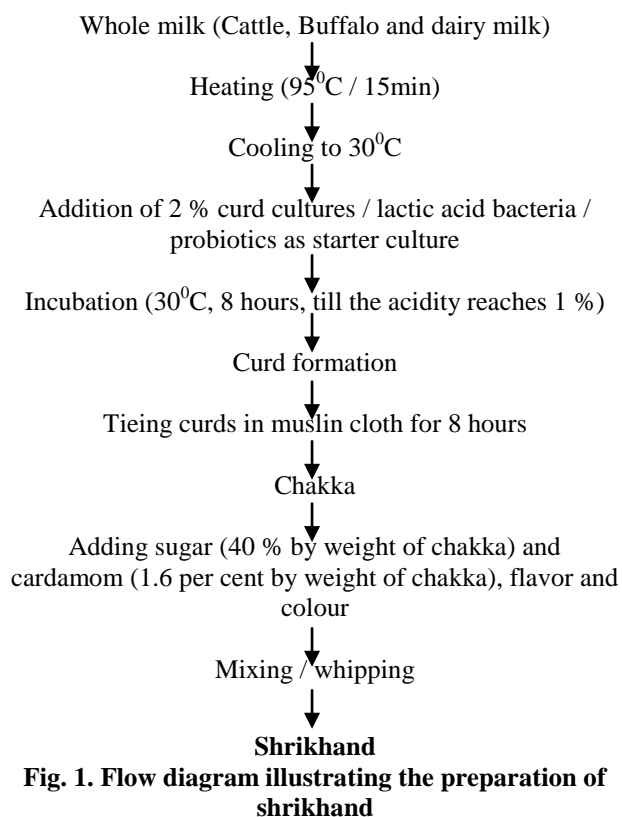


Fig. 1. Flow diagram illustrating the preparation of shrikhand

CHARACTERIZATION OF LAB ISOLATES AND CHEMICAL COMPOSITION OF SHRIKHAND

Lactic acid bacterial isolates were characterized based on the growth in MRS broth using biochemical tests *i.e.* Gram staining technique using 24 hours old culture, catalase production, fermentation of carbohydrates and spore production as suggested by Aneja (2). dextran production according to Garvie (6)., gelatin hydrolysis by Harrigan and Mc Cance (8) and growth on neutral red chalk agar medium as suggested by Chalmers (4). The shrikhand samples were analyzed for pH, titrable acidity, total sugars and reducing sugars. The pH of shrikhand samples was measured using digital pH meter of Anolog Model (Corin. Research USA).

TOTAL TITRABLE ACIDITY

Samples (10g) were diluted with 30 ml of water and were titrated against 0.1N NaOH using phenolphthalein as an indicator. The acidity was calculated by using the following formula and expressed in per cent according to Srivastava and Kumar (19).

$$\% \text{ Total acid} = \frac{\text{Titre value} \times N \text{ of alkali} \times \text{Volume made up} \times \text{equivalent wt. of acid} \times 100}{\text{Volume of the sample taken}} \times 1000$$

REDUCING SUGAR

It was estimated by following the method described by Shaffer-Somogyi micro method by Association of Official Analytical Chemists (1)

$$\text{Reducing sugars (\%)} = \frac{\text{Dextrose (mg)} \times \text{vol. made up} \times 100}{5 \times \text{wt. of sample taken} \times 1000}$$

TOTAL SUGARS

A shrikhand sample of 0.1ml was taken in a series of test tubes, added 1.0 ml of distilled water to each test tube. Pipetted out one ml of distilled water into another test tube to serve as blank. Added 0.5 ml of phenol reagent to each test tube and mixed well. Then five ml of sulphuric acid added to each test tube and allowed to stand for 10 min. The solution was then mixed well and placed in a water bath at 25-30°C for 10 -20 min. The absorbance of the solution was recorded at 490 nm according to Dubios (5)

$$\text{Total sugar as invert sugar} = \frac{\text{Dextrose (mg)} \times \text{volume made up} \times 100}{\text{Titer value} \times \text{weight of sample taken} \times 1000}$$

$$\% \text{ Sucrose} = (\% \text{ Total invert sugar} - \% \text{ reducing sugar}) \times 0.95$$

$$\% \text{ Total sugars} = \% \text{ reducing sugar} + \% \text{ Sucrose}$$

SENSORY EVALUATION

Shrikhand prepared using cattle milk, buffalo milk and dairy milk by adding curd, lactic acid bacterial isolates and probiotics as stater culture were evaluated by a selected panel of 10 members, which was based mainly on the appearance, colour, aroma, taste and overall acceptability of the product. Prior to tasting, each sample was coded and placed in a random manner, different shrikhand samples were placed along with water (to rinse the mouth) in the laboratory, and panelists were instructed to evaluate each sample by blind tasting as per the standard score card for organoleptic evaluation.

RESULTS AND DISCUSSION

Lactic acid bacterial isolates formed characteristic white, submerged colonies on Mann, Rogosa and Sharpe's (MRS) agar medium. Result revealed that (Table 1), all the lactic acid bacterial isolates were Gram +ve, rods and cocci. The lactic acid bacterial isolates were catalase -ve, Colonies appeared after 48 hrs on neutral red chalk agar, which were very small, deep red, surrounded by a clear zone formed due to the dissolution of CaCO₃ in the

medium, could not hydrolyse gelatin and they showed that isolates C₃, B₁, B₂, D₂, D₃ and D₄ changed the colour from red to yellow due to production of acids but C₁, C₄ and D₁ showed no change in colour. Collection of gas in durham's tube was observed in the tubes inoculated with the isolates C₁, C₄ and D₁ acid and small colonies appeared after 24 hrs on sucrose agar which were creamish to yellowish in colour. Isolates D₂ and D₃ could produce enormous deposition of slimy substances on the colonies, dextran was observed. These findings support the reports of Kebede (2007) reported that isolation, characterization and identification of lactic acid bacteria involved in traditional

fermentation of Borde, an Ethiopian cereal beverage. Similar results were also reported by Rao et al. (2000). Lactic acid bacteria can assimilate glucose as carbon source. All the isolates in this study showed assimilation of glucose in MRS broth. Glucose is a flavoured source which is an indication of glycolysis and is a major carbon utilizing pathway. These results are similar to those of Samelis et al.(1994) who also reported that gas (CO₂) production by glucose was determined in modified MRS broth containing inverted Durham tubes, with diammonium citrate replaced by ammonium sulphate.

Table 1: Characterization of isolates from shrikhand

| Isolates | Shape | Gram's reaction | Catalase activity | Glucose utilization | | Dextran production | Gelatin hydrolysis | Spore production | Growth on neutral red chalk agar |
|----------------|------------|-----------------|-------------------|---------------------|---|--------------------|--------------------|------------------|----------------------------------|
| | | | | A | G | | | | |
| C ₁ | Short rods | + | - | - | + | - | - | - | - |
| C ₂ | Short rods | + | - | - | - | - | - | - | - |
| C ₃ | Short rods | + | - | + | - | - | - | - | - |
| C ₄ | Long rods | + | - | - | + | - | - | - | - |
| B ₁ | Short rods | + | - | + | - | - | - | - | - |
| B ₂ | Long rods | + | - | + | - | - | - | - | - |
| B ₃ | Short rods | + | - | - | - | - | - | - | - |
| B ₄ | Short rods | + | - | - | - | - | - | - | - |
| D ₁ | Short rods | + | - | - | + | - | - | - | - |
| D ₂ | Cocci | + | - | + | - | + | - | - | + |
| D ₃ | Cocci | + | - | + | - | + | - | - | + |
| D ₄ | Long rods | + | - | + | - | - | - | - | - |

Note: A: Acid production, G: Gas production C: LAB isolated from cattle milk, D: LAB isolated from Dairy milk B: LAB isolated from Buffalo milk

CHEMICAL COMPOSITION OF SHRIKHAND

pH, residual sugars, total sugars and titrable acidity of shrikhand prepared using cattle milk, buffalo milk and dairy milk with different probiotics P₁ (*Lactobacillus sporogenes*), P₂ (*L. acidophilus*) and P₃ (*L. rhamnosus*) and in different combinations of P₁ x P₂ (*L. sporogenes* x *L. acidophilus*) along with different lactic acid bacterial isolates viz., Cattle milk (C₁, C₃& C₄), buffalo milk(B₁) and dairy milk (D₁, D₂ & D₄) were estimated and are presented in (Table 2, 3, 4 and 5). The treatments P₁x P₃, P₂ x P₃ and P₁ x P₂ x P₃ along with different lactic acid bacterial isolates are not presented in the result as they produced undesirable odour and not showed good quality with respect to appearance, taste, texture, colour and overall acceptability, hence the treatments were discarded.

EFFECT PH ON SHRIKHAND

The shrikhand prepared using lactic acid bacterial isolate C₁ had the highest pH in P₁ x P₂ (3.59) followed by P₂ (3.58), P₁(3.54), P₃(3.47) and the lowest pH was found

in C (3.43), C₃ had the highest pH in P₁ x P₂ (3.71) followed by P₁(3.61), P₃ (3.60), P₂(3.56) and the lowest pH was found in C (3.49), C₄ had the highest pH in P₁ x P₂ (3.66) followed by P₁ (3.63), P₃ (3.61), P₂ (3.53) and the lowest pH was found in C (3.50), D₂ had the highest pH in P₁ x P₂ (3.68) followed by P₃ (3.62), P₁ (3.50), P₂ (3.49%) and the lowest pH was found in C (3.46), D₄ had highest pH was found in P₁ x P₂ (3.72) followed by P₂ (3.62), P₃ (3.65), P₁(3.59) and the lowest pH was found in C (3.55), D₁ had the highest pH in P₁ x P₂ (3.70) followed by P₃ (3.67), P₁ (3.62), P₂ (3.60) and the lowest pH was found in C (3.56) and B₁ had highest pH was found in P₁ x P₂ (3.73) followed by P₂ (3.58), P₃ (3.54), P₁ (3.52) and the lowest pH was found in C (3.60) This may be due to the conversion of lactose into lactic acid and other organic acids by the starter cultures that reduced the pH of shrikhand and the lactose content of shrikhand is dependent on the extent of lactose degradation, moisture content of shrikhand Boghra et al. (2000) and Reddy et al. (1984).

Table 2: pH of shrikhand samples prepared using lactic acid bacterial isolates with probiotics

| Treatments | pH | | | | | Mean |
|----------------|--------------------------------|----------------|----------------|----------------|--------------------------------|------|
| | Lactic acid bacterial isolates | | | | | |
| | C | P ₁ | P ₂ | P ₃ | P ₁ xP ₂ | |
| C ₁ | 3.43 | 3.54 | 3.58 | 3.47 | 3.59 | 3.54 |
| C ₃ | 3.49 | 3.61 | 3.56 | 3.60 | 3.71 | 3.62 |
| C ₄ | 3.50 | 3.63 | 3.53 | 3.61 | 3.66 | 3.60 |
| D ₂ | 3.46 | 3.50 | 3.49 | 3.62 | 3.68 | 3.58 |
| D ₄ | 3.55 | 3.59 | 3.65 | 3.64 | 3.72 | 3.66 |
| D ₁ | 3.56 | 3.62 | 3.60 | 3.67 | 3.70 | 3.65 |
| B ₁ | 3.60 | 3.67 | 3.69 | 3.70 | 3.73 | 3.69 |
| Mean | 3.51 | 3.52 | 3.58 | 3.54 | 3.68 | |

Note: C : Control, P₁ : *Lactobacillus sporogenes*, P₂ : *L. acidophilus*, P₃ : *L. rhamnosus*, P₁xP₂: *L. sporogenes* +*L. acidophilus*, B₁: LAB isolated from buffalo milk, D₁, D₂ & D₄ : LAB isolated from dairy milk, C₁, C₃& C₄ : LAB isolated from cattle milk

Table 4: Total sugar (%) of shrikhand prepared using lactic acid bacterial isolates with probiotics

| Treatments | Total sugar(%) | | | | | Mean |
|-------------------|--------------------------------|----------------|----------------|----------------|--------------------------------|-------|
| | Lactic acid bacterial isolates | | | | | |
| | C | P ₁ | P ₂ | P ₃ | P ₁ xP ₂ | |
| C ₁ | 50.95 | 54.07 | 54.70 | 53.57 | 54.86 | 53.85 |
| C ₃ | 50.92 | 53.10 | 53.92 | 51.02 | 53.94 | 52.99 |
| C ₄ | 50.89 | 53.16 | 52.93 | 53.75 | 54.18 | 53.67 |
| D ₂ | 52.86 | 53.97 | 54.20 | 53.19 | 54.45 | 53.96 |
| D ₄ | 52.05 | 53.12 | 54.75 | 54.80 | 54.94 | 54.10 |
| D ₁ | 50.56 | 53.41 | 53.85 | 53.44 | 54.01 | 53.41 |
| B ₁ | 52.79 | 54.69 | 54.94 | 54.96 | 55.07 | 54.62 |
| Mean | 51.57 | 53.64 | 54.18 | 53.53 | 54.49 | |
| Source | S.Em ± CD @ 5% | | | | | |
| Probiotics (P) | 0.82 | | 1.63 | | | |
| Treatments (T) | 1.25 | | 2.49 | | | |
| Interaction (P×T) | 1.025 | | 4.06 | | | |

Note: C : Control, P₁ : *Lactobacillus sporogenes*, P₂ : *L. acidophilus*, P₃ : *L. rhamnosus*, P₁xP₂ : *L. sporogenes* +*L. acidophilus*, B₁ : Lactic acid bacteria isolated from buffalo milk, D₁, D₂ & D₄ : Lactic acid bacteria isolated from dairy milk, C₁, C₃& C₄ : Lactic acid bacteria isolated from cattle milk

Table 3: Residual sugars (%) of shrikhand prepared using lactic acid bacterial isolates with probiotics

| Treatments | Residual sugars (%) | | | | | Mean |
|----------------|--------------------------------|----------------|----------------|----------------|--------------------------------|-------|
| | Lactic acid bacterial isolates | | | | | |
| | PC | P ₁ | P ₂ | P ₃ | P ₁ xP ₂ | |
| C ₁ | 22.23 | 22.36 | 22.43 | 22.46 | 22.53 | 22.45 |
| C ₃ | 22.16 | 22.30 | 22.48 | 22.44 | 22.60 | 22.43 |
| C ₄ | 22.20 | 22.36 | 22.40 | 22.40 | 22.46 | 22.40 |
| D ₂ | 22.23 | 22.33 | 22.36 | 22.40 | 22.44 | 22.42 |
| D ₄ | 22.10 | 22.30 | 22.40 | 22.36 | 22.46 | 22.37 |
| D ₁ | 22.24 | 22.36 | 22.36 | 22.38 | 22.54 | 22.44 |
| B ₁ | 22.27 | 22.40 | 22.50 | 22.50 | 22.66 | 22.52 |
| Mean | 22.20 | 22.34 | 22.42 | 22.42 | 22.52 | |
| Source | S.Em± CD @ 5% | | | | | |

| | | |
|------------------|------|------|
| Probiotics (P) | 0.13 | 0.26 |
| Treatments (T) | 0.20 | 0.40 |
| Interaction(PxT) | 0.02 | 0.10 |

Note: C : Control, P₁:Lactobacillus sporogenes, P₂: L. acidophilus, P₃: L. rhamnosus, P₁xP₂: L. sporogenes +L. acidophilus, B₁: Lactic acid bacteria isolated from buffalo milk, D₁, D₂ & D₄: Lactic acid bacteria isolated from dairy milk, C₁, C₃& C₄: Lactic acid bacteria isolated from cattle milk

Table 5: Titrable acidity (%) of shrikhand samples prepared using lactic acid bacterial isolates with probiotics

| Treatments | Titrable acidity (%) | | | | | Mean |
|-------------------|--------------------------------|----------------|----------------|----------------|--------------------------------|----------|
| | Lactic acid bacterial isolates | | | | | |
| | C | P ₁ | P ₂ | P ₃ | P ₁ xP ₂ | |
| C ₁ | 1.21 | 1.24 | 1.22 | 1.23 | 1.24 | 1.23 |
| C ₃ | 1.20 | 1.23 | 1.21 | 1.24 | 1.25 | 1.23 |
| C ₄ | 1.17 | 1.26 | 1.20 | 1.22 | 1.30 | 1.24 |
| D ₂ | 1.18 | 1.22 | 1.26 | 1.23 | 1.29 | 1.26 |
| D ₄ | 1.20 | 1.25 | 1.24 | 1.26 | 1.28 | 1.25 |
| D ₁ | 1.18 | 1.26 | 1.26 | 1.25 | 1.30 | 1.26 |
| B ₁ | 1.22 | 1.29 | 1.27 | 1.29 | 1.30 | 1.28 |
| Mean | 1.21 | 1.25 | 1.23 | 1.24 | 1.26 | |
| Source | S.Em± | | | | | CD @ 5 % |
| Probiotics (P) | 0.02 | | | | | 0.03 |
| Treatments (T) | 0.03 | | | | | 0.05 |
| Interaction (PxT) | 4.00 | | | | | 1.68 |

Note: C : Control, P₁: Lactobacillus sporogenes, P₂:L. acidophilus, P₃: L. rhamnosus, P₁xP₂:L.sporogenes+L.acidophilus, B₁: Lactic acid bacteria isolated from buffalo milk, D₁, D₂ & D₄: Lactic acid bacteria isolated from dairy milk, C₁, C₃& C₄: Lactic acid bacteria isolated from cattle milk.

Table 6- Organoleptic evaluation of shrikhand prepared using lactic acid bacterial isolates as starter culture

| Treatments | Appearance | Aroma | Texture | Taste | Overall acceptability |
|----------------|------------|-------|---------|-------|-----------------------|
| C ₁ | 3.8 | 3.8 | 3.9 | 3.7 | 3.8 |
| C ₃ | 3.8 | 3.8 | 3.7 | 3.8 | 3.8 |
| C ₄ | 3.8 | 3.9 | 3.3 | 3.8 | 3.9 |
| B ₁ | 3.6 | 3.4 | 3.6 | 3.6 | 3.2 |
| D ₄ | 3.8 | 3.5 | 3.3 | 3.5 | 3.5 |
| D ₂ | 3.7 | 3.5 | 3.2 | 3.5 | 3.7 |
| D ₁ | 3.7 | 3.7 | 3.6 | 3.6 | 3.6 |

Note: Each value is an average of 10 replications (10 persons), * Scale: 5- Liked extremely; 4-Liked; 3-Neither liked nor disliked; 2-Disliked; 1-Disliked extremely, B₁: Lactic acid bacteria isolated from buffalo milk. D₁, D₂ & D₄: Lactic acid bacteria isolated from dairy milk, C₁, C₃& C₄: Lactic acid bacteria isolated from cattle milk

EFFECT OF RESIDUAL SUGARS (%) ON SHRIKHAND

The shrikhand prepared using lactic acid bacterial isolate C₁ recorded highest residual sugars in P₁ x P₂ (22.53 %) followed by P₃ (22.46 %), P₂ (22.43 %), P₁ (22.36 %) and the lowest residual sugars was found in C (22.23 %), C₃ had highest residual sugars was found in P₁ x P₂ (22.60 %) followed by P₂ (22.48 %), P₃ (22.44 %), P₁ (22.30 %) and the lowest residual sugars was found in C (22.16 %), C₄ had the highest residual sugars was found in P₁ x P₂ (22.46 %) followed by P₂ (22.40 %), P₃ (22.40 %), P₁ (22.36 %) and the lowest residual sugars was found in C (22.20 %), D₂ had highest residual sugars was found in P₁ x P₂ (22.44 %) followed by P₃ (22.40 %), P₂ (22.36 %), P₁ (22.33 %) and the lowest residual sugars was found in C (22.23 %), D₄ had highest residual sugars was found in P₁ x P₂ (22.46 %) followed by P₂ (22.40 %), P₃ (22.36 %), P₁

(22.30 %) and the lowest residual sugars was found in C (22.10 %), D₁ recorded the highest residual sugars in P₁ x P₂ (22.54 %) followed by P₃ (22.38 %), P₂ (22.36 %), P₁ (22.36 %) and the lowest residual sugars (%) was found in C (22.24 %) and B₁ had the highest residual sugars in P₁ x P₂ (22.66 %) followed by P₂ (22.50 %), P₃ (22.50 %), P₁ (22.40 %) and the lowest total sugars (%) was found in C (22.27 %). This may be due to its influence in sugar utilization by lactic acid bacteria Osman and Razig (2010); Khurana and Kanawjia (2007).

EFFECT OF TOTAL SUGAR (%) ON SHRIKHAND

The shrikhand prepared using lactic acid bacterial isolate C₁ had the highest total sugar in P₁ x P₂ (54.86 %) followed by P₂ (54.70 %), P₁(54.07 %), P₃ (53.57 %) and the lowest total sugar was found in C (50.95 %), C₃ had highest total sugar was found in P₁ x P₂ (55.07 %) followed

by P₂ (53.92 %), P₁ (54.07 %), P₃ (51.02 %) and the lowest total sugar in C (50.92 %), C₄ had highest total sugar was found in P₁ x P₂ (54.18 %) followed by P₃ (53.75 %), P₁ (53.16 %), P₂ (52.93 %) and the lowest total sugar was found in C (50.89 %), D₂ had highest total sugar in P₁ x P₂ (54.45 %) followed by P₂ (54.20 %), P₁ (53.97 %), P₃ (54.20 %) and the lowest total sugar was found in C (52.86 %), D₄ had highest total sugar in P₁ x P₂ (54.94 %) followed by P₃ (54.80 %), P₂ (54.75 %), P₁ (53.12 %) and the lowest total sugar was found in C (52.05 %) D₁ had the highest total sugar was found in P₁ x P₂ (54.01 %) followed by P₂ (53.85 %), P₃ (53.44 %), P₁ (53.41%) and the lowest total sugar was found in C (50.56%) and B₁ recorded the highest total sugar in P₁ x P₂ (55.07 %) followed by P₃ (54.96 %), P₂ (54.94 %), P₁ (54.69 %) and the lowest total sugar was found in C (52.79 %). That may be due to the blending of sugar with chakka. Sugar slowed down the chemical changes due to fermentation Boghra et al. (2000); Navita, 2009.

EFFECT OF TITRABLE ACIDITY ON SHRIKHAND

The shrikhand prepared using lactic acid bacterial isolate C₁ had highest titrable acidity in P₁ x P₂ (1.24 %) followed by P₁(1.24 %), P₃ (1.23 %), P₂ (1.22 %) and the lowest titrable acidity was found in C (1.21 %), C₃ had highest titrable acidity was found in P₁ x P₂ (1.25%) followed by P₃ (1.24%), P₁ (1.23 %), P₂ (1.21 %) and the lowest titrable acidity (%) was found in C (1.20 %), C₄ had highest titrable acidity was found in P₁ x P₂ (1.30 %) followed by P₁ (1.26 %), P₃ (1.22 %), P₂ (1.20 %) and the lowest titrable acidity (%) was found in C (1.17 %), D₂ had highest titrable acidity was found in P₁ x P₂ (1.29 %) followed by P₂ (1.26 %), P₃ (1.23 %), P₁ (1.22 %) and the lowest titrable acidity was found in C (1.18 %), D₄ had highest titrable acidity in P₁ x P₂ (1.28 %) followed by P₃ (1.26 %), P₁ (1.25 %), P₂ (1.24 %) and the lowest titrable acidity was found in C (1.20 %). In the shrikhand prepared using lactic acid bacterial isolate D₁, the highest titrable acidity was found in P₁ x P₂ (1.30 %) followed by P₁ (1.26 %), P₂ (1.26 %), P₃ (1.25 %) and the lowest titrable acidity (%) was found in C (1.18 %) and B₁ had the highest titrable acidity in P₁ x P₂ (1.30 %) followed by P₁ (1.29%), P₃ (1.29%), P₂ (1.27 %) and the lowest titrable acidity was found in C (1.22 %). This may be due to gradual increase in titrable acidity with the storage period Ghatak and Dutta (1998).

ORGANOLEPTIC EVALUATION

The Shrikhand samples prepared using lactic acid bacterial isolates were tested for organoleptic characteristics of the product are given in (Table 6). Results revealed that, the highest score was recorded with LAB isolates i.e. C₁, C₃ & C₄ obtained from cattle milk (3.80, 3.80 and 3.90 out of 5.00), followed by lactic acid bacterial isolates obtained by buffalo milk i.e. B₁ (3.20 out of 5.00) in terms of colour, appearance, aroma, texture, taste and overall acceptability of the product. Similar

observation were recorded by Rameshwar (2006), who also reported that the shrikhand has a typical semi solid consistency showing a characteristic firmness and pliability contributing to its suitability for consumption with puri and bread. Similar results were obtained by Salunke et al. (2006), and Patel and Chakraborty (1985) who reported that varying levels of fat, moisture and sugar had the least effect on colour but had a profound effect on appearance, flavour, body and textural properties of shrikhand.

REFERENCES

- O. A. C., 1980, Official methods of analysis, 13th Edn. Washington D. C., The Association of Official Analytical Chemists.
- ANEJA, K. R., 1996, Experiments in microbiology, plant pathology, tissue culture and mushroom cultivation. II Edition, 60-217pp.
- Boghra V. R. and Mathur, O. N., 2000, Physico-chemical status of major milk constituents and minerals at various stages of shrikhand preparation. J. Food Sci. Technol., 37:111-115.
- Chalmers, C. H., 1962, Bacteria in relation to the milk supply, Ed. Edward Arnold, 4th Edition, Landon.
- Dubios, M., Gills, K. A., Hamilton, J. K., Rebers, P. A. and Smith, F. M., 1996, Colorimetric methods for the determination of the sugars and related substances, Analy. Chem., 28: 350-356.
- Garvie, E. L., 1960, *Leuconostoc* and its nomenclature. J. Dairy. Res., 27: 283- 284.
- Ghatak P. N. and Dutta, S., 1998, Effect of admixing of cow and buffalo milk on the composition and sensory qualities of shrikhand. Indian. J. Nutr. Dietet., 35: 43-46.
- Harrigan, W. F. and MC Cance, E. M., 1976, The microbiological examination of foods. In; Laboratory Methods in Microbiology, A.P., London and New York.
- Kebede, A., 2007, Isolation, characterization and identification of lactic acid bacteria involved in traditional fermentation of borde, an Ethiopian cereal beverage. Afr. J. Biotechnol., 6:1469-1478.
- Khurana H. K. and Kanawjia, S. K., 2007, Recent Trends in Development of Fermented Milks. Curr. Nutr. Food Sci., 3: 91-108
- Mann, DE, J. C., Rogosa, M. and Sharpe, M. E., 1960, A medium for the cultivation of *Lactobacillus*. J. Appl. Bacteriol., 23: 130 -135
- Navita, N., Rashmi, S. and Upadhayay, P.K., 2009, Incorporation of *chakka* by papaya pulp in the

manufacture of *shrikhand*. J. Dairying, Foods & H.S., 28: 115-118.

- Osman M. M. D. and Razig, K. A. A., 2010, Quality Attributes of Soy-yoghurt during Storage Period. Pak. J. Nutr. 9: 1088-1093.
- Patel, R.S. and Chakraborty, B.K., 1985, Standardization of shrikhand manufacturing process of lactic fermentation. Le-lait., 65:55-56.
- Rameshwar, S, 2006, Characteristics and technology of traditional Indian cultured dairy products. Indian Dairyman., 58:49-51.
- Reddy K. K., Pasha Ali, M., Rao, B. V. and Jagannadha Rao, T., 1984. Studies on production and quality of shrikhand from buffalo milk. Indian. J. Dairy. Sci., 37:293-296.
- Salunke, P., Hasmukh, A. P. and Thakar, P. N., 2006, Sensory profile of market shrikhand sold in Maharashtra state. Indian. J. Dairy. Sci., 59: 363-368.
- Samelis, J., Murogenakis, F. and Metaxopoulos, J., 1994, Characterization of lactic acid bacteria isolated from naturally fermented Greek dry salami. Int. J. Food. Microbiol., 23: 179-196.
- Srivastava, R. P. and Kumar, S., 1993, Important methods for analysis of fruit or vegetable and their products. In: Fruits and vegetable preservation-principles and practices, II edition, 321-339 p.