

# Preparation of Shrikhand from goat milk using probiotic cultures

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## ABSTRACT

Shrikhand is a traditional fermented milk product of Indian origin and is popular in western parts of India. Starter culture is known to play vital role in the development of body, texture, flavor and aroma in the finished products and contributes significantly towards overall acceptance of products by the consumer. In investigation the probiotic cultures used in preparation of shrikhand were *Lb. acidophilus* and *Lb. delbrueckii ssp. bulgaricus* alone and in combination and LF-40 culture alone as a control. Probiotic cultures viz., *Lb. acidophilus* and *Lb. bulgaricus* alone and in combination containing viable lactobacilli counts more than  $10^7$  c.f.u./g in fresh as well as in stored shrikhand. The results revealed that organoleptically acceptable shrikhand from goat milk can be prepared by using probiotic cultures viz., *Lactobacillus acidophilus* and *Lactobacillus delbrueckii ssp. bulgaricus* alone and in combination. There was non significant difference observed for sensory evaluation for all treatments. Flavour score observed highest in (8.00) stored shrikhand prepared by combination of *Lactobacillus acidophilus* and *Lactobacillus delbrueckii*. The probiotic shrikhand can be prepared and stored at 7 + 1 C for a week and possessing good organoleptic quality with therapeutic quality.

**Key words:** Shrikhand, probiotic, goat milk product.

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## INTRODUCTION

Goat milk possesses medicinal and therapeutic value. Goat milk is good for human health. The fermented milk products have reputation due to their nutritional and therapeutic properties (Singh, 1996). Probiotics are live microorganisms that confer health benefits to the host when administered in certain quantities (FAO/WHO, 2001). They are essential for the proper functioning of the digestive tract, commonly referred to as 'friendly', 'beneficial' or 'good' bacteria (Jha *et al*, 2015). The interest in probiotic foods has been increasing in recent years, and has inspired invention and driven the development of new products around the world, in order to maintain the gut health with addition of probiotic bacteria into foods (Rajani *et al* 2016). In India dahi is traditionally prepared using mesophilic starter cultures. The rate of acid production is typically slower in dahi compared to yoghurt fermentation, which is typically completed within 5 hours. Therefore, commercial processors are increasingly using yoghurt cultures

(*Streptococcus thermophilus* and *Lactobacillus delbrueckii* supsp *bulgaricus*) to prepare dahi (Korasapati *et al*, 2016). Dahi is used as a base material for preparation of Shrikhand, indigenous butter lassi etc (Mistry, 2001, Aneja *et al.*, 2002).

Considering the importance of Shrikhand in human diet, the importance of goat milk and the importance of lactic acid bacteria in the human health, the present study was conducted to determine the suitability of probiotic strains *Lactobacillus delbrueckii* and *Lactobacillus acidophilus* to make an acceptable quality probiotic Shrikhand from goat milk.

## MATERIAL AND METHODS

Fresh goat milk was obtained from Osmanabadi goat unit, Post graduate institute, M.P.K.V., Rahuri. The milk was standardized to 6 % fat. The standardized goat milk was heated to 85 °C for 10 min. and then cooled to 30 °C. As per the treatment the cultures were inoculated @ of 1 percent. T<sub>0</sub> : LF- 40; T<sub>1</sub> : *Lactobacillus acidophilus*; T<sub>2</sub> : *Lactobacillus delbrueckii ssp. Bulgaricus*; T<sub>3</sub> : Combination of *Lb.*

*acidophilus* and *Lb. delbrueckii ssp. Bulgaricus* (1:1).

The inoculated cultures were properly mixed and then incubated at 37 °C for 8-10 hrs. After incubation period curd were broken and tied in muslin cloths separately and then tied in hanging position to drain whey for 15-16 hrs. Weight of chakka were recorded and ground sugar (@ 45 % by weight of chakka ) was added. The chakka and sugar was kneaded to smooth paste and filled in polystyrene cups and then stored at 7 ± 1°C temperature for one week. The prepared shrikhand samples were taken for organoleptic evaluation, chemical and microbial analysis. Freeze dried pure culture of LF-40, *Lactobacillus acidophilus*, *Lactobacillus delbrueckii ssp. Bulgaricus* were obtained from the National Dairy Research Institute, Karnal, Haryana. These cultures were maintained separately in sterilized 10 ml reconstituted skim milk in test tubes. The test tubes were autoclaved at 121°C / 15 lb / 15 min.

#### Analytical Techniques

Goat milk was analyzed for fat by Gerber method IS: 1224, Part I. Samples of Shrikhand were analyzed for total solids by gravimetric method (IS: 1166 (part II), 1973, fat as per the Gerber method IS: 1224 (Part – II) 1977 and Protein by Kjeldahl method and acidity as per IS-1166 (Part – II) 1973.

#### Enumeration of viable lactobacilli count

The MRS agar having pH 6.4 with double layer was used for enumeration of viable lactobacilli counts of T<sub>1</sub>, T<sub>2</sub>, & T<sub>3</sub> samples of shrikhand. Ten grams of shrikhand samples were taken separately in 90 ml sterilized phosphate buffer solution and then subsequently diluted using 9 ml phosphate buffer. The 7<sup>th</sup> and 8<sup>th</sup> dilution were taken in duplicate into

pertiplates and then MRS agar having pH 6.4 was added and mixed well. The plates were allowed to solidify. The plates were again overlaid with same agar and incubated at 37°C / 48 hr and then lactobacilli counts were recorded.

#### Enumeration of lactic culture (LF-40) counts

The lactose purple agar (LPA) having pH 7.4 ± 0.1 was used for the enumeration of lactic acid culture (LF-40) counts of control (T<sub>0</sub>).

#### Organoleptic quality of shrikhand

The fresh and refrigerated stored shrikhand (at 7 ± 1°C for a week) samples were subjected to the organoleptic evaluation. Shrikhand samples were provided to the panel of seven semi-trained judges for sensory evaluation. Samples were evaluated for flavour, body and texture, colour and appearance and overall acceptability by using 9-point Hedonic Scale.

#### Statistical Analysis

The data were subjected to Completely Randomized Design (CRD) as described by Snedecor and Cochran (1967).

## RESULTS AND DISCUSSION

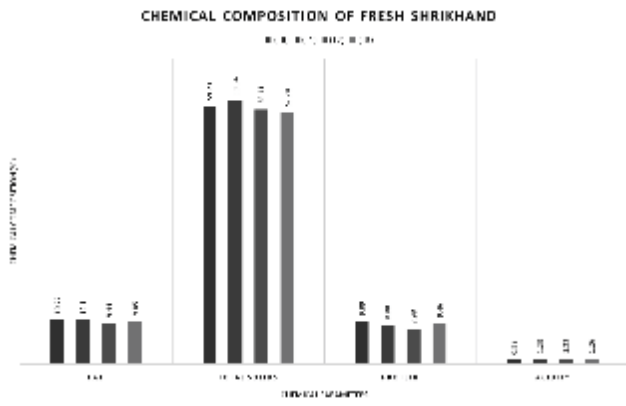
The fresh goat milk used for preparation of Shrikhand was containing on an average 4.05 per cent fat and 3.17 per cent protein. There was non-significant difference in the flavour scores of all fresh shrikhand samples. The samples T<sub>2</sub> prepared by using *Lb. delbrueckii ssp. Bulgaricus* @ 1 per cent secured highest flavour score (8.31) than other types of Shrikhand. The mean flavour score of treatment T<sub>0</sub>, T<sub>1</sub>, T<sub>3</sub> were 8.00, 7.94, and 8.00 respectively. Kiran et al (2012) reported flavor of *Brevibacillus brevis* MMB 12 fermented product scored 7.0 to 7.1

**Table 1.** Chemical composition and sensory quality of fresh Shrikhand.

Treatment	Chemical composition (%)				Sensory evaluation			
	Fat	Total solids	Protein	Acidity	Flavour	Body & Texture	Colour & appearance	Overall acceptability
Control (T <sub>0</sub> )	10.22	59.21	9.89	0.98	8.00	7.63	7.88	7.81
<i>Lb. acidophilus</i> (T <sub>1</sub> )	10.10	60.34	8.88	1.23	7.94	7.81	7.13	8.06
<i>Lb. delbrueckii ssp</i> (T <sub>2</sub> )	9.44	58.41	7.98	1.33	8.31	7.81	7.63	8.31
Combination (T <sub>3</sub> )	9.85	57.74	9.46	1.26	8.00	7.88	7.13	8.80
S.E. +	0.167	0.339	0.056	0.027	--	---	--	--
C.D.	0.514	1.044*	0.174	0.084	NS	NS	NS	NS

**Table 2.** Acidity and sensory quality of stored shrikhand

Treatment	Acidity	Sensory evaluation			
		Flavour	Body & texture	Colour & appearance	Overall acceptability
Control (T0)	0.992	7.13	7.56	7.50	7.88
<i>Lb. acidophilus</i> (T1)	1.290	7.71	8.19	8.00	8.06
<i>Lb. delbrueckii</i> ssp. (T2)	1.333	6.25	6.88	7.13	6.81
Combination (T3)	1.308	8.00	8.06	8.30	8.63

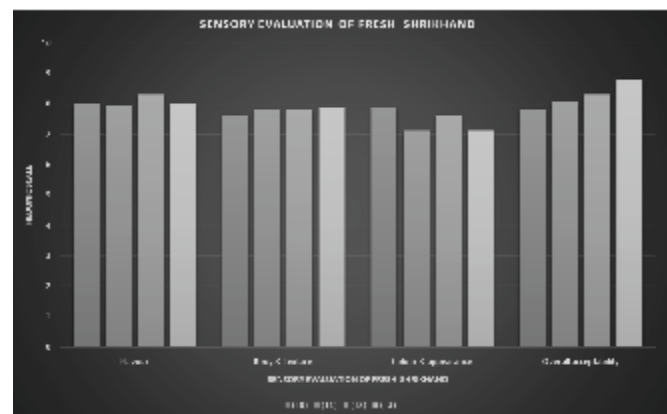


between 24 to 192 hours of preservation, and *Lb. delbrueckii* Subsp. *Bulgaricus* curd had maximum score of 7.0 which decreased to 6.0 after 96 hrs. Fadela et al (2009) also stated that the rate of acidification depends on amount and type of starter culture, thus affecting the sequence of gelation and determining the characteristic of casein matrix. There was considerably effect on body and texture characteristics of Shrikhand samples prepared by using various starter cultures. Mean score for body and texture of (T<sub>3</sub>) Shrikhand was higher (7.88) because body was uniform and smooth than rest of the samples (T<sub>0</sub>), which recorded lower score (7.63). The score was observed to be minimum as body was weak; texture was gritty and had definite curd particles. The samples T<sub>1</sub> and T<sub>3</sub> scored lower than rest of the treatment for colour and appearance. The treatment T<sub>3</sub> secured highest score (8.8) for overall acceptability than T<sub>0</sub>, T<sub>1</sub> and T<sub>2</sub> types, as it had

smooth, uniform body and sweetish- sour taste.

#### Chemical quality of Shrikhand

The result revealed that, fat content in all treatments was in the range of 8.5 to 10.30 per cent (Table -1) which coincided with the results of Zariwala and Sharma (1976). The maximum total solids content was recorded in T1 (60.34 per cent) sample. The treatment T0, T2 and T3 recorded 59.21, 58.41 and 57.74 respectively. The total solids content of samples coincide with the results of Patil (1991), Rachakonda (1995) and Bhogra and Mathur (2000). The table revealed that the protein content in control (T<sub>0</sub>) was 10 per cent which was slightly higher than (8.81 %), which was also reported by Karthikeyan (1993). The significant difference was observed in the protein content for all treatments. Acidity in all treatment was observed in the range of 0.98 to 1.26 %. And as the treatment level increases, acidity also increases. Kiran et al (2012) reported that curd

**Table 3.** Total counts of lactobacilli in fresh and refrigerated stored shrikhand. (Mean of four samples)

Name of product	Counts of lactobacilli (c.f.u./g)		
	T1	T2	T3
Fresh shrikhand	55 X 10 <sup>7</sup>	75 X 10 <sup>7</sup>	100 X 10 <sup>7</sup>
Stored shrikhand	30 X 10 <sup>7</sup>	38 X 10 <sup>7</sup>	53 X 10 <sup>7</sup>

**Table 4.** Lactic culture (LF-40) counts of fresh and refrigerated stored shrikhand. (Mean of four samples)

Name of product	Lactic culture (LF-40) counts (c.f.u./g)
Fresh shrikhand	25 X 10 <sup>7</sup>
Stored shrikhand	15 X 10 <sup>7</sup>

sample fermented with *Brevibacillus brevis* observed lower percentage of acidity (0.58% to 0.66%) after 12 hrs, than *Lb. delbrueckii* (0.98 % to 1.68 %). The result of high acid production for *Lb. delbrueckii Subsp. bulgaricus* culture agreed with findings of Kamruzzaman et al. (2002). These were slightly increases in acidity of all stored samples. All stored Shrikhand samples were fulfilled the BIS standard of Shrikhand (maximum 1.4 % lactic acid).

#### Sensory quality of stored shrikhand

The sensory evaluation of Shrikhand samples stored at  $7 \pm 1^{\circ}\text{C}$  was performed on 8<sup>th</sup> day. This day was selected on the basis of lactobacilli count, to have their beneficial effect with maximum viability. As day progresses, the viability of lactobacilli drastically lowered down during storage. The results of organoleptic quality of Shrikhand are showed in table -2. Mean score for flavour of stored Shrikhand of T<sub>3</sub> (8.0) was significantly higher (8.0) than T<sub>0</sub> and T<sub>2</sub> and it was at par with the score of T<sub>1</sub> (7.71). The results revealed that the stored Shrikhand of T<sub>2</sub> prepared by using *Lb. delbrueckii spp. bulgaricus* shown significantly lower score (6.88) for body and texture than all other samples as it because slightly grainy. For colour and appearance the score ranges between 7.13 to 8.30. There was significant difference observed for colour and appearance. Overall acceptability score of Shrikhand for T<sub>2</sub> was significantly lower (6.81) while that of shrikhand T<sub>3</sub> (8.30). It was clear from the table no.2 that all the cultures gave the Shrikhand of acceptable quality even after one week of refrigerated (at  $7 \pm 1^{\circ}\text{C}$ ) storage. It is revealed that fresh Shrikhand was more acceptable than stored Shrikhand and sensory score was declined on storage. Chemical state of milk proteins of products of their breakdown imparts characteristics physical, chemical and sensory properties to most of the dairy products (Sikorski et al 2001). The result for high colour score for *Lb. delbrueckii Subsp bulgaricus* was also observed by

Kiran et al (2012). Lower values for acidity was observed for curd prepared by using *Lb. acidophilus* by Patil et al (2015).

#### Acidity of stored shrikhand

Acidity was significantly lowered in control stored Shrikhand (T<sub>0</sub>) prepared by using LF-40 culture than other treatments (table-2). All stored Shrikhand samples were fulfilled the BIS norm for maximum per cent lactic acidity.

#### Microbial analysis

*Lactobacilli counts of fresh and refrigerated stored shrikhand:* In order to obtain maximum therapeutic value, the fermented milk product should contain population of viable cells of probiotic cultures more than 10<sup>7</sup>c.f.u. at the time of consumption (Speck, 1976;Tamine et al. 1995). It was observed (Table-3) that T<sub>3</sub> showed higher viable lactobacilli count (100 x 10<sup>7</sup>c.f.u./g) than fresh Shrikhand samples (T<sub>1</sub> and T<sub>2</sub>). On refrigerated storage, viable lactobacilli count of shrikhand (T<sub>3</sub>) decreased up to 53 x 10<sup>7</sup>c.f.u./g. The viable counts of *Lactobacillus acidophilus* observed in the study were slightly higher than that reported by Patil et al. (2015) and Sheikh et al (1990). All samples showed the required beneficial viable lactobacilli count in product when fresh and showed decreasing trend in refrigerated storage. Agnihotri and Pal (1997) noted the similar observation.

#### Lactic culture (LF-40) counts

The lactic culture count of shrikhand prepared by using LF-40 culture was recorded. In fresh shrikhand it was 25 x 10<sup>7</sup> and in stored shrikhand (T<sub>0</sub>) it was 15 x 10<sup>7</sup> c.f.u./g, respectively (Table - 4).

#### CONCLUSION

Delicious shrikhand with characteristics flavour can be prepared by using probiotic cultures viz., *Lb. acidophilus* and *Lb. bulgaricus* alone and in combination containing viable lactobacilli counts more than 10<sup>7</sup>c.f.u. /g when fresh as well as stored. The probiotic shrikhand can be prepared and stored

at  $7 \pm 1^{\circ}\text{C}$  for a week and possessing good organoleptic quality with therapeutic quality.

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