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## EVALUATION OF SOME FERMENTED MILK PRODUCTS

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### ABSTRACT

The preservation method achieved by dairy industry is storage under refrigeration condition. Many dairy products are stored at refrigeration temperature by wholesale dealer and retailers but at this refrigeration temperature, milk and milk products allow the growth of psychrotrophic organisms. Considerable variations in incidence of psychrotrophs in fresh raw milk have been reported by different observers. Some difference may be regional or seasonal and some are associated with the methods of cleaning and disinfection of equipments in dairy. The aim of the present study was to monitor culturable mesophilic and psychrotrophic communities in fermented milk products collected from nine dairies included in Nasik division, that may influence milk and fermented milk products shelf-life. Enumeration of bacteria from these samples showed the presence of mesophilic as well as psychrotrophic bacteria in each of the sample and there was a considerable variation in mesophilic and psychrotrophic bacterial content amongst the samples. Chemical examination was carried out to decide the nutritive quality of fermented milk products. The chemical examination of fermented milk products includes determination of pH, acidity (in terms of lactic acid), fat, total solids, SNF, proteins, sucrose and ash contents. Chemical contents of all fermented milk products were within the range mentioned by Bureau of Indian Standards.

**Keywords:** Fermented milk products, Shelf life, Psychrotrophs, Chemical composition

## 1. INTRODUCTION

Milk and milk products are the integral part of our rituals and we have the largest liquid milk consuming population of the world. The first mention of milk trading occurred during Mahabharata times (nearly 2500 BC) when butter (milk fat) is taken out of milk to ease movement from Gokul to Mathura. Lord Sri Krishna has been considered as a true cow savior. Milk and milk products have properties that are beneficial to a person's lifelong health. The rich package of conventional nutrients as well as specialized bioactive components makes milk as an important part of the overall human diet. Milk should not contain any contaminant at a level that jeopardizes the appropriate level of public health protection when presented to the consumer. Contamination of milk from animal and environmental sources during primary production should be minimized. The quality of the starting raw milk has a very definite effect on the yield and quality of products made from it. The compositional quality, the hygienic quality, the health of the cow and the level of contaminants present can all have an impact on the yield and quality, and hence financial return from products made from milk. Milk is an ideal and perfect medium for growth of bacteria and therefore it gets contaminated very easily and readily. It is extremely perishable in nature and its shelf life is limited to 3 to 4 hours depending upon the temperature of storage (Kumar A and Seth R, 2008). Extension of shelf life from hours to months has been a prime objective of the dairy industry for many years to meet the demands for increasing distribution times and distances (Goff HD and Griffiths MW 2006). Rapid cooling and cold storage of raw milk favour the growth of psychrotrophic bacteria in milk (Barbano DM, Ma Y and Santos MV, 2006). Psychrophiles (<math><20^{\circ}\text{C}</math>) and psychrotrophs (<math><37^{\circ}\text{C}</math>) are distinguished according to their range of temperature adaptation (Russell NJ, 1990). They thrive in cold environments due to unique features like cold shock proteins, short and unsaturated fatty acids in membranes, enzymes with high specific activity, thermolability and genetic changes to thermal shifts (Margesin et al., 2007). Lei et al., (2019) reported the isolates of *Pseudomonas*, *Acinetobacter*, *Flavobacterium*, *Chryseobacterium*, *Serratia* and *Aeromonas* were not only the most predominant in raw milk samples, but also proved to be high enzyme producers (Lei et al., 2019). Subramanian, P. and Ora, P.K. (2005) found that the total number of mesophilic bacteria surviving in the raw milk collected from individual medium size six dairy farms were found to be higher, ranging from 6.04 to 7.50 log<sub>10</sub> cfu/ml milk during the summer months. The number declined to 5.93 to 6.71 log<sub>10</sub> cfu/ml and to 5.30 to 5.53 log<sub>10</sub> cfu/ml milk during rainy and winter months respectively (Subramanian P and Ora PK, 2005).

**Aim of the study:**

1. To study the difference in chemical composition of some fermented milk products which includes determination of pH, fat, protein, salt content, acidity (in terms of lactic acid), total solids, SNF, ash and sucrose content.
2. Enumeration of psychrotrophic and mesophilic bacteria from some fermented milk products.

**2. MATERIALS AND METHODS****2.1 Collection and coding of Samples:**

In present work fermented milk samples constituting lassi and butter were collected in different seasons from nine different dairies in Nasik region of Maharashtra and were appropriately coded which includes nine samples of lassi namely WTL, WRL, RShL, RAL, RGL, SShL, SSL, SNL and SGL and two samples of butter namely RVB and RSB.

**2.2 Chemical Examination of samples:**

**Determination of pH:** The pH value of all samples was determined electrometrically with a pH meter (Toshcon Industries Private Ltd.). The pH meter was standardized with standard buffer solutions of pH 4 and pH 7 before use.

**Determination of Titrable acidity:** The total titrable acidity of all samples was determined according to the method of AOAC (2005). All samples were analyzed in triplicate (A.O.A.C. 2005).

Calculations: Titrable acidity as Lactic acid =  $\frac{9 AN}{W}$

Where A = Volume of standard NaOH required for titration; N = Normality of Standard NaOH solution; W = weight of the sample taken for test

**Determination of Fat Content:** Fat content of all the samples was determined by Gerber's method by using Gerber's butyrometer (FAO, 1977).

**Determination of Total Solids:** Total solids present in samples were determined by Gravimetric method using thermostatically controlled oven set at  $100 \pm 1^\circ\text{C}$  (FAO, 1977). All samples were analyzed in triplicates. Solids not fat (SNF) content was determined using the following formula: SNF content (%) = TS (%) - Fat (%).

### Detection of Sodium Chloride (Salt):

Modified Mohr method was used for the detection of sodium chloride present in the samples. Results were interpreted after analysis of samples in triplicates (A.O.A.C.2005).

Calculations: -Sodium Chloride Percent (w/w) =  $5.85 N (V1 - V2) / W$

Where N = Normality of Silver Nitrate; V1 = volume of silver Nitrate in sample titration.

V2 = Volume of silver Nitrate in blank titration; W = weight in gm of the sample.

**Determination of Sucrose Content:** Sucrose content was determined by Lane-Eynon (1923) Method (A.O.A.C. 2005).

**Determination of Protein Percentage:** Percentage of proteins present in all samples were determined by Kjeldahl's method (A.O.A.C. 2000). All samples were analyzed in triplicates

**Determination of Total Ash:** The total ash was determined gravimetrically by igniting the dried milk samples in a muffle furnace in which the temperature was slowly raised to 550°C. The sample was ignited until carbon (black colour) disappears or until the ash residue becomes white (Richardson, G. H.,1985).

### 2.3 Enumeration of psychrotrophic and mesophilic bacteria from lassi and butter:

Serial dilution technique was used for the enumeration of psychrotrophic and mesophilic bacteria present in samples as described by standard methods of the American Public Health Association (Vanderzant C. and Splittstoesser, D.F.1992). Samples were diluted ( $10^{-1}$  to  $10^{-10}$ ) using normal saline as a diluent and 0.1 ml of each diluted sample was plated on sterile 10% milk agar plate. Pour plate technique was used for plating and plates were incubated at 30°C for 24 hours and at 7°C for 10 days for growth of mesophilic and psychrotrophic bacteria respectively. After incubation appropriate plates were selected and results were expressed as  $\log_{10}$  cfu/ml.

### 3.RESULTS AND DISCUSSION

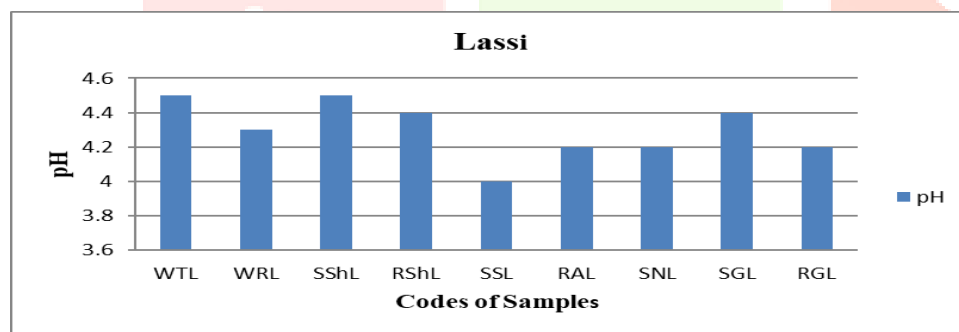
#### 3.1 Chemical Examination of samples:

##### Chemical Examination of Lassi:

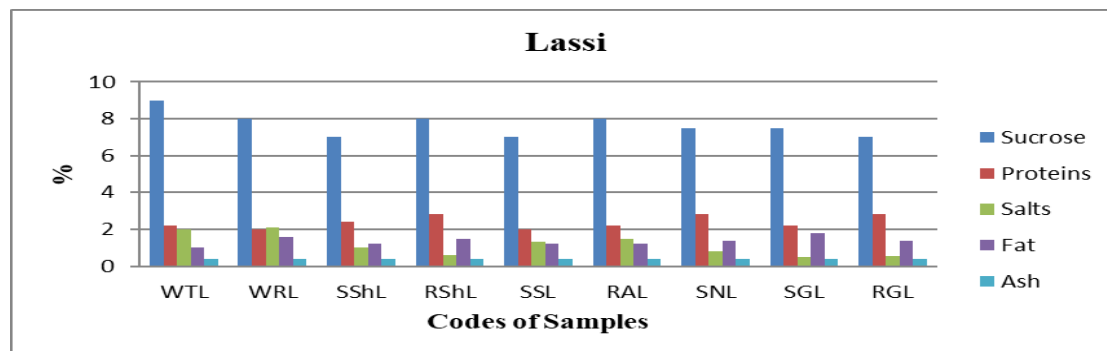
Chemical parameters such as pH, titrable acidity, salt (sodium chloride), fat, total solids, SNF, proteins, sucrose and ash percentage were determined for lassi samples.

It can be seen from the **Graph 1** that the pH of lassi samples was in a range between 4 to 4.5. The lowest pH value i.e. 4 amongst the samples was reported for a sample namely SSL and the highest pH value i.e. 4.5 was reported for the two samples namely WTL and SShL. Remaining six samples showed the pH values in this range. Acidity (in terms of lactic acid) of lassi ranged from 0.39% to 0.47%. Higher acidity (0.47%) was found in lassi sample SSL while less acidity (0.39%) was found in lassi sample SShL as compared to the lassi samples collected from other dairies. Savadogo et al., (2004) reported pH value of lassi in a range between 4.00 to 5.86 (Savadogo, 2004) while Bagal et al., (2007) reported pH value of lassi in a range between 4.13 to 4.45 (Bagal et al, 2007).

**Graph 1:**

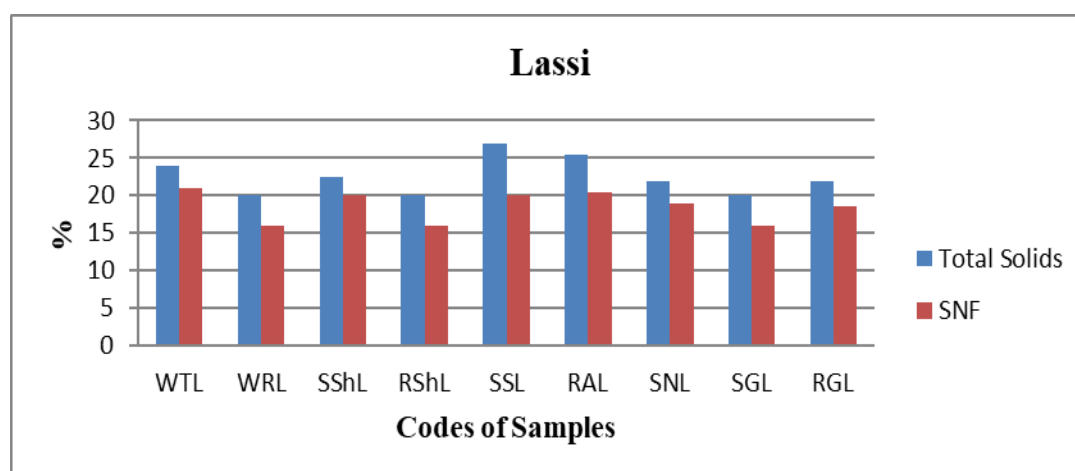


**Graph 2:**



Sucrose content in lassi samples was in a range between 7% to 9% (**Graph 2**). Higher percentage i.e. 9% of sucrose content was found in lassi sample namely WTL while lower percentage i.e.7% of sucrose content was found in three lassi samples namely SShL, SSL and RGL. Remaining five samples showed the sucrose content in between this range. The composition of lassi varies considerably since there is no standard method available for the preparation of this product. The factors affecting the composition of lassi are the type of milk used, extent of dilution during churning and efficiency of fat removal (Yadav et al., 1993). The protein content of lassi samples was in a range between 2% to 2.8%. Higher percentage i.e. 2.8% of protein content was found in three lassi samples namely SNL, RShL and RGL while lower percentage i.e. 2% of protein content was found in two lassi samples namely WRL and SSL. Remaining four samples showed the protein content in between this range. Jadhav et al., (2014) reported that the protein content of lassi was 2.70% (Jadhav et al., 2014). The salt content of lassi samples was in a range between 0.50% to 2.08%. High salt content i.e. 2.08% was found in lassi sample namely WRL while lower percentage 0.50% of salt was found in lassi sample namely SGL. Remaining seven samples showed salt content in between this range. The fat content of lassi samples was in a range between 1% to 1.8%. Higher percentage i.e.1.8% of fat content was found in lassi sample namely SGL while lower percentage i.e. 1% of fat content was found in lassi sample namely WTL. Remaining seven samples showed fat content in between this range. The ash content in lassi samples was in a range between 0.39% to 0.41%. Lower percentage i.e.0.39% of ash content was found in lassi sample namely SShL and higher percentage i.e. 0.41% of ash contents was found in two lassi samples namely SSL and SGL. Remaining six samples namely WTL, WRL, RShL, RAL, SNL and RGL showed ash contents of 0.40%. Jangle et al., (2011) reported that the protein and fat content of lassi was 2.70% and 3.21% respectively (Jangle et al., 2011).

**Graph 3:**



The total solids found in lassi samples was in a range between 24% to 27% (**Graph 3**). Higher percentage i.e. 27% of total solids was found in lassi sample namely SSL while lower percentage i.e. 24% of total solids was found in lassi sample namely WTL. Remaining seven samples showed total solids content in this range. The SNF content in lassi samples was in a range between 16% to 21%. Lower percentage i.e. 16% of SNF content was found in three lassi samples namely WRL, RShL and SGL while higher percentage i.e. 21% of SNF content was found in lassi sample namely WTL. Remaining five samples showed SNF content in between this range. Processing technology report for value addition of milk by NDRI, Karnal reported the SNF content of lassi to be 19%. Physico-chemical compositions of custard apple lassi were determined by Jangle et al., (2011). They have reported that the best quality custard apple lassi can be obtained by incorporation of 5% custard apple pulp. The product had 3.21% fat, 24.42% total solids, 75.55% moisture, 0.867% acidity and 2.70% protein (Jangle et. Al., 2011).

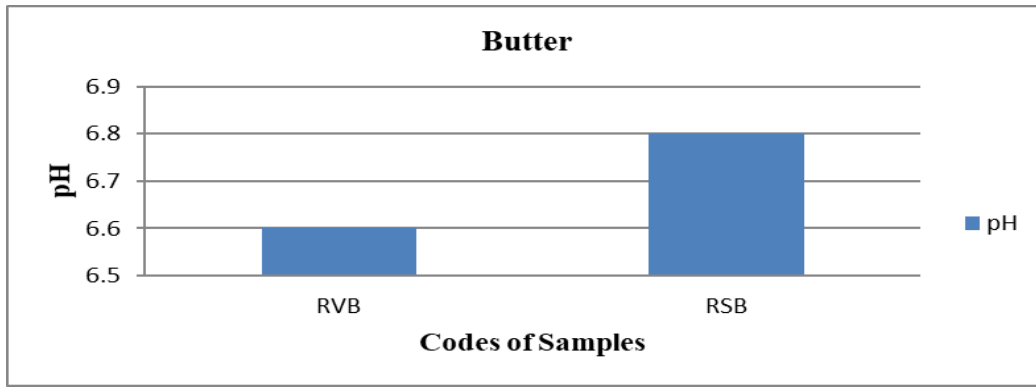
### **Chemical Examination of Butter:**

Chemical parameters such as pH, salt content (sodium chloride), fat, total solids, SNF, proteins, sucrose and ash content were determined for butter samples.

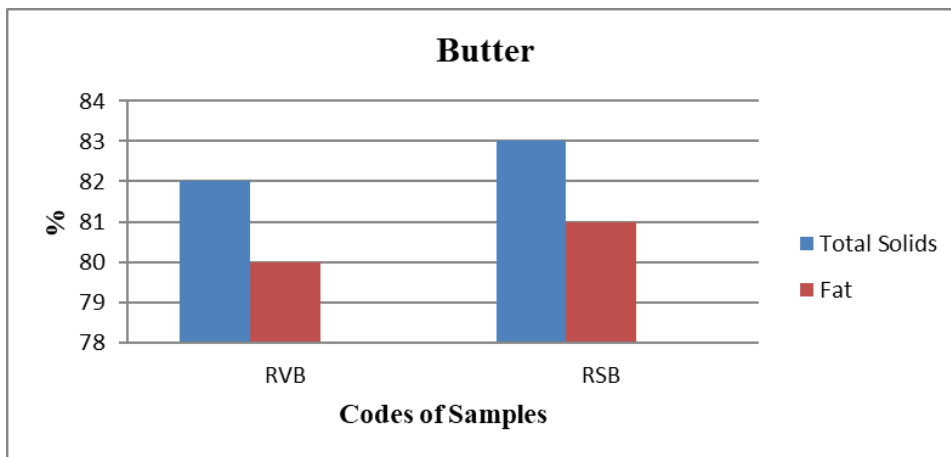
It can be seen from the **Graph 4** that the pH of two butter samples namely RVB and RSB was 6.6 and 6.8 respectively. The total solids content found in both the butter samples namely RVB and RSB was 82% and 83% respectively (**Graph 5**). Salt was not found in both butter samples collected from two dairies. The fat content in two butter samples namely RVB and RSB was found to be 80% and 81% respectively. SNF content of butter samples namely RVB and RSB was 1% and 1.5% respectively (**Graph 6**). Protein content in butter samples namely RVB and RSB was 0.5% and 1.0% respectively. Ash contents of both the butter samples was 0.40%.

Chemical composition of butter was determined by Enb et al, (2009). They have reported the values as pH (6.65), fat (79.40%), total solids (82.50%) and ash (0.25%) in butter (Enb et al., 2009).

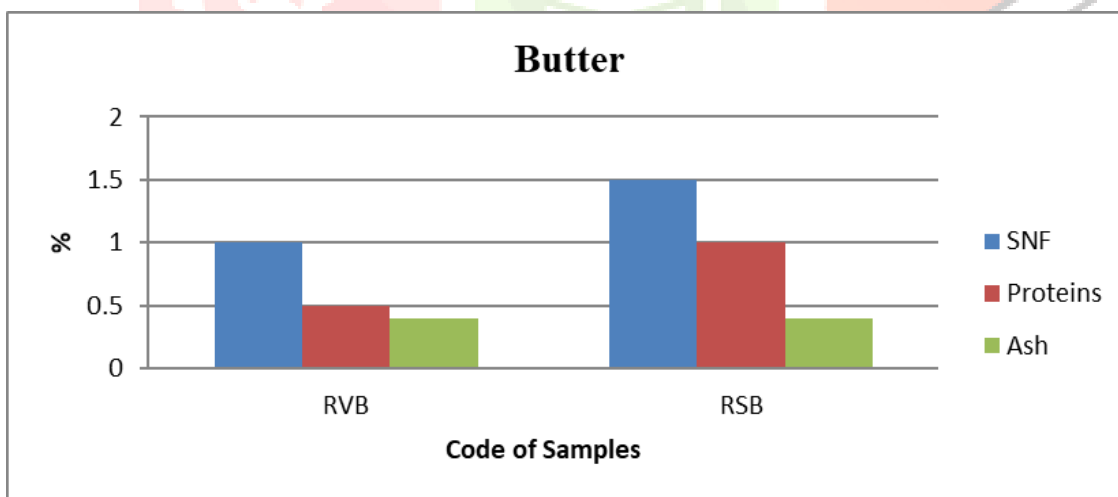
**Graph 4:**



**Graph 5:**



**Graph 6:**





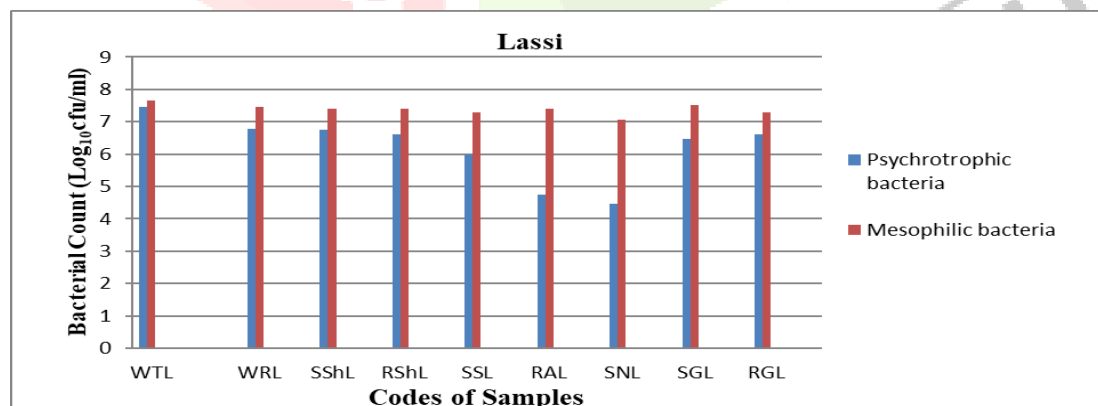
### 3.2 Enumeration of psychrotrophic and mesophilic bacteria from lassi and butter:

#### Enumeration of psychrotrophic and mesophilic bacteria from lassi:

It can be seen from the **Graph 7**, the average number of psychrotrophic bacteria present in lassi samples was in a range between 4.47 to 7.47  $\log_{10}$  cfu/ml. It was found that the lassi sample namely WTL showed higher number of psychrotrophic bacteria i.e. 7.47  $\log_{10}$  cfu/ml while lassi sample namely SNL showed less number of psychrotrophic bacteria i.e. 4.47  $\log_{10}$  cfu/ml. The number of psychrotrophic bacteria found in remaining samples was in this range. Ibrahim et al., (2005) reported the bacteriological examination shown that the mean psychrotrophic counts/ml in examined raw cow's milk, UHT milk, plain yoghurt, fruit yoghurt and white soft cheese samples were  $1.9 \times 10^3$ ,  $5.5 \times 10^2$ ,  $1.1 \times 10^3$ ,  $1.9 \times 10^3$  and  $1.7 \times 10^3$  respectively (Ibrahim, 2005).

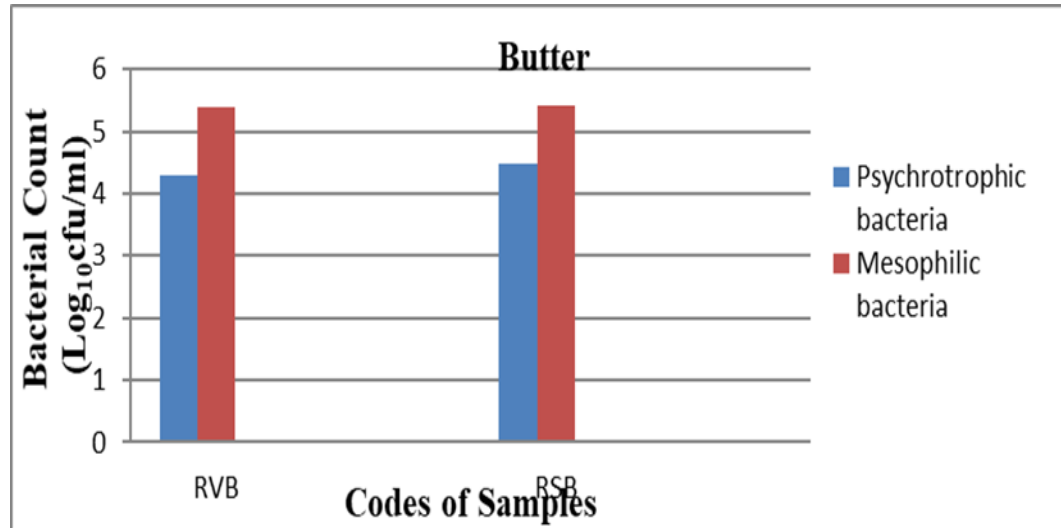
Average number of mesophilic bacteria present in lassi samples was in a range between 7.07 to 7.65  $\log_{10}$  cfu/ml. It was found that the lassi sample namely WTL showed higher number of mesophilic bacteria i.e. 7.65  $\log_{10}$  cfu/ml while lassi sample namely SNL showed less number of mesophilic bacteria i.e. 7.07  $\log_{10}$  cfu/ml. The number of mesophilic bacteria found in remaining samples was in this range. Mane et al., (2010) reported that average numbers of thermophilic psychrotrophs present in five lassi samples were  $36.73 \times 10^3$ ,  $25.31 \times 10^3$  and  $41.23 \times 10^3$  in summer, rainy and winter seasons respectively (Mane N.V. and Gandhi M.B., 2010).

**Graph 7:**



## Enumeration of psychrotrophic and mesophilic bacteria from butter:

Graph 8:



It can be seen from the **Graph 8** that the average number of psychrotrophic bacteria present in butter sample namely RVB was found to be 4.30 log<sub>10</sub> cfu/ml while the average number of psychrotrophic bacteria present in butter sample namely RSB was found to be 4.47 log<sub>10</sub> cfu/ml. These results indicate that butter samples of both the dairies showed presence of measurable number of psychrotrophic bacteria. The average number of mesophilic bacteria present in butter sample namely RVB was found to be 5.39 log<sub>10</sub> cfu/ml while the average number of mesophilic bacteria present in butter sample namely RSB was found to be 5.41 log<sub>10</sub> cfu/ml. These results indicate that both the butter samples showed presence of measurable number of mesophilic bacteria which was comparatively higher than the psychrotrophic bacterial count of respective butter samples. Kasana et al., (2002) reported the average TVC of mesophilic bacteria of butter samples from Ludhiana was 3.1 x 10<sup>5</sup> cfu/ml which was less than the average number of mesophilic bacteria reported in this work (Kasana et al., 2002). Elionora, H.Z. and Malka, H., (2007) reported the average percentage of the psychrotrophic bacterial population out of the total mesophilic population was 14.7%+6.4% in milk collected from Israel and psychrotrophs can cause about 10% lose in milk fats and proteins (Elionora HZ and Malka H, 2007).

## CONCLUSION:

The consumption of milk and milk products, including dairy based functional foods, is now more confidently and scientifically linked to enhanced consumer health and reduced risk of diseases. Chemical contents of different samples were within the range mentioned by Bureau of Indian Standards. Enumeration of bacteria from these samples showed the presence of mesophilic as well as psychrotrophic bacteria in each of the samples and there was a considerable variation in mesophilic and psychrotrophic bacterial content amongst the samples. Growth mesophilic

and psychrotrophic bacteria may result in development of undesirable flavours and spoilage of milk and milk products. Spoilage of milk and milk products may be avoided or prevented by controlling mesophilic and psychrotrophic organisms in milk and milk products.

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