

Effect of Probiotic Strains on Sensory Attributes of Buffalo Milk Cheddar Cheese

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Abstract Probiotics are living micro-organisms which impart certain health benefits like anti-oxidant, anti-hypertensive, immune-modulatory, opioid, anti-inflammatory, anti-microbial and consumer related inherit nutrition with the utilization in recommended quantity. The study aimed to determine the effect of *Lactobacillus acidophilus*, *Bifidobacterium bifidum* and *Lactobacillus helveticus* and their adjuncts on sensory attributes of cheddar cheese. The cheddar cheese having commercially available mesophilic starter cultures and with probiotics strains were prepared and assessed for acetic acid production, proteolysis and descriptive sensory analysis. The probiotics adjuncts *Lb. acidophilus* + *Bifidobacterium bifidum* (La + Bb) and *Lb. acidophilus* + *Bifidobacterium bifidum* + *Lb. helveticus* (La + Bb + Lh) showed remarkable results in production of acetic acid 489.33 (ppm) and as compared to control respectively. Among probiotic strains, the probiotic strain *Lactobacillus helveticus* showed the highest water-soluble nitrogen as 28.50 (%) and free amino acid 5220 µg/g. non-significant effect was observed for overall acceptability for control and probiotic adjuncts. Higher rates of organic acids and proteolysis was observed in probiotic adjuncts as compared to control samples. The current study reveals that the addition of probiotics adjuncts to cheddar cheese does not affects its overall acceptability.

Keywords: probiotics, sensory, cheddar cheese, proteolysis, functional foods, buffalo milk

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1. Introduction

Cheese is the fresh or ripened dairy product prepared by coagulation of casein and drainage of whey protein. It is one of the most conventional approaches for nutrients preservation of milk [1]. For cheese production, appropriate starter bacteria and rennet are utilized for casein coagulation and acid production [2]. More than 2000 varieties of cheese are available in the market depending on taste, texture, composition, manufacturing technology, ripening temperature and ripening period [3].

Cheddar is an example of hard-ripened cheese. All essential nutrients like protein, carbohydrates, fat, amino acids, fatty acids, minerals (phosphorus, zinc, calcium) vitamins are abundantly available in its matrix [4]. Three basic biochemical reactions known as glycolysis, lipolysis, and proteolysis initiates during cheese ripening. Typical

flavor, texture and sensory attributes of cheese are associated with these reactions during ripening [5].

Indigenous milk enzymes, starter, non-starter lactic acid bacterial enzymes, and rennet initiate proteolysis. Proteolytic enzymes contribute to decompose casein into peptides, amino acids, free amino acids, and other metabolic products [6]. The cheddar cheese flavor is associated to release of these compounds.

Sensory attributes play a major role in the marketing and sales of any food products. In the past, multiple conventional techniques were adopted in the industry to judge the sensory attributes of food and dairy products. Currently different descriptive sensory analysis techniques like spectrum, texture analyzer, flavor profile techniques [7] free choice profiling and quantitative descriptive analysis are applied for the said purpose [8]. Descriptive sensory analysis is the most reliable and effective tool for sensory assessment of dairy products. For the purpose a panel of 6-12 people is properly educated, trained and

mock sensory trials are conducted for different sensory aspects like flavor, texture, aroma, and appearance of cheese [9].

Probiotics are living microbes having positive health impact against diseases apart from their inherent nutritional characteristics [10]. Probiotics are being introduced into a variety of food products like juices, drinks, ice-cream, fermented and non-fermented dairy products to avail their health benefits [11].

Keeping in view the compositional and technological characteristics, cheese offers the best delivery potential for probiotics into the human gastrointestinal tract [12]. For consumer acceptance, probiotic strain incorporated in cheese must maintain or improve its sensory characteristics and viability as of traditional one [13]. *Lactobacillus* is a probiotic adjunct in cheese upturn the rate of proteolysis resulting in flavor intensification. Some of the probiotic adjuncts may lead to the production of bitter peptides in cheese [14].

Starter lactic acid bacteria convert lactose into lactic acid by lowering the pH of cheddar from 5.1-5.4 during glycolysis. During cheese manufacturing process, almost 98 % of lactose is converted into lactic acid or drained out. Along with ripening, the rest of the lactose is converted into L-lactate and it is further metabolized into acetic acid and carbon dioxide by starter bacteria, non-starter lactic acid bacteria and probiotic adjuncts [15]. Probiotic adjuncts like *Lactobacillus* and *Bifidobacterium* promote the production of acetate. Acetate plays a key role in the flavor production of cheese. Increased concentrations of acetate may lead to off-flavor in the end product which is not desirable [16].

In current study, one sample of cheddar cheese is prepared from commercial cheddar starter culture and seven by using three probiotic adjuncts (*Lactobacillus acidophilus*, *Bifidobacterium bifidum*, and *Lactobacillus helveticus*) along with their combinations. The probiotic adjuncts were selected keeping in view their pathogenic and metabolic behavior, anti-biotic resistance, viability and stability of strain (s) and their ability to tolerate and retain bile environment. All of the selected probiotic strains maintained their recommended viability during ripening for 120 days at 4°C. The purpose of the present study was to determine the effect of probiotic adjuncts on sensory attributes of the cheddar cheese after 120 days of ripening at 4°C.

2. Materials and Methods

Raw milk for Probiotic and control samples were purchased from Hafiz Dairy Farm Sargodha, Pakistan. Commercially available mesophilic Cheddar cheese starter cultures and probiotic adjuncts (Chr. Hansan Ireland Ltd. Rohan Industrial Estate, Little Island, Co. Cork, Ireland) were used for control and probiotic cheddar cheese production. Milk was firstly analyzed to check the suitability for cheese production.

2.1. Preparation of Probiotic Cheddar Cheese

Control and probiotic samples of cheddar cheese were prepared as described by Ong *et al.*, [15]. Seven probiotic

samples were produced by three probiotic adjuncts in combinations. The sample plan is described below in Table 1. All cheese samples were ripened for 120 days at 4 °C.

Table-1. Plan of study

| Sr. | Samples | Starter Cultures |
|-----|--------------|---|
| 1 | Control | <i>L. Lactis ssp. Lactis</i> and <i>L. Lactis ssp. cremoris</i> (Mesophilic Cheddar cheese starter cultures) (MCSC) |
| 2 | La | MCSC + <i>Lb. acidophilus</i> |
| 3 | Bb | MCSC + <i>Bifidobacterium bifidum</i> |
| 4 | Lh | MCSC + <i>Lb. helveticus</i> |
| 5 | La + Bb | MCSC + <i>Lb. acidophilus</i> + <i>Bifidobacterium bifidum</i> |
| 6 | La + Lh | MCSC + <i>Lb. acidophilus</i> + <i>Lb. helveticus</i> |
| 7 | Bb + Lh | MCSC + <i>Bifidobacterium bifidum</i> + <i>Lb. helveticus</i> |
| 8 | La + Bb + Lh | MCSC + <i>Lb. acidophilus</i> + <i>Bifidobacterium bifidum</i> + <i>Lb. helveticus</i> |

* MCSC: Mesophilic cheese starter culture.

2.2. Physio-chemical Analysis of Cheddar Cheese

Probiotic and control cheddar cheese were analyzed for moisture, fat, protein and pH by following the standard methods of AOAC [17].

2.3. Acetic Acid Production

Acetic acid production was assessed by using high-performance liquid chromatography (HPLC) as described by Akalin *et al.* [18] after 120 days of ripening at 4 °C.

2.4. Assessment of Proteolysis

Water-soluble nitrogen (WSN) and total free amino acids (TFAA) were assessed for both control and probiotic adjuncts after 120 days of ripening at 4 °C as described by Rulikowska *et al.* [19].

2.5. Descriptive Sensory Assessment

2.5.1. Panel Selection and Training

For descriptive sensory assessment ten faculty members and students from the Institute of Food Science and Nutrition (IFSN), University of Sargodha (Pakistan) were selected after taking their consent. Five briefing sessions for basic sensory techniques, sensory parameters, and sensory methods were arranged for effective sensory assessment of the product [20]. In training and briefing sessions, the panelists were trained for their capacity building to judge precisely the creamy, sweet, salty, sour and rancid flavor and odor. The panelist was provided access to deionized water and unsalted soda crackers.

2.5.2. Descriptive Sensory Assessment

The descriptive sensory analysis was conducted after 120 days of ripening at 4°C. For descriptive sensory assessment, all cheese samples were taken out from the refrigerator and divided into uniform pieces (1.5 x 1.5

x1.5 cm) at kept at room temperature (about 25°C) one hour before evaluation. All samples were prepared in triplicates and coded with four-digit numbers. Panelists were asked to evaluate their perception towards all samples in triplicate and record their liking and disliking ranging from 0-100 for all given parameters of cheese. All the samples were assessed in triplicate and descriptive scores were allotted to all sensory parameters within 100.

2.6. Statistical Analysis

The results obtained by the sensory analysis were analyzed using a one-way analysis of variance (ANOVA). The means were compared by Turkey test [21]. The data were presented as mean values \pm standard error (SE).

3. Results and Discussions

3.1. Physio-chemical Analysis of Cheese

The composition of control and probiotic cheese samples were analyzed for pH, moisture, fat, and protein after 120 days of ripening at 4°C. Results are summarized in Table 2. Non-significant difference ($P > 0.05$) was observed within samples for pH, moisture, fat and protein contents. Keeping in view the results, it was concluded that the addition of all three probiotic adjuncts and their combinations did not affect the cheese composition

which confirms the findings of Gardiner et al., [22] and Ong et al., [23].

3.2. Acetic Acid Production

Acetic acid and diacetyl are produced during glycolysis. Lactose is converted into lactic acid and eventually into acetic acid by pyruvate metabolism through lactic dehydrogenase and pyruvate formate lyase. Acetic acid behaves as a flavoring compound in cheddar cheese [24]. Concentrations of acetic acid in control and probiotic adjuncts after 120 days of ripening at 4 °C are described in Table 3.

The results indicate probiotic adjuncts produced significantly higher ($P < 0.05$) quantities of acetic acid as compared to the control sample (430 \pm 17.32 ppm). The maximum acetic acid production (516.7 \pm 25.17 ppm) was recorded in probiotic adjunct having *Bifidobacterium bifidum*. All probiotic adjuncts having *Bifidobacterium bifidum* (sample no. 03, 05 and 07) produced higher quantities of acetic acid as compared to other ones.

Bifidobacterium bifidum is capable to produce acetic acid and lactic acid from lactose by fructose-6-phosphate shunt pathway [25]. Acetic acid produces vinegary flavor in the final product. The increased quantity of acetic acid in probiotic adjuncts was traced in sensory scores during descriptive sensory analysis. All probiotic cheese samples secured higher vinegary flavor as compared to control but the maximum scores were obtained by probiotic adjuncts having *Bifidobacterium bifidum*. The overall acceptability of all probiotic cheese samples was within acceptable limits.

Table 2. Effect of Probiotic adjuncts on composition and proteolytic pattern of Cheddar cheese after 120 days of ripening at 4°C

| Samples | Moisture (%) | Fat (%) | Protein (%) | pH | Acetic acid (ppm) | WSN (%) | FAA (μ g/g) |
|--------------|-------------------|-------------------|-------------------|------------------|-----------------------|-------------------|-----------------------|
| Control | 36.41 \pm 0.11A | 30.48 \pm 0.07A | 26.54 \pm 0.14A | 4.90 \pm 0.06A | 430 \pm 17.32 D | 23.77 \pm 0.19D | 4480 \pm 65.57D |
| La | 36.42 \pm 0.09A | 30.47 \pm 0.08A | 26.51 \pm 0.18A | 4.90 \pm 0.04A | 450 \pm 26.46 CD | 27.76 \pm 0.10C | 5069.7 \pm 52.54D |
| Bb | 36.42 \pm 0.09A | 30.47 \pm 0.08A | 26.55 \pm 0.12A | 4.89 \pm 0.05A | 516.7 \pm 25.17A | 27.78 \pm 0.14C | 5123.3 \pm 37.86BC |
| Lh | 36.40 \pm 0.17A | 30.46 \pm 0.11A | 26.53 \pm 0.17A | 4.89 \pm 0.09A | 451.7 \pm 20.21 BCD | 28.50 \pm 0.13A | 5220 \pm 36.06A |
| La + Bb | 36.4 \pm 0.11A | 30.47 \pm 0.13A | 26.52 \pm 0.12A | 4.9 \pm 0.04A | 489.33 \pm 16.77 AB | 27.77 \pm 0.11C | 5136.7 \pm 47.26ABC |
| La + Lh | 36.41 \pm 0.06A | 30.47 \pm 0.17A | 26.54 \pm 0.14A | 4.91 \pm 0.05A | 458 \pm 20.30 BCD | 28.14 \pm 0.10B | 5203.3 \pm 49.33AB |
| Bb + Lh | 36.41 \pm 0.08A | 30.47 \pm 0.15A | 26.53 \pm 0.12A | 4.90 \pm 0.05A | 470.7 \pm 25.32 BC | 28.19 \pm 0.14B | 5214 \pm 39.85A |
| La + Bb + Lh | 36.4 \pm 0.08A | 30.49 \pm 0.11A | 26.52 \pm 0.12A | 4.89 \pm 0.03A | 456.7 \pm 20.82 BCD | 28.10 \pm 0.11B | 5183.3 \pm 51.32AB |

* Sample codes are described in Table 1, ** WSN: Water soluble nitrogen, *** FAA: Free amino acids

The results are expressed as mean \pm standard error of means. ABC means in column with like superscript do not differ ($P > 0.05$)

Table 3. Sensory Scores for flavor obtained by Probiotic adjuncts of Cheddar cheese after 120 days of ripening at 4°C

| Samples | Creamy | Sweet | Salty | Vinegary | Bitter | Soapy | Smoky | Rancid |
|--------------|--------------------|--------------------|--------------------|-------------------|--------------------|-------------------|--------------------|------------------|
| Control | 19.26 \pm 0.14A | 10.2 \pm 0.09A | 19.25 \pm 0.10AB | 8.36 \pm 0.07D | 7.13 \pm 0.04C | 3.62 \pm 0.06A | 3.97 \pm 0.07AB | 4.23 \pm 0.05B |
| La | 18.92 \pm 0.08C | 10.11 \pm 0.07AB | 19.17 \pm 0.06B | 8.51 \pm 0.09C | 7.30 \pm 0.08AB | 3.44 \pm 0.06BC | 3.99 \pm 0.07A | 4.20 \pm 0.09B |
| Bb | 19.06 \pm 0.05BC | 10.02 \pm 0.13B | 19.5 \pm 0.08A | 9.02 \pm 0.08A | 7.35 \pm 0.07A | 3.44 \pm 0.07BC | 3.85 \pm 0.07C | 4.43 \pm 0.06A |
| Lh | 19.21 \pm 0.05AB | 10.07 \pm 0.09AB | 19.22 \pm 0.07AB | 8.5 \pm 0.08C | 7.21 \pm 0.07BC | 3.39 \pm 0.07BC | 3.87 \pm 0.05BC | 4.25 \pm 0.07B |
| La + Bb | 19.21 \pm 0.07AB | 9.82 \pm 0.09C | 19.23 \pm 0.09AB | 8.85 \pm 0.07B | 7.30 \pm 0.08AB | 3.44 \pm 0.06BC | 3.96 \pm 0.09ABC | 4.37 \pm 0.05A |
| La + Lh | 19.17 \pm 0.08AB | 10.14 \pm 0.14AB | 19.23 \pm 0.09AB | 8.44 \pm 0.06CD | 7.29 \pm 0.08AB | 3.51 \pm 0.09AB | 3.93 \pm 0.05ABC | 4.24 \pm 0.08B |
| Bb + Lh | 18.99 \pm 0.07C | 9.76 \pm 0.06C | 19.16 \pm 0.09B | 8.83 \pm 0.07B | 7.33 \pm 0.06AB | 3.49 \pm 0.10B | 3.92 \pm 0.08ABC | 4.37 \pm 0.05A |
| La + Bb + Lh | 18.92 \pm 0.13C | 10.02 \pm 0.06AB | 19.12 \pm 0.10B | 8.50 \pm 0.09C | 7.23 \pm 0.09ABC | 3.36 \pm 0.06C | 3.86 \pm 0.06BC | 4.24 \pm 0.6B |

The results are expressed as mean \pm standard error of means. ABC means in column with like superscript do not differ ($P > 0.05$)

3.3. Proteolytic Pattern of Probiotic Cheddar Cheese

Proteolysis directly contributes to flavor, texture and sensory attributes of cheddar cheese [26]. The structure of the para casein matrix is hydrolyzed and the water-binding capacity of curd improved during proteolysis resulting in a modification in structure and texture respectively [27].

Indigenous milk enzymes, starter bacteria, non-starter lactic acid bacteria, and coagulants participate during proteolysis. Water-soluble nitrogen (WSN) fractions maintain the balance of whey protein, amino acids, and peptides of all sizes in curd. Secondary proteolysis metabolites (Free amino acid and small molecular weight peptides) are measured by water-soluble nitrogen percentage [28]. Along with the ripening period, the digestibility of casein is continually improved and quantities of free amino acid and bioactive peptides are increased [29].

The proteolysis of control and probiotic adjuncts were analyzed after 120 days of ripening at 4 °C by measuring the water-soluble nitrogen percentage (WSN %) and total free amino acids (TFAA). A significant difference ($P < 0.05$) was observed between control and probiotic adjuncts for water-soluble nitrogen percentage and total free amino acid. Water-soluble nitrogenous components of casein directly add to cheese flavor and texture.

The levels of water-soluble nitrogen percentage and Total free amino acid was observed the highest in probiotic adjunct having *Lactobacillus helveticus* 28.50±0.13 % and 5220±36.06 µg/g respectively. *Lactobacillus helveticus* possess higher proteolytic activity as compared to other probiotic adjuncts used in this study. All probiotic adjuncts having *Lactobacillus helveticus* showed higher water-soluble nitrogen percentage and total free amino acid contents as compared to other probiotic adjuncts. The results indicate that probiotic adjuncts ripened faster as compared to the control cheese sample.

During the descriptive sensory evaluation, higher cheddar scores were expected for probiotic adjuncts. However, according to results, a non-significant relation was observed within samples. Excessive proteolysis results in the bitterness of the product [30]. The result of this study reveals a positive correlation between scores of bitterness and proteolysis.

3.4. Descriptive Sensory Evaluation of Probiotic Cheddar Cheese

3.4.1. Flavor Description of Probiotic Cheddar Cheese

Taste, smell, and chemical irritation are three basic flavor-developing parameters of any product [31]. Taste is developed in all dairy products by organic acids and non-volatile sugars present in it, while the smell is produced by volatile compounds and chemical irritation of gaseous components present in it [32].

Descriptive sensory judgment for control and probiotic cheddar cheese after 120 days of ripening at 4°C was performed for its creamy, sweet, salty, vinegary, bitter, soapy, smoky and rancid flavor. A mean square table for the flavor of probiotic cheddar cheese is described in Table 3. A significant ($P < 0.05$) effect was observed

between control and all probiotic cheese samples. The control cheese sample secured the highest (19.26±0.14) scores for its creamy flavor. It was concluded by the present study that selected probiotic adjuncts to reduce the creamy flavor of cheddar cheese. Cheddar cheese having a combination of all three probiotic adjuncts secured the minimum creamy scores (18.92±0.13) among all samples.

A significant difference ($P < 0.05$) was observed for sweet flavor between control and probiotic adjuncts. The highest scores (10.2±0.09) for sweet flavor were awarded to control cheese sample. Cheddar cheese sample having probiotic adjunct *Bifidobacterium bifidum* and *Lactobacillus acidophilus* secured the minimum sensory scores (9.76 ±0.06) for sweetness. The salty, vinegary, bitter and rancid flavor was observed maximum in probiotic adjunct having *Bifidobacterium bifidum* scoring 19.5±0.08, 9.02±0.08, 7.35±0.07 and 4.43±0.06 respectively.

It has been observed in previous research that probiotic adjuncts may improve the cheddar cheese flavor. Overall acceptability of control and probiotic adjuncts is described in Table 3. Non-significant difference ($P > 0.05$) was observed in the overall acceptability of the control and probiotic adjuncts. The present study revealed that the addition of selected probiotic adjuncts did not affect the overall acceptability of probiotic cheddar cheese. The lowest overall acceptability scores were specified for probiotic adjuncts having *Bifidobacterium bifidum*.

Cheddar cheese sample having probiotic adjuncts achieved higher scores for salty, vinegary, bitter, smoky and rancid flavor. El Soda et al. found a higher bitterness level in probiotic cheddar cheese as compared to control support the results of the present study [33]. He associated the cheese bitterness with peptidase systems within the cheese matrix.

The flavor of probiotic cheese mainly depends on the probiotic strain, processing technology, processing conditions and ripening time and temperature. Every probiotic strain has its metabolic activities [34]. It has been established by multiple researches works that suitable probiotic strains did not affect the overall acceptability of cheese. Cheese varieties having *Bifidobacteria* produce higher concentrations of acetic and lactic acid as compared to control [35]. The increased amounts of acetic and lactic acid may affect the sensory characteristics of the product. It is evident in the present study that *Bifidobacterium bifidum* containing probiotic samples possess more bitterness and vinegary flavor.

Lactobacillus acidophilus in fresh cheese did not affect the sensory attributes (texture and flavor) during 15 days of ripening at 5 °C [29]. Bruiti et al. worked on fresh cheese and observed similar findings to the current study that overall acceptability, flavor, and odor did not affect significantly the addition of *Lactobacillus acidophilus* in cheese varieties [9].

Flavor acceptability of probiotic cheddar cheese might be lower as compared to control because of continuous break down of protein into peptides and free amino acids by lactic acid and starter bacteria and their respective enzymes [36].

Sensory characteristics of probiotic cheese are much influenced by the quantity of probiotic strain used during manufacturing. Higher amounts of probiotic strains

accelerate the proteolytic pattern, acid production, and peptide formation. Gomes et al. used increasing amounts of probiotic strains (0, 0.4 and 0.8 g/L) during cheese manufacturing and observed the response towards sensory attributes. A negative correlation was observed between sensory attributes and the number of probiotic strains (s). The increasing amount of probiotic strains reduced the sensory scores for flavor, bitterness and overall acceptability of cheese. [37].

It has been observed in a study by Katsiari et al. that probiotic strains *Lactobacillus casei ssp. Rhamnosus* along with *Lactobacillus lactis ssp. lactis* and *Lactococcus lactis ssp. cremoris* in combination are capable to improve the sensory characteristics of probiotic cheese as control after 90 and 180 days of ripening in [38]. The peptides produced by *Lactococcus lactis ssp. cremoris* improves to flavor and other sensory attributes of cheese [39].

Table 4. Sensory Scores for Texture obtained by Probiotic adjuncts of Cheddar cheese after 120 days of ripening at 4°C

| Samples | Crumbly | Firmness | Grainy |
|--------------|---------------|-------------|------------|
| Control | 15.64±0.12BC | 28.05±0.06A | 8.71±0.06A |
| La | 15.84±0.12A | 28.03±0.06A | 8.75±0.07A |
| Bb | 15.55±0.11C | 28.04±0.05A | 8.77±0.07A |
| Lh | 15.69±0.08ABC | 28.04±0.04A | 8.76±0.05A |
| La + Bb | 15.65±0.08BC | 28.05±0.06A | 8.76±0.07A |
| La + Lh | 15.74±0.12AB | 28.04±0.04A | 8.75±0.06A |
| Bb + Lh | 15.68±0.09ABC | 28.04±0.07A | 8.77±0.07A |
| La + Bb + Lh | 15.69±0.08ABC | 28.05±0.06A | 8.76±0.05A |

The results are expressed as mean ± standard error of means. ABC means in column with like superscript do not differ (P>0.05).

3.4.2. Texture of Probiotic Cheddar cheese

Texture plays a significant role in the overall acceptability of food products [40]. It is interlinked with the quality of the product. The texture is a functional and sensory description by senses (vision, hearing, touching) and kinetics for the physical, mechanical and surface properties of foodstuff [41]. Texture perception may vary from person to person because of variable factors like tongue pH, the temperature of mouth and composition of salivary glands [26].

Proteolysis plays a key role in the texture development of cheese [42]. The smooth and firm texture could be obtained by a balanced breakdown of casein into peptides and amino acids. Mesophilic and thermophilic lactic acid bacteria produce acetaldehyde. Elevated production of acetaldehyde gives rise to texture defects. It has been observed that *Bifidobacterium ssp.* is capable to produce higher amounts of acetaldehydes as compared to other strains of probiotic and normal starter bacteria [24].

The second major factor in cheese texture is the rate of acidification. All probiotic adjuncts produce more amounts of lactic acid as compared to the control sample. The amount of acid production in cheese depends upon probiotic strain (s) selection, the quantity of probiotic strain used, proteolytic and ripening conditions of the product [43].

The probiotic and control cheddar cheese were accessed for crumbly, firmness and grainy texture after 120 days of ripening at 4°C. Means square for the texture is summarized in Table 4. The highest scores (15.84±0.12) for crumbly texture were given to probiotic adjunct having

Lactobacillus acidophilus. A non-significant effect (P>0.05) was observed for grainy and firm texture between control and all probiotic adjuncts.

Table 5. Overall acceptability of Probiotic adjuncts of Cheddar cheese after 120 days of ripening at 4°C

| Samples | Overall Acceptability |
|--------------|-----------------------|
| Control | 6.41±0.09A |
| La | 6.40±0.08A |
| Bb | 6.34±0.07A |
| Lh | 6.34±0.08A |
| La + Bb | 6.38±0.08A |
| La + Lh | 6.37±0.08A |
| Bb + Lh | 6.35±0.10A |
| La + Bb + Lh | 6.36±0.07A |

The results are expressed as mean ± standard error of means. ABC means in column with like superscript do not differ (P>0.05).

The results of Dinakar and Mistry endorse the present study. They worked on cheese and recorded non-significant difference in texture between control and probiotic adjuncts. Some of the probiotic strains influence the texture properties of cheese by accelerating the proteolytic activity and acidification [44]. Ong et al. worked on cheddar cheese and observed lesser hardness scores in probiotic adjuncts (*B. longum*, *Lb. Casei*, and *Lb. paracasei*) as compared to control [45].

Rapid acidification in cheese by some probiotic strains may lead to demineralization of casein Heller 2003. Rapid acidification tends to rise in moisture content ultimately affecting the texture and quality of cheese [46]. Intensive lactic acid produced by probiotic adjuncts weakens the calcium to casein bond resulting in brittle texture in cheese.

Another factor affecting cheese texture is the quantity of salt used during its manufacturing. Salt contents directly influence the acidification and proteolysis of all cheese varieties. Free water in cheese is drained by salt. Gomes and Malkata (1998) studied the effect of salt concentrations on the texture of probiotic goat cheese. He observed decreased protein matrix volume with an increase in salt concentration [47].

4. Conclusion

Cheese could be a better carrier for probiotic adjuncts as compared to other food products because of its higher potential for maintaining the viability and stability of probiotic adjuncts. At consumption, the viability of probiotic adjuncts should not lesser than 10⁶ CFU/g without having any adverse effect on the sensory attribute of the product. In the present study, the non-significant effect was observed in the overall acceptability of all control and probiotic adjuncts. Fortunately, by selecting suitable probiotic strain (s), processing and ripening conditions probiotic cheddar cheese having excellent sensory attributes are produced. In the global market, certain probiotic cheddar cheese varieties owing to good sensory attributes are already available. In the future, it should be focused on the development of probiotic cheese varieties with multiple strains having excellent sensory attributes, more stability, and viability of probiotic adjuncts.

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