



A Comparison of the Biophysical Characteristics and Survival of Probiotic Bacteria in Yogurt Produced from Cattle, Goat, and Camel Milk

Najmeh Akrami¹, Seyyed Ali Yassini Ardakani², Mohammad Daneshi^{3*}

¹ Graduate student, Department of Food Science and Technology, Yazd Branch, Islamic Azad University, Yazd, Iran

² Associate Professor, Department of Food Science and Technology, Yazd Branch, Islamic Azad University, Yazd, Iran.

³ Assistant Professor, Department of Food Science and Technology, Yazd Branch, Islamic Azad University, Yazd, Iran.

*Corresponding Author

Abstract: Due to special therapeutic effects and nutritional value, nowadays, probiotic dairy products paved the ground for extensive researches about the methods of production and maintenance of these products and investigation of their therapeutic and nutritional aspects. Improving the growth of probiotic bacteria in milk is one of the solutions to the problems of the manufacturing technology of these products. This study aimed to investigate the difference of survival time in the probiotic bacteria and the biophysical characteristics of yogurt produced from different milk (cattle, goat, and camel milk) and to find out the preservation of probiotic bacteria and its biophysical characteristics during storage condition at 4 ± 2 °C. In the production of probiotic yogurt, *Lactobacillus acidophilus* and *Bifidobacterium* bacteria were directly used at 37 °C and the number of bacteria and biophysical characteristics and sensory properties during the three-week storage period was investigated. The comparison between the samples was done by using analysis of variance (ANOVA) and Fisher's exact test. The results showed that there is no significant difference between the two samples of probiotic bacteria (*Lactobacillus acidophilus* and *Bifidobacterium*) at the level of 0.05.

Keywords: *Lactobacillus*, *Acidophilus*, *Bifidobacterium Lactis*, Probiotic, Yogurt.

INTRODUCTION

Yogurt is a product of fermented milk and has been produced for several thousands of years, and because of our nutritional benefits, it is of particular importance to human health (Aziznia, Khosrowshahi and Rahimi, 2008). Yogurt has been known as a product with favorable effects for consumers. In recent years, there has been a significant increase in the popularity of yogurt as a food product; especially the use of some starter in the production of yogurt increases its nutritional and physiological value (Lourent-Hattingh and Viljoen, 2001). Due to the presence of probiotics and its beneficial effect on the microflora of the digestive system, yogurt is considered as one of the most important and successful fermented dairy products in the field of functional foods. One of the characteristics of dairy products is to provide the necessary medium for the growth of many microorganisms. Therefore, it is possible to increase their nutritional value by adding one or more beneficial bacteria to dairy products (Kneifel, Jaros and Erhard, 1993). Probiotic bacteria are microorganisms that can grow in milk and yogurt. Probiotics are live microorganisms intended to provide

health benefits when consumed, generally by improving or restoring the microflora and has beneficial effects on the health of the consumer (Shrutt, 1999). One of the probiotic types is *Lactobacillus acidophilus*. The characteristics of probiotic bacteria and their culture medium are different, so, changes in milk fermentation conditions or optimization of this process can increase the number of probiotic bacteria and their lifetime in yogurt (IDF, 1998). Two-stage fermentation is another method used to produce and reproduce probiotic bacteria during the milk fermentation process. The method is to inoculate the probiotic bacteria separately into the milk and after one or two hours, they add starter bacteria to it. This results in the probiotic bacteria being able to fit better with their growth environment (Sodin, 1998).

Cow's milk contains 87.4% water and 12.6% solids (total solids). Solids are composed of 3.9% fat, 3.2% protein, 4.6% lactose (no water) and 0.9% other materials such as minerals, vitamins, and so on. Non-water compounds of milk are in various forms such as lactose, colloids (proteins) and emulsions in water (lipids or milk fat). The mentioned physical characteristics of the milk can be used to facilitate commercial separation and decomposition of the main composition of milk. Take (Çakmakçi et al., 2012). Chemically, cow's milk composition resembles goat's milk, while the amount of the compositions varies. Goat milk benefits from more dry matter, total protein and casein, fat and minerals which determines its nutritional value. Besides, goat's milk contains more Vitamin A, Vitamin B6, Sodium, and Niacin are higher when it is compared with the cow's milk. The composition of the fatty acid of the goat's milk is also different, as it is rich in volatile fatty acids (caproic, caprylic and capric) which indicate the taste and smell of the dairy products. When the amount of the medium-chain fatty acids is high, there is a longer bacistrostatic phase (Karim et al., 2006). On the other hand, camel's milk contains a small amount of short-chain fatty acids and a small amount of carotene which may make the camel's milk white. The camel's milk contains a variety of vitamins, including E, D, C, A and B. It is rich in vitamin C and can be considered as a good source of vitamins in desert areas where there are not enough vegetables and fruits. The amount of niacin in camel's milk is higher than that of the cow's milk. The amount of vitamin A and riboflavin in camel milk is lower than that of the cow's milk. The concentration of pantothenic acid, folic acid and B12 in the camel's milk is much higher than that of the cow's milk. However, the concentration of vitamin E in the camel's milk is very close to that in the cow's milk (Boycheva, et al., 2011).

In this regard, a few types of research have been done. Çakmakçi et al. (2012) examined probiotic characteristics, sensory quality, and stability of probiotic fruit yogurt with banana flavor. The results showed that the number of *Streptococcus Thermophilus*, *Lactobacillus Delbrueckii*, and *Bifidobacterium* bacteria decreased during storage. After seven days, the sensory quality and probiotic properties of the specimens decrease. Control yogurt and yogurt containing *Bifidobacterium Bifidum* received the highest score (Çakmakçi et al., 2012). Zubeirç et al. (2012) examined the qualitative characteristics of production, chemical, and acceptability of yogurt obtained other than cattle's milk (sheep, goat, and camel). The results showed that there was no difference in pH between the yogurts obtained from the three species of milk. The study of yogurt composition also showed that in yogurt produced from sheep's milk, fat content was significantly high but in yogurt produced from camel milk, fat and protein contents were low compared to other yogurts. In terms of sensory properties, the results of the evaluation showed that the yogurts obtained from the three species of milk have very high acceptability (Zubeirç et al., 2012).

Moayednia and Mazaheri (2011) showed that *Lb. Acidophilus* has activity and high growth rates compared to *B. Lactis* when they are cultivated pure in milk. The study on the milk used in the intercropping showed that *Lb. Acidophilus* had a stimulus effect on the growth rate of *B. Lactis*. However, their growth rate was not influenced by other bacteria (Moayednia and Mazaheri, 2011). Zhao et al. (2006) found that casein hydrolyzate reduced the yogurt fermentation time and increased the number of probiotic bacteria, but the total number is reduced during storage. Accordingly, in this research, the difference in survival time of probiotic bacteria and our biophysical characteristics of the yogurt in a product of different milk (cattle, goat, and camel) will be investigated.

Materials and Method

In this study, a yogurt's starter namely CH1 and YC-X11 from Kristin Hansen, Denmark, and BBL prebiotic starter for probiotic yogurts produced from *Bifidobacterium* as well as AC12 starter were used to produce probiotic yogurts from *acidophilus* manufactured by Micromilk Company in Italy. The culture medium of MRS BILE AGAR in German Merck Company was used to take a N/9 normal Caustic Soda, and Phenol-phthalene in this study.

The experiments were done by using the following equipment: pH meter, model; 004 manufactured by EON Corporation of Bulgaria, Texture analyzer, model; 10CTM 350 manufactured by ROCHDALE ENGLAND, and Brookfield Viscometer, model; I + PRO DV-I, manufactured in USA, and Centrifuge, model ; HB-108 Manufactured by Dian Azma Exploration Company.

In this study, the production and packaging of yogurt samples were done at Morvarid-e-Sahar Dairy Company in Yazd City in Iran. Microbiological and synergistic tests and acidity and pH were performed at the Laboratory of Standard and Industrial Research Center in Karaj (Idegostar-e-Razi) and Textile Testing and Viscosity Test were conducted at a laboratory in the Department of Agriculture, University of Tehran.

The initial stages of production were done by pasteurizing the milk using the LTLT method. Thus, cattle's milk with 3.3 % fat, goat's milk with 3.5 % fat and camel's milk with 3% fat without passing through the cream separator were introduced into the pasteurization step, and at this stage, they were placed at 65 °C for 30 minutes until pasteurization was performed. Then, it was cooled to 40 °C and inoculated with YC-X11 and CH1-BBL starter to produce probiotic yogurts containing *Bifidobacterium* and AC12 starter to produce probiotic yogurt containing *acidophilus* with 0.01 wt, which were previously prepared in non-fat sterile milk. In the next, inoculated milk was poured in polypropylene containers and their doors were closed with the aid of a packing machine with polystyrene foils. The incubation procedure was carried out for clot formation for 3.5 hours to achieve acidity of 70 °D at 37 °C. Finally, they were kept at 4 °C for 24 hours to increase firmness. The samples of probiotic yogurt were kept in a refrigerator at 4 °C for the duration of the microbial, physicochemical tests. After preparing probiotic yogurt samples, sampling of each treatment was carried out on days 7, 14, and 21 after the production for physicochemical and microbiological tests. By directly inserting the electrode of the pH meter into the homogeneous yogurt, the pH was measured according to National Iranian Standard No. 2852 (cited in the thesis of this paper). The acidity was measured and reported in NaOH (N/g). The specimen tissue test was carried out without removing yogurt from the container and a texture analyzer with a 13 mm diameter probe at 6 °C.

To measure WHC, 20 g of samples were first centrifuged for 30 minutes at 1250×g and 20 °C (height = 4.8 cm). The removed water (WE) was then separated and weighed, and the WHC was calculated according to equation 1 (Gosman-Gonzales et al., 1999).

Eq. 1

$$WHC(\%) = 100 \frac{(y - WE)}{y}$$

The count of *Acidophilus* and *Bifidobacterium* bacteria were performed according to the methods mentioned in the Iranian National Standard No. 9616 and 13772, respectively. Analysis of variance (ANOVA) was used to evaluate the acidity and pH differences between *Lactobacillus Acidophilus* and *Bifidobacterium*.

Analysis of variance was used to evaluate the acidity and pH differences between *Lactobacillus acidophilus* and *bifidobacterium*. In this method, the Fisher test statistic was used to test the hypothesis. If the value of this statistic is higher than the related value in the Fisher table, then the zero hypothesis is rejected. Besides, to test the hypothesis, a significance probability value can be used. Thus, when the significant value is lower than 0.05, the zero hypothesis is rejected.

Finding

Table 1: The probably significant value of inter-group and intra-group acidity and pH tests in different types of milk

		Sum of squares	Degrees of freedom	Average of squares	F-test statistic	The probably significant value
Acidity test in cattle's milk	inter-group	140.167	1	140.167	1.665	0.266
	intra-group	336.667	4	84.167		
	Total	476.833	5			
Acidity test in goat's milk	inter-group	160.167	1	160.167	1.441	0.296
	intra-group	444.667	4	111.167		
	Total	604.833	5			
Acidity test in camel milk	inter-group	0.000	1	0.000		
	intra-group	0.000	4	0.000		
	Total	0.000	5			
pH test in cattle's milk	inter-group	0.002	1	0.002	0.083	0.788
	intra-group	0.098	4	0.024		
	Total	0.100	5			
pH test in goat's milk	inter-group	0.014	1	0.014	0.327	0.598
	intra-group	0.171	4	0.043		
	Total	0.185	5			
pH test in camel milk	inter-group	0.000	1	0.000	0.143	0.725
	intra-group	0.000	4	0.000		
	Total	0.000	5			

According to Table 1, the significant value in all tests is greater than 0.05 and has led to the acceptance of the zero hypotheses. Therefore, there was no statistically significant difference in average acidity and pH in *Lactobacillus Acidophilus* and *Bifidobacterium* samples in all three milk types.

The investigation of differences between *Acidophilus* and *Bifidobacterium* samples

Table 2: The probably significant value of inter-group and intra-group of average texture in *Lactobacillus Acidophilus* and *Bifidobacterium* samples from cattle's milk

	Sum of squares	Degrees of freedom	Average of squares	F-test statistic	The probably significant value
inter-group	0.044	1	0.044	3.908	0.119
intra-group	0.045	4	0.011		
Total	0.090	5			

According to Table 2, the significant value in all tests is greater than 0.05 and has led to the acceptance of the zero hypotheses. Therefore, there was no statistically significant difference in average texture in *Lactobacillus Acidophilus* and *Bifidobacterium* samples.

Table 3: The probably significant value of inter-group and intra-group of average texture in *Lactobacillus Acidophilus* and *Bifidobacterium* samples from camel's milk

	Sum of squares	Degrees of freedom	Average of squares	F-test statistic	The probably significant value
inter-group	0.034	1	0.034	2.612	0.181
intra-group	0.051	4	0.013		
Total	0.085	5			

According to Table 3, the significant value in all tests is greater than 0.05 and has led to the acceptance of the zero hypotheses. Therefore, there was no statistically significant difference in average texture in *Lactobacillus Acidophilus* and *Bifidobacterium* samples.

Table 4: The probably significant value of inter-group and intra-group of average texture in *Lactobacillus Acidophilus* and *Bifidobacterium* samples from goat's milk

	Sum of squares	Degrees of freedom	Average of squares	F-test statistic	The probably significant value
inter-group	0.012	1	0.012	3.845	0.121
intra-group	0.013	4	0.003		
Total	0.025	5			

According to Table 4, the significant value in all tests is greater than 0.05 and has led to the acceptance of the zero hypotheses. Therefore, there was no statistically significant difference in average texture in *Lactobacillus Acidophilus* and *Bifidobacterium* samples.

Table 5: The probably significant value of inter-group and intra-group of viscosity in *Lactobacillus Acidophilus* and *Bifidobacterium* samples from cattle's milk

	Sum of squares	Degrees of freedom	Average of squares	F-test statistic	The probably significant value
inter-group	0.273	1	0.273	2.237	0.209
intra-group	0.488	4	0.122		
Total	0.761	5			

According to Table 5, the significant value in all tests is greater than 0.05 and has led to the acceptance of the zero hypotheses. Therefore, the average viscosity in *Lactobacillus Acidophilus* and *Bifidobacterium* samples are not statistically significant.

Table 6: The probably significant value of inter-group and intra-group of viscosity in *Lactobacillus Acidophilus* and *Bifidobacterium* samples from camel's milk

	Sum of squares	Degrees of freedom	Average of squares	F-test statistic	The probably significant value
inter-group	0.113	1	0.113	0.543	0.502
intra-group	0.835	4	0.209		
Total	0.948	5			

According to Table 6, the significant value in all tests is greater than 0.05 and has led to the acceptance of the zero hypotheses. Therefore, the average viscosity in *Lactobacillus Acidophilus* and *Bifidobacterium* samples are not statistically significant.

Table 7: The probably significant value of inter-group and intra-group of viscosity in *Lactobacillus Acidophilus* and *Bifidobacterium* samples from goat's milk

	Sum of squares	Degrees of freedom	Average of squares	F-test statistic	The probably significant value
inter-group	0.005	1	0.005	0.005	0.947
intra-group	3.650	4	0.912		
Total	3.654	5			

According to Table 7, the significant value in all tests is greater than 0.05 and has led to the acceptance of the zero hypotheses. Therefore, the average viscosity in *Lactobacillus Acidophilus* and *Bifidobacterium* samples are not statistically significant.

Table 8: The probably significant value of inter-group and intra-group of number in *Lactobacillus Acidophilus* and *Bifidobacterium* samples from cattle's milk

	Sum of squares	Degrees of freedom	Average of squares	F-test statistic	The probably significant value
inter-group	1.560	1	1.560	3.439	0.137
intra-group	1.814	4	0.454		
Total	3.374	5			

According to Table 8, the significant value in all tests is greater than 0.05 and has led to the acceptance of the zero hypotheses. Therefore, the average number in *Lactobacillus Acidophilus* and *Bifidobacterium* samples are not statistically significant.

Table 9: The probably significant value of inter-group and intra-group of number in *Lactobacillus Acidophilus* and *Bifidobacterium* samples from camel's milk

	Sum of squares	Degrees of freedom	Average of squares	F-test statistic	The probably significant value
inter-group	0.921	1	0.921	2.332	0.201
intra-group	1.580	4	0.395		
Total	2.502	5			

According to Table 9, the significant value in all tests is greater than 0.05 and has led to the acceptance of the zero hypotheses. Therefore, the average number in the sample is not statistically significant.

Table 10: The probably significant value of inter-group and intra-group of number in *Lactobacillus Acidophilus* and *Bifidobacterium* samples from goat's milk

	Sum of squares	Degrees of freedom	Average of squares	F-test statistic	The probably significant value
inter-group	0.760	1	0.760	1.282	0.321
intra-group	2.372	4	0.593		
Total	3.132	5			

According to Table 10, the significant value in all tests is greater than 0.05 and has led to the acceptance of the zero hypotheses. There was no statistically significant difference between *Lactobacillus Acidophilus* and *Bifidobacterium* samples.

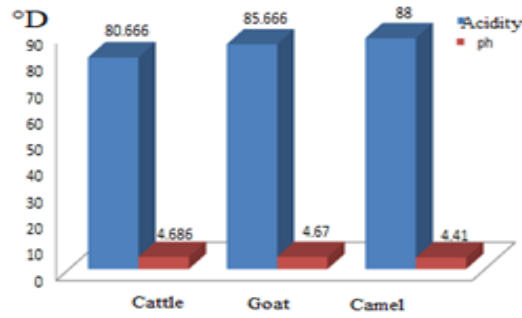


Chart 1: Acidity and pH changes in probiotic samples of *Lactobacillus Acidophilus* prepared from three types of milk

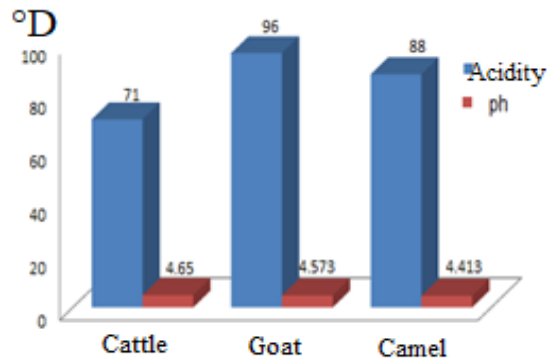


Chart 2: Acidity and pH changes in probiotic samples of *Bifidobacterium* prepared from three types of milk

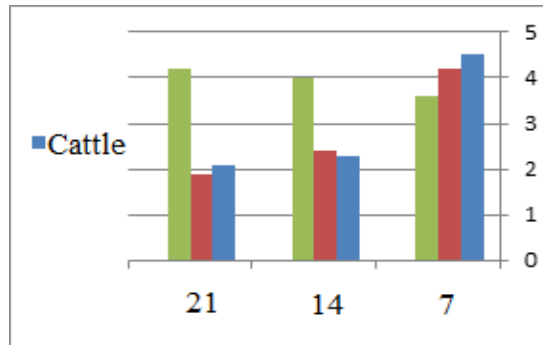


Chart 3: Viscosity changes in probiotic yogurt samples prepared from three types of milk (cattle, goats, camels) during three weeks of storage (Day 1 results not mentioned)

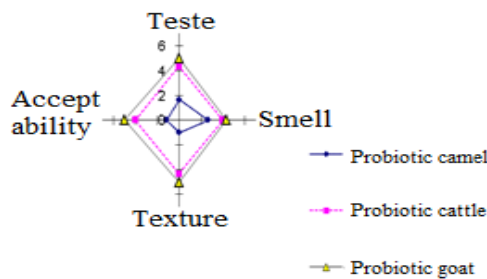


Figure 4: Sensory evaluation of samples on the 7th day (camel or cattle is in the chart means their milk, the statistical output cannot be changed in this figure)

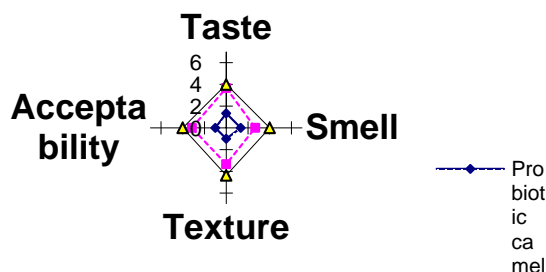


Figure 5: Sensory evaluation of samples on the 14th day

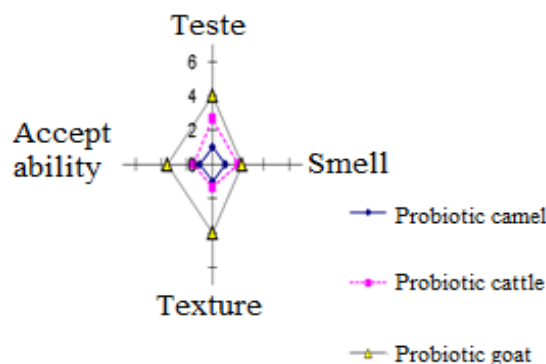


Figure 6: Sensory evaluation of samples on the 21st day (prebiotic milk)

Discussion

Physicochemical properties

- pH change

The results showed that the acidity in samples of probiotic yogurt with *Lactobacillus Acidophilus* starter was higher than *Bifidobacterium*. Bonczar et al. (2002) showed that the amount of milk fat affects the characteristics of common and probiotic yogurts such as pH, acidity and free fatty acid content. It was also shown that samples with higher fat percent had higher pH than those with lower fat percent (Abu-Taraboush, AL-Dagal and AL-Royli, 1998). Shaker et al. (2000) studied the rheological properties of yogurt with four levels of fat during the fermentation process and concluded that increasing milk fat increases the viscosity and decreases the production of acid by starter bacteria.

In this study, samples from goat's milk have higher acidity due to the presence of fat above other milk. During experiments conducted by Brownell in 1993, it was shown that the calcium excretion from the micelle begins at pH=5.1-5.3, at pH=4.8, the created Casein particles resemblance and create larger Casein resemblance and with the gradual decrease of pH increases the fusion and the viscosity of the milk. The increase in viscosity refers to the gradual formation of the gel and the gradual conversion of the incubation phase from the liquid form to gel form, and the best pH for the completion of the gel network is reported to be 4.1-4.6.

According to Walstra and Guerts, in 1999, with decreasing pH, the repulsive force between casein micelles was reduced, and pH = 4.6-4.7, casein micelles are highly unstable in terms of thermodynamics and with van der Waals attraction join together and the fusion phenomenon occurs. Decreasing pH and increasing acidity leads to the dissolution of colloidal calcium phosphate (CCP) or calcium phosphate micelle, resulting in an increase in calcium ion in milk water (Walstra et al., 1999).

- **Changes in acidity**

During the fermentation process, some lactose in the base milk was converted to lactic acid under various enzymes of thermophilic lactic bacteria. It caused a significant reduction in pH, which led to the decomposition of colloidal calcium phosphate, casein instability and even the release of isolated casein molecules. When a maximum population was reached; casein casings started to swell at pH below 5.5, since almost all colloid calcium phosphate was decomposed and precipitated. The sediment significantly reduced the population of casein casings and the formation of clusters and chains that were connected in order to form a gel. The gel consisted of a three-dimensional network with a serum containing proteins and lactose, and salts enclosed in aqueous phase (serum proteins depend on thermal treatment).

According to the results, there is a reverse relationship between pH and acidity in all samples, which means that pH has decreased with increasing acidity. Also, the results show that the curve slope is different in samples with various solids content so that the curve slope is lower for samples with lower solids content (cattle and camels) than for higher solids content (goats). The other result is that pH and initial acidity (in the first week) for samples with a higher solids content (goat) are lower than the two other samples. These phenomena can be explained by the fact that concentrated milk increases the concentration of the natural lactic acid of milk, like other components. It is also observed that for special pH, the acidity of the sample is more with higher solid content. Ozer et al. (1998) observed that the amount of pH in concentrated yogurt (16% solid content) reached 4.3 after 240 minutes, while the rate of acidity increase in the yogurt with the solids content was slower, due to higher buffer capacity of concentrated milk.

- **Texture review**

As shown in the Charts, the yogurt produced from probiotic *Lactobacillus Acidophilus* starter has an increasing trend in strength, except for cattle's milk. In the samples produced by the *Bifidobacterium* starter in three types of milk, an increasing trend was observed. Among them, the highest increase was observed in goat's milk, which improved the shelf-life during storage, due to the increase in the solids of the primary milk, the growth of the starters in the goat's milk (10.2 non-fat solids matters), and the increase in the production of extracellular polysaccharides by probiotic bacteria in longer fermentation conditions (Augustin, Cheng and Clarke, 1999). In 1999, the other study by Usi Mirin was conducted to investigate the effect of goat breeding and animal husbandry on the concentration of yogurt produced. Yogurt produced from goats fed on lush meadows and meadows is more concentrated, and on the contrary, milk products, namely yogurts, produced from the milk of cattle and goats that feed on grass and domestic concentrates or, in other words, dry fodder, have fewer concentrations and fewer solids. Improvement of texture properties by increasing the amount of fat may be due to the increase in total solids content and, consequently, the product rigidity (Augustin, Cheng and Clarke, 1999) because the increase in dry matter causes the gel network to remain stable and increases the water binding capacity (Güzel-Seydim et al., 2005). The results of studies by Marakoudakis et al. (2006) showed that the hardness of yogurt samples prepared with *Lactobacillus* strain increased over time.

- **Measurement of viscosity**

Known factors that affect the viscosity of yogurt include differences in milk composition especially fat and protein content that varies in different breeds of goats. According to Rassick and Koorman in 1978, the ratio of fat to non-fat solids and how to starter production is a major factor affecting the viscosity of yogurt. Typically, the responsible starter for producing polysaccharides from commercial starters of *Lactobacillus Delbrueckii* is the *Bulgaricus* subspecies. Goat's milk yogurt is less viscosity than cattle's milk yogurt in the market. The concentration of cattle's milk yogurt is usually increased by adding up to 16% of the total solids by adding special powders or Casein. Lucy et al. found that a higher coagulation temperature (42 °C compared to 30 °C) led to a loose gel production. The higher temperatures will reduce fermentation time and increase rheological properties before and after mixing.

- **Evaluation of syneresis**

Investigation of changes in the syneresis of samples during storage showed that the highest amount related to the samples during the first week of storage due to increased acidity and severe shrinkage of the gel network by cooling. In the sample prepared from camel milk during storage, syneresis increases, which may be due to the acidity increase during storage.

Regarding the change in syneresis during storage, the different results reported by Supavititpatana et al. (2010) showed that in corn milk yogurt, the amount of syneresis increased during storage. Barrantes et al. (1996) reported that syneresis decreases during storage. Mahdian and Mazaheri Tehrani (2007) showed that with the increasing solids matter, the amount of yogurt syneresis significantly decreased.

- **Evaluation of sensory**

The accumulation and presence of acetaldehyde in the product depending on the presence or absence of active enzymes to convert this composition to other metabolites. The presence of alcohol dehydrogenase in this bacterium causes acetaldehyde to be metabolized (acetaldehyde is produced from threonine by the activity of threonine aldolase, and subsequently metabolized to ethanol by the activity of alcohol dehydrogenase). This finding explains the lack of flavor in the milk of *Lactobacillus Acidophilus*. The acetaldehyde taste threshold is lower than ethanol, and the conversion of acetaldehyde to ethanol reduces the flavor and taste of the product. Therefore, preventing the activity of the alcohol dehydrogenase (active in *Lactobacillus Acidophilus*) causes the production of fermentation products with sensory properties like yogurt. In the case of products containing *Bifidobacterium*, this bacterium during fermentation produces lactic acid and acetic acid at the ratio of 3:2, and a product with vinegar-like flavor, which is completely undesirable.

Microbial properties

- **Counting *Bifidobacterium***

The results showed that on the seventh day, the number of samples obtained from the cow's milk under the starter *Lactobacillus*

which on the 14th day reached to 74.28 CFU/g. While the number of acidophilus in the camel's milk on the seventh day was measured 18.8 CFU/g and on the 14th day was 7.78 CFU/g. However, in the goat's milk, this number on the 14th day reached to 9.065 CFU/g which shows an increase in the number of bacteria, and it possessed the highest number of bacteria.

In this study, the number of probiotic bacteria in three milk types was counted on days 7, 14 and 21, and in the yogurt sample prepared with AC12 starter and cattle's milk on the 14th day, we have the maximum number of bacteria with *Lactobacillus Acidophilus* and on the twenty-first day, the number of bacteria decreased, but the number was higher than the seventh day, while in yogurt sample with BBL starter from cattle's milk, there is no significant difference between the 14th and the 21st days.

- **Counting *Lactobacillus Acidophilus***

Acidophilus requires acetate, riboflavin, pantothenic acid, calcium, niacin, and folic acid for growth. *Lactobacillus Acidophilus* requires a complete nutrient medium for growth that contains many amino acids, vitamins, growth factors, and fermentable carbohydrates (Khosravi Darani and Kushki, 2008). According to the above, these compounds were higher in goat milk than other milk, and the highest number of *Lactobacillus Acidophilus* was associated with yogurt produced from goat milk.

Conclusion

The results of this study showed that the number of probiotic bacteria was maintained during storage. The most commonly defined range for the density of living probiotic bacteria is 1×10^6 to 5×10^8 CFU per gram of product. It is better to reproduce the probiotic bacteria during fermentation and produces to a defined extent, thus, the bacteria will be replicated in a consistent environment and will survive longer in the half-life of the final product. Milk and dairy products, due to having many essential nutrients, are an acceptable environment for the growth of probiotic bacteria. Also, since the past, treatment of these foods has been

considered necessary and acceptance by the activity of bacteria as a fermented food. However, as a result of the single inoculation of these bacteria into milk, growth and acidification are very slow. The presence of some nutrients in inadequate or inaccessible amounts is a major factor of the low growth of these bacteria. On the one hand, the deficiency of amino acids and peptides in milk and on the other hand, the weakness of these bacteria in proteolytic activity, tend to slow the growth and prolongation of fermentation time.

Reference

1. Abu-Taraboush, H.M., AL-Dagal, M.M., and AL-Royli, M.A. (1998). Growth, Viability and Proteolytic Activity of Bifidobacteria in Whole Camel Milk. *Journal of Dairy Science*, 81:354-361.
2. Augustin, M.A., Cheng, L.J., Clarke, P.T. 1999. Effects of preheat treatment of milk powder on the properties of reconstituted set skim yogurts. *Int. Dairy J.* 9:415-416
3. Aziznia, S., Khosrowshahi, A. and Rahimi, J. (2008). Whey Protein Concentrate and Gum Tragacanth as Fat Replacers in Nonfat Yogurt: Chemical, Physical, and Microstructural Properties. *Journal of Dairy Science*. 91: 2545–2552.
4. Barrantes, E., Tamime, A.Y., Sword, A.M., Muir, D.D. & Kaláb, M., 1996, The manufacture of set-type natural yogurt containing different oils - 2: rheological properties and microstructure. *International Dairy Journal*, 6, 827-837.
5. Bonczar, G., Wszolek, A. & Siuta, A. (2002). The effects of certain factors on the properties of yogurt made from ewe's milk. *Food Chemistry*, 79, 85-91.
6. Boycheva, S., Dimitrov, T., Naydenova, N. and Mihaylova, G. (2011): Quality characteristics of yogurt from goat's milk, supplemented with fruit juice. *Czech J. Food Sci.*, 29: 24-30.
7. Çakmakçi, S., Cetin, B., Turgut, T., Gurses, M. Erdogan, A. (2012). Probiotic properties, sensory qualities, and storage stability of probiotic banana yogurts. *Turk. J. Vet. Anim. Sci*, 36(3): 231-237.
8. Çakmakçi, S., Cetin, B., Turgut, T., Gurses, M. Erdogan, A. (2012). Probiotic properties, sensory qualities, and storage stability of probiotic banana yogurts. *Turk. J. Vet. Anim. Sci*, 36(3): 231-237.
9. Güzel-Seydim Z, Wyffels, Jennifer T., Seydim, Atif C and Greene, Annel K. 2005. Turkish kefir and kefir grains: microbial enumeration and electron microscopic observation, *International Journal of Dairy Technology*, 5(1), 25-29.
10. International Dairy Federation. (1998). Fermented milk: science and technology. *IDF Bulletin*, 227.
11. Karim, G., Diani, Dardashty, A., Khalaji, A. (2006). *Milk and Its Quality*, Second Edition, Tehran University Press.
12. Kneifel W, Jaros D, Erhard F (1993) Microflora and acidification properties of yogurt and yogurt-related products fermented with commercially available starter cultures. *Int J Food Microbiol* 18: 179-189.
13. Lourent-Hattingh, A. and Viljoen, C. (2001). Yogurt as probiotic carrier food, *International Dairy Journal*, 11:1-17.
14. Mahdian, E. & Mazaheri Tehrani, M., 2007, Evaluation the effect of milk total solids on the relationship between growth and activity of starter cultures and quality of concentrated yoghurt., *American-Eurasian Journal of Agricultural & Environmental Science*, 2 (5), 587-592.
15. Marakoudakis. Petros A. Chiristos Miaris, Paval Rojez, Nikolas Manalis, Flora Magkanari, George. Kalantzopoulos, (2006) Effie Tsakalidou. *Int Dairy J.*(16)52-60.
16. Moayednia, N., Mazaheri, A.F. (2011). Growth and Activity of Selected Probiotic Strains in UHT Milk. *Journal of Food Biosciences and Technology*, 1: 63-69.
17. Ozer, B. H., Bell, A. E., Grandison, A. S. & Robinson, R. K. (1998). Rheological properties of concentrated yogurt. *J. Tex Stu*, 29, 67-79.
18. Shaker, R.R., Jumah, R.Y. & Abu-Jdayil, B., 2000, Rheological properties of plain yogurt during the coagulation process: impact of fat content and preheat treatment of milk. *Journal of Food Engineering*, 44, 175-180.

19. Shrtt, C. (1999). The probiotic century historical and current perspectives. *Trends Food Sci Technol.*, 10 (12), 411-417.
20. Sodin, F. (1998). Effect of continuous fermentation of milk with on immobilized cell bioreactor on fermentation kinetics curd properties.
21. Supavititpatana, P., Raviyan, P and Wirjantoro, T.I. 2010. Characteristics and shelf-life of Corn Milk Yogurt, *Chiang Mai University Journal of Natural Sciences* 9(1):133-150.
22. Walstra, P., Guerts, T.J., Noomen, A., Jemella, A., Van Boekel, M.A. (1999). *Dairy Technology, Principles of milk properties and processes.* Marcel Dekker, Inc.
23. Zhao, Q.Z., Wang, J.S., Zhao, M.M., Jiang, Y.M. and Chun, C. (2006). Effect of Casein Hydrolysates on Yogurt Fermentation and Texture Properties during Storage. *Food Technology and Biotechnology*, 44: 429–434.
24. Zubeirç, I., Basher, M., Alameen, M.H., Mohammed, M. and Shuiep, E.S. (2012).The processing properties, chemical characteristics, and acceptability of yogurt made from non-bovine milk. *Livestock Research for Rural Development*, 24 (3).