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*Full Length Research Paper*

# Studying microbial, physiochemical and sensory properties of directly concentrated probiotic yoghurt

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**Skim milk with 8.5% total solids was concentrated to 15 and 20% total solids by vacuum evaporation and inoculated with probiotic *Lactobacillus*. Yoghurts were incubated at 42$^{o}$C and stored at 4$^{o}$C. Survival of *Lactobacillus*, physiochemical (pH, acidity, synersis, and hardness) and sensory properties (taste and texture) of probiotic yoghurts were evaluated every 7 days to 21 days. Results showed that, increasing the total solid concentration of milk increased the survival of *Lactobacillus acidophilus*, acidity and hardness of yoghurt and reduced the pH and synersis. However, the survival of probiotic *Lactobacillus* decreased throughout the storage period at 4$^{o}$C. This work shows the importance of total solid concentration of milk on survival probiotic strains, physiochemical and sensory properties of yoghurt.**

**Key words:** probiotic yogurt, total solids, probiotic survival, physiochemical properties, sensory properties.

## INTRODUCTION

Probiotic bacteria, defined as "living micro-organisms", which upon ingestion in certain numbers, exert health benefits beyond inherent basic nutrition" (Guarner and Shaafsma, 1998) have become a major topic of lactic acid bacteria research over the past 20 years. The main "probiotic effects" attributed to these bacteria are: enhancement of immunity against intestinal infections, and improvement in lactose utilization, prevention of diarrholeal diseases, colon cancer, hypercholesterolaemia and upper gastrointestinal tract diseases, stabilization of the gut mucosal barrier (Kailasapathy and Rybka, 1997), formation or reconstruction of a well-balanced indigenous intestinal microflora, improvement of calcium absorption and vitamin synthesis and predigestion of proteins (Nakasawa and Hosono, 1992; Wood, 1992). *Lactobacillus acidophilus* LA5, having characteristics as inducing balance in intestinal microbial flora, increasing the immunity of body and protecting against traveler's diarrhea (Founden et al., 2000) is an important health promoting probiotic strain. Recently, in a small study, the effect of yoghurt enriched with *L. acidophilus* LA5, on fatty metabolites and microbial flora of Iranian people was studied (Niazmand et al., 2005).

Different types of products were proposed as carrier foods for probiotic microorganisms by which consumers can take in large amounts of probiotic cells for the therapeutic effect. Yoghurt has long been recognized as a product with many desirable effects for consumers, and it is also important that most consumers consider yoghurt to be "healthy". Yoghurt consumption has increased significantly, presumably because of its perceived health benefits (Wood, 1992). Concentrated yoghurt has higher nutritional value, better keeping quality, better taste and texture than normal yoghurt; so it has more acceptability among consumers. To extract water from plain yoghurt, in traditional method cloth bags are used. Due to the main setbacks of this method which are: lower yield, labor intensiveness and unhygienic conditions during the straining stage, some efforts have been made to develop other novel methods. Concentrating by vacuum evaporation of milk is studied by different researchers. Although there are no set standards concerning the population of the probiotic organisms in yoghurt at the end of the product shelf life, it is usually considered that anywhere between 5 and 8 logCFU g$^{-1}$ is an acceptable final population (Shah et al., 1995). Factors related to technological

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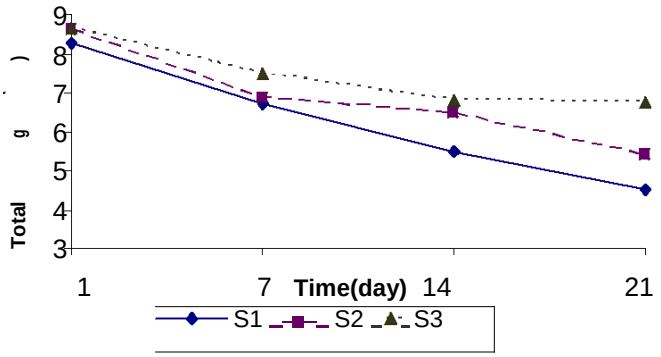


**Figure 1.** Total count of *L. acidophilus* in different total solid concentration of milk in 21days.

and sensory aspects of the probiotic food products are of utmost importance since only by satisfying the demands of consumers can the food industry succeed in promoting the consumption of functional products in the future (Mattila-Sandholm et al., 2002).

In this study we will examine the effect of increasing total solid concentration of milk and making concentrated yoghurt by vacuum evaporation of milk on survival of *L. acidophilus*, physiochemical and sensoryproperties of probiotic yoghurt during refrigerated storage.

## MATERIALS AND METHODS

In this study, cow's milk (0.09% fat, 8.5% total solids) was obtained from Pegah factory (mashahd, Iran). Probiotic culture *L.acidophilus* LA5, (freeze-dried DVS) was obtained from Chr. Hansen's company's representative (Tehran, Iran) and prepared according to manufacture's instructions.

### Production of concentrated yoghurt

Skim milk with 8.5 g/100g total solids was heated at 90 – 95$^{o}$C for 5 min and then concentrated by a batch evaporator (-0.8 bar and 55$^{o}$C) to 15 and 20 g/100g total solids and cooled to 42$^{o}$ C. After inoculation with 5% *L. acidophilus*, milk was distributed to 100 mL plastic retail containers, sealed and incubated at 42$^{o}$C until acidity reached 0.9%, it was then cooled and stored at 4$^{o}$C. At time intervals of 1, 7, 14 and 21 days, yoghurt samples were subjected to microbiological, physiochemical and sensory analysis. Three replicates of yoghurt were made.

### Microbial, physiochemical and sensory analysis of yoghurt

For microbiological analysis, samples (1.0 mL) of yoghurts were decimally diluted in sterile peptone water (Merck, Germany) (0.1%) and 0.1 ml aliquot dilutions were spread over the surface of plates of MRS agar (Merck, Germany) and incubated in 37$^{o}$C for 3 days (Vinerola and Reinhemir, 1999). After 3 days Colony Forming Units (CFU) were counted by a colony counter.

Regarding the physiochemical analysis of yoghurt, pH measure-ents were carried out using a digital pH-meter (Metrohm 691, Swiss), while acidity was titrated by M/10 NaOH solution. Degree of synersis, expressed as proportion of free whey, was measured by a small modification of method used by Al-Kadamany (Al- Kadamany et al., 2003). A 10 g sample of concentrate yoghurt was layered on 10 cm Whatman paper (#2) that was filtered into a Buchner funnel, and vacuum filtered for 6 min. The proportion of free whey was calculated as follows: Free whey (g/100g) = (weight of initial sample-weight of sample after filtration /weight of initial sample) *100.

Texture analyzer (Model: QTS, CNS Farnell, America) was used to measure the hardness of concentrated yoghurts. The probe penetrated the samples to a depth of 15 mm at a speed of 1.0 m/s and the force exerted on the probe was automatically recorded (Yazici and Akgun, 2004). All determinations were carried out in triplicate and at 18 ± 2$^{o}$C and average readings were taken. Yoghurt samples were subjected to sensory evaluations (taste and texture) by educated seven-member panel from Food Science and Technology Students of Ferdowsi University of Mashhad.

### Statistical analysis

Statistical analysis of data for effects of the factors on pH, acidity, synersis, hardness, survival of probiotic strain and sensory properties was performed by one-way and two factor randomized complete block design using MSTAT C software. The factors were: (1) total solid (8.5%, 15%, 20%), (2) storage time (1,7,14 and 21 days).The mean differences were analyzed using Dunkan's test at P<0.05 (O'Mahony, 1986).

## RESULTS AND DISCUSSION Survival

### of *Lactobacillus acidophilus*

During the 21 days, the survival of *L. acidophilus* decreased significantly (P<0.05) (Figure 1) . By increasing the total solid concentration of milk, the number of *L. acidophilus* increased. This result shows that we can have a better survival of *L. acidophilus* in concentrated yoghurts than normal yoghurt. Results showed that re-commended number for *L. acidophilus* by some authors (Shah et al., 1995) was just observed in $S_2$ and $S_3$. Increasing the total solid concentration of milk from 16 to 23%, improves the growth of *Lactobacillus bulgaricus* (Ozer and Robinson, 1999).

## Physiochemical properties,

Results showed that there was no significant change (P<0.05) in pH to 14$^{th}$ day, but decreased from 14$^{th}$ to 21$^{st}$ day. The initial pH values for the different yoghurt types ranged from 3.93 to 4.21.The lowest pH were observed for $S_3$. Better survival of *L. acidophilus* and production of more lactic acid during fermentation could be the reason for this low pH. PH values decreased approximately 0.1, 0.03, 0.02 unit in $S_3$, $S_2$ and $S_1$, respectively (Figure 2). Acidity increased significantly (P<0.05) in 21 days of storage. Higher acidity was observed in yoghurts with higher total solid concentrations, 1.08, 1.2 and 1.32 in $S_1$, $S_2$ and $S_3$, respectively (Figure 3). Higher lactose content and higher number of *L. acidophilus* in $S_2$ and $S_3$ is the main reason for higher lactic acid production. By increasing the total

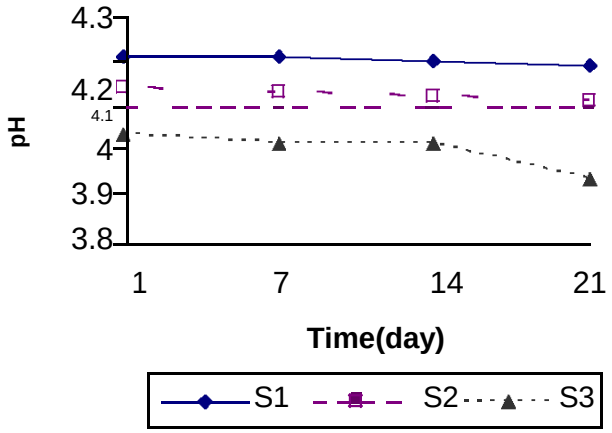


**Figure 2.** Changes in pH values in different total solid concentration of milk in 21days.

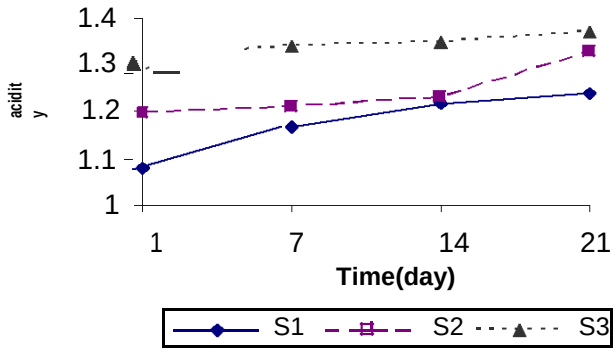


**Figure 3.** Changes in acidity values in different total solid concentration of milk in 21days.

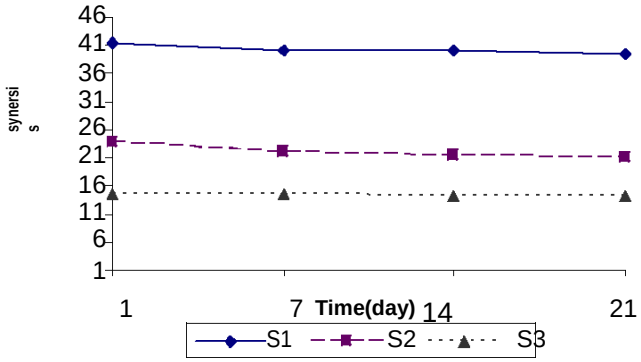


**Figure 4.** Changes in synersis values in different total solid concentration of milk in 21days

solid concentration of milk, the synersis decreased, mean values for the synersis in the first day was 41.54, 23.56 and 14.58%, respectively, which decreased to 39.54, 21.16 and 14.2%, respectively after 21 days. Changes of synersis values during 21 days, was 2, 2.4 and 0.38 unit in $S_1$, $S_2$ and $S_3$, respectively, the

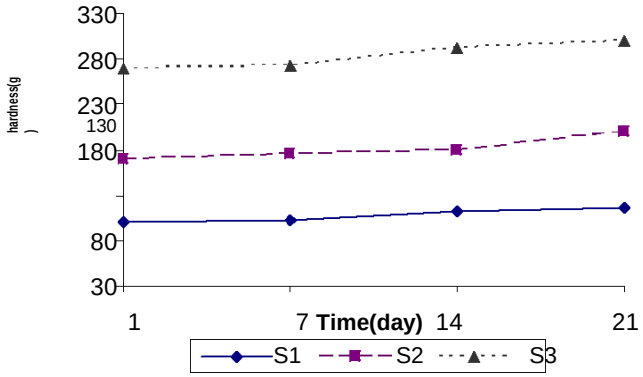


**Figure 5.** Changes in hardness values in different total solid concentration of milk in 21days

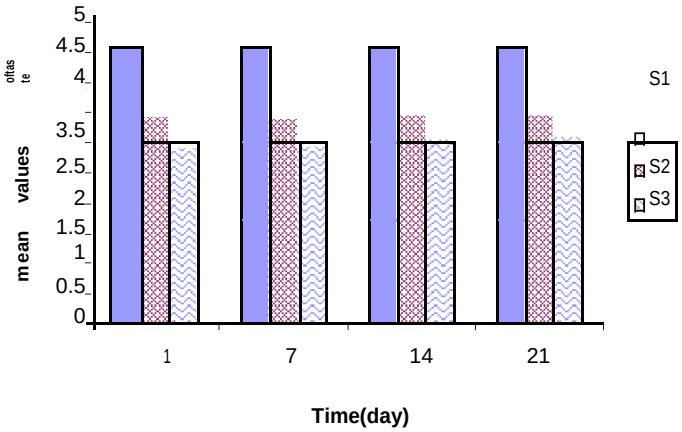


**Figure 6.** Changes in taste values in different total solid concentration of milk in 21days.

regression coefficient between time and synersis was about 0.86, 0.88 and 0.91 for $S_1$, $S_2$ and $S_3$respectively. This results show that there is a good correlation between time and synersis in higher total solid concen-trations. (Figure 4) by increasing the total solid concen-tration of milk from (8.5 to 20%) hardness increased significantly ( <0.05). Hardness increased 15.67, 30.33 and 30.67 units in $S_1$, S2 and $S_3$, respectively (Figure 5). Results show that there was correlation between synersis and hardness, as hardness increases the synersis decreased. The correlation values were 0.83, 0.87 and 0.97 for $S_1$, $S_2$ and $S_3$, respectively. This results show that the correlation increased as total solid concentration increased.

## Sensory properties

Figures 6 and 7 shows the effect of total solid concentration of milk on taste and texture of yoghurt in 21 days of storage. $S_1$ had the best taste and texture among other yoghurts. No significant change (P<0.05) was observed

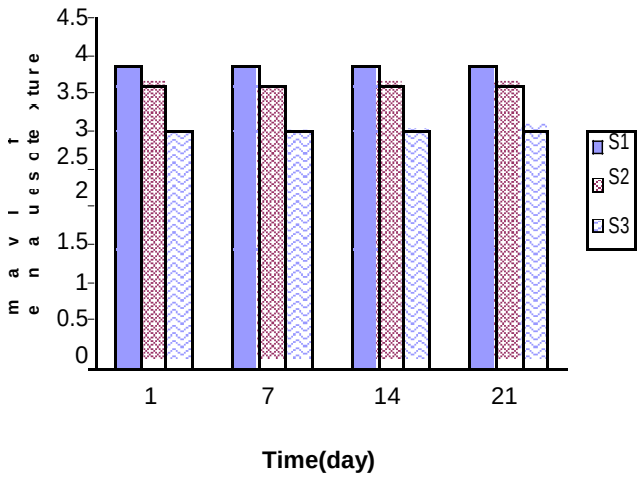


**Figure 7.** Changes in texture values in different total solid concentration of milk in 21days.

for taste during 21 days storage, but texture was highly scored by panelists in day 21.

## Conclusion

Results showed that by increasing the total solid concentration of milk and producing concentrated yoghurt, the survival of *L. acidophilus*, acidity and hardness increased and pH and synersis decreased. Among yoghurt samples $S_1$ get the best score for taste and texture. In all samples the survival of *L.acidophilus*, pH and synersis, decreased during the 21 days, but acidity and hardness increased. No significant change ( <0.05) was observed for taste during 21 days storage, but texture was highly scored by panelists in day 21.

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