Journal of Food Science and Technology (Iran)

Homepage:www.fsct.modares.ir

Scientific Research

Evaluation the impact of lactic isolates from local dairy products as adjunct culture and ripening period on physico-chemical properties of Feta cheese

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ARTICLE INFO

Article History:

Received:2025/06/23

Accepted:2025/08/20

Keywords:

Feta cheese,

Lactic acid isolate,

Local dairy products.

DOI: 10.48311/fsct.2025.84052.0.

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ABSTRACT

In this research, the effect of five different treatments as adjunct culture, including different combinations of lactic acid bacteria strains (isolated from cheese curd, fresh cheese, Lighvan ripened cheese, and yogurt) on the physicochemical parameters (% dry matter, % acidity, pH, % salt, % fat, % total nitrogen, proteolysis (% soluble N to total N), and lipolysis of feta cheese prepared by the traditional methods were investigated during 60 days. Five Treatments in this study include: T1: *Lactiplantibacillus plantarum+Lb. helveticus*, T2: *Enterococcus faecium* + *Lb. helveticus*, T3: *Lactococcus lactis* + *Streptococcus thermophilus* + *Lb. delbruckeii*, T4: *Lb.plantarum*+ *Ent.faecium* and T5: (control): *Lac.lactis* ssp *lactis*+ *Lac.lactis* ssp *cremoris*. The results showed that as the storage time progressed, the pH of all treatments decreased (from 6.3 to 5.9) and the acidity increased (from 0.27 to 0.66) significantly ($p<0.01$). Among treatments, the highest pH belonged to T1(6.21) and the lowest pH, belonged to T5 (5.94). Also dry matter of all treatments experienced declining trend (from 38.46 to 34.008) significantly ($p<0.01$) with storage time. The highest amount of % dry matter, % total N and % salt, was related to T1 on day 60. The measurement of the proteolysis and lipolysis indices showed that the highest amount at the end of the storage period was related to the T3, 10.40 and 10.199, respectively. In general, the outcomes of this research showed that with the help of T1, (*Lb. plantarum* and *Lb. helveticus*) to feta cheese formulation, a product with desirable physicochemical characteristics can be produced.

1-Introduction

From the Food and Agriculture Organization (F.A.O) and the World Health Organization (W.H.O) point of view, cheese is the fresh or ripened solid or semi-solid product which is produced from milk, skim milk or curd by the action of rennet or other suitable coagulants. This product contains high-quality protein and is very rich in essential amino acids. Cheese is well-known as a functional food, so that its use has a direct and significant impact on health. Cheese is a food which is rich in whole and full protein that can be consumed instead of meat. Cheese is considered as a source of bioactive peptides, during the cheese-making process, protein hydrolysis occurs, the resulting peptides act as bioactive molecules [1].

Cheese, as the most important milk-derived product, is a rich source of essential nutrients such as fat, fatty acids, proteins, peptides, amino acids, vitamins, and minerals. Nowadays, many types of cheese are produced with different taste, texture, and appearance characteristics. This diversity in cheese production has given cheese a specific rank in the diets of worldwide communities [2].

Feta cheese is one of the most famous types of brine cheese in the world, manufactured by traditional and industrial methods. As a genuine representative of brine cheeses, this type of cheese has originated in Greece. Feta cheese has a white color and a soft texture that has a relatively salty, slightly acidic taste, and a pleasant aroma and flavor. This cheese is often made from sheep and goat milk, but in higher demand cases for feta cheese, cow's milk may also be used [2].

Feta cheese is a soft cheese popular in many countries in Africa, Europe, and elsewhere. It was traditionally made from goat's milk, but these days, a variety of milks, including sheep's, cow's, and buffalo's milk, are used to make feta cheese. Traditionally, feta cheese was made from raw milk; however, more recently, due to changes in the flavor and characteristics of the cheese over the years, the milk is pasteurized before being used to make feta cheese. The natural microbiota, found in raw milk results in a short ripening time and intense flavor, but can also lead to undesirable defects in the cheese's characteristics or pose a health risk to the consumer due to the presence of pathogenic bacteria.

Consequently, traditional feta cheese is manufactured from pasteurized milk to guarantee consumer safety and to maintain the typical characteristics of the cheese throughout the year. However, feta cheese produced from pasteurized milk has several challenges due to the lack of flavor intensity compared to feta cheese made from raw milk [3, 4, 5]. As a result, several studies have focused on the production of feta cheese from pasteurized milk using different starter cultures, additives, different milk sources, and changes in processing conditions to induce flavor and texture compared to raw milk feta cheese. The conventional process of feta cheese production is carried out using lactic acid bacteria (LAB). LAB in feta cheese usually increases with increasing acidity at the beginning of ripening, when stored at 5–7°C, and then the number of LAB remains constant for up to 60 days [5]. On the other hand, mesophilic starter cultures decrease during early ripening of feta cheese, especially in the presence of higher salt content (6–8%) and pH less than 5. Therefore, thermophilic bacteria and probiotics have been used as adjunct starter cultures to improve the flavor of feta cheese [5].

Industrial production of this cheese needs pasteurization of milk and addition of starter cultures, but the use of commercial cultures results in loss of the unique characteristics of traditional cheeses, as the natural complex microbiota is replaced by commercially defined cultures. Thus, the use of indigenous starter cultures consisting of LAB isolated from traditional soft cheeses with proven technological capabilities can both improve product quality and maintain the identifying characteristics of traditional cheeses [6,7]. The objective of present study was to investigate the effect of storage time or ripening period and the type of adjunct culture and their interaction on the physicochemical properties of the resulting feta cheeses.

2-Materials and Methods

Strain Preparation

The lactic acid bacteria isolates used in this study included: *Lactiplantibacillus plantarum* which have been isolated from fresh Lighvan cheese, *Lactococcus lactis* from Lighvan cheese curd, *Enterococcus faecium* from ripened Lighvan cheese, *Streptococcus thermophilus*, *Lb. helveticus*, *Lb. delbrueckii* isolated from yogurt were used for the

preparation of cheese treatments. [8, 9] The commercial starter cultures for the production of the control cheese sample included *Lactococcus lactis* ssp *lactis* and *Lactococcus lactis* ssp *cremoris*, which were obtained from Pegah Khorasan Company.

To activate the strains that were stored in a freezer at -80°C, they were first removed from the freezer and each strain was cultured in its respective culture medium, which was MRS culture medium for *Lb. plantarum*, *Lactobacillus helveticus*, *Enterococcus faecium*, and BHI culture medium for *Lactococcus lactis*, *Streptococcus thermophilus*, and *Lb. delbrueckii*. They were incubated at 37°C for 24 hours and a suspension of 10^8 cfu/ml of each strain was used for inoculation in cheese milk, using the McFarland standard [8].

The milk used for cheese production was obtained from Khorasan Razavi Dairy Factory, and its physicochemical characteristics are presented in Table (1).

Table 1. Milk Characteristics used in cheese production.

Characteristics of Milk	)Lactic Acid %(Acidity	pH	Fat (%)	Density	Non Fat Solids(NFS)
Amounts (Mean ± STD)	14.4±0.0	6.73±0.01	3.325±0.09	1.03±0.0	8.25±0.04

Cheese-making

The production of Feta cheese was carried out according to the standard method No. 2344 [10]. Pasteurization of the milk was performed at 72°C for 15 seconds in a water bath. Then, to reach a microbial population of 10^8 cfu/ml, starter cultures of the desired lactic strains were separately prepared as a suspension with a concentration of 10^8 cfu/ml using the McFarland method, and 1% v/v of this microbial suspension was added to the milk. This was done at a temperature of 32–34°C, and the mixture was kept at this temperature for 20 minutes.

In the next step, calcium chloride was incorporated at a concentration of 15 grams per 100 liters of milk. Then, rennet was added at a rate of 1 gram per 25 liters of milk. After about one hour, the formed curd was separated from the whey and placed inside a cloth. The completion of the process was determined based on the initial clot amount and in consultation with a cheese producer. The curds were then pressed using weights and cut into pieces measuring 2 to 3 cubic centimeters with a sterile knife. Subsequently, the curds were salted with a 10% brine solution and stored at a temperature of 10 to 12°C for two months. During this period, sensory evaluation and sampling were performed by panelists on days 15, 30, and 60. The control sample was prepared under the same conditions using only commercial starter cultures.

The bacterial strains used in the treatments prepared in this study are listed in Table 2.

Table 2. Treatments used in this study.

Treatments	Strains Used
T1	& *Lb. helveticus* Commercial starter +*Lb. plantarum*
T2	*Lb. helveticus* Commercial starter +*Enterococcus faecium* &
T3	Commercial starter +*Streptococcus thermophilus* & *Lactococcus lactis* & *Lb. delbrueckii*
T4	& *Enterococcus faecium* Commercial starter + *Lb. plantarum*
T5(Control)	*& Lactococcus lactis ssp lactis* Commercial starter *(Lactococcus lactis ssp cremoris)*

Physicochemical properties of cheese

Measurement of Dry matter (%)

The measurement of dry matter in cheese was performed using a moisture analyzer (Sartorius Ltd., Epsom, UK). One gram of the cheese sample was evenly spread on a paper and placed in the device. After the water evaporated, the device displayed the percentage of dry matter [11].

Determination of % acidity

According to the method of the National Standards Organization, No. 2852 was carried out [12]. First, the required solutions were prepared: 0.1 N sodium hydroxide, phenolphthalein, 1% solution (1 gram of phenolphthalein in 100 milliliters of alcohol), 0.1 N potassium hydroxide, and neutral ethyl alcohol. The cheese sample was completely homogenized in a mortar or blender and transferred into sealed containers. Twenty grams of the sample was weighed in a beaker, then dissolved in a certain amount of carbon dioxide–free distilled water and diluted to 250 milliliters in a volumetric flask. After filtration, 25 milliliters of the clear solution were transferred into a beaker. 0.5 milliliters of phenolphthalein was added and titrated with 0.1 N sodium hydroxide. The titration was continued until a faint pink color appeared and remained stable for 5 seconds.

$$\frac{N\times 0/009\times 100}{M1}\times\frac{\text{Volume of volumetric flask used}}{V}$$

=Acidity

N = milliliters of 0.1 N sodium hydroxide used in titration, M_1 = weight of the sample, equal to 20 grams,

V = test solution volume, equal to 25 milliliters

pH Measurement

pH was measured using a pH meter (Metrohm, Germany). In this step, the electrode was directly inserted into the homogenized cheese matrix to obtain the pH value [12].

Measurement of Salt Content

To measure the amount of salt in cheese samples, the Mohr method was used. Two grams of cheese were mixed with distilled water in a 250-mL volumetric flask. Then, 25 mL of the diluted solution along with 1 mL of potassium chromate indicator was titrated with 0.1 M silver nitrate solution. The percentage of salt was calculated using the following formula [13].

Salt percentage = (V × 0.1 × 5.58) / 2

In this equation, V is the volume of silver nitrate consumed, expressed in millilitres.

Fat Measurement

The fat content of the cheese was determined using the Gerber method. For this purpose, 3 g of cheese were weighed into a butyrometer. Then, 5 mL of warm water was gently added and mixed until the cheese was completely dissolved. Subsequently, 10 mL of 90% sulfuric acid was carefully added along the wall of the butyrometer. Next, 1 mL of amyl alcohol was added, followed by enough water to bring the volume of the mixture up to the narrow part of the butyrometer. The butyrometer was then placed in a water bath at 65 °C for 5–10 minutes. After that, it was centrifuged for 5 minutes at 1100 rpm. Finally, the butyrometer was placed again in a water bath at 65 °C, and after 10 minutes the fat content was read directly from the butyrometer scale [14].

Total Nitrogen

The total nitrogen content of the cheese treatments was determined using the Kjeldahl method. The sample amount for each cheese treatment was 0.5 g. The total protein content was calculated by multiplying the measured nitrogen content by the conversion factor of 6.38 [15].

$$\text{Total Nitrogen} = \frac{1.4007\times 0.1\times \text{ml}\ \ Hcl\ \text{consumed}}{\text{sample weight}}$$

Proteolysis Evaluation

Determination of Soluble Nitrogen at pH 4.6

The preparation and extraction of soluble nitrogen at pH 4.6 were carried out according to the modified method of Kuchroo and Fox (1982). Thirty grams of cheese samples were homogenized with the addition of 60 mL of distilled water. The pH of the samples was then adjusted to 4.6 using 0.1 N HCl and NaOH solutions. The samples were left to stand for 30 minutes, after which the pH was readjusted to 4.6 and the mixture was centrifuged at 4000 × g for 30 minutes. After centrifugation, the samples were filtered through Whatman No. 42 filter paper, and the soluble nitrogen content was determined using the Kjeldahl method [16].

Lipolysis Evaluation

Cheese samples (10 g) were thoroughly homogenized with 6 g of anhydrous sodium sulfate and transferred to a screw-cap container with 60 mL of diethyl ether. The mixture was stirred thoroughly with a magnetic stirrer and then filtered through Whatman No. 1 filter paper. The remaining residue was washed twice consecutively, each time with 20 mL of diethyl ether. The filtrate was titrated with 0.1 N ethanolic KOH in the presence of phenolphthalein indicator. After titration, the solvent was evaporated under a fume hood. The residual fat was weighed, and the total free fatty acids in the cheese were expressed as milliequivalents per 100 g of fat (meq/100 g) [17].

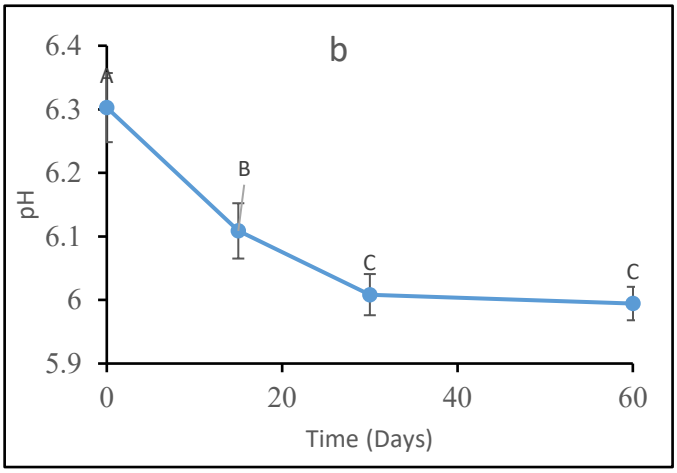
Statistical Analysis

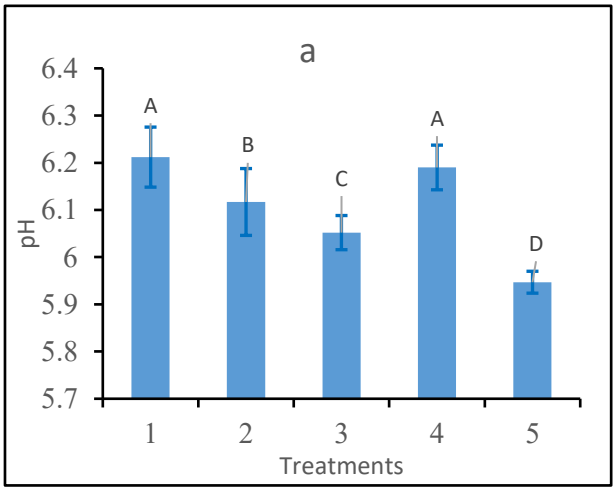
Analysis of variance (ANOVA) was performed using the General Linear Model, defining 5 treatments from different combinations of lactic acid bacteria (as adjunct starters) and 4 evaluation times or ripening intervals (day 0, day 15, day 30, and day 60) in the model. After running the model, the normality of the residuals (observation errors) for all traits under study was confirmed. Mean comparisons were conducted using Tukey's test at a 5% significance level. All statistical analyses were performed using Minitab software, version 21.

3-Results and Discussion

pH

The analysis of variance showed that the effects of treatments (different types of adjunct starters), time, and the interaction between treatments and time on pH were significant ($P < 0.01$). Comparison of mean pH values under different treatments indicated that, overall, this variable ranged from 4.6 (in treatment 1) to 8.5 (in treatment 5) (Figure 1-a). Treatments 2, 3, and 4 showed pH values slightly different from those of treatments 1 and 5. Figure 1-b illustrates the changes in pH during the ripening period of Feta cheese. The interaction between factors indicated that in treatments 1, 2, and 3, a decreasing trend was observed until day 30, followed by an increasing trend. In treatment 4, an almost consistent decreasing trend was observed, whereas treatment 5 showed a relatively irregular trend, likely due to differences in the strains used in this treatment (Figure 1-c).





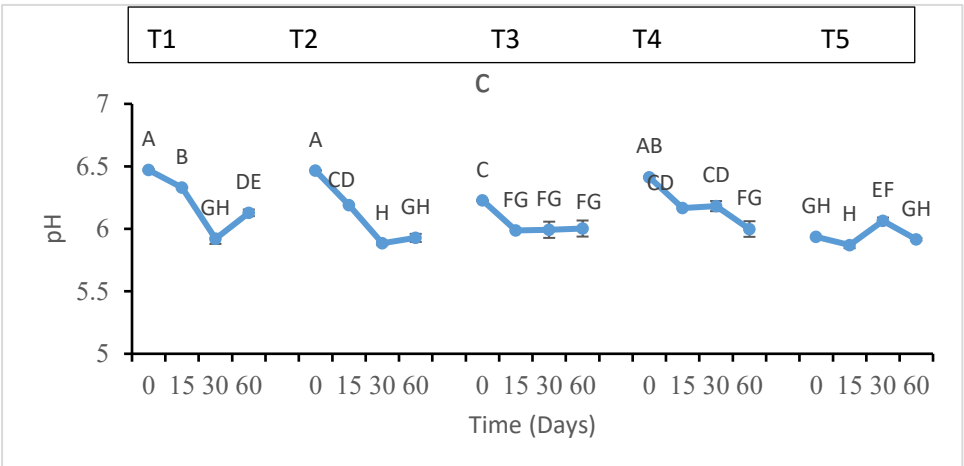


Figure 1. Effects of different treatments on pH of Feta cheese (a). Treatment 1(T1): *Lb.plantarum* & *Lb.helveticus*, Treatment 2(T2):*Ent.faecium* & *Lb.helveticus*; Treatment 3(T3):*Lb.delbrueckii* & *Str.thermophilus*, Treatment 4(T4): *Lb.plantarum* & *Ent.faecium*, Treatment 5(T5): Control. Effects of ripening period on pH of Feta cheese (b). Effect of interaction of treatments and ripening period on pH of Feta cheese (c).

The results of the study showing the pH of different cheese treatments compared to the control sample are presented in Figure 1-a. The findings indicate that the pH of all the treatments under investigation was higher than that of the control sample (treatment 5). Among the treatments, the highest and lowest pH values were observed in treatment 1 (*Lactobacillus plantarum* and *Lactobacillus helveticus*) and treatment 5 (control), respectively. Additionally, based on another part of the statistical analysis, pH significantly decreased ($p \leq 0.05$) with increasing storage time (Figure 1-b).

pH is one of the factors that has a significant impact on cheese stability, microbial growth conditions, enzymatic activity, and the rate of biochemical reactions during cheese ripening. The decrease in pH during the ripening period is mainly related to the conversion of lactose to lactic acid by lactic acid bacteria, and, to some extent pH increase during prolonged ripening, is associated with the production of amino acids and fatty acids through proteolysis and lipolysis [18]. A rapid decrease in pH during the early stages of cheese production is essential for curd formation and for preventing or reducing the growth of undesirable microorganisms in milk [19]. Other researchers have also reported a decrease in the pH of Iranian white cheese during the ripening period [20, 21]. Additionally, Gholamhoseinpour et al. (2022) and Montazarani et al. (2018) reported a decrease in the pH of Feta cheese containing different starters during the storage period [22, 23].

In this study, throughout the storage period, all treatments had higher pH values than the control sample. Among the different samples, those containing *Lactobacillus plantarum–Lactobacillus helveticus* and *Enterococcus faecium–Lactobacillus helveticus* showed higher pH than the other samples. This is likely due to the low ability of these bacteria to ferment lactose and produce lactic acid [24].

The main characteristic of lactic acid bacteria is lactose fermentation and lactic acid production; however, not all lactic acid bacteria acidify milk to the same extent. Some rapidly lower the pH of milk, while others do so slowly. Enterococci belong to the lactic acid bacteria group and generally have low to moderate acidifying capacity [24]. Several studies have reported that Enterococcus strains exhibit weak acidification, with only a small percentage capable of producing enough acid to reduce the pH below 4.5–5 after incubation at 37 °C for 16–24 hours.

In agreement with the results of this study, Siousia et al. (2013) reported that the pH of Cheddar cheese containing *Lactobacillus plantarum* isolated from Caciocavallo cheese was slightly higher than that of the control sample [25]. Kayaoglu (2006) investigated the effect of *Lactobacillus brevis* and *Lactobacillus paracasei* as adjunct starters and reported that on day 30 of ripening, the pH of cheese containing these adjunct starters was higher than that of the control cheese, which contained *Lactococcus lactis* subsp. *lactis* and *Lactococcus lactis* subsp. *cremoris*. This difference was attributed to the type of microorganisms present in the adjunct starter as well as the ratio of viable cells [26].

Acidity

The analysis of variance showed that the effects of treatments (different types of adjunct starters), time, and the interaction between treatments and time on acidity were significant ($P < 0.01$). Acidity exhibited a significant increasing trend during the storage period (Figure 2-a). Comparison of mean acidity values under different treatments indicated that, overall, this variable ranged from 0.4 (in treatments 1 and 4) to 0.69 (in treatment 5) (Figure 2-b). The statistical findings of acidity in different cheese treatments during the storage period are presented in Figure 2-c. The results showed that at different storage intervals, the acidity of the control sample was higher than that of all treatments ($P < 0.01$).

According to another part of the findings, increasing the storage time had a significant increasing effect ($P < 0.01$) on the acidity of the different treatments and the control sample, with the highest acidity observed at the end of the storage period.

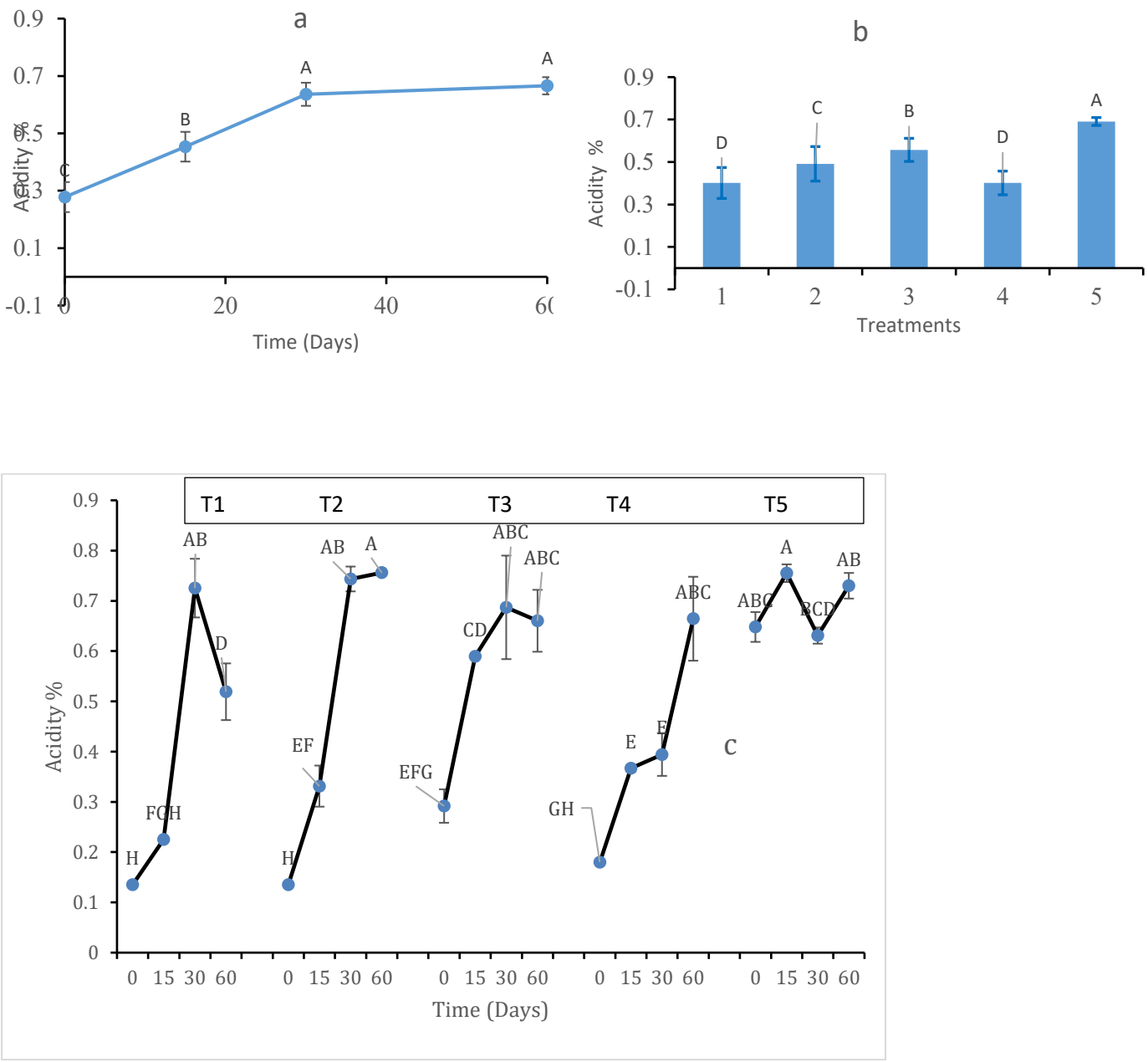


Figure 2. Effects of different treatments on acidity of Feta cheese (a). Treatment 1(T1): *Lb.plantarum* & *Lb.helveticus*, Treatment 2(T2):*Ent.faecium* & *Lb.helveticus*; Treatment 3(T3):*Lb.delbrueckii* & *Str.thermophilus*, Treatment 4(T4): *Lb.plantarum* & *Ent.faecium*, Treatment 5(T5): Control. Effects of ripening period on acidity of Feta cheese (b). Effect of interaction of treatments and ripening period on acidity of Feta cheese (c
).

The results showed that the increasing trend in acidity of the cheese samples was similar to the decreasing trend in pH. Acidity and pH have an inverse relationship, and during storage, due to the production of lactic acid and hydrogen ions, acidity increases while pH decreases. The titratable acidity and pH of cheese are of high importance because of their effects on microbial growth, enzymatic activity during ripening, rheological properties of cheese, and sensory evaluation [27].

In agreement with the results of this study, Kendili et al. (2002) reported that the acidity of Kashar cheese increases during storage due to lactic acid production and pH reduction [28]. Similarly, Sadeghi and Nateghi (2020) reported that during ripening, the activity of bacteria increases the acidity of brined white cheese [29]. In line with this, Gholamhoseinpour et al. (2022) reported that during ripening, the acidity of Feta cheese samples containing various commercial starters (SafeIT 2, FRC-65, and R-704) increases [22]. These researchers

attributed the increase in acidity to the formation of lactate, as well as the production of free fatty acids and amino acids through lipolysis and proteolysis.

In another study, Montazarani et al. (2018) reported that the acidity of Feta cheese containing *Lactobacillus paracasei* isolated from kefir and the control sample increased with storage time [23]. According to another part of the results, the cheese treatment containing *Lactobacillus delbrueckii*, *Lactobacillus lactis*, and *Streptococcus thermophilus* had higher acidity than the other treatments during ripening. This was likely due to greater lactic acid production and the more advanced progress of lipolysis and proteolysis by these microorganisms (which aligns with the results of the lipolysis and proteolysis tests discussed later in this study).

In this regard, Hiyaloglu et al. reported in studies conducted in 2005 and 2013 both significant and non-significant effects of starters on the acidity of various cheeses [30, 31]. Contrary to the results of this study, Gholamhoseinpour et al. (2022) reported that the type of starter did not have a significant effect on the acidity of Feta cheese, which could be due to differences in the type of cheese used in that study [22].

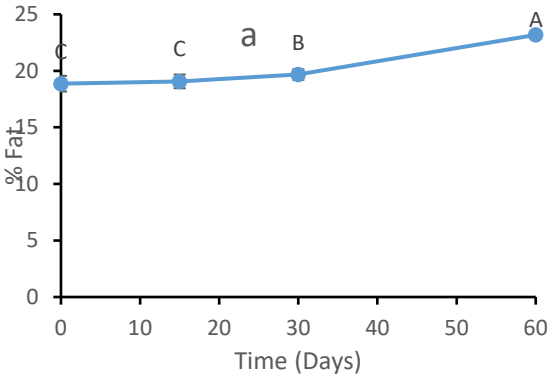
Fat content

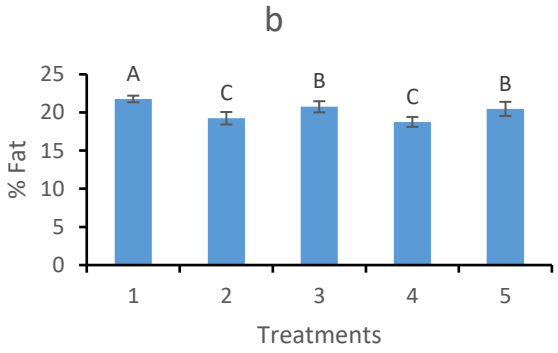
The analysis of variance for fat content showed that the effects of time, treatments (different types of adjunct starters), and the interaction between time and treatments on fat were significant ($P < 0.01$). Comparison of means indicated that the highest fat percentage (21.77%) was observed in treatment 1, while the lowest percentage (18.75%) was observed in treatment 4. Treatments 5, 3, and 2 had fat percentages that were among those of treatments 1 and 4, respectively (Figure 3-b).

Changes in the fat content of different cheese treatments during the 60-day storage period showed a significant increasing trend ($P < 0.01$) (Figure 3-a). Figure 3-b illustrates the effect of different Feta cheese treatments on fat percentage, which was also significant ($P < 0.01$).

The findings indicated that among the different cheese treatments, treatment 1 (21.77%) had significantly higher fat content ($P \leq 0.01$) than the other treatments. The next highest fat content was observed in treatments 3 and 5, which also differed significantly ($P \leq 0.01$) from the remaining treatments. The lowest fat content was found in treatments 2 and 4. No significant difference was observed between treatments 2 and 4, while both differed significantly ($P \leq 0.01$) from the other treatments.

The interaction effect of time and treatments on fat content is shown in Figure 3-c. As observed, treatment 1 exhibited an increasing-decreasing-then-increasing trend, treatments 2 and 3 showed a continuous increasing trend, and treatment 4 showed a slight initial decrease followed by an increase.





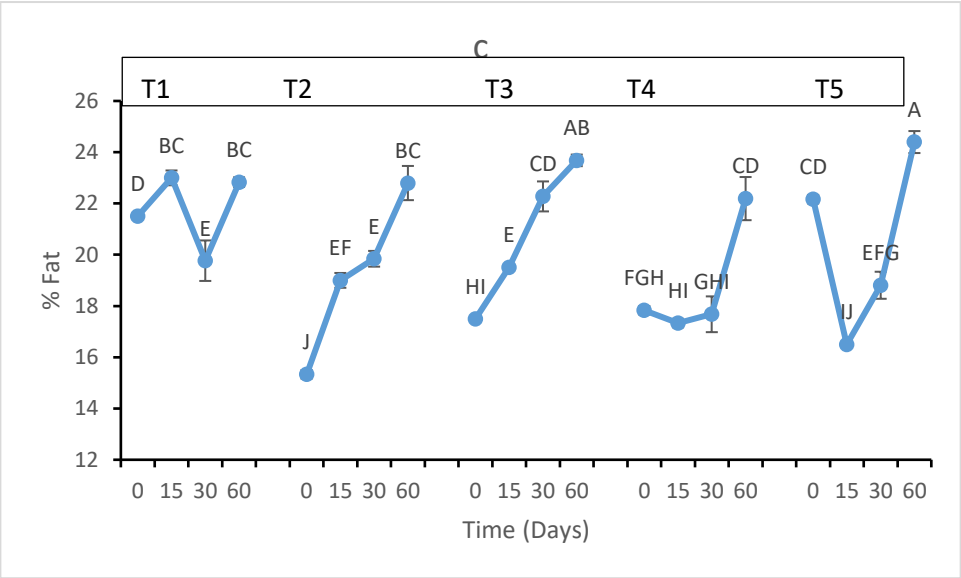


Figure 3. Effects of different treatments on % Fat of Feta cheese (a). Treatment 1(T1): *Lb.plantarum* & *Lb.helveticus*, Treatment 2(T2):*Ent.faecium* & *Lb.helveticus*; Treatment 3(T3):*Lb.delbrueckii* & *Str.thermophilus*, Treatment 4(T4): *Lb.plantarum* & *Ent.faecium*, Treatment 5(T5): Control. Effects of ripening period on % Fat of Feta cheese (b). Effect of interaction of treatments and ripening period on % Fat of Feta cheese (c).

The decrease in fat content during storage in treatment 4 (containing *Lactobacillus plantarum–Enterococcus faecium*) and the control sample can be attributed to the progress of lipolysis and the hydrolysis of fat into free fatty acids [22]. Various studies have also reported changes in fat concentration in different cheeses during ripening. Milci et al. (2005) and Shahab Lavasani et al. (2012) observed that with increasing ripening time, the fat content of cheese samples decreased, mainly due to the hydrolysis of fat into free fatty acids and volatile compounds [32, 33].

In contrast, Aly (1995) observed that the fat content of Feta cheese increased during ripening and explained this phenomenon by a decrease in moisture over time [34]. Moisture increase leads to a decrease in fat content during storage; in other words, moisture and fat have an inverse relationship. Similarly, Sadeghi and Nateghi (2020) reported that with increased moisture in Feta cheese during storage, fat and protein content decreased, showing an inverse relationship between moisture and fat [29].

In another study, consistent with part of the results of this research, Milci et al. (2005) and Lavasani et al. (2012) reported that increased ripening time led to a reduction in fat content [32, 33]. Consistent with this study, Gholamhoseinpour et al. (2022) reported that Feta cheese containing different starters exhibited different fat patterns during ripening, with some treatments showing a decrease in fat while others showed an increase; however, these changes were not statistically significant [22].

Dry matter percentage

The results of the analysis of variance showed that the influence of treatments (different types of co-starters), time, and the interaction effect of treatments × time on dry matter were significant ($P < 0.01$). Figure 4-a illustrates the changes in the percentage of dry matter over the storage period of Feta cheese, which showed a significant decreasing trend ($P < 0.01$). Figure 4-b presents the analysis of variance results of the dry matter percentage of different cheese treatments compared to the control sample. Mean comparisons indicated that the highest dry matter percentage (37.79%) was observed in Treatment 1, while the lowest percentage (33.38%) was in Treatment 5.

Figure 4-c shows the interaction effect of time and different Feta cheese treatments. With increasing storage time, the dry matter content of all treatments exhibited a decreasing trend, except for Treatments 1 and 3, which showed a decreasing trend until day 30, followed by a slight increase from day 30 to day 60. The rate

of decrease, however, varied statistically among the different treatments.

Comparisons of different treatments on day 0 of storage showed that the dry matter content of Treatment 1 was significantly higher ($P \leq 0.01$) than that of other treatments, and significant differences ($P \leq 0.01$) were observed among the other treatments and the control sample. On day 15 of storage, the dry matter content of Treatments 1, 2, and 4 was higher than that of the control, unlike Treatment 3.

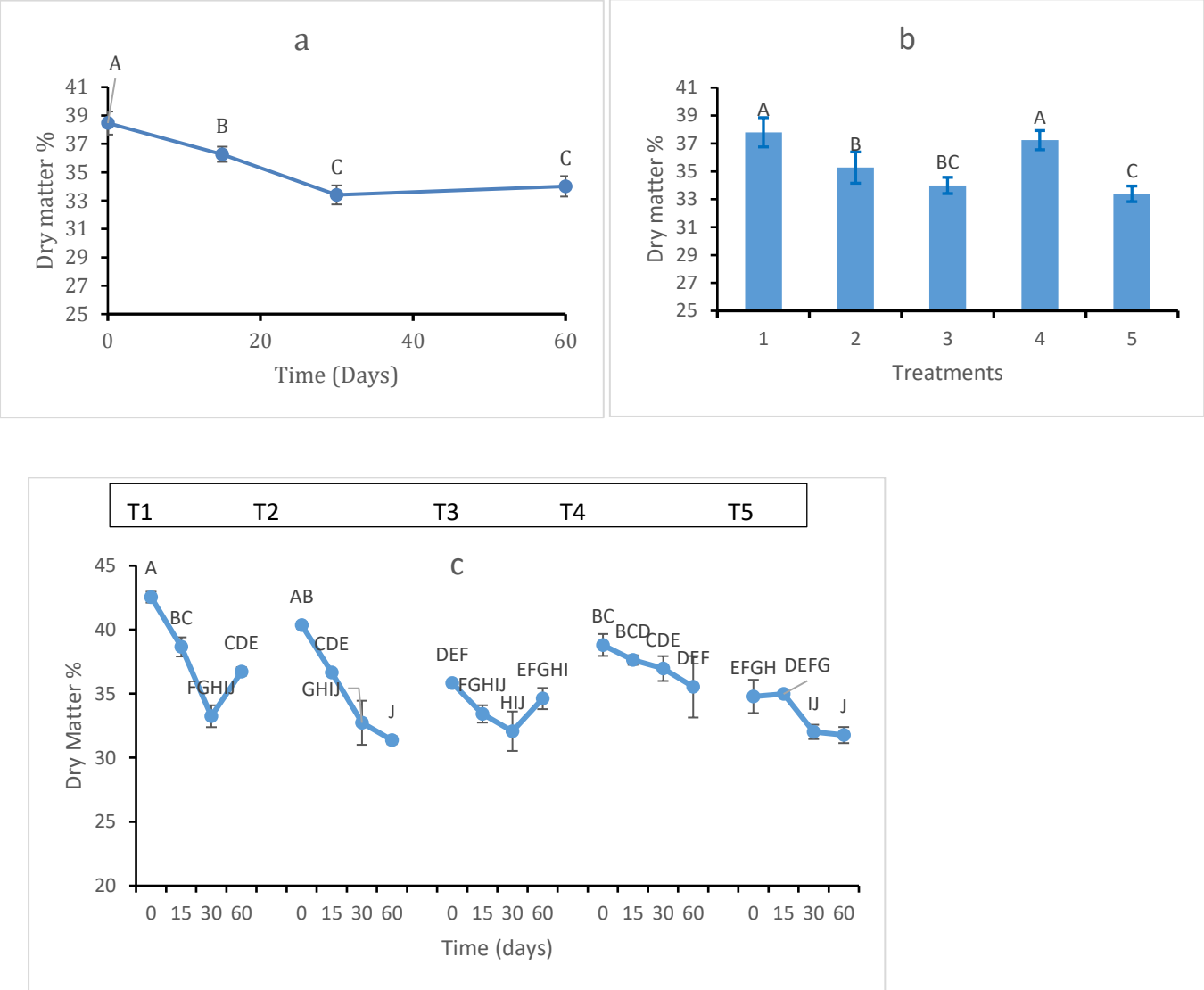


Figure 4. Effects of different treatments on % Dry Matter of Feta cheese (a). Treatment 1(T1): *Lb.plantarum* & *Lb.helveticus*, Treatment 2(T2):*Ent.faecium* & *Lb.helveticus*; Treatment 3(T3):*Lb.delbrueckii* & *Str.thermophilus*, Treatment 4(T4): *Lb.plantarum* & *Ent.faecium*, Treatment 5(T5): Control. Effects of ripening period on % Dry Matter of Feta cheese (b). Effect of interaction of treatments and ripening period on % Dry Matter of Feta cheese (c).

As mentioned, with an increase in storage time, the dry matter content of different treatments decreased, and this decrease was significant in some samples. There are conflicting reports regarding the effect of storage time on the solid content of cheeses in different varieties. Aly (1995), Azarnia et al. (1997), Al-Otaibi and Wilbey (2004), and Lopez et al. (2007) reported that with increased ripening time, the solid content increases. They stated that high storage temperatures of the curd, which affect casein hydration, lead to an increased degree and intensity of syneresis due to higher salt concentration in the curd. Water evaporation during storage and minimal water absorption in cheeses with low pH could also explain this increase.

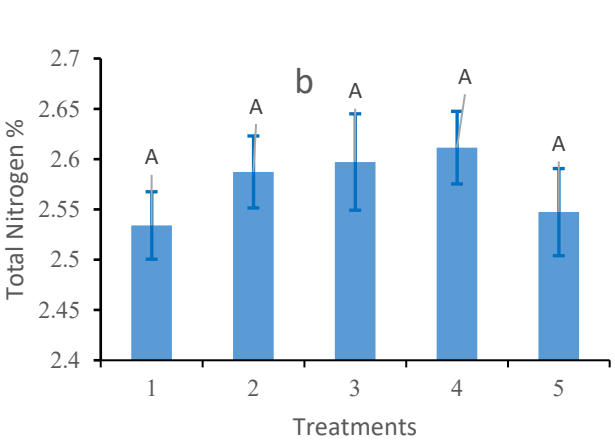
Conversely, Milci et al. (2005) and Shahab Lavasani et al. (2012) reported a decrease in cheese solid content during storage. The migration of water-soluble proteins and peptides from the curd to the surrounding brine, lipolysis, and the transfer and migration of free fatty acids from the cheese matrix to the brine have been suggested as the main factors causing this reduction in cheese dry matter.

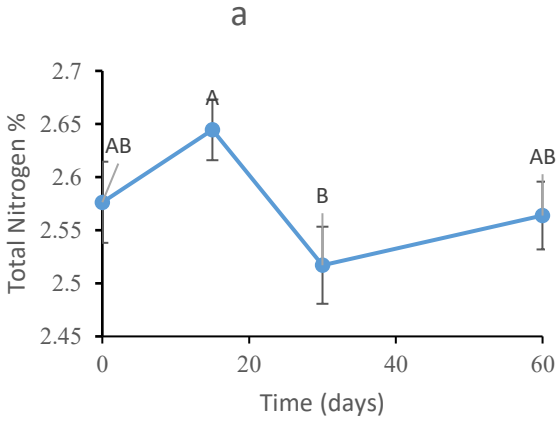
Consistent with the results of this study, Lavasani et al. (2010), while examining changes in the physicochemical characteristics of Liqvan cheese, reported that the dry matter content of the cheese decreased over the ripening period and sometimes remained constant. During the storage period, the treatment containing *Lactobacillus plantarum–Lactobacillus helveticus* (treatment 1) had higher dry matter (lower moisture) than other treatments, which could probably be due to the weaker ability of *Lactobacillus plantarum–Lactobacillus helveticus* bacteria to retain water in the casein network, resulting in more whey loss.

In a study by Sobhi Sarabi et al. (2014), in agreement with part of this study's findings, it was reported that there was no significant difference in the dry matter content of sheep milk cheese with the addition of *Lactobacillus casei–Lactobacillus plantarum* strains isolated from traditional Liqvan cheese. Gholamhosseinpour et al. (2022) also reported that the type of starter culture did not have a significant effect on the dry matter content of Feta cheese after ultrafiltration. Similarly, Haines et al. (2003) and Hayaoglu et al. (2005) reported comparable results.

Protein (Total Nitrogen)

The results of the analysis of variance showed that the effects of treatments (different types of adjunct starters), time, and the interaction of treatments and time on total nitrogen content were not significant. Statistical findings on changes in the protein content of different cheese treatments compared with the control sample during the storage period are presented in Figure 5. With increasing storage time, the protein content of sample 1 showed a continuous upward trend, while samples 2, 4, and 5 displayed a similar pattern—an initial increase up to day 15 followed by a decrease up to day 60. On day 0 of storage, the protein content of treatments 2, 3, and 4 was higher than that of the control sample, although the differences were not significant ($p \leq 0.01$). However, the protein content of treatment 1 was significantly lower on day 0. Moreover, on day 15 of storage, the protein content of all treatments showed no significant differences ($p \leq 0.05$) from one another.





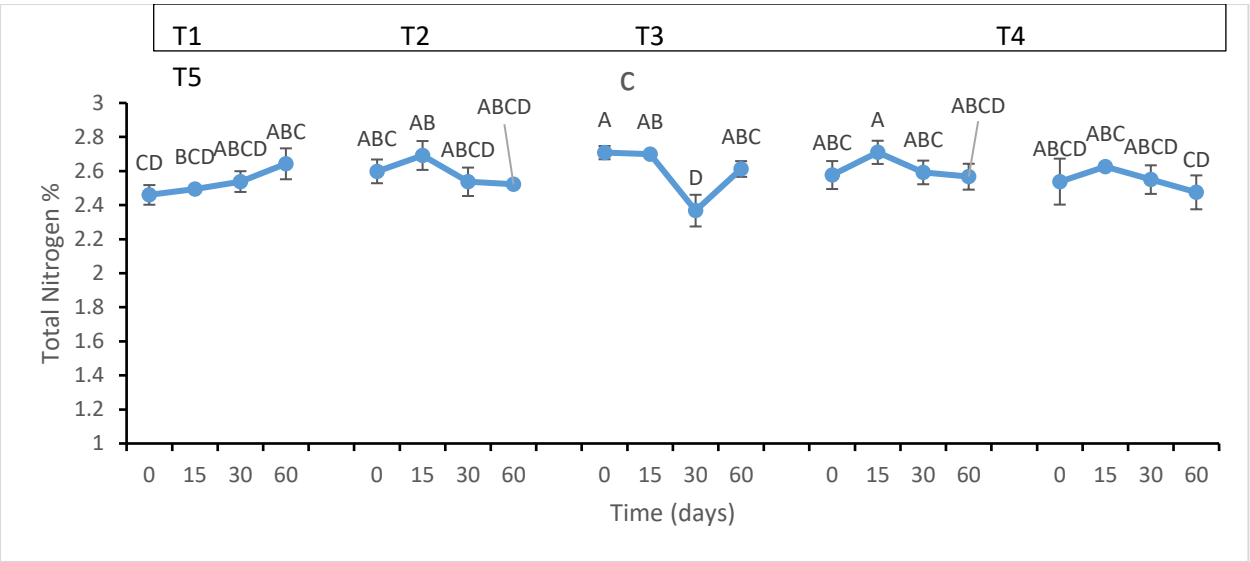


Figure 5. Effects of different treatments on % Total Nitrogen of Feta cheese (a). Treatment 1(T1): Lb.plantarum & Lb.helveticus, Treatment 2(T2):Ent.faecium & Lb.helveticus; Treatment 3(T3):Lb.delbrueckii & Str.thermophilus, Treatment 4(T4): Lb.plantarum & Ent.faecium, Treatment 5(T5): Control. Effects of ripening period on % Total Nitrogen of Feta cheese (b). Effect of interaction of treatments and ripening period on % Total Nitrogen of Feta cheese(c).

As mentioned, with the increase in storage time, the protein content of all treatments and the control sample exhibited different trends (Figure 4-C). In this regard, Gholamhosseinpour et al. (2022) reported that the protein content during ripening at 4 °C decreased until day 40 and then increased until the end of the ripening period [22]. The initial decrease in protein at the beginning of storage may be due to proteolysis reactions, while the subsequent increase is likely related to the rise in dry matter content.

On day zero of storage, comparison among treatments showed that the third treatment had the highest protein content, while the first treatment had the lowest. By day 15, the fourth treatment exhibited a higher protein content than the others, although this difference was not statistically significant (Figure 4-C). This observation is probably attributable to the lower water absorption of this treatment compared to the others during the storage period.

Salt content (%)

The results of the analysis of variance showed that the effect of treatments (different types of adjunct starters) and time on salt percentage were significant ($P<0.01$), while the interaction effect of treatments × time on salt percentage was not significant. Comparison of the mean salt content under the influence of treatments indicated that, overall, this variable ranged from 3.61% (in treatment 1) to 2.71% (in treatment 3) (Figure 6-b).

The effect of time on salt content is shown in Figure 6-a, where illustrates that with increasing storage time, the salt content increases. The statistical findings regarding the salt content of different cheese treatments during the storage period are presented in Figure 6-c. On day zero of storage, the salt content of treatment 1 (T1) was significantly higher ($P<0.01$) than that of the other treatments. Among the treatments, the highest and lowest salt contents belonged to treatment 1 and treatment 3, respectively (Figure 6-b). On day 15 of storage, treatment 1 still had the highest salt content, while no significant differences were observed among the other treatments and the control sample.

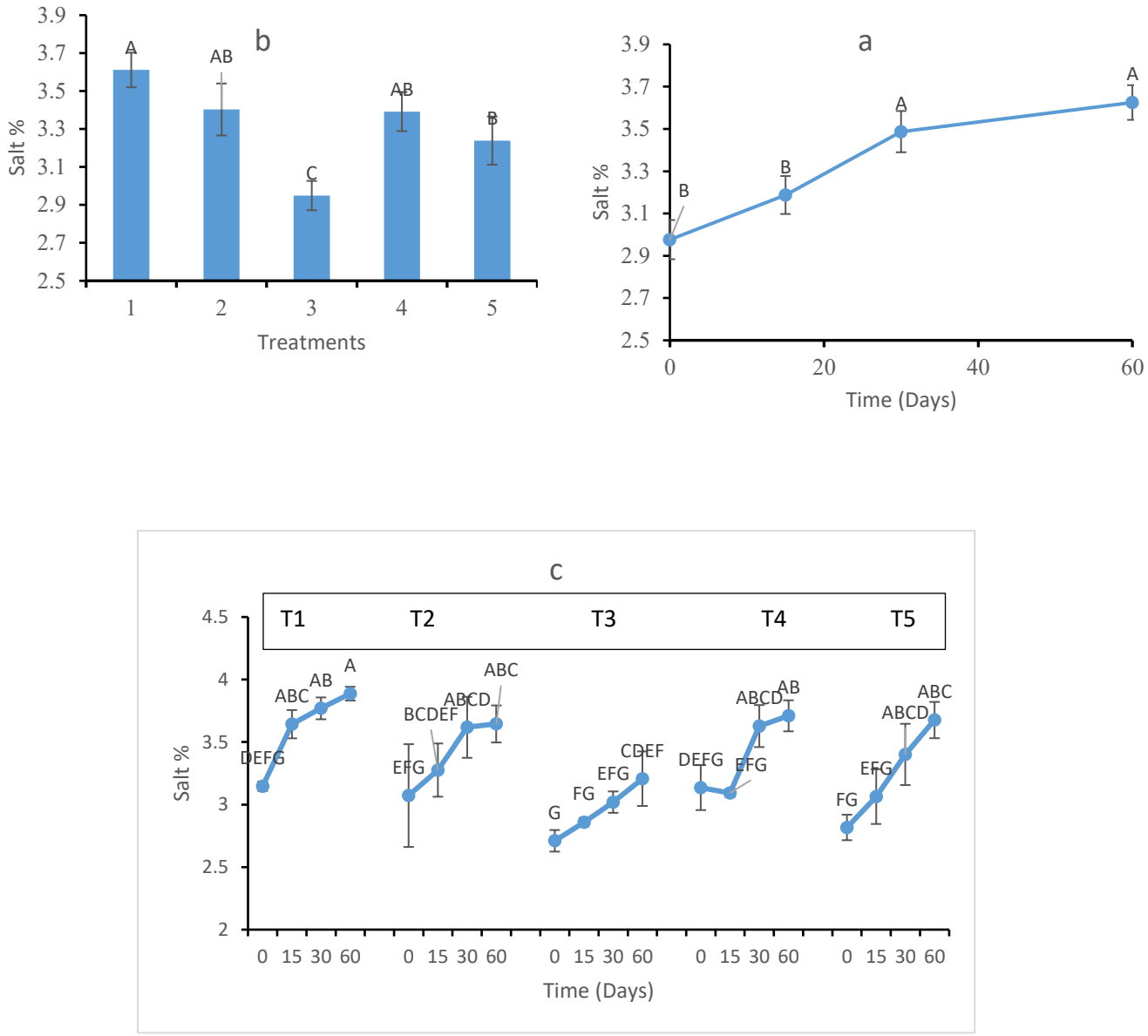


Figure 6. Effects of different treatments on % Salt of Feta cheese (a). Treatment 1(T1): Lb.plantarum & Lb.helveticus, Treatment 2(T2):Ent.faecium & Lb.helveticus; Treatment 3(T3):Lb.delbrueckii & Str.thermophilus, Treatment 4(T4): Lb.plantarum & Ent.faecium, Treatment 5(T5): Control. Effects of ripening period on % Salt of Feta cheese (b). Effect of interaction of treatments and ripening period on % Salt of Feta cheese (c).

As mentioned earlier, no significant differences were observed among the treatments during the storage period; in other words, as stated above, the interaction effect of time × treatment on salt percentage was not significant. An increase in storage time led to higher salt content in some treatments; however, this increase was significant only in treatment T1, which contained *Lactobacillus plantarum–Lactobacillus helveticus*. Azarnia et al. (1997) stated that this increase may be due to water loss and the gradual diffusion of salt from the brine into the cheese curd during storage [35].

In line with the results of the present study, Gholamhosseinpour et al. (2022) reported that during the ripening period, the salt content of ultrafiltration Feta cheese samples containing different starters increased, which was likely due to the gradual diffusion of salt from the surrounding brine into the cheese matrix during ripening [22]. According to some researchers (Hynes et al., 2003; Muir et al., 1996), the effect of the starter on the salt content of various cheeses was not significant [39, 40]. In contrast, Hayaloglu et al. (2013) reported a significant effect of starter type on the salt content of Gokceada cheese [31].

Different results have been reported regarding changes in salt concentration during cheese ripening. No significant increase in salt content

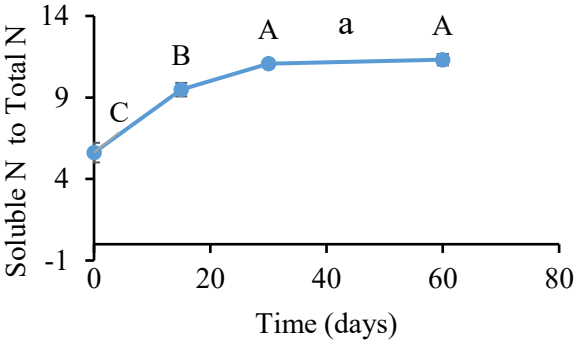
during cheese ripening was observed by Azarnia et al. (1997) and Shahab Lavasani et al. (2012) [33, 35]. However, Karimi et al. (2012) reported a non-significant decrease in salt content [41].

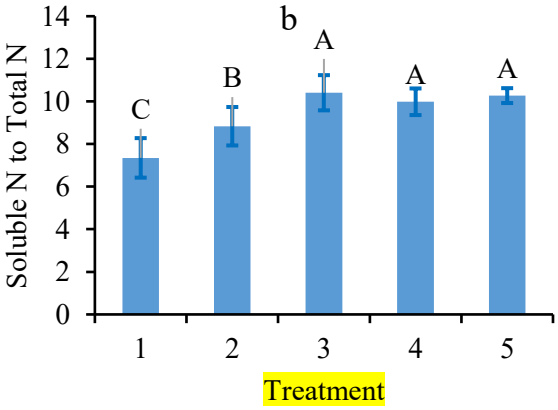
Ratio of soluble nitrogen to total nitrogen

The results of the analysis of variance showed that the effect of treatments (different types of adjunct starters), time, and the interaction of treatments × time on the ratio of soluble nitrogen to total nitrogen were significant ($P<0.01$). The ratio of soluble nitrogen to total nitrogen in different cheese treatments compared to the control sample during the storage period is presented in Figure 7. With increasing storage time, the proteolysis index of all treatments and the control sample increased significantly ($p\leq0.05$), indicating the progression of proteolytic reactions.

On day zero of storage, all treatments had lower proteolysis indices than the control sample. Among the treatments, the highest and lowest values of this index belonged to treatment 3 and treatment 1, respectively. On day 15 of storage, treatments 3 and 4 showed higher proteolysis indices than the control, while treatments 1 and 2 had lower values than the control.

The effect of adjunct and starter cultures on the ratio of soluble nitrogen to total nitrogen depends on the strain type and whether the strain is thermophilic or mesophilic in nature. An increase in the ratio of soluble nitrogen to total nitrogen during the ripening of ultrafiltration Feta cheese has also been observed in previous studies [42, 43, 44].





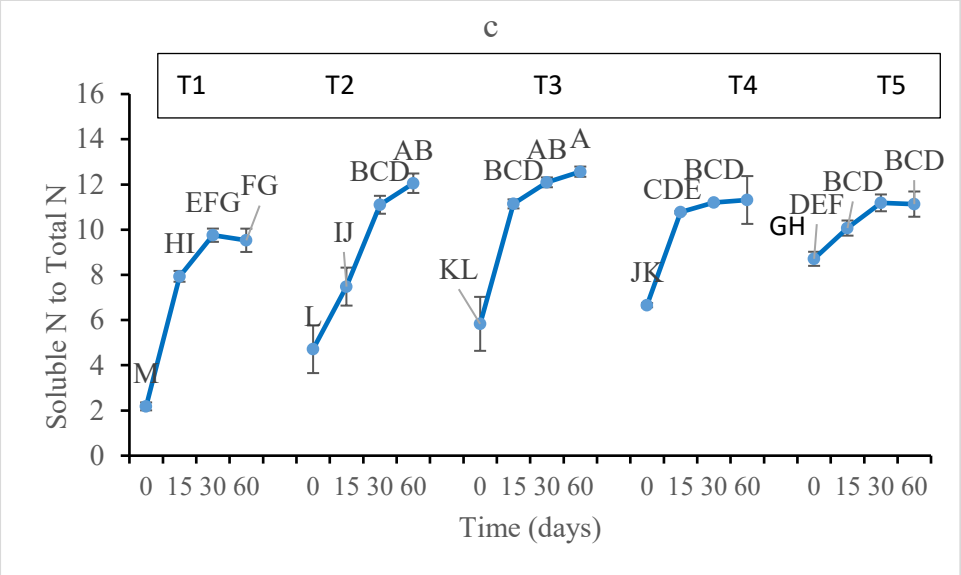


Figure 7. Effects of different treatments on % Soluble N to Total N of Feta cheese (a). Treatment 1(T1): Lb.plantarum & Lb.helveticus, Treatment 2(T2):Ent.faecium & Lb.helveticus; Treatment 3(T3):Lb.delbrueckii & Str.thermophilus, Treatment 4(T4): Lb.plantarum & Ent.faecium, Treatment 5(T5): Control. Effects of ripening period on % Soluble N to Total N of Feta cheese (b). Effect of interaction of treatments and ripening period on % Soluble N to Total N of Feta cheese (c).

The amount of soluble nitrogen only provides information about the intensity of primary proteolysis and does not reveal details regarding the composition of soluble nitrogen. Therefore, the total amount of soluble nitrogen may be similar in different samples, but due to the production of different proteolysis products, diverse flavors and aromas may be generated [45]. Primary proteolysis of cheese proteins mainly results from residual rennet activity and endogenous proteinases, while the complex proteolytic and peptidolytic systems of starter and non-starter microorganisms lead to secondary proteolysis [46].

Enterococcus species play an important role in cheese ripening through casein degradation via their proteolytic and peptidolytic activities. In this regard, proteolytic activity has been reported for *Enterococcus faecalis*, *Enterococcus faecium*, and *Enterococcus durans* isolated from various cheeses. It is noteworthy that Enterococcus strains possess stronger proteolytic activity compared to other lactic acid bacteria, which makes them important in cheese ripening [47].

Lactobacillus species have been reported as dominant organisms in cheeses made from raw milk, as they are able to grow under harsh selective conditions and, due to their excellent proteolytic properties, play a key role in developing the sensory characteristics of the product [48]. As mentioned earlier, at the beginning of the storage period, the cheese treatments had lower soluble protein content compared to the control, whereas at the end of storage, the results were reversed. In this context, Lane and Fox (1996) stated that the addition of non-starter lactic acid bacteria (NSLAB) does not play a major role in casein breakdown [49], and the primary proteolytic ability of NSLAB is generally weak and variable [50].

In another study, Di Cagno et al. (2006), investigating the effect of mesophilic lactobacilli on proteolysis in Caciotta cheese, reported that although NSLAB contribute little to primary proteolysis, their peptidases play a major role in secondary proteolysis by acting on peptides produced during primary proteolysis [51]. Similarly, Hynes et al. (2002), in a study on the use of different starter cultures and adjunct Lactobacillus strains in washed-curd cheese after 28 days of ripening, reported that the soluble nitrogen content of adjunct-containing samples was equal to or slightly higher than that of the control [52].

Higher levels of secondary proteolysis in Cheddar cheese containing adjunct Lactobacillus strains have also been demonstrated by Ong et al. (2007) and Sihufe et al. (2013), who attributed the observed increase to the higher peptidase activity in cheeses made with adjunct cultures [53, 25]. It

was found that the ratio of soluble nitrogen to total nitrogen increased during the storage period, which is consistent with the findings of Franco et al. (2001), Atasoy et al. (2008), and Gholamhosseinpour et al. (2022) [22, 54, 55].

Lipolysis

Figure 8 shows the lipolysis index of different cheese treatments compared to the control sample during the storage period. With increasing storage time, the lipolysis index increased significantly ($p \leq 0.05$) in all treatments as well as in the control. On day zero of storage, treatments 3 and 4, in contrast to treatments 1 and 2, had higher lipolysis indices than the control. On day 15 of storage, the lowest lipolysis index was observed in the control sample, while all treatments showed higher lipolysis indices than the control. Among the treatments, the highest and lowest lipolysis indices were related to treatment 3 and treatment 5, respectively.

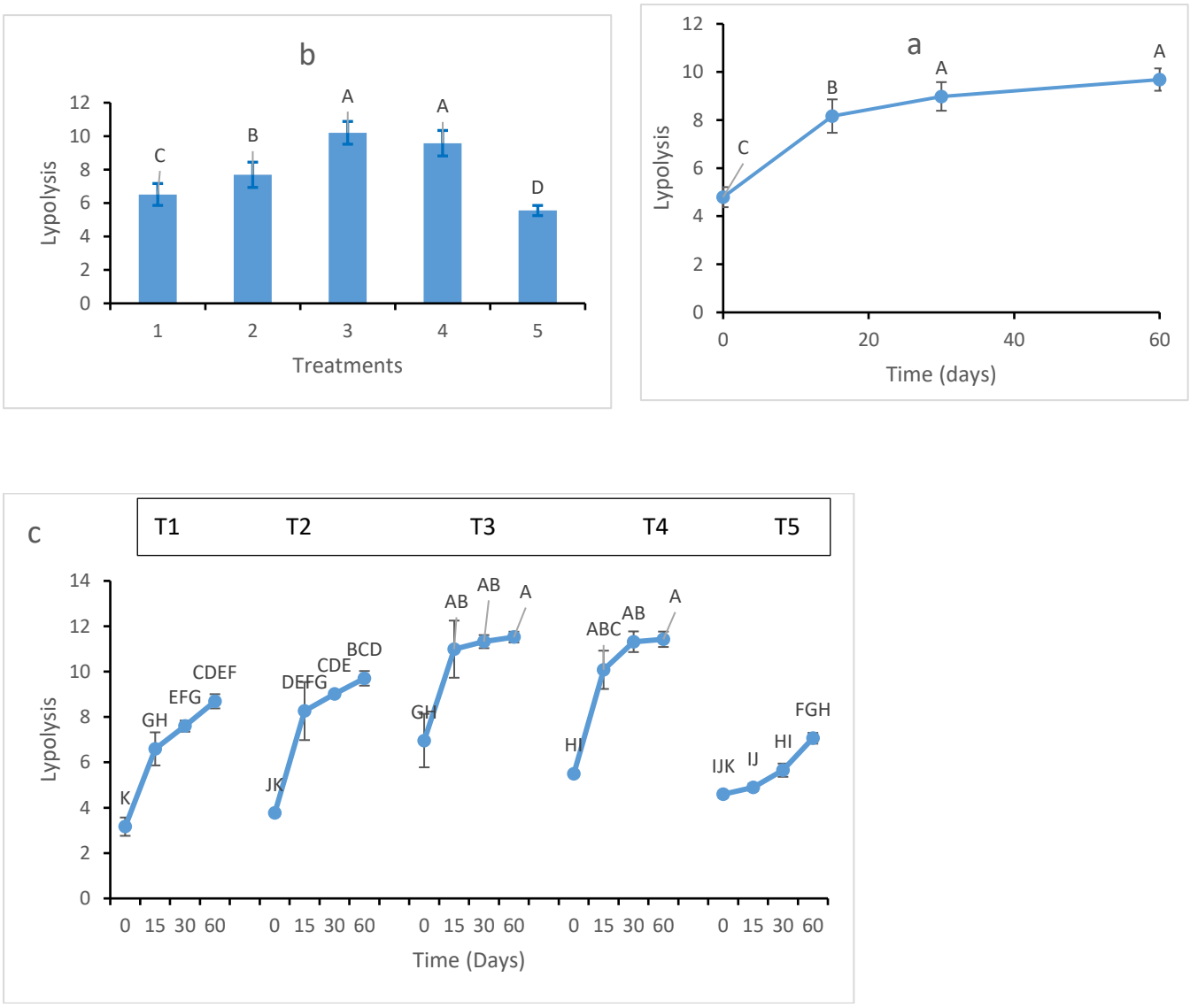


Figure 8. Effects of different treatments on Lypolysis of Feta cheese (a). Treatment 1(T1): Lb.plantarum & Lb.helveticus, Treatment 2(T2):Ent.faecium & Lb.helveticus; Treatment 3(T3):Lb.delbrueckii & Str.thermophilus, Treatment 4(T4): Lb.plantarum & Ent.faecium, Treatment 5(T5): Control. Effects of ripening period on Lypolysis of Feta cheese (b). Effect of interaction of treatments and ripening period on Lypolysis of Feta cheese ©.

The hydrolysis of milk fat during cheese production and ripening is caused by milk's endogenous lipase, lipolytic enzymes of starter and non-starter bacteria, and lipases produced by psychrotrophic bacteria. Lipolysis plays an important role in cheese ripening. It leads to the formation of free fatty acids, which contribute to flavor development and serve as precursors

for compounds such as methyl ketones, alcohols, and lactones. Major factors affecting the extent of lipolysis include the type and quality of milk, milk pasteurization temperature used for cheesemaking, the starter culture, storage and ripening temperature, brine concentration, and milk lipase [45].

Lipase in cheese originates from six sources: milk, rennet, starter cultures, adjunct cultures, non-starter bacteria, and exogenous lipases if used [56]. It has been shown that when the levels of non-starter lactic acid bacteria in cheese increase significantly, these bacteria may balance lipase and esterase activities and contribute to the release of free fatty acids during ripening [51].

Hynes et al. (2002) studied the effect of different starter cultures and adjunct Lactobacillus strains (five strains of Lactobacillus plantarum, one strain of Lactobacillus pentosus, and four strains of Lactobacillus casei) on the ripening of washed-curd cheese [52]. They reported that regardless of the type of commercial starter used, after 28 days of ripening, the level of lipolysis and free fatty acid content in all cheeses containing adjunct cultures was higher than in the control samples. These findings are in agreement with the results of the present study.
As mentioned earlier, in all treatments, lipolysis increased during the storage period, which is consistent with the findings of Xanthopoulos et al. (2000), Atasoy et al. (2008), and Lavasani (2011) [33, 55, 57].

4-Conclusion

In this study, the effect of different lactic isolates (obtained from curd, fresh cheese, ripened Lighvan cheese, and yogurt), in the form of five different treatments, on the physicochemical properties of traditionally produced Feta cheese was investigated. The results showed that with increasing storage time, pH decreased while acidity increased, with the highest and lowest pH values belonging to treatment (1) containing *Lactobacillus plantarum–Lactobacillus helveticus* and treatment (5) (control), respectively.
The analysis of dry matter indicated that with increasing storage time, the dry matter content of the control and all treatments decreased, and the highest dry matter at the end of the ripening period belonged to treatment (1) containing *L. plantarum–L. helveticus*. Moreover, the highest total nitrogen and salt contents at the end of ripening were also observed in treatment (1). The higher salt content at day 60 could potentially act as an inhibitory factor against spoilage microorganisms in cheese.
The evaluation of proteolysis and lipolysis indices of different treatments and the control sample during storage showed that the highest values of these indices at the end of storage were related to treatment (3), containing *Lactobacillus delbrueckii–Lactococcus lactis–Streptococcus thermophilus*.
Overall, the findings of this study demonstrated that the addition of the lactic isolates *Lactobacillus plantarum–Lactobacillus helveticus* to the formulation of Feta cheese can lead to the production of a product with desirable physicochemical properties.

5-Acknowledgments

The authors would like to express their sincere gratitude to the Vice Chancellor for Research and Technology of Ferdowsi University of Mashhad for their support and financial assistance. The research of the corresponding author was financially supported by Ferdowsi University of Mashhad under research project code (57902/3). The authors also wish to thank Astan Quds Razavi Dairy Company and Pegah Khorasan Razavi Dairy Company for their cooperation in cheese production, as well as the Central Laboratory and the Molecular Biotechnology Laboratory of Ferdowsi University of Mashhad for their collaboration in conducting the experiments related to this article and thesis.

6-References

[1]. López-Expósito, I., Amigo, L., & Recio, I. (2012). A mini-review on health and nutritional aspects of cheese with a focus on bioactive peptides.

[2]. Ghoddoosi, H., B. Habibi Najafi, M. B. Mazaheri Tehrani, M. and Razavi, M. A. 2004. Production of Feta Cheese by Industrial and Traditional Methods. Ferdowsi University of Mashhad Publications.

[3]. Ahmed, M. E., Rathnakumar, K., Awasti, N., Elfaruk, M. S., & Hammam, A. R. (2021). Influence of probiotic adjunct cultures on the characteristics of low-fat Feta cheese. *Food Science & Nutrition*, *9*(3), 1512-1520.

[4]. Hamdy, A. M., Ahmed, M. E., Mehta, D., Elfaruk, M. S., Hammam, A. R. A., & El- erwy, Y. M. A. (2020). Enhancement of low-fat Feta cheese characteristics using probiotic bacteria. *Food Science & Nutrition*, *8*(11), fsn3.1889. https://doi.org/10.1002/fsn3.1889.

[5]. Bintsis, T., & Robinson, R. (2004). A study of the effects of adjunct cultures on the aroma compounds of Feta-type cheese. *Food Chemistry*, *88*(3), 435–441. https://doi.org/10.1016/j.foodc hem.2004.01.057

[6]. W.C. Meijer, H.B. Brandsma: Application of Lactic Acid Bacteria in Dairy Starters. In: *Food Fermentation*, R.M.J. Nout, W.M. de Vos, M.H. Zweitering (Eds.), Wageningen Academic Publishers, Wageningen, The Netherlands (2005) pp. 61–67.

[7]. Leboš Pavunc, A., Beganović, J., Kos, B., Uroić, K., Blažić, M., & Šušković, J. (2012). Characterization and application of autochthonous starter cultures for fresh cheese production. *Food Technol Biotechnol*, *50*(2), 141-51.

[8]. Edalatian, M. R., Najafi, M. B. H., Mortazavi, S. A., Alegría, Á., Nassiri, M. R., Bassami, M. R., & Mayo, B. (2012). Microbial diversity of the traditional Iranian cheeses Lighvan and Koozeh, as revealed by polyphasic culturing and culture-independent approaches. Dairy science & technology, 92, 75-90.

[9]. Hajimohammadi Farimani, R., Habibi Najafi, M. B., Fazly Bazzaz, B. S., Edalatian, M. R., Bahrami, A. R., Flórez, A. B., & Mayo, B. (2016). Identification, typing and functional characterization of dominant lactic acid bacteria strains from Iranian traditional yoghurt. European Food Research and Technology, 242, 517-526.

[10] Iran Standard no. 2344 (1402)., Brined Cheese. Specification and test methods. *National Standards Authority of Iran, Tehran, Iran*.

[11]. Iran Standard no. 1753 (1398)., Cheese and processed cheese products: Determination of Total solids. Specification and test methods. *National Standards Authority of Iran, Tehran, Iran*.

[12]. Iran Standard no.2852. (1401) Milk and milk products. Determination of titrable acidity and pH - Test method. *National Standards Authority of Iran, Tehran, Iran.*

[13]. Iran Standard no.1809. (1399). Determination of chloride content. *National Standards Authority of Iran, Tehran, Iran.*

[14]. Iran Standard no.8785. Cheese – Determination of fat content. *National Standards Authority of Iran, Tehran, Iran.*

[15]. Iran Standard no 9188-1. (1394). Milk and milk products — Determination of nitrogen content —Part 1: Kjeldahl principle and crude protein calculation. *National Standards Authority of Iran, Tehran, Iran.*

[16]. Kuchroo, C.N., and Fox, P.F. (1982). Soluble nitrogen in cheddar cheese. comparison of extraction procedures. Michwissenchaft, 937, 331-335.

[17]. Marshal, R.T. (1992). Standard methods for the examination of dairy products. pp: 271-272.

[18]. 1. Fox, P. F., Mc. Sweeney, P. L., Cogan, T. M., and Guinee, T. P. (Eds.), 2004. Cheese: Chemistry, Physics and Microbiology: General Aspects (Vol. 1). Academic Press.

[19]. Sarantinopoulos, P., Kalantzopoulos, G., & Tsakalidou, E. (2002). Effect of Enterococcus faecium on microbiological, physicochemical and sensory characteristics of Greek Feta cheese. *International Journal of Food Microbiology*, *76*(1-2), 93-105.

[20]. Rahimi, J., Khosrowshahi, A., Madadlou, A., & Aziznia, S. (2007). Texture of low-fat Iranian white cheese as influenced by gum tragacanth as a fat replacer. *Journal of dairy science*, *90*(9), 4058-4070.

[21]. Khosrowshahi, A., Madadlou, A., zadeh Mousavi, M. E., & Emam-Djomeh, Z. (2006). Monitoring the chemical and textural changes during ripening of Iranian White cheese made with different concentrations of starter. Journal of dairy science, 89(9), 3318-3325.

[22]. Gholamhosseinpour, A., & Zare, S. (2024). Investigating the effect of Persian gum on physicochemical, textural and rheological characteristics of UF white brined cheese during ripening. *International Dairy Journal*, *155*, 105952.

[23]. Mantzourani, I., Terpou, A., Alexopoulos, A., Chondrou, P., Galanis, A., Bekatorou, A.,... & Plessas, S. (2018). Application of a novel potential probiotic lactobacillus paracasei strain isolated from kefir grains in the production of feta-type cheese. Microorganisms, 6(4), 121.

[24]. Terzić-Vidojević, A. , Veljović, K. , Popović, N. , Tolinački, M. , & Golić, N.

(2021). Enterococci from raw-milk cheeses: current knowledge on safety, technological, and probiotic concerns. Foods, 10(11), 2753.
[25]. Ciocia, F., McSweeney, P. L., Piraino, P., & Parente, E. (2013). Use of dairy and non-dairy Lactobacillus plantarum, Lactobacillus paraplantarum and Lactobacillus pentosus strains as adjuncts in cheddar cheese. *Dairy Science & Technology*, *93*, 623-640.
[26]. Kayagil, F. (2006). Effect of traditional starter cultures on quality of cheese (Master's thesis, Middle East Technical University).
[27]. Nelson, B. K. , & Barbano, D. M. (2004). Reduced-fat Cheddar cheese manufactured using a novel fat removal process. Journal of dairy science, 87(4), 841-853.
[28]. Kondyli, E. , Katsiari, M. C. , Masouras, T. , & Voutsinas, L. P. (2002). Free fatty acids and volatile compounds of low-fat Feta-type cheese made with a commercial adjunct culture. Food Chemistry, 79(2), 199-205.
[29]. Sadeghi, F. and Nateghi, L. (2020). Investigating the sensory properties and antimicrobial effect of frankincense and shallots essential oil on Listeria monocytogenes bacteria in white brined cheese. *Iranian Food Science and Technology Research Journal*, *16*(4), 343-356. doi: 10.22067/ifstrj.v16i4.80935
[30]. Hayaloglu, A., Guven, M., Fox, P. & McSweeney, P. (2005). Influence of starters on chemical, biochemical, and sensory changes in Turkish white-brined cheese during ripening. *Journal of Dairy Science,* 88, 3460-3474.
[31]. Hayaloglu, A., Tolu, C. & Yasar, K. (2013). Influence of goat breeds and starter culture systems on gross composition and proteolysis in Gokceada goat cheese during ripening. *Small Ruminant Research,* 113, 231-238.
[32]. Milci, S. , Goncu, A. , Alpkent, Z. A. F. E. R. , & Yaygin, H. (2005). Chemical, microbiological and sensory characterization of Halloumi cheese produced from ovine, caprine and bovine milk. International Dairy Journal, 15(6-9), 625-630.
[33]. SHAHAB LAVASANI, A. R., Ehsani, M. R., Mirdamadi, S., & EBRAHIM ZADEH MOUSAVI, M. A. (2012). Changes in physicochemical and organoleptic properties of traditional Iranian cheese Lighvan during ripening. International journal of dairy technology, 65(1), 64-70.
[34]. Aly, M. E. (1995). An attempt for producing low-sodium Feta-type cheese. *Food chemistry*, *52*(3), 295-299.
[35]. Azarnia, S. , Ehsani, M. R. , & Mirhadi, S. A. (1997). Evaluation of the physico-chemical characteristics of the curd during the ripening of Iranian brine cheese. International Dairy Journal, 7(6-7), 473-478.
[36]. Al-Otaibi, M. M., & Wilbey, R. A. (2004). Effect of temperature and salt on the maturation of white-salted cheese. International journal of dairy technology, 57(1), 57-63.
[37]. Lopez, C., Camier, B., & Gassi, J. Y. (2007). Development of the milk fat microstructure during the manufacture and ripening of Emmental cheese observed by confocal laser scanning microscopy. *International Dairy Journal*, *17*(3), 235-247.
[38]. Sobhi Sarabi, Y., Hesari, J., Peyghamdoost, S. H. and Raft, A. (2014). Physicochemical and sensory properties of pasteurized sheep cheese with the addition of Lactobacillus casei and Lactobacillus plantarum strains isolated from traditional Lighvan cheese. Food Industry Research (Agricultural Science), 24(3): 413-423.
[39]. Hynes, E. , Ogier, J. C. , Son, O. , & Delacroix-Buchet, A. (2003). Influence of starter and adjunct lactobacilli culture on ripening of miniature washed-curd cheeses. Le Lait, 83(1), 17-29.
[40]. Muir, D. D. , Banks, J. M. , & Hunter, E. A. (1996). Sensory properties of Cheddar cheese: effect of starter type and adjunct. International Dairy Journal, 6(4), 407-423.
[41]. Karimi, R. , Mortazavian, A. M. , & Karami, M. (2012). Incorporation of Lactobacillus casei in Iranian ultrafiltered Feta cheese made by partial replacement of NaCl with KCl. Journal of dairy science, 95(8), 4209-4222.
[42]. Karami, M., Ehsani, M., Mousavi, S., Rezaei, K. & Safari, M. (2009b). Changes in the rheological properties of Iranian UF-Feta cheese during ripening. Food chemistry, 112, 539-544.
[43]. Fathollahi, I., Hesari, J., Azadmard, S., & Oustan, S. (2010). Influence of proteolysis and soluble calcium levels on textural changes in the interior and exterior of Iranian UF white cheese during ripening. *World Acad. Sci. Eng. Technol*, *66*, 844-849.
[44]. Miočinović J. Puđa, P. , Radulović, Z. , Pavlović, V. , Miloradović, Z. , Radovanović, M. , & Paunović, D. (2011). Development of low fat UF cheese technology. Mljekarstvo, 61(1), 33.
[45]. De Wit, M., Osthoff, G., Viljoen, B. C., & Hugo, A. (2005). A comparative study of lipolysis and proteolysis in Cheddar cheese and yeast-inoculated Cheddar cheeses during

ripening. *Enzyme and Microbial Technology*, *37*(6), 606-616.
[46]. Hesari, J. , Ehsani, M. R. , Khosroshahi, A. , & McSweeney, P. L. (2006). Contribution of rennet and starter to proteolysis in Iranian UF white cheese. Le Lait, 86(4), 291-302.
[47]. Franz, C. M., Holzapfel, W. H., & Stiles, M. E. (1999). Enterococci at the crossroads of food safety?. *International journal of food microbiology*, *47*(1-2), 1-24.
[48]. Torres-Llanez, M. J., Vallejo-Cordoba, B. , Díaz-Cinco, M. E. , Mazorra-Manzano, M. A. , & González-Córdova, A. F. (2006). Characterization of the natural microflora of artisanal Mexican Fresco cheese. Food control, 17(9), 683-690.
[49]. Lane, C. N. , & Fox, P. F. (1996). Contribution of starter and adjunct lactobacilli to proteolysis in Cheddar cheese during ripening. International Dairy Journal, 6(7), 715-728.
[50]. Swearingen, P. A. , O'sullivan, D. J. , & Warthesen, J. J. (2001). Isolation, characterization, and influence of native, nonstarter lactic acid bacteria on Cheddar cheese quality. Journal of Dairy Science, 84(1), 50-59.
[51]. Di Cagno, R., Quinto, M., Corsetti, A., Minervini, F., & Gobbetti, M. (2006). Assessing the proteolytic and lipolytic activities of single strains of mesophilic lactobacilli as adjunct cultures using a Caciotta cheese model system. International Dairy Journal, 16(2), 119-130.
[52]. Hynes, E. , Ogier, J. C. , Lamberet, G. , & Delacroix-Buchet, A. (2002). The influence of starter and adjunct lactobacilli culture on the ripening of washed curd cheeses. Brazilian Journal of Chemical Engineering, 19, 397-402.
[53]. Ong, L., Henriksson, A., & Shah, N. P. (2007). Proteolytic pattern and organic acid profiles of probiotic Cheddar cheese as influenced by probiotic strains of Lactobacillus acidophilus, Lb. paracasei, Lb. casei or Bifidobacterium sp. International Dairy Journal, 17(1), 67-78.
[54]. Franco, I., Prieto, B., Urdiales, R., Fresno, J. M., & Carballo, J. (2001). Study of the biochemical changes during ripening of Ahumado de Aliva cheese: a Spanish traditional variety. *Food Chemistry*, *74*(4), 463-469.
[55]. Atasoy, A. F., & Türkoğlu, H. (2008). Changes of composition and free fatty acid contents of Urfa cheeses (a white-brined Turkish cheese) during ripening: Effects of heat treatments and starter cultures. *Food Chemistry*, *110*(3), 598-604.
[56]. Holland, R. , Liu, S. Q. , Crow, V. L. , Delabre, M. L. , Lubbers, M. , Bennett, M. , & Norris, G. (2005). Esterases of lactic acid bacteria and cheese flavour: Milk fat hydrolysis, alcoholysis and esterification. International Dairy Journal, 15(6-9), 711-718.
[57]. Xanthopoulos, V. , Polychroniadou, A. , Litopoulou-Tzanetaki, E. , & Tzanetakis, N. (2000). Characteristics of Anevato cheese made from raw or heat-treated goat milk inoculated with a lactic starter. *LWT-Food Science and Technology*, *33*(7), 483-488.

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مقاله علمی_پژوهشی

بررسی تاثیر جدایه‌های لاکتیکی حاصل از فراورده‌های لبنی بومی به عنوان کمک آغازگر و دوره رسیدگی بر خصوصیات فیزیکو شیمیایی پنیر فتا

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اطلاعات مقاله

تاریخ های مقاله :

تاریخ دریافت: ۱٤۰٤/۰٤/۰۲

تاریخ پذیرش: ۱٤۰٤/۰۵/۲۹

کلمات کلیدی:

پنیر فتا،

جدایه لاکتیکی،

فرآورده‌های لبنی بومی

DOI: 10.48311/fsct.2025.84052.0.

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چکیده

در این تحقیق تأثیر پنج تیمار مختلف به عنوان کمک اغازگر شامل ترکیبات مختلف باکتری‌های اسید لاکتیک (جدا شده از دلمه پنیر، پنیر تازه، پنیر رسیده لیقوان و ماست) بر ویژگی‌های فیزیکوشیمیایی پنیر فتا شامل درصد ماده خشک، درصد اسیدیته، pH ، درصد نمک، درصد چربی، درصد ازت کل، پروتئولیز(نسبت نیتروژن محلول به نیتروژن کل)، لیپولیز، طی مدت زمان ٦۰ روز دوره نگهداری، بررسی شد. تیمارهای مورد استفاده در این تحقیق عبارت بودند از: تیمار ۱ : *لاکتی پلانتی باسیلوس پلانتاروم* و *لاکتوباسیلوس هلوتیکوس* ، تیمار ۲ : *انتروکوکوس فاسیوم* و *لاکتوباسیلوس هلوتیکوس* ، تیمار ۳ :*لاکتوکوکوس لاکتیس، استرپتوکوکوس ترموفیلوس* و *لاکتوباسیلوس دلبروکی*، تیمار ٤ : *لاکتی پلانتی باسیلوس پلانتاروم* و *انتروکوکوس فاسیوم* و تیمار ۵ (نمونه شاهد): *لاکتوکوکوس لاکتیس* زیرگونه *لاکتیس* و *لاکتوکوکوس لاکتیس* زیرگونه *کرموریس* . نتایج نشان داد که با افزایش زمان نگهداری، pH تمامی تیمارها کاهش(از ٦/۳ به ۵/۹) و اسیدیته (از ۰/۲۷ به ۰/٦٦) افزایش معنی داری ($P<0.01$) یافت. در بین تیمارها نیز بالاترین pH متعلق به تیمار ۱ (٦/۲۱) وکمترین آن به تیمار ۵ (۵/۹٤) بود. همچنین با افزایش زمان نگهداری، میزان درصد ماده خشک نمونه شاهد و تمامی تیمارها کاهش معنی داری ($P<0.01$) از ۳۸/٤٦ به ۳٤/۰۰۸ می‌یابد. نتایج اندازه گیری شاخص پروتئولیز و لیپولیز تیمارهای مختلف و نمونه شاهد در طول مدت نگهداری نشان داد که بیشترین میزان این شاخص‌ها در پایان زمان نگهداری مربوط به تیمار۳ به ترتیب۱۰/٤۰ و ۱۰/۱۹۹ بود. به طور کلی، نتایج این تحقیق نشان داد که با افزودن سویه‌های *لاکتی پلانتی باسیلوس پلانتاروم* و *لاکتوباسیلوس هلوتیکوس* به فرمولاسیون پنیر فتا می‌توان محصولی با ویژگی‌های فیزیکوشیمیایی مطلوب تولید کرد.