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Research Article

Physicochemical Properties of Fatty and Low-Fat White Cheese Produced with Cultures Forming Exopolysaccharide

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Abstract

In this study, the possibilities of the usage of Exopolysaccharide (EPS)-producing cultures in White cheese production have been researched. For this purpose, physical, chemical, were investigated at days 1, 15, 30, 60 and 90 of cheese that produced two replicas with fat and non-fat cow milk via different cultures. It has been detected that the EPS has different effects on full fat and non- fat cheeses. The yields value was he cheese produced with EPS-producing cultures.). In all cheeses, the dry matter ratio was increased during ripening period (p>0.0.5), fat was stable (p>0.0.5), faded proportion was decreased (p>0.0.5), total nitrogen ratio was decreased; water-soluble and 12% TCA-soluble nitrogen ratio and index of ripening and treatable acidity were increased (p>0.0.5 were increased. Also, EPS producing culture was exhibited positive effect on texture and aroma in full-fat cheeses.

Introduction

Cheese is a fermented dairy product, which is rich in proteins, bioactive peptides, amino acids, fats, fatty acids, minerals and vitamins, and is important for adequate and balanced nutrition. Its structure, texture and aroma vary according to technological developments, cultures used and maturation conditions [1,2]. Starter cultures, which form the basis of the production of fermented foods, are defined as beneficial microorganisms that limit the development of harmful microorganisms in dairy technology, provide the formation of product-specific taste, aroma and structure, and are added to milk in order to obtain a standard product as much as possible [3,4]. Among the starter cultures, Lactic Acid Bacteria (LAB) play an important role in milk technology because of their rapid acid development, microbial stabilization of milk, product-specific aroma development, structure-forming capacity and beneficial effects on health depending on the amount and incubation period [5,3,6]. Some strains of LAB produce polysaccharides with different structures in suitable environments [7-10,3,6]. Polysaccharides secreted out of bacteria are defined as exocellular polysaccharides or Exopolysaccharides (EPS) [9]. EPSs are synthesized by the producer strain in order to be protected against the dangers that may occur in the environment such as drying of the environment, phage attacks, protozoa plunder, toxic compounds, antibiotics and osmotic stress. They also help the cell to adhere to the surface environment and form colonies [11]. EPSs, which consist of monosaccharide units linked by glycosidic bonds, can be composed of a single type of monosaccharide (homopolysaccharide) or different monosaccharides (heteropolysaccharide) [12,13,14]. The majority of mesophilic and thermophilic LABs used in dairy technology are capable of producing heteropolysaccharide group EPS. Lactococcus lactis ssp. lactis, Lactobacillus delbrueckii ssp. bulgaricus and Streptococcus thermophilus are the main LABs that produce heteropolysaccharides [12,13,14].

The amounts of polysaccharides produced by LAB vary based on the incubation time, bacterial strain, substrate amount and composition. Eps production of bacterial strains is not stable due to the above-mentioned reasons. While exopolysaccharide production increases in growth at low incubation temperatures, it also increases at near-neutral pH or with an increase in the carbon/nitrogen ratio in the growth medium [15,16]. In order to improve the texture and aroma of low-fat cheese, another recommended method, apart from modifying processing techniques, is the selection of starter cultures. The use of LAB cultures that produce EPS is seen as one of the possible ways in this regard. Since EPS provides functional properties such as increasing viscosity, stabilization and water binding in foods, the use of bacteria producing these polymers can provide an alternative to commercial stabilizers and fat replacers in cheese technology [12,9,17]. In this study, the effects of commercial starter cultures used in cheese production that provide acidity and aroma development, as well as the addition of EPS producing starter as a support culture on the physicochemical properties of white cheese produced from skim and fat cow's milk were investigated.

Materials and Methods

In the white cheese production, cow's milk, LYOFAST MOS 062 E (culture containing Lactococcus lactis ssp. lactis - Streptococcus thermophilus) as commercial liquid microbial rennet, DELVO-ADD 100F DIRECT SET (culture consisting of the EPS-producing strain of Streptococcus thermophilus), calcium chloride and salt were obtained from the markets in Kayseri marketplace. The trial produced 8 types of cheese. Cheese sample (A1) made with skimmed milk (0.6% fat) without adding culture (negative control group for skimmed cheese), Cheese sample (A2) made with fatty milk (3.1% fat) without adding culture (positive control group for fatty cheese), Cheese sample (negative control group for skim cheese) (B1) made using commercial cheese culture LYOFAST MOS 062 E (culture containing Lactococcus lactis ssp. lactis and Streptococcus thermophilus) with skim milk (0.6% fat), Cheese sample (B2) made using commercial cheese culture LYOFAST MOS 062 E (culture containing Lactococcus lactis ssp. lactis and Streptococcus thermophilus) with fatty milk (3.1% fat) (positive control group for fatty cheese), Cheese sample (1% + 1%) (C1) made with the combination of skimmed milk (0.6% fat) commercial cheese culture (culture containing LYOFAST MOS 062 E Lactococcus lactis ssp. lactis and Streptococcus thermophilus) and

EPS producing culture (culture consisting of EPS producing strain of DELVO-ADD 100F DIRECT SET Streptococcus thermophilus), Cheese sample (1% + 1%) (C1) made with the combination of skimmed milk (3.1% fat) commercial cheese culture (culture containing LYOFAST MOS 062 E Lactococcus lactis ssp. lactis and Streptococcus thermophilus) and EPS producing culture (culture consisting of EPS producing strain of DELVO-ADD 100F DIRECT SET Streptococcus thermophilus), Cheese sample (D1) made with skim milk (0.6% fat) using an EPS-only culture (culture consisting of an EPS-producing strain of DELVO-ADD 100F DIRECT SET Streptococcus thermophilus), Cheese sample (D2) made with whole milk (3.1% fat) using EPS-only culture (culture consisting of an EPS-producing strain of DELVO-ADD 100F DIRECT SET Streptococcus thermophilus).

Preparation and activation of starter culture

Reconstituted milk with 12% dry matter from skimmed milk powder was transferred to test tubes in 20 ml and heat treatment was applied in an autoclave at 90°C for 30 minutes. Lyophilized starter cultures were inoculated (at the level of 2%) into the cooled milk and incubated at 30–37Co until coagulated. These inoculation and incubation procedures were repeated two more times and the cultures were activated. Then, only LYOFAST MOS 062 E (2%) to one tube, only DELVO-ADD 100F DIRECT SET (2%) to another tube, and LYOFAST MOS 062 E and DELVO-ADD 100F DIRECT SET (1%+1%) to the last tube were inoculated in equal proportions. In this way, it is ensured that especially combined cultures adapt to each other. Activated cultures were inoculated at a rate of 2% separately into heat-treated (30 min at 90°C) reconstituted milk in 1-liter flasks and then turned into operating cultures to be used in production after incubation (Figure 1).

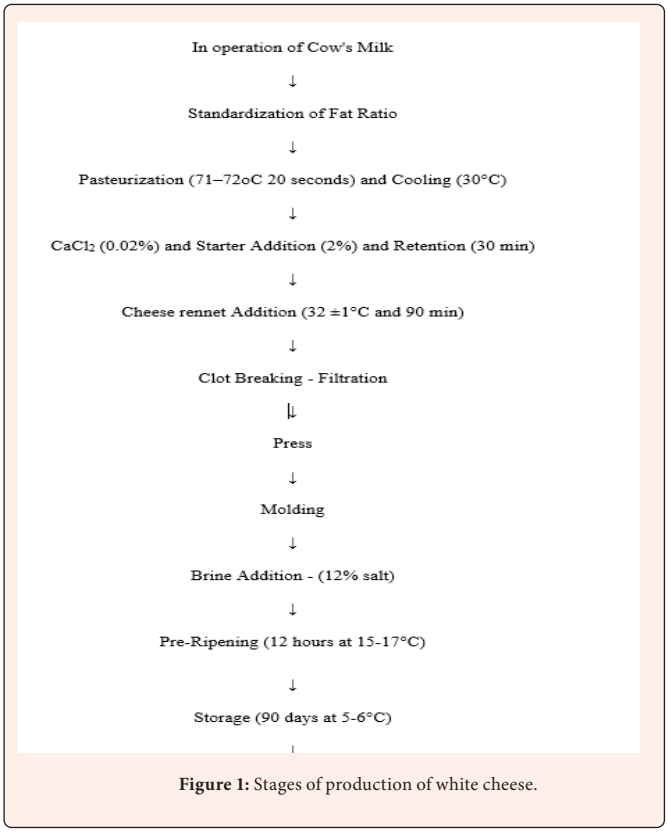

Figure 1: Stages of production of white cheese.

Physiochemical analyses

The pH values of the cheese samples were measured using a benchtop pH meter (Hanna-Instrument pH microprocessor pH211, Romania). The electrode of the pH meter is immersed in different points of the cheese molds and the average of the values read is taken, % acidity values of cheeses were determined by titration method according to the methods specified by [18]. The amount of dry matter, gravimetric method; the amount of fat was determined as % fat by the Van gulik method using a Gerber cheese butyrometer [19]. The water ratio in the fat-free cheese mass was found by dividing the water ratio by the fat-free cheese mass ratio; salt contents were determined by the Mohr method [18]. The ash amount of the cheese samples was determined by the method given by [19]. The total nitrogen content of the cheese samples was determined by the Kjeldahl method using the Kjeltec nitrogen determination device. The value found was multiplied by a factor of 6.38 and expressed as % protein [18]. For SCA analysis, 20 g of homogenized white cheese samples were weighed into polyethylene stomacher bags and 100 ml of distilled water (40°C) was added to them. The mixture is homogenized for 15 minutes. Then it was centrifuged (Nüve NF800R) for 30 minutes at 5°C and 5000 rpm and the oil was removed by filtration through glass wool. Some of the obtained filtrate was stored at -20°C to be preserved until the analysis time [20]. Nitrogenous substance determination was made via Kjeldahl method by taking 5 ml from the other part [21,18]. For this purpose, 25 ml of filtrate obtained from water-soluble nitrogen determination and 25 ml of 24% trichloroacetic acid (TCA) solution were mixed in a 1:1 ratio and kept at room temperature for 2 hours and then filtered using Whatman grade 42 filter paper. Nitrogenous substances were determined via Kjeldahl method by taking 25 ml of the obtained filtrate [18].

Statistical analyses

The experimental design of this study was created as follows: Two raw materials (fat and skim milk) x 4 cultures (without added culture, commercial cheese culture, combination of commercial cheese culture (1:1) + EPS producing culture and cultured cheeses producing EPS) x 5 storage times (1, 15, 30, 60 and 90 days) x 2 repetitions. Statistical evaluations of the research findings were carried out using the SPSS 18.00 package program. One-way analysis of variance and Tukey's multiple comparison test were used to compare the parameters of cultures (groups). Analysis of variance was applied in repeated measurements to evaluate the change according to days in each group.

Discussion and Conclusion

PH values of starter cultures

In the study, LYOFAST MOS 062 E (culture containing Lc lactis ssp Lactis-S thermophilus) obtained from Mayasan as commercial cheese starter culture, and DELVO-ADD 100F DIRECT SET (culture consisting of EPS producing strain of S. thermophilus) obtained from IMCD as EPS producing starter culture were used. The pH development values made to determine the activation of starter cultures used in cheese production are summarized in (Table 1). pH values of different cultures were measured at different time intervals in a 24-hour period, and it was determined that the pH decreased in all cultures.

Table 1: pH development values of the cultures used in the study.

Duration (Hours)	pH		
	LYOFAST (MOS 062 E)	DELVO ADD (100 F)	MOS 062 E+100F (1/1)
0	6,40±0,02	6,41±0,02	6,39±0,01
3	4,60±0,04	4,66±0,02	4,72±0,01
6	4,59±0,01	4,60±0,01	4,56±0,01
9	4,58±0,00	4,57±0,00	4,54±0,01
12	4,58±0,00	4,52±0,01	4,53±0,02
24	4,53±0,00	4,39±0,01	4,50±0,00

Cheese Yield Values

When the yield values of cheese varieties are compared; It was determined that the yield values of cheeses made with skimmed milk with or without culture added were higher than those made with skim milk ($p>0.05$). In cheeses made with eps culture, it was found that yield increased even at a low rate ($p>0.05$). Under normal circumstances; considering the water retention capacity of EPS, an increase in yield is expected. It is thought that the lower than expected increase in yield in the study may be

due to the low EPS production in the first few hours, depending on the characteristics and environmental conditions of the EPS producing cultures. In similarly qualified studies [22], reported that cultural differences did not make a significant difference in yield in the first place.

Physicochemical analysis results

The pH values obtained in cheese samples are given in (Table 2). In general, pH values decreased during maturation in all cheese groups (p>0.05). It is thought that the differences seen in the initial pH values between groups may be due to the lack of culture in negative control groups and the use of different starter cultures in other groups [23]. also found a decrease in pH values in general in their study investigating the effects of different starter cultures and maturation in white cheese.

Table 2: PH values of cheese samples during storage.

Cheese Groups		Storage Time					ÖD (Storage Duration)
		Day 1	Day 15	Day 30	Day 60	Day 90	
Low-fat	A1	$6,35^{fA}$±0,03	$6,11d^{BC}$±0,01	$5,96^{cC}$±0,01	$5,88^{eC}$±0,01	$5,84^{dC}$±0,01	p<0,05
	B1	$5,43^{cd}$±0,02	5.30 b±0.03	5.32 b±0.03	5.07b±0.02	5.21b±0.01	ns
	C1	5.34 b±0.03	5.31 b±0.01	5.32 b±0.01	$5,32^{cd}$±0,01	$5,28^{bc}$±0,02	ns
	D1	$5,50^{d}$±0,01	$5,38^{c}$±0,00	5.38 b±0.01	$5,37^{d}$±0,01	$5,32^{c}$±0,01	ns
fatty	A2	$6,27^{eA}$±0,00	$6,13^{dBA}$±0,01	$5,99^{cB}$±0,01	$5,98^{fB}$±0,01	$5,90^{dB}$±0,01	p<0,05
	B2	5.14 a±0.00	5.17 a±0.01	$5,16^{a}$±0,00	$5,10^{a}$±0,01	5.04a±0.03	ns
	C2	$5,44^{d}$±0,03	5.34bc±0.03	5.33 b±0.03	$5,33^{cd}$±0,00	$5,28^{bc}$±0,01	ns
	D2	$5,36^{bc}$±0,01	5.31 b±0.01	5.31 b±0.02	$5,30^{bc}$±0,00	$5,28^{bc}$±0,04	ns
ÖD (Groups according to)		p<0,01	p<0,01	p<0,01	p<0,01	p<0,01	

[24], on the other hand, stated that, contrary to these studies, pH values of all cheese samples increased at the end of 90 days of maturation in white cheeses made from reduced fat milk. stated that the pH of cheese samples decreased. In another study, it was reported that the pH values of Karish cheese made with EPS producing cultures generally decreased during the maturation process [25]. While some researchers argue that starter cultures are effective on the change in pH values during the maturation process [26], some researchers argue that the pH change is not caused by the difference in the starter [27]. The titration acidity values in cheese samples determined during storage are given in (Table 3) in terms of lactic acid (%). At the end of 90 days of storage, acidity values increased in all samples (p>0.05). The titration acidity values determined in cheese samples made by adding commercial culture were found to be higher than the cheeses made with EPS added culture. This can be caused by the fact that lactotoxes can develop in low temperature and salt concentration environments and have the ability to produce high levels of acid from lactose [2].

Table 3: Titration acidity values of cheese samples during storage (lactic acid %).

Cheese Groups		Storage Time					(Storing Time)
		Day 1	Day 15	Day 30	Day 60	Day 90	
Low-fat	A1	$0,108^{a}$±0,001	$0,115^{a}$±0,003	$0,122^{a}$±0,004	$0,145^{b}$±0,005	$0,149^{b}$±0,001	ns
	B1	$0,397^{e}$±0,010	$0,395^{c}$±0,004	$0,420^{d}$±0,001	$0,422^{f}$±0,003	$0,422^{e}$±0,003	ns
	C1	$0,324^{d}$±0,00	$0,364^{c}$±0,004	$0,365^{c}$±0,004	$0,333^{d}$±0,005	$0,344^{d}$±0,006	ns
	D1	$0,257^{b}$±0,01	$0,288^{b}$±0,011	$0,287^{b}$±0,002	$0,285^{c}$±0,001	$0,287^{c}$±0,005	ns
fatty	A2	$0,104^{a}$±0,001	$0,112^{a}$±0,006	$0,117^{a}$±0,002	$0,118^{a}$±0,004	$0,122^{a}$±0,003	ns
	B2	$0,394^{e}$±0,008	$0,433^{d}$±0,004	$0,447^{e}$±0,004	$0,488^{g}$±0,006	$0,489^{f}$±0,004	ns
	C2	$0,292^{c}$±0,012	$0,352^{b}$±0,046	$0,354^{c}$±0,005	$0,360^{d}$±0,001	$0,364^{d}$±0,004	ns
	D2	$0,394^{e}$±0,006	$0,394^{c}$±0,005	$0,410^{d}$±0,001	$0,402^{e}$±0,006	$0,408^{e}$±0,004	ns
ÖD (Groups according to)		p<0,01	p<0,01	p<0,01	p<0,01	p<0,01	

It is known that as a result of the fermentation that occurs during the ripening of semi-soft and soft cheeses, lactose turns into lactic acid, this conversion is fast initially, and then slows down to the degree of cessation due to the decrease in lactose [28,29]. In the study, it is thought that the fluctuations in titration acidity during maturation may be caused by the formation of alkaline substances in the environment due to proteolysis and the inclusion of them in the composition and the differences in dry matter [22]. The amount of dry matter (%) determined during maturation in cheese samples is given in (Table 4). On the first day, % dry matter values varied between 38.45% and 39.16% in non-fat cheeses and between 39.76% and 41.61% in fatty cheeses. At the end of storage, it showed a change between 36.93-38.61% in non-fat cheeses and 37.35-42.02% in fatty cheeses. It is thought that the difference in the initial dry matter values of the cheeses may be due to the different effects of different cultures on the formation of cheese curd, as well as the production of cheese with fat and skim milk (p<0.01).

Table 4: Drying matter of cheese samples during storage (%).

Cheese Groups		Storage Time					(Storing Time)
		Day 1	Day 15	Day 30	Day 60	Day 90	
Low-fat	A1	$39,16^{b}$±0,00	38.85b±0.35	38.91ab±0.08	39.12b±0.05	$39,61^{d}$±0,18	ns
	B1	38.55a±0.0	38.07a±0.71	38.24a±0.46	38.04b±0.04	38.61b±0.24	ns
	C1	$38,45^{a}$±0,00	38.13a±0.03	$38,34^{a}$±0,008	38.12b±0.18	37.92ab±0.01	ns
	D1	38.80aA±0.06	38.48bAB±0.09	38.81bAB±0.01	$38,44^{ab}$±0,04	37.73aB±0.09	p<0,05
fatty	A2	$41,61^{de}$±0,00	41.29d±0.52	$41,61^{d}$±0,07	$41,71^{e}$±0,22	$42,02^{e}$±0,31	ns
	B2	40.91dA±0.05	$38,14^{aB}$±0,07	$38,09^{aB}$±0,00	37.81aB±0.12	37.35aB±0.21	p<0,05
	C2	$39,76^{c}$±0,08	38.35a±0.14	38.72b±0.27	$38,75^{cd}$±0,01	38.67b±0.11	ns
	D2	$40,31^{d}$±0,00	$39,84^{c}$±0,18	$39,72^{c}$±0,27	$39,91^{d}$±0,01	$39,21^{d}$±0,06	ns
GALL(by groups)		p<0,01	p<0,01	p<0,01	p<0,01	p<0,01	

In D1 and D2 samples, which were prepared only with EPS producing culture, a decrease in dry matter was detected during storage. During the ripening period of white cheese made with pasteurized milk [21], determined the dry matter contents of cheese samples made with fatty and commercial cheese culture as 39.80% and 41.12% on the 1st and 90th days, respectively. In this study, it was observed that the changes in the dry matter content of the cheeses made with skimmed and fat milk, without added culture and commercially cultured during the maturation period were proportionally compatible with the values found by [21]. The dry matter ratio in cheese obtained from skimmed milk was found to be statistically significantly lower than the dry matter ratio of cheese made with EPS producing culture (p<0.01). EPS produced by LAB increases the water holding capacity of cheese [30,1]. Dry matter ratios decreased during the 90-day maturation period in the experimental groups obtained with EPS producing culture. This can be explained in such a way that the resulting EPS holds the cheese juice more, which reduces the rate of drying. In a study conducted in support of this, it was determined that the water rate in Karish cheese made with EPS-producing culture increased by 2% during storage compared to the control group cheese made with culture that did not produce EPS [1]. The fat ratios obtained in all experimental cheese groups are given in (Table 5). In general, no significant difference was found between the 1st and 90th days in the evaluation of the fat ratios obtained in all cheese groups according to the cultures and maturation period (p>0.05). When the cheeses made using fat and skim milk were compared, a statistically significant difference was found (p<0.01).

Table 5: Fat amounts of cheese samples during storage (%).

Cheese Groups		Storage Time					ÖD (Storage Time)
		Day 1	Day 15	Day 30	Day 60	Day 90	
Low-fat	A1	4,0^{b}±0,00	4,0^{b}±0,00	4,0^{b}±0,00	4,7^{b}±0,40	4,5^{b}±0,00	ns
	B1	4,0^{b}±0,00	4,0^{b}±0,00	4,7^{b}±0,40	4,5^{b}±0,00	4,5^{b}±0,00	ns
	C1	4,0^{b}±0,00	4,0^{b}±0,00	4,7^{b}±0,40	4,7^{b}±0,40	4,7^{b}±0,40	ns
	D1	4,0^{b}±0,00	4,0^{b}±0,00	4,0^{b}±0,00	4,7^{b}±0,40	4,0^{b}±0,00	ns
fatty	A2	18,0^{a}±0,00	18.5a±0.70	18.5a±0.40	18,0^{a}±0,40	18.5a±0.70	ns
	B2	18,0^{a}±0,70	18,0^{a}±0,00	18,0^{a}±0,70	17.5a±0.70	17.5a±0.70	ns
	C2	18.5a±0.70	18,0^{a}±0,00	18,0^{a}±0,00	17,0^{a}±0,00	18,0^{a}±0,70	ns
	D2	18,0^{a}±0,00	18,0^{a}±0,70	18,0^{a}±0,00	18,0^{a}±0,00	18,0^{a}±0,40	ns
ÖD (Groups according to)		p<0,01	p<0,01	p<0,01	p<0,01	p<0,01	

[31]. reported in a study that the fat ratios in Penela cheese were not significantly affected by the storage time and the amount of EPS formed. determined that the amount of fat (0.5%) did not change during storage in low-fat Karish cheese they made with EPS producing and non-EPS producing cultures. The water content in the fat-free cheese mass % or water fat free (WFF) content in the fat-free cheese mass determined during maturation in cheese samples are given in (Table 6). On the first day, WFF ranged between 63.38% and 64.11% in non-fat cheeses and between 71.20 and 73.91% in fatty cheeses. When WFF changes in fatty and non-fat cheeses were compared, it was found to be statistically significant (p<0.05). Likewise, on the first day, WFF rates of cultured cheeses were found to be higher in fatty and fat cheeses than in non-cultured ones (p<0,05). Cheeses produced with commercial culture had a higher WFF ratio than non-cultured cheeses, while samples of cheese produced with EPS producing culture had a higher WFF content than cheese samples produced with and without added culture (p<0,01).During maturation, it was determined that the proportion of cheeses produced with eps producing culture decreased in drying. As a result of the increase in water contents, the increase in WFF rates was considered normal. Many previous studies on this subject support these results [32,30,12,33,1].

Table 6: Water ratios of cheese samples in fat cheese mass during storage (%).

Cheese Groups		Storage Time					(Storing Time)
		Day 1	Day 15	Day 30	Day 60	Day 90	
Low-fat	A1	63,38^{a}±0,00	63.69a±0.10	63.63a±0.47	63.74a±0.57	63,23^{a}±0,30	ns
	B1	64.01a±0.01	64.51a±0.77	64.67a±0.41	64.77a±0.07	64.28a±0.22	ns
	C1	64,11^{a}±0,00	64.44a±0.01	64.56a±0.15	64.79a±0.22	65.06a±0.23	ns
	D1	63.75a±0.02	64.08a±0.02	63.73a±0.01	64.46a±0.15	64.86a±0.12	ns
fatty	A2	71,20^{c}±0,00	72.03b±1.42	71.64b±0.26	71.08b±0.07	71.14b±1.14	ns
	B2	72,06^{cd}±0,65	75,43^{cd}±0,09	75.05c±0.75	75,38^{c}±0,59	75,93^{c}±1,03	ns
	C2	73.91±0.89	75,18^{d}±0,19	74,73^{c}±0,37	74,24^{c}±0,02	74.79c±0.61	ns
	D2	72,79^{d}±0,20	73.81bc±0.50	73,51^{c}±0,37	73,28^{c}±0,02	74,13^{c}±0,47	ns
ÖD(Groups according to)		p<0,01	p<0,01	p<0,01	p<0,01	p<0,01	

The amounts of salt detected in cheese samples during maturation are given in (Table 7). On the first day, salt contents were determined between 3.21% and 3.62% in fat cheeses and between 3.42% and 3.85% in fatty cheeses. It has been observed that the amount of salt varies in relation to the fat content. Salt rates increased in all groups during maturation. It was determined that the starter culture difference did not make a significant difference in salt ratios. The salt ratio in fresh cheese was determined as 4% by [34,35], and as 4.64% by [36]. in ripened cheese [35,37], found 3.34% [35], between 5.80% and 6.10% [38], 8.75% and [39] 4.86% - 5.42%. In this study, the increase in salt content of test cheese samples during ripening was also parallel to other studies [1]. found that the salt content of Karish cheese made with EPS-producing cultures increased from 5.00% to 5.80% in cheese made with cultures that did not produce EPS, and from 5.70% to 6.50% in cheese made with EPS-producing cultures during maturation.

Table 7: Amounts of salt in cheese samples during storage (%).

Cheese Groups		Storage Time					(Storing Time)
		Day 1	Day 15	Day 30	Day 60	Day 90	
Low-fat	A1	3,61^{cA}±0,01	4.08aB±0.04	4,84^{bC}±0,032	5,01^{cC}±0,008	5,05^{dC}±0,004	p<0,05
	B1	3.21aA±0.06	4,81^{cB}±0,29	4,92^{cB}±0,035	4,97^{cB}±0,002	4,93^{cB}±0,053	p<0,05
	C1	3,43^{b}±0,001	4.51b±0.157	4,69^{b}±0,006	4,72^{b}±0,018	4,74^{b}±0,024	ns
	D1	3,62^{cA}±0,06	4,07^{aB}±0,014	4.73bBC±0.13	4,90^{cC}±0,001	4,90^{cC}±0,010	p<0,05
fatty	A2	3.42bA±0.02	4,61^{bB}±0,02	4,68^{bB}±0,010	4,69^{bB}±0,009	4,67^{bB}±0,004	p<0,01
	B2	3,85^{cA}±0,03	4,92^{cA}±0,04	4,23^{aB}±0,046	4,35^{aB}±0,052	4,32^{aB}±0,006	p<0,05
	C2	3,82^{dA}±0,01	4,17^{aB}±0,07	4,29^{aB}±0,011	4,34^{aBC}±0,03	4,39^{aC}±0,003	p<0,05
	D2	3.42bA±0.04	4,17^{aB}±0,01	4.54bBC±0.16	4,69^{bBC}±0,07	4,75^{bC}±0,006	p<0,05
ÖD(By Group)		p<0,01	p<0,01	p<0,01	p<0,01	p<0,01	

Ash rates detected in cheese samples during ripening are given in (Table 8). The ash contents of cheese samples ranged from 4.51% to 5.58% in fat cheeses and 3.70% to 4.53% in fatty cheeses on day 1 [24]. reported that the amount of ash increased in all groups during the 90-day ripening period in the white cheeses made with the control group and EPS producing culture. In another study, it was determined that the amount of ash in Karish cheese made with EPS-producing and non-EPS-producing cultures increased in both cheese types. This case revealed that EPS has no effect on the ash content in particular. Likewise [23], reported that the amount of ash varies during maturation in White cheeses made with different starter cultures.

Table 8: Amounts of ash in cheese samples during storage (%).

Cheese Groups		Storage Time					Öd (Storing Time)
		Day 1	Day 15	Day 30	Day 60	Day 90	
Low-fat	A1	5,58^{c}±0,059	5,75^{d}±0,326	5,61^{c}±0,333	5,63^{b}±0,114	5,56^{d}±0,103	ns
	B1	4,98^{c}±0,157	5,31^{cd}±0,080	5,27^{bc}±0,154	5,34^{ab}±0,091	5,38^{cd}±0,022	ns
	C1	4,61^{b}±0,298	5,17^{bc}±0,103	5,20^{c}±0,015	5,04^{ab}±0,090	5,20^{bc}±0,142	ns
	D1	4.51b±0.182	5,30^{cd}±0,013	5,41^{bc}±0,071	5,33^{ab}±0,103	5,37^{cd}±0,010	ns
fatty	A2	4,46^{b}±0,355	5,03^{ab}±0,108	4,83^{a}±0,200	4,75^{a}±0,032	4,73^{a}±0,025	ns
	B2	4,03^{b}±0,117	4,47^{a}±0,325	4,70^{a}±0,153	4,97^{ab}±0,026	4,99^{ab}±0,018	ns
	C2	4.53b±0.214	4,73^{ab}±0,103	4,73^{a}±0,042	4,87^{a}±0,033	4,86^{a}±0,075	ns
	D2	3,70^{aA}±0,027	4.58aAB±0.083	4.77aB±0.750	4,99^{abB}±0,018	4,95^{abB}±0,026	p<0,05
according to ÖD(Gr.)		p<0,01	p<0,01	p<0,01	p<0,01	p<0,01	

In this study, the total nitrogen contents detected during maturation in cheese samples are summarized in (Table 9). The total amount of nitrogen ranged from 2.92% to 3.48% in fat cheeses on the first day and 2.08% to 2.36% in fatty cheeses (p<0,01). On the 90th day, there was a decrease in all groups compared to the first day (p<0.05 for the D1 group).

Table 9: Total nitrogen amounts in cheese samples during storage (%).

Cheese Groups		Storage Time					ÖD (Storing Time)
		Day 15	Day 30	Day 60	Day 90		
Low-fat	A1	3,48^{c}±0,004	3,35^{c}±0,006	3,10^{d}±0,018	2,92^{c}±0,054	2,97^{f}±0,009	ns
	B1	3,43^{c}±0,013	3,49^{f}±0,022	3,16^{d}±0,049	2,72^{d}±0,044	2,81^{c}±0,060	ns
	C1	2,96^{d}±0,004	2,82^{d}±0,054	2,57^{c}±0,021	2,57^{c}±0,017	2,68^{d}±0,006	ns
	D1	2,92^{dA}±0,006	2,85^{dA}±0,004	2,69^{cB}±0,004	2,62^{cdB}±0,002	2,63^{dB}±0,004	p<0,01
-fatty	A2	2,36^{c}±0,003	2,14^{b}±0,004	2,04^{ab}±0,083	2,09^{b}±0,012	2,12^{c}±0,004	ns
	B2	2,35^{c}±0,004	2,30^{c}±0,011	1,88^{a}±0,034	1,86^{a}±0,061	1,90^{a}±0,009	ns
	C2	2,21^{b}±0,003	2,23^{c}±0,005	2,09^{b}±0,006	1,97^{ab}±0,011	2,00^{b}±0,020	ns
	D2	2,08^{a}±0,0042	1,97^{a}±0,018	1,92^{ab}±0,067	1,97^{ab}±0,011	1,97^{ab}±0,006	ns
ÖD (Groups according to)		p<0,01	p<0,01	p<0,01	p<0,01	p<0,01	

Determined that the total nitrogen amount was 3.09%-2.19% on the 1st and 90th days, respectively. The results obtained in this study were similar to the results of. It is also stated by some researchers that there is a decrease in the amount of total nitrogen during maturation [40,41]. It was observed that the total nitrogen amount was low in cheese with high fat content. It is thought that this may be due to an inverse relationship between the fat content of the milk to be processed into the cheese and the total nitrogen content of the cheese [24]. In this study, the % protein values determined in all cheese groups are given in (Table 10).

Table 10: Amounts of protein in cheese samples during storage (%).

Cheese Groups		Storage Time					ÖD (Storing Time)
		Day 1	Day 15	Day 30	Day 60	Day 90	
Low-fat	A1	22,20^{e}±0,02	21,37^{e}±0,03	19,77^{d}±0,11	18,62^{e}±0,34	18,94^{f}±0,05	ns
	B1	21,88^{e}±0,08	22,26^{f}±0,14	20,16^{d}±0,31	17,35^{d}±0,28	17,92^{e}±0,38	ns
	C1	18,88^{d}±0,02	17,99^{d}±0,34	16,39^{c}±0,13	16,39^{c}±0,11	17,09^{d}±0,04	ns
	D1	18,6^{dA}±0,03	18,1^{dA}±0,02	17,16^{cB}±0,02	16,71^{cC}±0,01	16,77^{dBC}±0,02	p<0,01
fatty	A2	15,05^{c}±0,01	13.65b±0.02	13.01a±0.53	13.33b±0.07	13,52c±0,02	ns
	B2	14,99^{c}±0,02	14,67^{c}±0,07	11.99ab±0.21	11.86a±0.38	12.12a±0.05	ns
	C2	14.09b±0.01	14,22^{c}±0,48	13.33b±0.03	12.56ab±0.06	12.76b±0.12	ns
	D2	13.27a±0.26	12.56a±0.11	12.24ab±0.42	12.56ab±0.07	12.56ab±0.03	ns
ÖD(Groups according to)		p<0,01	p<0,01	p<0,01	p<0,01	p<0,01	

On the first day, protein content in fat cheeses ranged from 18.62% to 22.20% and in fatty cheeses from 13.27% to 15.05%. The difference between fatty and fat cheeses was found to be statistically significant (p<0,01). In eps-producing cultured cheese samples, protein contents were found to be less than non-cultured and commercially cultured cheese samples (p<0,01). It is thought that this decrease in protein amounts may be due to the decomposition of the milk protein casein by enzymes and the passage of the decomposition products to the brine [22]. It has also been supported by various researchers that the total protein amount in brined white cheese decreases during ripening [42-45]. determined that the protein content of white cheese produced by using different starter cultures, although it varies according to the starter cultures, decreased during maturation in some samples and increased in some cheese samples. However, they emphasized that this difference is not due to starter culture but may be due to salt absorption and/or the passage of certain soluble substances into water. The amounts of water-soluble nitrogen (WSN) (%) determined during maturation in cheese samples are given in (Table 11). In this study, the lowest and highest WSN amounts were 0.203% and 0.079% and 0.189% respectively on the first day for cheese groups produced from skimmed and fatty milk. While the amount of WSN determined during the ripening period was low in cheeses produced without culture additives, an increase was observed in cheeses produced with commercial and EPS producing cultures at similar rates [46]. found that the amount of nitrogen dissolved in water increased during the 90-day maturation of cheese samples. In addition [47], determined that WSN amounts of cheeses increased from 0.21% to 0.36% during the storage period (105 days) in traditional feta cheese. reported an increase in the amount of WSN during storage at different rates according to the starter culture type. The values obtained in this research are similar to previous researches.

Table 11: Amounts of water-soluble nitrogen (%) specified in cheese samples during storage.

Cheese Groups		Storage Time					ÖD (Storing Time)
		Day 1	Day 15	Day 30	Day 60	Day 90	
Low-fat	A1	0,084^{ab}±0,004	0,105^{a}±0,006	0,119^{a}±0,002	0,188^{a}±0,020	0,206^{a}±0,006	ns
	B1	0,167^{d}±0,002	0,291^{c}±0,008	0,311^{c}±0,001	0,345 ^{b}±0,037	0,376^{b}±0,008	ns
	C1	0,203^{f}±0,010	0,348^{c}±0,009	0,392^{d}±0,010	0,458^{c}±0,020	0,639^{b}±0,237	ns
	D1	0,112^{c}±0,006	0,192^{b}±0,006	0,188^{b}±0,004	0,226^{a}±0,027	0,241^{a}±0,006	ns
fatty	A2	0,079^{a}±0,004	0,106^{a}±0,006	0,106^{a}±0,008	0,179^{a}±0,040	0,213^{a}±0,008	ns
	B2	0,179^{de}±0,002	0,314^{c}±0,040	0,392^{d}±0,039	0,420^{b}±0,003	0,426^{b}±0,006	ns
	C2	0,102^{bcA}±0,003	0,174^{bA}±0,006	0,384^{dB}±0,019	0,355^{bB}±0,022	0,369^{bB}±0,003	p<0,05
	D2	0,189^{c}±0,006	0,200^{b}±0,011	0,180^{b}±0,006	0,235^{a}±0,006	0,244^{a}±0,006	ns
ÖD(Group-according to the		p<0,01	p<0,01	p<0,01	p<0,01	p<0,01	

12% TCA-soluble nitrogen provides information about medium small peptides, amino acids and amines that occur during proteolysis, and smaller nitrogenous compounds such as urea and ammonia [18]. In this study, the amount of nitrogen dissolved in 12% TCA (%) determined in all cheese groups during maturation is given in (Table 12).

Table 12: TE amounts determined in cheese samples during storage (%).

Cheese Groups		Storage Time					ÖD (Storing Time)
		Day 1	Day 15	Day 30	Day 60	Day 90	
Low-fat	A1	0,058^{a}±0,010	0,073^{a}±0,006	0,091^{a}±0,004	0,112^{a}±0,012	0,145^{b}±0,008	ns
	B1	0,125^{b}±0,006	0,237^{c}±0,003	0,250^{d}±0,005	0,258^{d}±0,008	0,277^{e}±0,009	ns
	C1	0,105^{b}±0,016	0,122^{b}±0,006	0,172^{bc}±0,010	0,171^{c}±0,030	0,186^{cd}±0,006	ns
	D1	0,121^{b}±0,010	0,118^{b}±0,002	0,167^{bc}±0,008	0,175^{c}±0,008	0,187^{d}±0,006	ns
fatty	A2	0,058^{a}±0,013	0,064^{a}±0,006	0,067^{a}±0,005	0,094^{a}±0,006	0,104^{a}±0,008	ns
	B2	0,102^{b}±0,007	0,136^{b}±0,010	0,148^{b}±0,006	0,163^{bc}±0,006	0,173^{c}±0,010	ns
	C2	0,121^{b}±0,004	0,132^{b}±0,001	0,178^{c}±0,011	0,196^{c}±0,015	0,203^{d}±0,004	ns
	D2	0,097^{b}±0,015	0,131^{b}±0,008	0,165^{bc}±0,006	0,164^{b}±0,014	0,177^{d}±0,006	ns
ÖD(Groups according to)		p<0,01	p<0,01	p<0,01	p<0,01	p<0,01	

On the 1st day of maturation, the amount of TCA varied between %0.058 and %0.125 in non-fat samples and between %0.058 and % 0.121 in fatty cheeses (p>0.05). During storage, TCA amounts increased in all samples and on the 90th day the ratios were determined as 0.145% to 0.277% in fat cheeses and 0.104% to 0.203% in oily cheeses. The amount of TCA is compared in terms of cultured and culture-free cheese groups; in groups where fatty or skimmed milks are used, culture has been significantly lower during all maturation of cheeses produced without additions compared to cultured groups (p<0,01). This is due to the effect of placenta, the proteolytic enzyme naturally found in milk, on the increased amounts of TCA in cheeses produced without culture additions. Since this enzyme is not easily affected by heat treatment, it breaks down proteins during maturation, increasing the amount of TCA. In addition, it is thought that the reason for the higher rate of cultured cheeses may be due to the use of culture. It has also been observed that different cultures exhibit similar differences in cheeses obtained from fat or skim milk [48]. in their study in which they examined the proteolysis formation during ripening in Ezine cheese, they determined that the amount of TCA increased during ripening in the samples. In the same way; Moatso et al. Same way [49]. found that the amount of TCA in Feta cheese increased during storage [50-52].

Conclusion and Recommendations

During storage, pH values decreased in all samples, but statistically insignificant (p>0.05). Titration acidity, on the other hand, increased overall in all samples during maturation (p>0.05). Amounts of dry matter decreased insignificantly in all cheeses at the end of 90 days of maturation, except for the cheese samples produced with fat milk and commercial culture, skimmed milk and cheese samples produced only with EPS producing culture (p>0.05). The amount of fat was found to be significantly higher (p<0.01) in cheeses made with whole milk, but low in those made with skim milk. However, the fat content in each group did not change significantly during the maturation process (p>0.05). The ratio of water in the fat-free cheese mass differed significantly between non-fat and fatty cheeses (p<0.01), but a slight increase was observed according to the maturation time (p>0.05). While the amount of salt in all samples varied significantly according to the groups in each period of storage (p<0.01), it was found to increase significantly during storage, except for the skim milk sample (p>0,05) and cheese made with commercial culture + EPS-producing culture (p<0.05). The amount of ash in cheese samples made with skim milk was significantly higher than those made with fat milk (p<0.01). There was no significant difference in the amount of ash in the cheese samples over time (p>0.05). In cheese samples made with EPS-producing cultures, the amount of ash was found to be lower than in those who did not produce EPS. During maturation, total nitrogen amounts decreased in all cheese samples. Total nitrogen amounts were found to be lower in cheeses obtained by

eps-producing culture than others (p<0,05). Protein content decreased significantly in general over the 90-day period. When comparing samples, it was found that the protein content of cheeses using EPS-producing culture was lower than the control samples (p<0,05). There has been an increase in the amount of WSN and the OEA. While there was no significant difference between commercial culture and EPS-producing cultures and cheeses, both parameters were found to be significantly lower in cheeses produced without culture additions (p<0,05). TCA amount and TCA in relation to OI showed a parallel course to the amount of nitrogen dissolved in water.

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