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Turkish artisanal Tulum cheese ripened in tripe: The importance of the milk type and changes with ripening

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ABSTRACT

Karin Kaymagi is a type of Tulum cheese traditionally produced from sheep milk and ripened in tripe following the addition of cream to the curd. In this study, it was traditionally produced by mixing certain proportions of raw sheep and cow milk (100:0, 75:25, 50:50, 25:75, and 0:100). The samples were allowed to ripen, and analyses were made for chemical, biochemical, electrophoretic, sensory, and textural properties at the 3rd, 30th, 60th, and 90th days of ripening. According to the results obtained, both cheese type and ripening time had significant effects on dry matter, fat, protein, salt, salt in dry matter, % lactic acid, pH, ripening rate, NPN ratio, and PPN ratio ($P < 0.05$). Only cheese type had a remarkable effect on salt in dry matter and fat in dry matter ($P < 0.05$). The highest lipolysis value was determined in the samples made of sheep milk only (KK2) ($P < 0.05$). Electrophoretic analysis showed that α_{S1}-casein and β-casein concentrations decreased until the end of ripening. Sensory analysis denoted that the KK2 sample was appreciated the most based on colour, appearance, structure, taste, and odor scores ($P < 0.05$).

Keywords: Tulum cheese, Karin Kaymagi cheese, ripening, sheep tripe, lipolysis, electrophoresis

INTRODUCTION

Although milk, owing to its rich content and high nutritional value, is a unique human food and a valuable breeding ground for microorganisms, it is a perishable food. It is possible to increase the shelf life of milk by processing it into dairy products and cheese, with around 2000 varieties known worldwide, a good alternative product that can be manufactured for this purpose (Tamime and Robinson, 1991). This diversity is because cheeses show a wide variety of tastes and textures depending on the difference in raw materials, the consumption habits of the nations, and even the local habits. Almost 200 traditional and modern cheese varieties are produced in Türkiye. Modernized living conditions have caused most of the traditional cheeses to be forgotten (Öründü and Tarakçı, 2021). A standard technique is not applied in the production of Tulum cheese in Turkey and it is produced with traditional methods in family companies and primitive dairy farms, usually between March and July. Goat and sheep milk is traditionally preferred in the production of Tulum cheeses; however, cow's milk is also used due to the increasing demand for Tulum cheese consumption recently. While traditional methods of Tulum cheese production are maintained, various new technological advantages have been incorporated into artisanal production over time (Demirci et al., 2021). One of the traditional cheeses with high nutritional value is Karin Kaymagi, a type of Tulum cheese. Karin Kaymagi Cheese, produced in Gümüshane, Erzurum, and Kars, Eastern provinces in Türkiye, is very rare in the market because it is traditionally produced in family businesses only to meet their needs. The name "Karin Kaymagi", meaning cream in the tripe, is derived from the fact that the cheese is ripened in either sheep or goat tripe for a long time after adding milk cream/butter. Although it is generally produced using sheep milk, cow milk is also used in times when sheep milk is scarce.

Karın Kaymagi Cheese is a kind of cheese similar to Tulum cheese, which has a very high-fat content, ripe, nutritious, unique in terms of its preparation, and preserved by pressing the tripe. Karın Kaymagi Cheese has a distinctive aroma owing to its high-fat content, unique production and ripening technique (Çakmakçı et al., 1995; Turgut et al., 2012). Despite all these exclusive features, studies on Karın Kaymagi Cheese are quite limited.

Therefore, considering the difficulty of access to sheep milk in recent years, this research aimed to determine the applicability of different combinations of sheep and cow's milk and their effect on the changes that occur during cheese ripening, especially lipolysis rate, casein fractionation and sensory properties.

MATERIALS AND METHODS

Materials

The raw sheep milk (dry matter 17.15 ± 0.06%, protein 4.76 ± 0.04%, fat 6.32 ± 0.12%, lactic acid 0.19 ± 0.002%, and pH 6.83 ± 0.04) and cow milk (dry matter 12.73±0.08%, fat 3.81 ± 0.02%, protein 3.33 ± 0.06%, lactic acid 0.14 ± 0.003%, and pH 6.56 ± 0.01) used in the research was obtained from the Karagöl Plateaus of Giresun. Cream produced from cow and sheep milk was obtained from local producers. A commercial liquid rennet (Rumeli Trade, İstanbul; 1 / 12,000 mcu/g) was used in milk coagulation. The sheep tripes used for packaging had been obtained from a butcher, cleaned thoroughly with hot water, and dried for a month. Finally, they were sewn into bags sized to hold approximately 1 kg of cheese before being filled with cheese.

Cheese production

Following filtration of the milk with a filter cloth, the raw milk from sheep and cows was mixed in certain proportions (KK1: 100% cow milk, KK2: 100% sheep milk, KK3: 75% cow milk + 25% sheep milk, KK4: 25% cow milk + 75% sheep milk and KK5: 50% cow milk + 50% sheep milk) and processed directly into the cheese at the temperature it was milked from an animal. The curd obtained by adding commercial rennet to the milk was cut in the size of chickpeas and the whey that released was filtered out with a cheese bag. The curd was pressed with a weight of about 10 kilograms for three days to remove the remaining whey completely. After crumbling, salt (3%, w/w) and cream (5%, w/w) were added to the curd. After thoroughly mixing, it was filled into a previously prepared tripe leaving no gaps.

Chemical analysis

Moisture determination of cheeses was analyzed by gravimetric method (IDF, 1982). The samples' salt and fat content were determined by the Mohr and Gerber methods, respectively. % Lactic acid was determined as percent lactic acid using the AOAC method (AOAC, 1996). The pH value was directly determined with a pH meter (Ohaus, Starter 3100) from a cheese macerate prepared in distilled water (1:1). Total nitrogen (TN) in cheese was determined by the Kjeldahl method (IDF, 1993) with a UDK-149 (VELP Scientifica, Usmate, Italy) distillation unit. Total protein (%) was calculated by multiplying the nitrogen percentage by a factor of 6.38.

Lipolysis determination

Lipolysis was determined using the Bureau of Dairy Industries (BDI) method and expressed as Acid Grade Value (ADV). BDI reagent (30 g triton X-100 and 70 g sodium tetraphosphate in 1 L of distilled water) was added to 10 g of sample in a lipolysis butyrometer. To extract the oil, it was placed in a boiling water bath. Following 1 minute of centrifugation, aqueous methanol was added until the oil reached the butyrometer's neck and it was centrifuged for another minute. The oil fraction was then transferred to a 50 ml flask and weighed and 5 ml of solvent (4: 1; petroleum ether: n-propanol) was added to the flask. Finally, it was titrated with 0.02N KOH, and total free fatty acid was calculated (Salji and Kroger, 1981).

Proteolytic analysis

A water soluble nitrogen fraction (WSN), a 5% phosphotungstic acid-soluble nitrogen fraction (PTA-SN) and a 12% trichloroacetic acid-soluble nitrogen fraction

(TCA-SN) were determined according to the Kjeldahl method outlined in IDF (1993). All nitrogen fractions were calculated as a percentage of total nitrogen.

Electrophoretic analysis

Electrophoresis analysis of cheese samples was applied using the method given by Creamer (1991) with minor modifications by Tarakci et al. (2004). A discontinuous gel system was used in this analysis. Protein bands were colored with a dye solution (1 g Coomassie Brilliant Blue R-250: 500 ml isopropanol: 200 ml glacial acetic acid in 2 liters) and excess dye was removed with a decolorization solution (200 ml isopropanol: 0.2 L acetic acid in 2 liters). Dye-Colored gels were scanned with a scanner (Epson L3151; Seiko Epson Corp., Nagano, Japan) and transferred to the computer. The intensities of the α_{s1}-casein and β-casein bands were appointed using densitometry software (Total Lab Limited, Newcastle, UK).

Textural analysis

Textural values of cheese samples were determined with TA-XT2 (Stable Micro Systems Ltd., Surrey, England). Before the analysis, the cheeses were brought to 20 ± 2 °C and cut into cubes (20x20x20 mm) with a cutting knife. Six parameters were examined in texture profile analysis. A total of six parameters, including hardness (g), adhesiveness (g/sec), cohesiveness, chewiness (g), springiness and gumminess (g) of the samples, were determined. Analysis was performed using a P/36 aluminum cylinder probe (36 mm diameter, AACC) and the load cell of 25 kg with the following conditions: test speed of 0.4 mm/sec, initial test speed of 1.0 mm/sec, final test speed of 0.4 mm/sec, 40% pressure, and retention time of 5 seconds (Everard et al., 2006; Kahyaoğlu, 2002). The analysis was performed in quadruplicate.

Sensory analysis

Sensory analysis was conducted by eight panelists selected from Ordu University faculty members and graduate students. The necessary preliminary training was given to the panelists before the evaluation. Randomly coded cheese samples were left at room temperature for 1 hour before sensory analysis. Panel members scored the samples for three sensory parameters, including color, appearance and texture (I), smell (II), and taste (III). Sensory values were scored on a hedonic scale from 1 (poor) to 5 (excellent).

Experimental design and statistical analysis

The research design was completely randomized with a factorial structure (4x5). The factors were "storage time" (3, 30, 60 and 90 days) and "milk type and ratio". The milk was divided into two batches and cheese production for the experiment was carried out in duplicate (n=4) on the same day with the same milk. All tests were applied in duplicates. SPSS (version 25.0) program was used for statistical analysis of the data and results are presented as the mean ± standard deviation. Significance was tested using one-way and two-way analysis of variance (ANOVA). Tukey's multiple comparisons test was applied to determine the important sources of variation that have significant effects. The differences have been considered statistically significant at P value < 0.05.

RESULT AND DISCUSSION

Chemical changes

The dry matter, % lactic acid, fat, fat in dry matter, protein, salt, salt in dry matter, pH and lipolysis values of Karin Kaymagi cheeses are shown in Table 1.

Titration acidity values varied between 0.55% and 0.63% in terms of percent lactic acid during the ripening period, which is in line with the standards (Turkish Standardization Institute, 1995). % Lactic acid between the periods were statistically significant (P < 0.05) only for the 30th day. % Lactic acid decreases until the 30th day of ripening. Then the acidity increases until the 90th day possibly due to the increase in the amount of free acids resulting from the decomposition of lactose into lactic acid and the hydrolysis of fats (Kurt and Caglar, 1993).

The average dry matter content increased from 56.41% to 81.44% during the ripening period. After 90 days of ripening, the highest dry matter ratio was found in KK2.

Table 1. Chemical changes during ripening in Karın Kaymağı cheese ripened in tripe

Variabl	Ripening time (day)	Cheese samples					$\bar{x}$ ± SD
		KK1	KK2	KK3	KK4	KK5	
Dry matter (%)	3	57.09±0.39^{A}	55.72±0.01^{A}	55.43±0.98^{A}	56.61±0.41^{A}	57.23±0.99^{A}	56.41±0.91^{A}
	30	74.56±1.08^{B}	75.41±1.23^{B}	70.40±0.17^{B}	73.10±2.15^{B}	71.42±0.98^{B}	72.97±2.19^{B}
	60	77.14±2.50^{BC}	78.76±0.0^{C}	76.99±0.40^{C}	79.96±1.43^{C}	75.68±0.36^{C}	77.70±1.85^{C}
	90	80.80±0.81^{a,C}	83.98±0.77^{b,D}	80.15±0.44^{a,D}	81.13±0.58^{a,C}	81.14±0.18^{a,D}	81.44±1.46^{D}
	$\bar{x}$ ± SD	72.39±9.80^{bc}	73.46±11.4^{c}	70.74±10.1^{a}	72.69±10.5^{bc}	71.36±9.48^{ab}	
Fat (%)	3	32.75±1.06^{A}	31.00±1.41^{A}	32.50±0.70^{A}	33.75±0.88^{A}	32.62±0.88^{A}	32.45±1.22^{A}
	30	43.75±0.35^{B}	41.50±0.71^{B}	42.75±1.06^{B}	41.00±1.41^{B}	41.50±0.71^{B}	42.10±1.26^{B}
	60	45.37±0.53^{B}	44.75±1.06^{BC}	45.50±0.71^{BC}	43.75±0.35^{BC}	43.50±0.35^{B}	44.57±0.99^{C}
	90	48.25±0.35^{C}	47.87±0.18^{C}	47.12±0.53^{C}	45.87±1.23^{C}	47.00±0.35^{C}	47.22±0.98^{D}
	$\bar{x}$ ± SD	42.53±6.29^{b}	41.28±6.82^{a}	41.96±6.10^{ab}	41.00±5.11^{a}	41.15±5.68^{a}	
Fat in dry matter (%)	3	57.35±1.46	55.63±2.55	58.61±0.23	58.95±1.98	56.99±0.55	57.51±1.74
	30	58.68±1.32	55.02±0.03	60.86±1.44	56.12±3.58	58.10±1.78	57.76±2.61
	60	58.83±1.22^{ab}	56.81±1.31^{ab}	59.09±0.60^{b}	54.72±1.42^{a}	57.47±0.19^{ab}	57.38±1.84
	90	59.70±0.16	56.99±0.31	58.78±0.33	56.54±1.93	57.92±0.56	57.99±1.39
	$\bar{x}$ ± SD	58.64±1.25^{bc}	56.11±1.40^{a}	59.33±1.13^{c}	56.58±2.42^{ab}	57.62±0.87^{abc}	
Protein (%)	3	21.27±0.34^{A}	20.73±0.14^{A}	20.93±0.14^{A}	20.92±0.41^{A}	20.92±0.41^{A}	20.95±0.30^{A}
	30	24.82±0.48^{a,B}	27.64±0.14^{d,B}	25.72±0.07^{ab,B}	26.95±0.28^{cd,B}	26.21±0.21^{bc,B}	26.26±1.04^{B}
	60	26.27±0.35^{a,B}	29.97±0.23^{c,C}	27.01±0.23^{a,C}	28.49±0.46^{b,C}	27.58±0.34^{ab,C}	27.86±1.37^{C}
	90	27.79±0.21^{a,C}	30.80±0.41^{d,C}	28.28±0.06^{ab,D}	29.57±0.21^{c,C}	29.07±0.20^{bc,D}	29.10±1.12^{D}
	$\bar{x}$ ± SD	25.04±2.59^{a}	27.28±4.23^{e}	25.48±2.97^{b}	26.48±3.58^{d}	25.94±3.29^{c}	
Salt (%)	3	2.32±0.05^{b,A}	2.32±0.01^{b,A}	2.16±0.04^{a,A}	2.19±0.02^{ab,A}	2.24±0.07^{ab,A}	2.24±0.07^{A}
	30	3.05±0.02^{bc,B}	3.15±0.07^{d,B}	2.77±0.10^{a,B}	2.87±0.03^{ab,B}	2.82±0.04^{a,B}	2.93±0.15^{B}
	60	3.17±0.01^{ab,BC}	3.34±0.07^{b,C}	3.04±0.09^{b,B}	3.18±0.01^{ab,C}	2.99±0.07^{a,BC}	3.14±0.13^{C}
	90	3.33±0.05^{b,C}	3.59±0.07^{b,D}	3.17±0.13^{a,B}	3.21±0.04^{a,C}	3.21±0.11^{a,C}	3.30±0.17^{D}
	$\bar{x}$ ± SD	2.96±0.41^{b}	3.10±0.51^{c}	2.78±0.42^{a}	2.86±0.44^{a}	2.81±0.39^{a}	
Salt in dry matter (%)	3	4.06±0.07	4.16±0.02	3.90±0.14	3.87±0.09	3.90±0.10	3.98±0.14
	30	4.09±0.09	4.18±0.06	3.94±0.14	3.93±0.06	3.94±0.11	4.01±0.13
	60	4.11±0.01	4.25±0.00	3.95±0.14	3.97±0.09	3.95±0.10	4.04±0.14
	90	4.12±0.10	4.27±0.03	3.96±0.14	3.97±0.03	3.96±0.14	4.05±0.15
	$\bar{x}$ ± SD	4.09±0.07^{b}	4.21±0.05^{b}	3.94±0.11^{a}	3.93±0.07^{a}	3.94±0.09^{a}	

Continued. Table 1

Variabl	Ripening time (day)	Cheese samples					$\bar{x}$ ± SD
		KK1	KK2	KK3	KK4	KK5	
pH	3	4.73±0.04^{b,A}	4.49±0.02^{a,A}	4.71±0.01^{b,A}	4.57±0.01^{a,A}	4.68±0.01^{b,A}	4.63±0.09^{A}
	30	4.81±0.01^{c,A}	4.63±0.04^{a,B}	4.83±0.02^{c,B}	4.70±0.01^{ab,B}	4.77±0.02^{bc,B}	4.75±0.08^{B}
	60	4.94±0.01^{c,B}	4.73±0.01^{a,B}	4.93±0.01^{c,C}	4.79±0.02^{b,C}	4.88±0.01^{c,C}	4.85±0.08^{C}
	90	5.11±0.01^{b,C}	4.89±0.02^{a,C}	5.08±0.04^{b,D}	4.92±0.03^{a,D}	5.04±0.03^{b,D}	5.01±0.09^{D}
	$\bar{x}$ ± SD	4.89±0.15^{d}	4.68±0.15^{a}	4.89±0.14^{d}	4.74±0.13^{b}	4.84±0.14^{c}	
Lactic acid (%)	3	0.61±0.029	0.58±0.005	0.59±0.001^{AB}	0.61±0.006	0.59±0.005^{A}	0.60±0.016^{B}
	30	0.55±0.018	0.57±0.006	0.56±0.006^{A}	0.56±0.012	0.56±0.005^{A}	0.56±0.012^{A}
	60	0.61±0.022^{ab}	0.63±0.012^{b}	0.57±0.012^{a,AB}	0.58±0.005^{ab}	0.63±0.006^{b,B}	0.60±0.028^{B}
	90	0.59±0.012	0.59±0.023	0.61±0.018^{B}	0.60±0.024	0.59±0.012^{A}	0.60±0.015^{B}
	$\bar{x}$ ± SD	0.59±0.031	0.59±0.024	0.58±0.023	0.59±0.024	0.59±0.026	
Lipolysis	3	0.51±0.002^{a,A}	0.46±0.002^{a,A}	0.49±0.03^{a,A}	0.47±0.002^{a,A}	0.60±0.02^{b,A}	0.51±0.05^{A}
	30	1.14±0.01^{b,B}	0.99±0.03^{b,B}	0.80±0.08^{a,A}	1.05±0.02^{b,B}	0.78±0.007^{a,A}	0.95±0.15^{B}
	60	1.64±0.02^{a,C}	3.67±0.04^{d,C}	1.95±0.15^{ab,B}	2.29±0.07^{bc,C}	2.59±0.09^{c,B}	2.42±0.73^{C}
	90	2.68±0.02^{a,D}	5.38±0.07^{c,D}	2.60±0.02^{a,C}	3.25±0.05^{b,D}	3.35±0.02^{b,C}	3.45±1.06^{D}
	$\bar{x}$ ± SD	1.49±0.84^{a}	2.62±2.13^{c}	1.46±0.91^{a}	1.76±1.15^{b}	1.83±1.25^{b}	

Values are means ± standard deviation
a–e letters point out differences ($P < 0.05$) between cheese samples
A–D letters point out differences ($P < 0.05$) between ripening days

The dry matter in this sample was significantly higher than in other cheese samples. This result is most likely caused by sheep milk's high protein and fat content. The average dry matter content was higher when compared with the studies on Tulum cheese (Celik and Tarakci, 2017; Öztürk and Akin, 2017; Tekin and Güler, 2019; Tekin and Güler, 2021) and similar to the data obtained by Turgut et al. (2012) for Karin Kaymagi. It can be said that the reason why the dry matter ratios are higher than the Tulum cheese samples is due to the fact that the tripe has a more porous structure compared to the Tulum (sheep or goat skin) (Hayaloglu et al., 2007). In addition, adding cream to cheese samples during production also increases the dry matter ratio.

The average fat content of cheese samples increased significantly from 32.45% to 47.22% during the 90-day ripening period. The fat contents have changed in parallel with the dry matter, and Hayaloglu (2003) similarly reported that the fat content of cheeses and fat in dry matter change in parallel with the change in dry matter ratios. The values for fat content were close to the fat content of Karin Kaymagi cheeses from the market analyzed by Turgut et al. (2012) (43.36%) and Çakmakçı et al. (1995) (39.00%).

However, the values were higher than those from studies on other Tulum cheese types. The main reason for this is that extra cream is added to the cheese during the production of Karin Kaymagi cheese. The effect of milk type and ration of sheep milk and cow milk on the fat content in the cheese was significant. Cheeses from only cow milk obtained higher fat content than cheeses from sheep milk and cheeses from 25% cow milk + 75% sheep milk and from 50% cow milk + 50% sheep milk.

Due to their fat in dry matter values exceeding 40%, these cheeses can be classified as full-fat Tulum cheeses (Turkish Standardization Institute, 1995).

Protein ratios in the samples increased from 20.95% to 29.10%, parallel with the increase in dry matter with ripening ($P < 0.05$). At the end of ripening, the significantly highest protein content was detected in the KK2 sample with a value of 30.80%, probably caused by the high protein content of sheep milk ($P < 0.05$). In this result, it is thought that the high protein ratio of sheep milk is effective. These values are higher than the average protein content (25.51% and 19.01%, respectively) determined in Karin Kaymagi by Turgut et al. (2012) and Çakmakçı et al. (1995) and in Afyon Tulum cheese (22.48%) by Kara and Akkaya (2015). Furthermore, these values are in accordance with the Erzincan Tulum cheese values (30.19-30.51%) determined by Uçar and Tekinşen (2004).

Salt ratios in the samples increased statistically from 2.24% to 3.30% on average during the ripening period. Cheese type also significantly affected the salt ratio with higher values in KK1 and KK2 cheeses than in the other cheese. The dry matter values of the samples were again effective in these differences.

During the ripening process, salt content in dry matter did not increase significantly. These values are lower than the value (6.15%) obtained in the study by Çakmakçı et al. (1995) on Karin Kaymagi cheese but more in accordance with the value (4.90%) determined by Tarakci and Durmus (2016) in Tulum cheese ripened in tripe.

With the ripening period, the average pH values of the samples increased significantly from 4.63 to 5.01. The highest value at the end of 90 days of ripening was determined in the KK1 sample with a value of 5.11, which was not significantly different from the value for the KK3 and KK5 cheeses. The mean value at the end of ripening is close to the pH values (pH 4.9) of Karin Kaymagi cheese analyzed by Turgut et al. (2012) and the pH value (pH 5.16) determined by Güven and Konar (1994) in Tulum cheese.

The average lipolysis values increased during the ripening period (0.51 – 3.45) and the highest lipolysis value at the end of ripening (5.38) was found in KK2 cheese produced entirely from sheep milk. This value was statistically higher than in the other cheeses. Ripening time and cheese type ($P < 0.05$) significantly affected lipolysis values. It is thought that the different milk combinations utilized, the biochemical changes that occur during the ripening period, and the different fat content of the samples play important roles in the emerging differences. The mean lipolysis (ADV) values determined in this research were found to be lower than the ADV values of Tulum cheese in the research of Sert et al. (2014).

Changes in nitrogen fractions

WSN/TN, PTA-SN/TN and TCA-SN/TN values were determined as indicators of the nitrogen compounds fractionation process (Table 2).

The one important index in determining the ripening progress in cheese is the amount of water-soluble nitrogen. There was a significant increase in water-soluble nitrogen values in the cheese samples during ripening ($P < 0.05$). Although water-soluble nitrogen values at the beginning of ripening were relatively higher for KK2 and KK4 cheeses, they were not statistically different ($P > 0.05$). At the end of ripening, the water-soluble nitrogen values varied between 23.73% and 28.02%, and the highest value was found in cheese produced from sheep milk (KK2). These values are higher than the value (5.58 ± 0.15%) of Tulum cheese ripened by Tarakci and Durmuş (2016) and the value of Erzincan Tulum cheese (21.07 ± 0.93%) by Çakır and Çakmakçı (2018), lower than the 30th-day value of Croatian cheese ripened in sheepskin by Rako et al. (2019) and similar to the average value (21.48%) obtained in Croatian cheese ripened in sheepskin by Vrdoljak et al. (2022).

It was determined that TCA-SN/TN values increased significantly during the ripening period ($P < 0.05$). At the end of the ripening period, the highest TCA values were determined in KK2 with 4.58%, which was statistically higher than the values for the other cheeses.

Table 2. Biochemical changes during ripening in Karın Kaymağı cheese ripened in tripe

Variabl	Ripening time (day)	Cheese samples					$\bar{x}$ ± SD
		KK1	KK2	KK3	KK4	KK5	
TCA-SN/TN (%)	3	2.40±0.14^{a,A}	2.73±0.04^{b,A}	2.35±0.07^{a,A}	2.53±0.01^{ab,A}	2.48±0.05^{ab,A}	2.50±0.15^{A}
	30	2.88±0.10^{a,B}	3.90±0.04^{b,B}	2.99±0.14^{a,B}	3.96±0.00^{b,C}	3.75±0.07^{b,B}	3.50±0.49^{B}
	60	3.09±0.10^{a,B}	4.05±0.07^{c,B}	3.35±0.03^{a,C}	3.90±0.00^{bc,B}	3.73±0.07^{b,B}	3.62±0.37^{C}
	90	3.27±0.07^{a,B}	4.58±0.14^{d,C}	3.61±0.00^{ab,C}	4.05±0.01^{c,D}	3.99±0.16^{bc,B}	3.90±0.47^{D}
	$\bar{x}$ ± SD	2.91±0.36^{a}	3.81±0.72^{d}	3.07±0.50^{b}	3.61±0.67^{c}	3.49±0.63^{c}	
PTA-SN/TN (%)	3	1.69±0.03^{a,A}	2.33±0.14^{b,A}	1.72±0.04^{a,A}	1.99±0.02^{a,A}	1.82±0.09^{a,A}	1.91±0.25^{A}
	30	2.13±0.01^{a,B}	2.53±0.07^{b,A}	2.81±0.09^{c,B}	2.85±0.02^{c,B}	2.58±0.02^{b,B}	2.58±0.27^{B}
	60	2.65±0.02^{a,C}	3.24±0.07^{c,B}	3.09±0.08^{bc,BC}	2.89±0.07^{ab,B}	2.94±0.09^{ab,C}	2.96±0.21^{C}
	90	3.07±0.12^{a,D}	3.98±0.02^{b,C}	3.28±0.08^{a,C}	3.29±0.02^{a,C}	3.29±0.07^{a,D}	3.38±0.33^{D}
	$\bar{x}$ ± SD	2.38±0.56^{a}	3.02±0.69^{c}	2.72±0.64^{b}	2.75±0.50^{b}	2.65±0.58^{b}	
WSN/TN (%)	3	5.79±0.23^{A}	6.90±0.29^{A}	5.89±0.37^{A}	6.60±0.13^{A}	6.13±0.78^{A}	6.26±0.54^{A}
	30	15.70±0.02^{B}	16.03±0.14^{B}	15.92±0.31^{B}	16.33±0.67^{B}	16.00±0.14^{B}	15.99±0.33^{B}
	60	24.14±0.76^{C}	23.11±0.65^{C}	23.47±0.22^{C}	22.55±1.18^{C}	23.57±0.12^{C}	23.51±0.90^{C}
	90	25.04±0.43^{ab,C}	28.02±0.60^{c,D}	23.73±0.43^{a,C}	26.77±0.18^{bc,D}	24.71±1.03^{ab,C}	25.65±1.68^{D}
	$\bar{x}$ ± SD	17.66±8.30^{ab}	18.51±8.50^{b}	17.25±7.77^{a}	18.06±8.12^{ab}	17.78±8.10^{ab}	

(WSN/TN): Water soluble nitrogen, (TCA-SN/TN): 12% trichloroacetic acid soluble nitrogen, and (PTA-SN/TN): 5% phosphotungstic acid soluble nitrogen.

Values are means ± standard deviation.

a–e letters point out differences ($P < 0.05$) between cheese samples.

A–D letters point out differences ($P < 0.05$) between ripening days.

Sample identification: KK1: 100% cow milk, KK2: 100% sheep milk, KK3: 75% cow milk + 25% sheep milk, KK4: 25% cow milk + 75% sheep milk and KK5: 50% cow milk + 50% sheep milk.

In contrast, the value for KK4 was statistically higher than the values for cheese KK1 and KK3. The value for KK5 was significantly higher than for KK1. The reason for these differences are related to the proportion of sheep milk in the samples.

The aminonitrogen ratio (PTA-SN/TN) also indicates the peptidase activity in cheese. In most cheeses, peptidases, especially aminopeptidases and proteinases, break down bitter peptides released from the breakdown of casein. This eliminates bitterness and improves the taste and aroma (Di Cagno et al., 2004). PTA values of cheese samples increased significantly during the ripening period. At the end of the 90th day, the value for KK2 was significantly higher than for any other cheese, while the value for the other cheeses were not significantly different. The values at the end of the ripening period were in accordance with the values (3.92-4.53%) found by Çakmakçı et al. (2011) in Tulum cheese at the end of the 90th day and higher than the value (% 2.06) found by Tarakci and Durmuş (2016) in Tulum cheese ripened in the tripe.

Electrophoretogram patterns

Raw milk contains cathepsin D and plasmin in its structure. Despite its relatively low activity, cathepsin D retains some of its activity after pasteurization (Fox et al.,

1993). Cheeses produced in this study were produced from raw milk, so the natural enzymes found in milk may have affected proteolysis rates.

Electrophoretograms for α_{s1}-CN and β-CN fractions, together with other degradation products, are given in Figure 1.

The first effect on α_{s1}-CN is shown by rennet. This enzyme hydrolyzes the α_{s1}-CN from the C-terminus of the region 24/25-199 to form a large molecule peptide called α_{s1} (Grappin et al., 1985). Different findings in the literature regarding the rate of α_{s1}-CN degradation in cheese are possible due to differences in milk used, production procedure, and ripening conditions (Tarakci and Durmuş, 2016).

As shown in Figure 1, α_{s1}-CN was broken down and its concentration decreased continuously during ripening. Residual values between 100% in unripened cheese and 83.63% after 90 days of ripening were determined for α_{s1}-CN (Figure 2). The highest decrease occurred in the KK2 sample, and the lowest decrease occurred in the KK1 sample, which is thought to be due to the milk type.

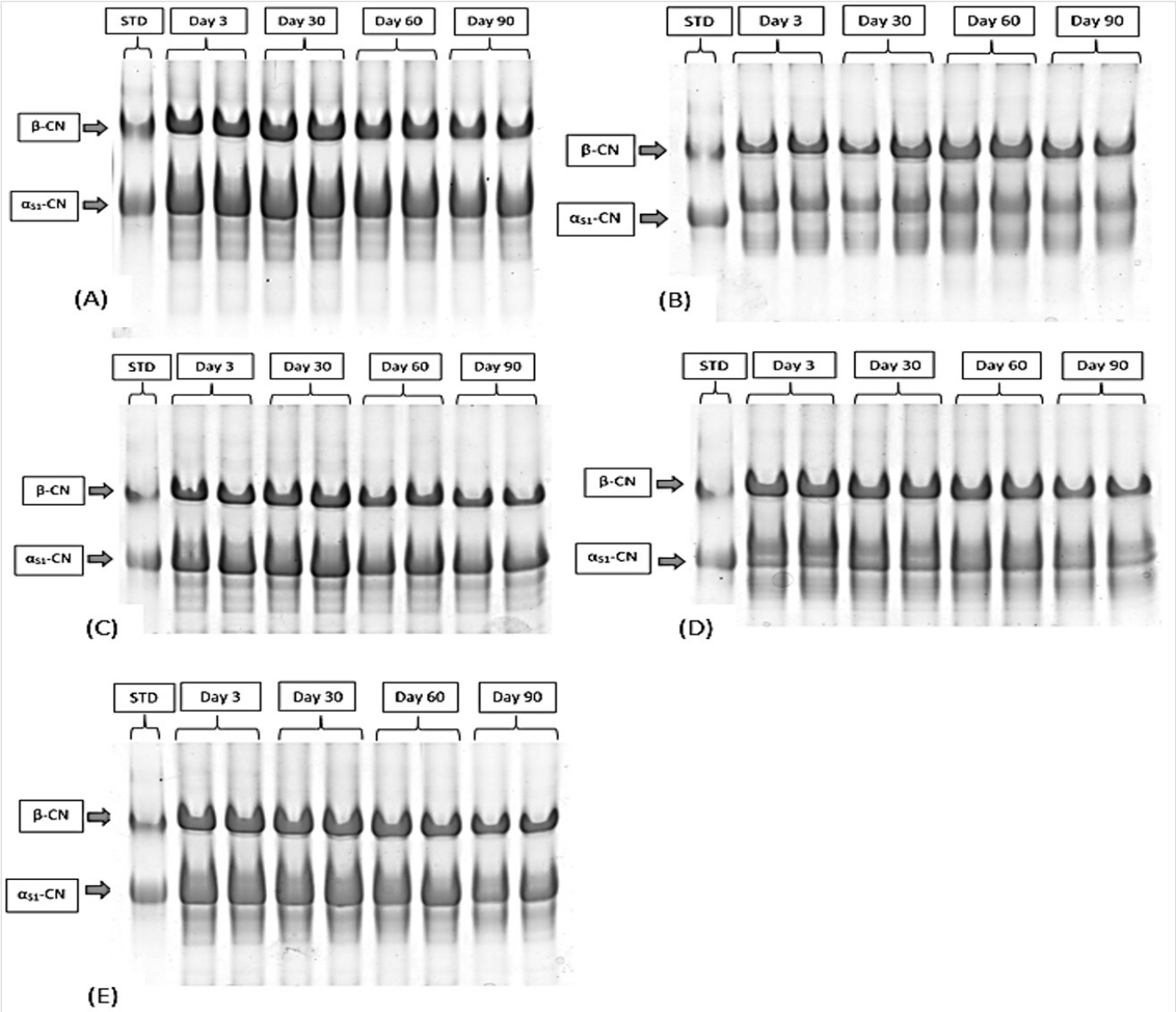

Figure 1. Urea-PAGE electrophoretograms of Karın Kaymağı cheeses: (A): KK1, (B): KK2, (C): KK3, (D): KK4 and (E): KK5, CN = casein

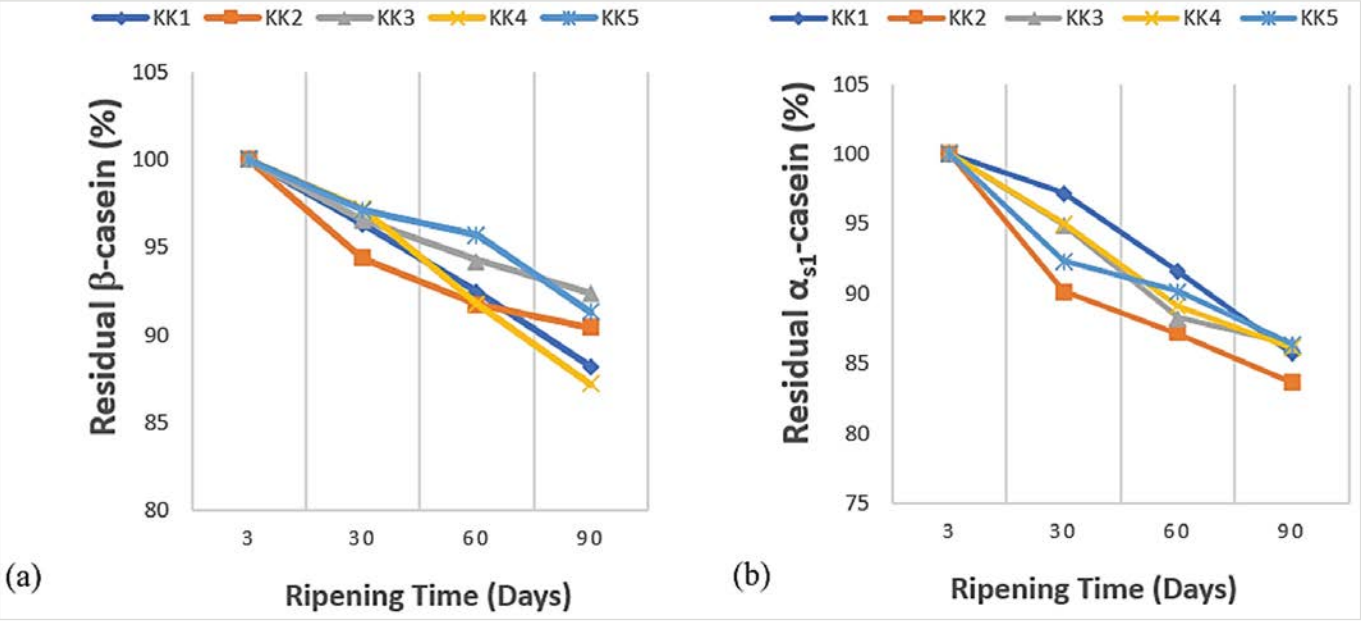

Figure 2. Changes in mean residual β-casein (a) and α_{s1}-casein (b) concentrations (%) in Karın Kaymağı cheeses during ripening

It has been determined that β-CN is more resistant than α_{s1}-CN to proteolysis by rennet, plasmin, or starter enzymes in the cheese matrix, due to its structure and tendency to coalesce. β-CN is broken down into β-1, β-2, and β-3 components with the help of rennet and γ-CN by the effect of plasmin at the beginning of ripening (Turin et al., 1995). The residual amount of β-CN in the cheese samples increased as the ripening period progressed, and values between 100% in unripe cheese and 87.12% after 90 days of ripening were determined (Figure 2). Reduction in the concentration of β-CN in different cheese types has been reported by many researchers (Hayaloglu, 2003; Kim et al., 2004).

Texture profile analysis

Hardness, cohesiveness, gumminess, springiness, adhesiveness, and chewiness values of cheese samples are shown in Figure 3.

The maximum force applied in the first compression of cheese is called hardness (Kim et al., 2004). Hardness in cheese is associated with moisture and salt. The hardness of cheese decreases with increasing moisture content, and increases with increasing salt content (Kaya, 2002). The hardness values also increased during the ripening period likely due to the increase in dry matter values. KK3 sample had the highest hardness value at the end of ripening, while KK1 samples had the lowest hardness value. When the averages of hardness values are examined, it is seen that these values are higher than the hardness values of Tulum cheese ripened on goat skin by Şengül et al. (2014) and the hardness values of Croatian cheese ripened in sheepskin by Rako et al. (2019). The main reason for this is thought to be that the tribe, which is used as a packaging material, is more permeable than animal skin and, accordingly, the increase in dry matter.

Cohesiveness reflects the degree to which the chewed mass sticks together in the mouth (Diezhandino et al., 2016). No significant change was observed in the cohesiveness values until the 60th day of ripening but significantly increased between the 60th and 90th days ($P < 0.05$). Considering the cheese types, no significant difference was detected ($P > 0.05$).

Gumminess is delineated as the shearing force required to prepare food for swallowing (Raphaelides et al., 1995) and is calculated as hardness × stickiness. Gumminess values increased during ripening. The highest value was determined in the KK3 sample and the lowest in the KK1 sample at the end of the ripening period. Gumminess values increased rapidly between the 60th and 90th days of ripening.

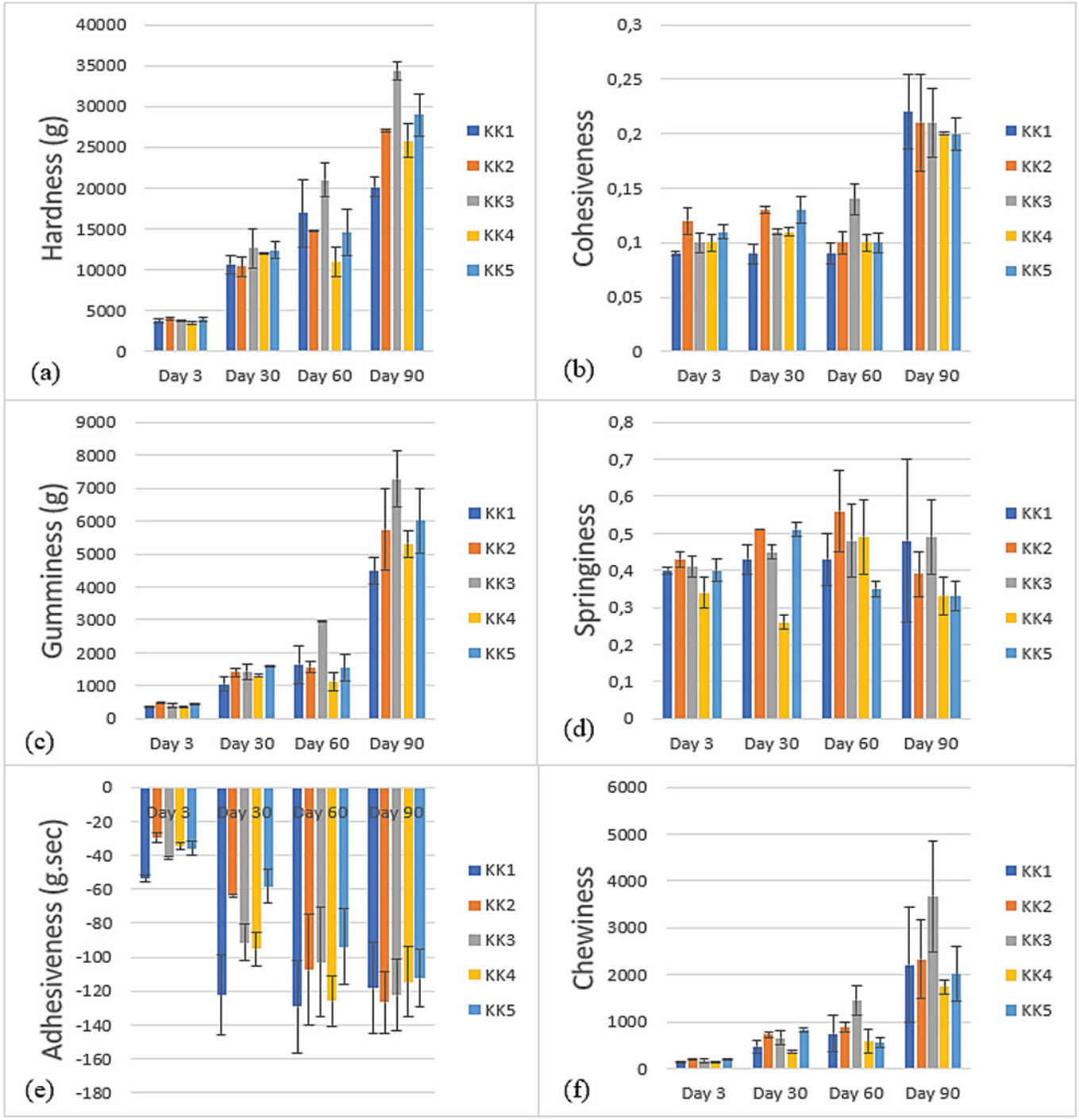

Figure 3. Mean values and standard deviations for hardness (a), cohesiveness (b), gumminess (c), springiness (d), adhesiveness (e), and chewiness (f) of Karın Kaymağı cheeses

Gumminess values were found to be lower when compared to the Tulum cheese ripened in goat skin by Şengül et al. (2014), but the rapid increase between the 60th and 90th days was similar to this study.

The degree of restoring of food during chewing is defined as springiness (Truong et al., 2002). Springiness values vary between 0.40 ± 0.11 and 0.47 ± 0.10. When the springiness values are examined during the ripening period, there is no regular pattern of increase or decrease. When the values are compared with other studies; It is similar to the values determined by Tomar et al. (2020) for Tulum cheese, and lower than the value determined by Şengül et al. (2014) for Tulum cheese and the value found by Rako et al. (2019) for Croatian cheese.

Adhesiveness is known as the stickiness felt in the mouth when chewing food. The adhesiveness values decreased during ripening. This decrease is thought to be caused by the increase in dry matter content. When Figure 3 is reviewed according to the cheese type, the lowest adhesiveness value was determined in the KK2 sample and the highest in the KK5 sample at the end of the ripening period. When the results are evaluated; It is seen that it is lower than the average values found by Tomar (2019) in Tulum cheese.

The act of chewing is the work required to get semi-solid food ready to be swallowed. The chewiness is defined as the product of gumminess and springiness, which is equal to gumminess × springiness. The chewiness values of cheese samples are given in Figure 3. As the samples ripened, chewiness values increased, and the lowest and highest values were found in samples KK4 and KK3 ($P < 0.05$).

Sensory analysis

Sensory results for cheese samples are given in Figure 4. When the means of section, appearance and structure scores according to ripening periods were examined, the highest nominal score was given to KK2 with a value of 3.90 ($P < 0.05$).

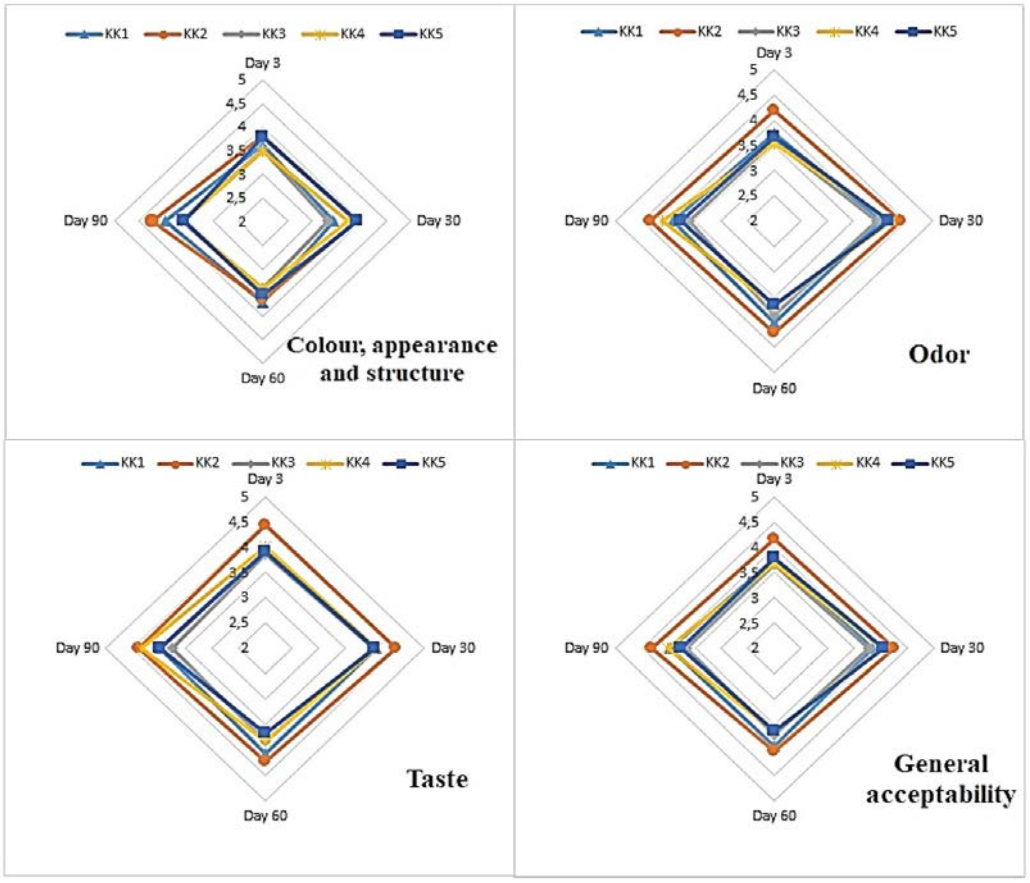

Figure 4. Sensory evaluation results of Karın Kaymağı cheeses

This value was statistically higher than the values obtained for samples KK3 and KK4 but not statistically different from the scores for samples KK1 and KK5.

When the averages of the odor values were examined according to the ripening time, these samples had a statistically better odor than the samples evaluated after 3 and 60 days but not better than scores obtained after 90 days of ripening. When compared in terms of sample types, the significantly highest score was obtained by KK2 ($P < 0.05$).

Overall, it would be proper to state that the utilization of sheep milk in the production enhances the sensory properties of Karin Kaymagi cheese. Samples evaluated after 30 days of ripening scored significantly better than samples evaluated after 60 days.

CONCLUSION

The data obtained in this study show that the investigated Karın Kaymağı cheeses were full-fat cheeses with normal salt and acidity rates and dry matter content. High dry matter, salt content and 90 days of ripening increase the safety of Karın Kaymağı cheese produced from raw milk. The fact that the tripe, which is used as the packaging material, is rather permeable is probably an important reason for the rapid increase in the dry matter of the cheeses. Also, the main reason for KK2 sample having the highest dry matter ratio could be attributed to the higher dry matter of sheep milk than cow milk. Again, the relatively higher lipolysis rate in the KK2 sample may indicate that the fat in sheep milk is more favorable for lipolytic activity. According to the sensory analysis results, the sample produced entirely from sheep milk (KK2) was more appreciated than the others. When all these data are examined, the use of sheep milk in the production of Karin Kaymagi both increases its nutritional value and makes it more appreciated by consumers.

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