Süleyman Demirel Üniversitesi
Fen Bilimleri Enstitüsü Dergisi
Cilt 23, Özel Sayı, 131-138, 2019

Süleyman Demirel University
Journal of Natural and Applied Sciences
Volume 23, Special Issue, 131-138, 2019

DOI: 10.19113/sdufenbed.538894

Aromatic and Sensorial Properties of Çökelek Cheese Produced by Different Methods

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(Alınış / Received: 23.05.2018, Kabul / Accepted: 07.11.2018, Online Yayınlanma / Published Online: 12.03.2019)

Keywords
Çökelek cheese,
Aroma active compound,
GC-Olfactometry

Abstract: In this study, it was aimed to reveal aroma-active compounds and sensory properties of Çökelek cheese made from milk, yogurt and Tuluk yogurt. For this purpose, aroma active compounds in Çökelek samples were determined by Gas Chromatography-Olfactometry. Furthermore, sensory properties of cheese samples were determined by *Spectrum*® analysis. As result, 17 identified and 2 unknown aroma active compounds were determined in Çökelek samples. Butyric acid, Furaneol® and sotolon were identified at high intensities in all cheeses. It was determined that the intensity of butyric acid was higher in Çökelek made from yogurt while the intensities of Furaneol® and sotolon were found to be higher in Çökelek made from milk. "Cooked", "Whey", "Creamy", "Rancid" "Fermented", "Yeast", "Sour", "Salty", "Sweet" and "Umami" were defined as characteristic sensory terms for Çökelek cheeses. It was determined that cooked, creamy and sweet were perceived in Çökelek made from yogurt while yeast aroma was found to be a higher in Çökelek made from Tuluk yogurt.

Farklı Yöntemlerle Üretilen Çökelek Peynirinin Aromatik ve Duyusal Özellikleri

Anahtar Kelimeler
Çökelek peyniri,
Aroma aktif bileşen,
GC-Olfaktometri

Özet: Bu çalışmada süt, yoğurt ve tuluk yoğurdundan yapılan Çökelek peynirinin aroma aktif bileşenleri ve duyusal özeliklerinin ortaya çıkarılması amaçlanmıştır. Bu amaçla, çökelek örneklerinin aroma aktif bileşenleri Gaz kromatografi Olfaktometri (GCO) yöntemi kullanılarak belirlenmiştir. Ayrıca, peynir örneklerinin duyusal özellikleri Spectrum® analizi kullanılarak tespit edilmiştir. Sonuç olarak, Çökelek örneklerinde toplam 17 adet tanımlanmış ve 2 adet de bilinmeyen aroma aktif bileşen belirlenmiştir. Bütirik asit, Furaneol® ve sotolon gibi bileşenlerin çökelek örneklerinde yüksek yoğunlukta olduğu tespit edilmiştir. Bütirik asitin aroma yoğunluğunun yoğurttan üretilen çökelekte en yüksek olduğu, Furaneol® ve sotolonun ise sütten yapılan çökelek peynirlerinde yüksek olduğu belirlenmiştir. Çökelek peynirleri için belirlenen karakteristik duyusal terimler "Pişmiş", "Peyniraltı suyu", "Kremamsı", "Ransit", "Fermente", "Maya", "Ekşi", "Tuzlu", "Tatlı" ve "Umami"dir. Yoğurttan yapılan çökelek peynirlerinde pişmiş, kremamsı ve tatlı özelliklerin daha yüksek algılandığı, Tuluk yoğurdundan yapılan çökeleklerde ise maya aromasının daha yüksek olduğu belirlenmiştir.

1. Introduction

Çökelek cheese is a type of cheese produced in almost every region in Turkey. Although, it can be produced from milk products such as milk and yogurt, it can also be obtained by filtering the curd produced from buttermilk (yayık ayran) [1, 2]. The curd produced by this method can be used as a supplementary raw

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material in the production of some Küp and skin Tulum cheeses. Çökelek cheese, is known as "Ekşimik" in Western Anatolia and Trakya region, "Keş", "Kesik" or "Urda" in the Mediterranean region, "Minzi" or "Minci" in the Eastern Black Sea region, and "Jaji", "Cacik" and "Torak" in the Eastern Anatolia Region [1, 3-5].

There were many studies on the physical, chemical and microbiological properties of the Çökelek cheeses produced in Turkey [1, 6, 7]. For instance, Ağaoğlu et al. [6] determined the average moisture, fat, protein, ash, acidity values and aerobic mesophilic bacteria and yeast counts of 25 Çökelek cheese samples produced in Van and its region as 81.84%, 1.2%, 8.04%, 0.94%, 1.2% lactic acid, $9.8x10^6$, and $1.3x10^5$cfu/g, respectively. The average chemical composition and microbiological properties of 14 Çökelek cheeses available in the market in Antalya and Burdur had 28.55% in dry matter, 17.48 ° SH acidity, 3.54% fat, 0.9% salt, $1x10^1$ cfu/g total bacteria, $2x10^3$ cfu/ g yeast-mold, $8.8x10^2$ cfu/g coliform bacteria, and $5.1x10^2$ cfu/g *S. aureus*. Moreover, 50% of the Çökelek samples examined were reported to contain *E. coli* [1]. Önganer and Kırbağ [7] studied fresh consumed Çökelek cheeses in Diyarbakır. it was reported that the total count of microorganisms in this cheese was 8.49 log cfu/g, the heterofermentative lactic acid bacteria were 8.58 log cfu/g, the psychrophilic bacteria were 8.51 log cfu / g, the proteolytic microorganisms were 8.49 log cfu/g, yeast-mold count was 6.67 log cfu/g, *S. aureus* was 7.53 log cfu/g, *Salmonella* spp. was 7.80 log cfu/g, *Bacillus spp*. was 7.70 log cfu/g, and *Clostridium* spp. was 7.81 log cfu/g.

There are limited studies investigating the aroma profile and sensory properties of Çökelek cheese-focused on studying the production technique and some physical, chemical and microbiological properties. The use of yogurt as a raw material in many regions, different storage conditions, and the use of different storage materials significantly affect the aromatic and sensory properties of the Çökelek cheese. Şimsek and Sağdıç [8] investigated that the effect of starter culture types and different temperature treatments on some properties of Çökelek cheese produced from goat milk. The researchers found that çökelek cheeses produced with yoghurt culture applying 95°C for 30 min had lowest saturated fatty acid content and the highest appearance and flavour scores. Another study by Güler [9], it was revealed that the profiles of organic acid and volatile compounds of Surk cheese which is artisanal çökelek with herbs and spices in Hatay region in Turkey. For Surk cheese, butanoic acid, hexanoic acid, octanoic acid, decanoic acid were found to be as major acids while carvacrol, γ - terpinene, p-cymene and eugenol were major phenolics

Several researchers also investigated aroma and sensory properties of some traditional cheeses like Çökelek Cheese in Turkey. In this context, Şimşek and Tuncer [10] investigated fresh and traditional Akcakatik Cheese made from by drying strained yogurt with or without adding cloves or black cumin. The researcher found that acetaldehyde, diacetyl, ethanol and ethyl acetate are major flavor compounds of Akcakatik cheese. Moreover, it was found that ripened Akcakatik cheese had a higher intensity of diacetyl and acetaldehyde than the fresh Akcakatik cheese samples. In a another study, volatile and sensory properties of traditional Dolaz (Tort) cheese made from whey by Okur and Güzel-Seydim [11], it was found that acetaldehyde, acetone, etanol, acetic acid, diacetyl and 1-butanol were major volatile compounds in Dolaz cheese while soft, yellowish-light brown color, granular structure were defined as characteristic appearance terms for Dolaz cheese. Hayaloglu et al. [12] determined about 100 volatile compounds in Tulum cheese ripened in Goat's Skin or Plastic Bags for 90 days. The researcher found that the main volatiles were short-chain fatty acids as butanoic acid, 2-methyl propanoic acid, 3-methyl butanoic acid, ketones as 2-butanone, 2-heptanone and diacetyl, and primary alcohols as 3-methyl 1-butanol, 2-butanone and 2-methyl 1-propanol, aldehydes as acetaldehydes and 2-propanal and esters as 3-methylbutyl acetate, ethyl butanoate and ethyl lactate.

It is important to characterize the aromatic profiles and sensory properties of foods [13], particularly when the properties governing flavor and the appearance and structural characteristics of a food play a significant role in its value [14]. The characterization of the aromatic profiles and sensory properties of dairy products such as cheese, yogurt, and kefir, can improve and/or standardize the methods of producing the product preferred by consumers and control the product's originality and authenticity, for example, the ripening period required in cheese or the determination of its quality [15].

This study aimed to determine flavor compounds profile and sensory properties of Çökelek cheeses produced by three different methods by using instrumental and sensory analysis.

2. Material and Method

In the present study, Çökelek cheese produced from cow milk, yogurt and Tuluk yogurt (cow milk kept in a goat skinbag (Tuluk) for 20 days) Aroma authentic standards used in the study were obtained from Sigma-Aldrich (St. Louis, Mo., USA), Merck (Darmstadt, Germany), Bedoukian Research Inc. (Danbury, CT, USA), and Aromsa (Gebze, Kocaeli). Milk Çökelek cheeses were symbolized as MC (milk

Çökelek), YC (yogurt Çökelek), and TC (Tuluk yogurt Çökelek).

2.1. Preparation of Tuluk

For the preparation of Tuluk, 6-7 months-old female goat skin prepared by shaving and dry salting method for at least 3 months were used. The excess fat and meat pieces on the skin were removed, feet and arm parts of the skin were tied, the bottom part was covered with rope and wooden rods using a special technique and readied for use (Figure 1 and Figure 2). The bottom part of the Tuluk is closed with rope and wooden rod by a special technique.

2.2. Cheese production

The Çökelek cheese is a lactic acid clot; therefore, its characteristics are different from other cheeses. The production of the Çökelek cheese was carried out in April-May. The production stages of Çökelek cheeses was shown in Figure 3. Cheese production was carried out in two replications.

2.3. Determination of aroma-active compounds

The samples were analysed by using solid phase microextraction (SPME) method [16] and gas chromatography-olfactometry [17]. 3 g of cheese sample was prepared with a PTFE / silicone septa cap and amber (Perkin-Elmer) coloured 40 mL vials were weighed and 1 g NaCl was added. The sample was kept in a 40°C water bath for 30 minutes to stabilize the flavor of the aroma substances in the headspace. Subsequently, SPME fiber (2 cm-50/30 µm DVB / Carboxen / PDMS stable flex, Supelco, Bellefonte, USA) was immersed in the vial crown and incubated at 40 °C for 30 min and stained with fiber GCO (6890, Agilent Technologies, Wilmington, and aroma compounds were determined by injection of SPME fiber to GC-O. HP-INNOWAX (30 m x 0.25 mm internal diameter (id) x 0.25 µm film thickness) column was used in the GCO (J & W Scientific, Folsom, CA, USA). GC conditions; carrier gas: Helium, inlet pressure: 11.54 psi, total flow: 54.1 ml/min, flame ionization detector temperature: 250°C, oven temperature program: Initial temperature and duration: 40°C, 3 minutes, Ramp: 10 C/ min, final temperature and duration: 200°C 25 minutes [18]. The GCO procedure was conducted by an experienced panelist in two replicates. The concentrations of the aroma active compounds in the Çökelek cheeses were measured on a 10-point scale (0: not present-10: very high). The definitions of the aroma active ingredients are provided by both the retention index determined under GCO conditions and by smelling the authentic standards under the same conditions [19].

2.4. Sensory analyses

The sensory properties of the Çökelek samples were determined by *Spectrum*® analysis [20]. Sensory evaluations were performed by a panel of 7 experienced (4 male and 3 female experts, aged 24-45 years) individuals. The intensities of the terms determined in the samples by panel members were evaluated on a 10-point scale (0 = very low, 10 = very intense). Cökelek samples were presented to the panelists randomly and in two replicates, encoded with 3 different codes in white plastic plates at room temperature. Reference materials and sensory terms used for the evaluation of cheese samples were presented in Table 1.

Table 1. Sensory terms and references (Meilgaard et al. [20]).

Descriptor	References
Cooked	Milk heated to 85°C for 30 min
Whey	Whey powder solution (5%)
Creamy	Dairy cream or butter
Rancid	10 µL butyric acid/methanol
Fermented	Fresh yogurt
Yeast	Dough
Fruity	Peach, apricot.
Sour	0.08% Citric acid
Salty	0.5% Sodium chloride
Sweet	2% saccharose
Umami	1% monosodium glutamate

2.5. Statistical analysis

One-way ANOVA analysis was used in the study to determine the differences in the aroma-active components and sensory characteristics of cheese samples. TUKEY multiple comparison (TUKEY-HDS) test was used to determine significant differences

Figure 1. Preparation of Tuluk (Goat Skin Bag)

Figure 2. Production of çökelek cheese

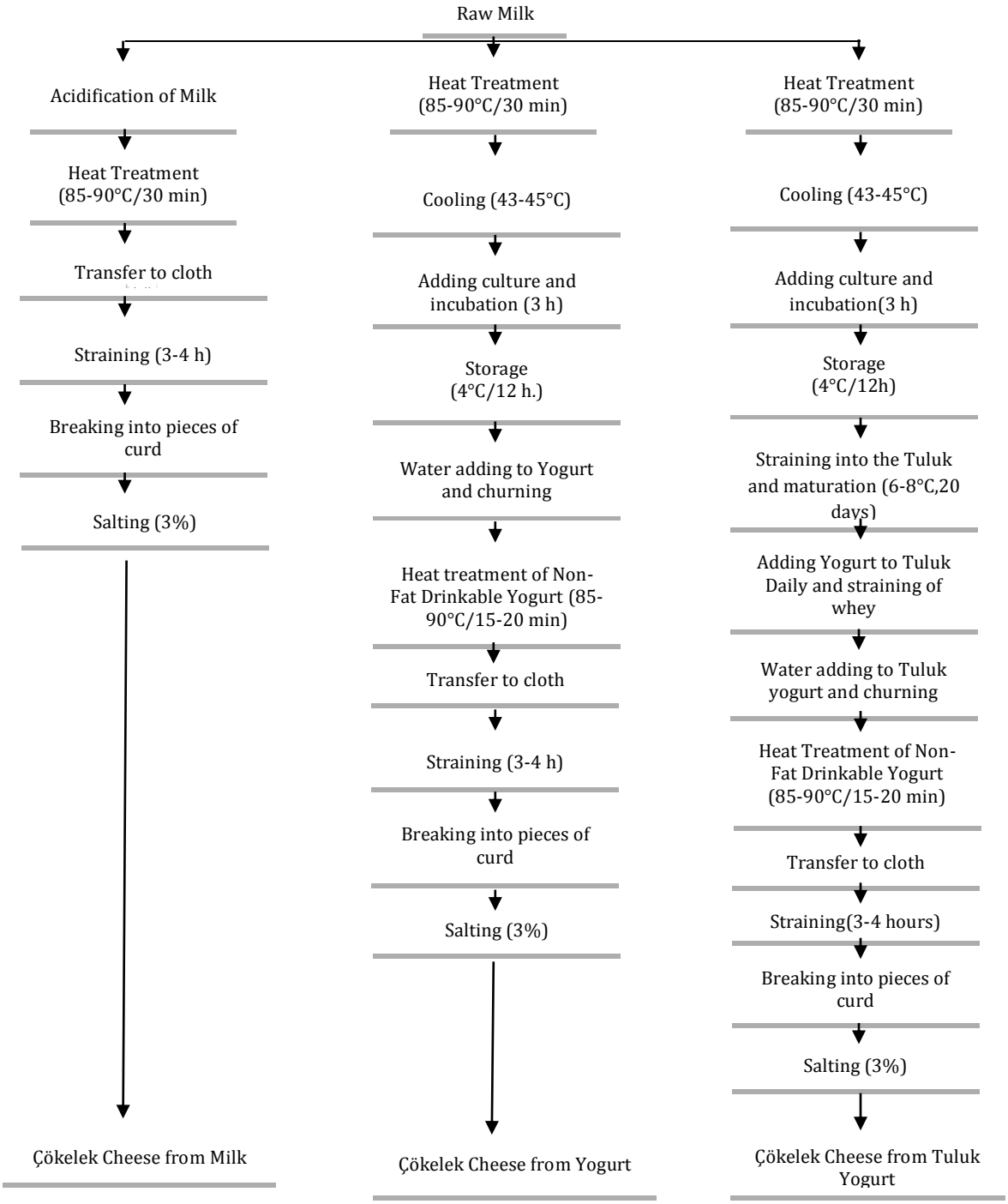

Figure 3. Production Steps of Çökelek Cheese made by different methods

between samples [21]. SPSS for Windows (version 20.0) statistical package program was used for the statistical analyses.

3. Results and Discussion

The flavor of fresh cheeses usually comes from the carbonyl compounds such as acetaldehyde and diacetyl, which are the result of lactose or citrate fermentation of starter bacteria used during production [22]. In this study, the intensity of aroma active compounds of freshly consumed and differently produced Çökelek cheeses were determined. Aroma active compounds determined in the Çökelek cheeses were presented in Table 2.

Seventeen active aroma compounds were detected in cheese samples. The specified aroma-active compounds including acid, aldehyde, ketone, ester, lactone, and sulphur type compounds. The two aroma components of the aroma components identified in the cheese leaves are not identifiable as chemical substances. The intensity of the common active aroma compounds of the samples produced by all three methods were also determined at different levels. The differences observed in the active aroma compounds of the Çökelek samples produced by different methods vary depending on the fermentation process and the method of production. It was determined that butyric acid (cheese, acid), Furaneol® (burned sugar) and sotolon (spice, burnt) were at higher intensity in comparison with other aroma active compounds in all Çökelek cheese samples (Table 2). The aroma intensity of butyric acid in MC was found to be lower (P<0.05) than YC. The aroma intensity of the butyric acid in Tuluk Çökelek was found to be similar to the density of the MC sample (P> 0.05). This differences can be based on hydrolysis rate of free fatty acids in Çökelek samples. It is well known butyric acid is formed by hydrolysis of free fatty acid and mostly identified in ripened cheeses [12, 15, 17].

Sotolon and Furaneol® are particularly susceptible to thermal degradation of sugars and Maillard reactions, both of which can be detected in heat-treated milk and dairy products (by thermal degradation of lactose). Fureneol® associated with caramel and / or burned sugar flavor [17, 24]. The intensity of Furaneol® in the samples of MC, YC, and TC was 4.38, 1.38, and 1.38, respectively. Furaneol® intensity of MC sample was higher than the other Çökelek samples (P<0.05). The intensity of the sotolon which is associated with the spice and burnt aroma was determined the highest in MC (3.5). There was no significant difference (P>0.05) between MC and YC samples in terms of *D*-decalactone (peach) (P<0.05). The intensity of this aroma in the TC sample was higher (1.08) compared with the MC an YC (P<0.05). This differences can be attribute to differences of the cheese making procedures.

Ethylbutyrate, ethylpentanoate and 2-phenylethanol (rose) were not detected in MC. However, the intensities of these aroma compounds were similar for YC and TC samples (P>0.05). Moreover, the intensity of hexanoic acid (sweaty, sour) in the YC sample was found to be higher than that in the TC sample (P<0.05). Conversely, 2-methyl butyric acid (sweaty, acid) and *p*-cresol (animal), which are

Table 2. Aroma active compounds of Çökelek cheeses

RI	Volatiles	Aroma Quality[a]	Aroma Intensity[b] (Mean± S.D) Çökelek Cheeses		
			MC	MC	MC
805	Dimetylsulfide	Sulphur	Nd	1.50±0.75	Nd
980	Unknown 1	Ester, apple	Nd	0.20±0.17	Nd
1051	Ethylbutyrate	Fruity, apple	Nd	0.50±0.00[a]	0.50±0.00[a]
1133	Ethylpentanoate	Bubble gum	Nd	0.38±0.21[a]	0.33±0.17[a]
1198	Ethylhexanoate	Fruity, sweet	Nd	0.38±0.21[a]	0.33±0.17[a]
1362	(Z)-1,5-octadien-3-one	Metallic, dirty	Nd	0.45±0.13[a]	0.25±0.03[b]
1501	Methional	Potatoes	1.45±0.38[b]	2.00±0.35	3.38±0.32[a]
1530	Butyric acid	Acid, sour	2.35±0.95[b]	4.13±0.19[a]	3.38±0.65[ab]
1559	2-methyl butyric acid	Sour, acid	Nd	Nd	2.25±0.65
1662	Phenyl acetaldehyde	Rose, flower	Nd	Nd	0.25±0.13
1769	2-acetyl 2-tiazoline	Popcorn	0.75±0.31	Nd	Nd
1859	2-phenylethanol	Rose, sour	Nd	1.25±0.65[a]	1.08±0.42[a]
1899	Hexanoic acid	Sweaty, sour	Nd	2.50±0.25[a]	0.75±0.28[b]
1964	Furaneol	Burnt sugar	4.38±0.94[a]	1.38±0.32[b]	1.38±0.21[b]
2055	Unknown 2	Fishy	0.50±0.00	Nd	Nd
2068	*p*-cresol	Barny	Nd	Nd	0.40±0.20
2145	Sotolon	Herb, burnt	3.50±0.56[a]	1.75±0.22[b]	1.88±0.32[b]
2249	D-decalactone	Peach, fruity	0.40±0.2[b]	0.50±0.31[b]	1.08±0.28[a]
2369	γ-dodecalactone	Sweet	1.13±0.45[b]	3.08±0.35[a]	1.63±0.21[b]

MC; Milk Çökelek, YC: Yogurt Çökelek, TC: Tuluk Çökelek RI: retention index on HP-INNOWAX column, [a]: aroma quality determined on olfactory port, [b] aroma intensity (10 point scale), S.D: standard deviation. Nd: Not Detected

expressed as the off-flavour in dairy products were detected only in MC. Unknown compound 6 was detected only in the TC sample. This unknown compound was associated with fishy aroma by GC-O panelists. Moreover, the unknow compound 2 that was associated with ester/apple aroma was detected at a significant level only in the YC sample. The active aroma active compounds of Çökelek cheese involve the esterification of ethyl butyrate, ethyl pentanoate, and ethyl hexanoate; and the degradation of methionine and 2-methyl butyric acid protein and amino acids with the degradation of butyric acid, hexanoic acid and lactone-derived decalactone, particularly fatty acids[18, 23, 24]. The 2-methyl butyric acid from the aroma components was found to be high only in the sample of TC. This was associated with the possibility that the fermentation can continue in tuluk and the proteins may be broken up at a later stage.

In general, the aroma detected in all three Çökelek samples is due to the active aroma components detected in many dairy products [17,18, 22, 24, 25]. In a study conducted by Ercan et al. [25], the intensity of butyric acid, decanoic acid (sour, candle), hexanoic acid, ethyl hexanoate (sour orange) and 2-butenedial (cheesy) aroma-active components in sepet cheese collected from the Aegean region were high. Avşar et al. [15] found that in the Erzincan tulum cheese, some of the Çökelek cheeses contained high levels of acetic acid (vinegar), butyric acid, 3-methyl butanoic acid (sweaty), propanoic acid (cheesy, sweaty), diacetyl (butter), hexanal, ethyl butanoate, ethyl hexanoate and methionine in the Erzincan Tulum cheese. Another study [26] reported that the intensity of diacetyl, butyric acid, hexanoic acid, ethyl butanoate, ethyl hexanoate, 3-methyl butyric acid (cheesecloth, oily) aromas was high in Gruyere, Manchego and Ragusano cheese ripened for 12 months.

The sensory properties determined by the panel members for the Çökelek cheeses were shown in Table 3. The aromatic terms specified for the Çökelek cheeses are "cooked", "whey", "creamy", "rancid", "fermented" and "yeasty". Taste terms are "sour", "salty", "sweet" and "umami". Significant differences were found between Çökelek samples in terms of cooked, creamy, fermented, yeast aromas and also sour, salty and sweet tastes. ($P<0.05$). According to this observations, the scores of cooked, creamy and sweet in MC were found to be a higher than the other Çökelek samples ($P<0.05$). Fermented aroma was perceived lower in MC samples than other samples ($P<0.05$), whereas the yeast aroma was perceived higher in TC samples than in other samples ($P<0.05$). These results probably based on the ripening of yogurt in Tuluk and its subsequent use in Çökelek cheese production.

Table 3. Sensory properties of Çökelek cheeses

Sensory properties	Çökelek Cheeses (Mean ± S.D)		
	MC	YC	TC
Cooked	4.23±0.24^{a}	2.46±0.22^{b}	2.68±0.28^{b}
Whey	2.70±0.25^{a}	2.65±0.22^{a}	2.78±0.32^{a}
Creamy	2.74±0.35^{a}	1.28±0.26^{b}	1.13±0.19^{b}
Rancid	0±0	1.04±0.26^{a}	0.70±0.26^{a}
Fermented	1.07±0.30^{b}	2.59±0.26^{a}	2.72±0.24^{a}
Yeast	0.04±0.04^{c}	0.73±0.32^{b}	1.50±0.36^{a}
Fruity	0.08±0.04^{b}	0.39±0.12^{a}	0±0
Sour	0.84±0.21^{b}	3.14±0.25^{a}	3.80±0.27^{a}
Salty	1.14±0.18^{b}	2.11±0.20^{a}	2.51±0.17^{a}
Sweet	2.03±0.31^{a}	0.89±0.13^{b}	0.82±0.13^{b}
Umami	0.08±0.05^{a}	0.10±0.06^{a}	0.18±0.02^{a}

MC; Milk Çökelek, YC: Yogurt Çökelek, TC: Tuluk Çökelek, S.D:standard deviation

TC samples were found to be saltier and sour than the other samples probably due to long fermentation stage of Tuluk yogurt. The sweetness intensity was determined in the Çökelek cheese produced from cow milk ($P<0.05$). The aroma and taste terms for the Çökelek cheeses are determined by panelists as the basic sensory terms in many dairy products, similar to aroma active ingredients [15, 18, 27]. Drake et al. [27] identified cooked, whey, creamy, sulphur, brothy, salty, sweet, sour and bitter taste characteristics in 11 different cheddar cheeses. In another study [28], sharp, milky, rennet, creamy and toasty aromatic properties were determined in Idiazabal, Manchego, Roncal and Zamorano cheeses produced in Spain. Leuven et al. [29] found creamy, fruity, nutty, pungent and animal aroma characteristics and sweet, salty sour and bitter (caffeine) taste characteristics in Gouda-type cheeses. The study determined that sweet, bitter flavor characteristics and fruity and nutty aromas were high in Gouda type cheeses made from raw milk and ripened in 6 weeks while the sour taste and sharp aroma were high in cheeses produced from pasteurized milk. The characteristic aroma and flavor characteristics of the Ezine cheese produced in Turkey are expressed as cooked, creamy, whey, rancid, goatish, animal, fermented, sour, salted and umami [30]. In addition to similar taste and aroma properties, sulphur, yeasty/ moldy, bitter (caffeine), acrid, nasal and tongue burner and tongue biting aroma and taste characteristics were determined in Erzincan Tulum cheese, while aroma and taste characteristics similar to Ezine cheese including sulphur, nutty, meat juice-like, mold and tongue biting sensory properties were determined in Kars Kaşar cheese [15].

Although there are many studies on the volatile components of traditional dairy products produced in Turkey, studies on the determination of the flavor components and their sensory properties had the highest effect on the aroma profile and a limited effect in that of the volatile components. Other widely consumed cheese varieties include white cheese, cheddar cheese, and Tulum cheese. In this study, it

was determined that the Çökelek cheeses made by using different production techniques and raw materials had a higher amount of active aroma components than those produced using milk and yogurt. In terms of sensory properties, it was determined that the MC samples had cooked, creamy and sweet characteristics, whereas the TC samples had a higher yeast aroma. The yogurt fermentation process, which is preserved in tuluk and produced by removing its serum, continues and the level of proteolysis changes. Similar studies in other traditional cheese types are also significant in terms of presenting the product quality and sensory qualities.

4. Conclusion

In this study, it was determined that sensory and aroma characterization of Çökelek cheese, which produced by different raw materials There were significant differences in profiles of aroma active compounds and sensory properties of Çökelek cheeses, and variations were observed among the cheeses based on the production methods. It was determined that Total 19 aroma-active compounds were found in the cheese samples. Ester type aroma active compounds were found in cheese samples at lower intensity while butyric acid, methional, γ-dodecalactone and Furaneol® major aroma-active compounds with higher intensity for Çökelek cheeses. Eleven sensory attributes were developed in Çökelek cheese by panel members. Cooked, Whey, Creamy and Sweet were the most characteristic terms for Çökelek cheese. Similar studies in other traditional cheese types are also significant in terms of presenting the product quality and sensory qualities.

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