

The Impact of Different Grape Pomaces from Winemaking Waste on the Ripening of İzmir Tulum Cheese: A Study on Fatty Acid Composition, Free Fatty Acids, and Volatile Compounds

Aysun ATALAY*¹ , Filiz YILDIZ AKGÜL¹ , Özer KINIK¹ 

¹Ege Üniversitesi, Agriculture Faculty, Department of Dairy Technology, İzmir, Türkiye

²Aydın Adnan Menderes Üniversitesi, Agriculture Faculty, Department of Dairy Technology Aydın, Türkiye

Abstract: This research aimed to examine the fatty acids, free fatty acids, and volatile compounds of İzmir Tulum cheeses ripened in brines prepared using different grape pomaces from wine production waste. For this purpose, four groups of İzmir Tulum cheese were produced: white wine waste: sultaniye grape pomace (W), red wine waste: petit-verdot grape pomace (R), red and white wine mixture waste: 1:1 mixture of sultaniye and petit-verdot (RW) and control: pomace-free group (C). Among the short chain fatty acids, butyric (C4), caproic (C6), caprylic (C8) and capric (C10) acids were determined in all cheese samples. Long chain fatty acids such as myristic (C14), palmitic (C16) and oleic (C18:1) acids were found in high amounts. The most abundant free fatty acids in all cheese samples were oleic acid (C18:1) followed by stearic acid. Butyric (C4) and caproic (C6) acid contents increased during storage and were highest in RW and R samples at the end of storage. A total of 32 volatile components (12 acids, 3 alcohols, 9 esters, 3 terpenes, 2 hydrocarbons, 3 other compounds) were detected in İzmir Tulum cheese samples during the ripening period. It was observed that the volatile components content varied according to the grape pomace type ($p<0,05$). Especially, the formation of volatile components was higher in red wine (R) and red-white (WR) blend waste samples and this difference was found to be statistically significant ($p<0,05$). It has been observed that red and mixed grape pomaces affect the taste and aroma of İzmir Tulum cheese during storage.

Keywords: Grape pomace, winemaking waste, fatty acids, volatile compound

Şarap Üretim Atıklarından Elde Edilen Farklı Üzüm Posalarının İzmir Tulum Peynirinin Olgunlaşmasına Etkisi: Yağ Asidi Bileşimi, Serbest Yağ Asitleri ve Uçucu Bileşikler Üzerine Bir Araştırma

Öz: Bu çalışmada, şarap üretim atıklarından elde edilen farklı üzüm posaları kullanılarak hazırlanan salamuralarda olgunlaştırılan İzmir Tulum peynirlerinin yağ asitleri, serbest yağ asitleri ve uçucu bileşenlerin incelenmesi amaçlanmıştır. Bu amaçla, dört grup İzmir Tulum peyniri üretilmiştir: beyaz şarap atığı: sultaniye üzüm posası (W), kırmızı şarap atığı: petit-verdot üzüm posası (R), kırmızı ve beyaz şarap atığı karışımı: sultaniye ve petit-verdot üzüm posalarının 1:1 karışımı (RW) ve kontrol: posa içermeyen grup (C). Kısa zincirli yağ asitlerinden bütirik (C4), kaproik (C6), kaprilik (C8) ve kaprik (C10) asitler tüm peynir örneklerinde tespit edilmiştir. Miristik (C14), palmitik (C16) ve oleik (C18:1) asitler gibi uzun zincirli yağ asitleri yüksek miktarlarda bulunmuştur. Tüm peynir örneklerinde en bol bulunan serbest yağ asitleri oleik asit (C18:1) ve ardından stearik asit olmuştur. Butirik (C4) ve kaproik (C6) asit içerikleri depolama sırasında artmış ve depolama sonunda RW ve R örneklerinde en yüksek seviyeye ulaşmıştır. İzmir Tulum peynirlerinde olgunlaşma sırasında toplam 32 uçucu bileşen (12 asit, 3 alkol, 9 ester, 3 terpen, 2 hidrokarbon, 3 diğer bileşikler) tespit edilmiştir. Oluşan uçucu bileşenlerin kullanılan üzüm posası türüne göre değiştiği gözlenmiştir ($p<0,05$). Özellikle kırmızı (R) ve kırmızı-beyaz (WR) karışım posalarını içeren örneklerde uçucu aroma bileşenlerinin oluşumunun daha yüksek olduğu ve bu farkın istatistiksel olarak önemli olduğu tespit edilmiştir ($p<0,05$). Kırmızı ve karışım üzüm posalarının İzmir Tulum peynirinde depolama süresince tat-aromayı etkilediği görülmüştür.

Anahtar Kelimeler: üzüm posası, şarapçılık atığı, yağ asitleri, uçucu bileşen

INTRODUCTION

The primary residual material generated from the winemaking process is known as grape pomace, which consists of the remnants left after pressing grapes (Kandyliş et al., 2021). This pomace primarily comprises grape skins, seeds, and stems. The proportion of grape pomace, which typically ranges from 15% to 30% of the total weight of crushed grapes, depends on factors such as grape variety, pressing methods, and fermentation techniques (Bordiga et al., 2019; García-Lomillo and Gonzalez-SanJos'e, 2017; Yu and Ahmedna, 2013). According to Organisation Internationale de la vigne et du vin (OIV, 2019), global production of grape pomace reached approximately 11.1 million tons in 2018. Such substantial quantities of by-products raise significant concerns within both industrial and

scientific communities due to the considerable environmental risks associated with their disposal. Around 82 million liters of wine were produced in Türkiye in 2021 (FAO 2023). Of this production, 20% is released as pulp. Therefore, 16,400 tons of waste was released in this production (Cabaroğlu, 2023).

Grape pomace has found a significant role in animal feed for over 40 years, garnering considerable attention from both scientific and industrial sectors in recent decades. Numerous

***Corresponding Author:** atalaraysun@gmail.com

This research was funded by Scientific Research Projects Department of Ege University (project no:22972).

The submitted date: December 15, 2023

The accepted date: December 25, 2023

studies have been conducted to assess the advantageous impacts of grape pomace on animals and animal-related goods (Bennato et al., 2020; Ianni & Martino, 2020). Additionally, a promising application of grape pomace involves fortifying various food products, imparting them with innovative properties. Recent literature highlights numerous studies examining the incorporation of grape derivatives into dairy, meat, cereal, and other food items (Frühbauerova et al., 2020; García-Lomillo and Gonzalez-SanJose, 2017).

Consumers are increasingly conscious of the direct impact of food on their health (Mollet and Rowland, 2002), and the dairy market is actively engaged in promoting health and wellness (Brockman and Beeren, 2011). Various biological activities associated with dietary fiber and polyphenols sourced from grape pomace have been documented, suggesting potential advantages in their utilization within dairy production processes and for enhancing product quality (Zhu et al., 2015). Factors such as environmental sustainability (Augustin et al., 2013) and effective waste management (Fontana et al., 2013) also significantly encourage the incorporation of non-dairy products as ingredients in the dairy industry.

Upon reviewing the literature, it is emphasized that new value-added product can be produced by using winemaking by-products (Kalli et al., 2018). An example of these is the use of black grape powder dried with different drying methods in yogurt production and so a functional food was produced (Demirkol, 2016). The study result on the phenolic content and in vitro antioxidant activity of solid by-products obtained from white and red wine production, it was found that winery by-products are very rich sources of antioxidant and polyphenols compared to other agricultural food solid wastes (Makris et al., 2007). In a study with PetitSuisse cheese produced with winery by-products, it was determined that it would be enriched with compounds effective on antioxidant activity (Deolindo et al., 2019). In many studies on pomace, it has been reported that pomace is an important source of phenolic substances and has high antioxidant activity in the study of Crespo and Brazinha (2010). As can be seen, grape pulp has been used in many dairy products in many studies. In these studies, functional properties, antioxidant capacity and phenolic substance contents of the product were emphasized. However, there is no study on how the addition of grape pomace affects fatty acids, free fatty acids and volatile components, which are important criteria in the formation of taste and aroma in cheese and consumer acceptability. İzmir Tulum cheese, which is one of the varieties of Tulum cheese, has an

important place among the cheeses produced in Türkiye (Akan, 2020). İzmir Tulum cheese is creamy white in color, but the production method, the type of milk and the amount of salt in the cheese affect the color. It is a semi-hard cheese in terms of its structure. In terms of pore structure, it is a full-fat cheese that contains many small pores and has a unique aroma and taste (Kamber, 2007).

The purpose of the present study was to examine the changes in FAs, FFAs and volatile components during storage of İzmir Tulum cheese in brine prepared using different grape pomaces from wine production waste.

MATERIALS AND METHODS

Materials

Raw cow's milk was sourced from Boyacıoğulları Ömür Dairy Products Co. Grape pomaces from winemaking waste (sultaniye, petit-verdot and a 1:1 v/v mixture grapes) utilized in the cheese brine were acquired from Sevilen Wine Inc. Fatty acid methyl esters (FAME) (Supelco 37 Component FAME Mix CRM47885), and individual free fatty acid standards (C4, C6, C8, C10, C12, C14, C16, C18, C 18:1, C 18:2, C 18:3) were procured from Sigma-Aldrich (Germany).

Methods

Production of İzmir Tulum Cheese

İzmir Tulum cheese was produced at Boyacıoğulları Ömür Dairy Products Co. (Tire, İzmir) in accordance with the operating process. Thermization (at 55°C for 1 minute) process was applied to the raw cow's milks. The milk was standardized to 45% fat in dry matter. Afterwards, the milk was cooled to fermentation temperature (36±4°C). Then, rennet (1/16000 force) and calcium chloride were added into the milk and incubated until the pH reached 6.20-6.40. After the appropriate clot firmness was achieved (45 minute), the clot breaking process was performed in the boiler and then was pressed. The end pH of pressing was 5.20-5.50. The filtered clot was cut both horizontally and longitudinally. In order to remove yellowing and bitter water from the cheeses, rock salt was sprinkled between cheeses. After dry salting, the cheeses were kept in cold storage for 2 days. Then the cheeses were transferred to 4.5 kg tins. 4 different specially prepared brines were added to the cheeses. The brine was prepared as follows: Three distinct brine solutions were created: Sultaniye grape pomace derived from white wine production waste, Petit-Verdot grape pomace from red wine production waste, and the 1:1 (w/w) blend of these two. The grape pomace was soaked in boiled and cooled water overnight (Figure 1). The brine's salt concentration was 12%.



Figure 1. Transferring the brine prepared with grape pomace onto Izmir Tulum cheese

The tins were then taken to the cold storage (4°C) of Ege University Faculty of Agriculture Dairy Technology Enterprise and ripened for 6 months. The closed tins were opened on the analysis days (1st, 90th and 180th days) and fatty acids, free fatty acids, volatile aroma compounds were performed on the cheese samples.

Fatty Acids of Izmir Tulum Cheeses

For the extraction of fatty acids from cheeses, the method proposed by Ackman (1998) was adopted. The fatty acid methyl esters (FAME) extracted were analyzed using a gas chromatograph (GC) (Agilent 7697A, Agilent Technologies, CA, USA) equipped with a flame ionization detector (FID). Separation of the FAME occurred on a capillary HP-FFAP column (J&W 19091F-433, Agilent Technologies; 30 m x 0.25 mm i.d.; 0.25 µm film thickness) with nitrogen as the carrier gas at a flow rate of 3 mL min⁻¹. The initial oven temperature was set at 100 °C and programmed to increase to 240 °C at a rate of 10 °C min⁻¹. Injection volume was 2 µL, an inlet temperature was 225 °C and a split ratio was 100:1. Identification of individual FAME was accomplished by comparing their retention times to those of a standard FAME mix (Supelco 37 Component FAME Mix CRM47885). The quantity of each FAMEs was expressed as a percentage of the total FAME. Each sample underwent two injections by the GC autosampler.

Free Fatty Acids of Izmir Tulum Cheeses

The extraction and quantification of free fatty acids (FFAs) followed the methods outlined by Deeth et al. (1983) with slight modifications as detailed by Guler (2008). An Agilent GC (Agilent 7697A, Agilent Technologies, USA) equipped with a capillary column (J&W 19091F – 433, Agilent Technologies, USA; 30 m x 0.25 mm i.d; 0.25 µm) was employed for analysis. The GC conditions were as follows: the GC oven temperature was programmed to increase from 120 to 230 °C at a rate of 10 °C min⁻¹, the injection volume was 2 µL, the injector and detector temperatures were set at

230 °C, and the split ratio was adjusted to 1:10. Nitrogen served as the carrier gas with a flow rate of 1 mL min⁻¹. Fatty acid standards purchased from Sigma were injected at concentrations of 50, 100, 150, 200, and 250 ppm for identification purposes. Results were expressed in parts per million (ppm), and each sample underwent analysis twice

Volatile compounds of Izmir Tulum Cheeses

Whetstone et al. (2003) method was used to injections into the gas chromatography system for the detection of volatile components in cheese samples on the 1st, 90th and 180th day, and for the identification of volatile components and determination of retention times.

Solid Phase Micro Extraction (SPME) method was used to extract volatile components found in cheeses (Stashenko and Martinez 2007). 50/30 µm Divinylbenzene/Carboxen/Polydimethylsiloxane (DVB/CAR/PDMS, Agilent, USA) fiber was used. The injection was made into Agilent brand GC 7890A (Agilent, USA) gas chromatograph, equipped with a mass spectrometer and flame ionization detector (FID). Volatile components separated on a DB Wax column (122-7032, Agilent Technologies, USA; 30 m x 0.25 mm i.d.; 0.25 µm film thickness) were determined by scanning in the 30-300 m/z range on a GC5975 C MSD mass spectrometer. Column temperature program in gas chromatography; It was applied as holding at 40°C for 5 minutes, heating with an increase of 10°C to 100°C, heating with an increase of 20°C to 200°C and keeping it at this temperature for 10 minutes. NIST/Flavournet libraries (2009) were searched to identify volatile components. The amounts of volatile components were calculated with the following formula, using the areas of 81 ppm 2-methyl-3-heptanone (Sigma-Aldrich, USA), which is the internal standard used during the extraction of the samples, and the areas of the volatile components obtained in gas chromatography.

Volatile Component Concentration (ppm)=[(% area of volatile component)/ (% area of internal standard)]*0.81

Statistical analyses

One Way Anova Variance analysis was applied in order to compare the characteristics of the ripening brine prepared using pomace from winemaking by-products, on certain days during the ripening of the cheeses. For this purpose, SPSS version 25.0 (SPSS Inc. Chicago, Illinois) statistical analysis package program was used. As a result of variance analysis, the significant data were tested at the p<0.05 level according to the Duncan multiple comparison test.

RESULTS AND DISCUSSION

Fatty Acids of cheese samples

The fatty acid values during the storage period of Izmir Tulum cheeses are given in Table 1. Among the short chain

Table 1. Fatty acids of İzmir Tulum cheeses (%)

Fatty Acids	Samples*	Ripening days		
		1	90	180
C ₄	C	1,54±0,01 ^{aA}	2,10±0,36 ^{aAB}	2,41±0,12 ^{aB}
	W	2,18±0,82 ^{aA}	2,42±0,10 ^{aA}	2,22±0,05 ^{aA}
	RW	1,65±0,07 ^{aA}	2,62±0,21 ^{aB}	2,37±0,02 ^{aB}
	R	1,63±0,08 ^{aA}	2,45±0,23 ^{aB}	2,35±0,06 ^{aB}
C ₆	C	1,09±0,00 ^{aA}	1,47±0,30 ^{aAB}	1,67±0,09 ^{aB}
	W	1,50±0,50 ^{aA}	1,71±0,09 ^{aA}	1,55±0,02 ^{aA}
	RW	1,17±0,05 ^{aA}	1,79±0,20 ^{aB}	1,66±0,02 ^{aB}
	R	1,16±0,05 ^{aA}	1,76±0,02 ^{aC}	1,62±0,03 ^{aB}
C ₈	C	2,73±0,13 ^{aA}	3,25±0,63 ^{aA}	3,67±0,20 ^{aA}
	W	3,57±1,14 ^{aA}	3,73±0,13 ^{aA}	3,46±0,08 ^{aA}
	RW	2,82±0,09 ^{aA}	3,89±0,38 ^{aB}	3,67±0,02 ^{aB}
	R	2,81±0,12 ^{aA}	3,85±0,10 ^{aB}	3,61±0,07 ^{aB}
C ₁₀	C	0,29±0,0 ^{aA}	0,37±0,07 ^{aA}	0,42±0,03 ^{aA}
	W	0,45±0,06 ^{bA}	0,20±0,29 ^{aA}	0,40±0,01 ^{aA}
	RW	0,31±0,01 ^{aA}	0,45±0,04 ^{aB}	0,43±0,01 ^{aB}
	R	0,31±0,01 ^{aA}	0,45±,01 ^{aC}	0,42±0,00 ^{aB}
C ₁₂	C	3,20±0,01 ^{aA}	3,61±0,78 ^{aA}	4,11±0,20 ^{aA}
	W	3,84±0,80 ^{aA}	4,07±0,02 ^{aA}	3,91±0,12 ^{aA}
	RW	3,31±0,08 ^{aA}	4,23±0,22 ^{aB}	4,09±0,02 ^{aB}
	R	3,31±0,07 ^{aA}	4,19±0,22 ^{aB}	4,09±0,08 ^{aB}
C ₁₄	C	11,53±0,35 ^{abA}	11,03±2,66 ^{aA}	13,37±0,37 ^{aA}
	W	12,09±0,23 ^{bA}	12,84±0,21 ^{aA}	12,99±0,55 ^{aA}
	RW	11,34±0,03 ^{aA}	12,83±0,73 ^{aB}	13,12±0,29 ^{aC}
	R	11,43±0,06 ^{aA}	12,71±0,06 ^{aB}	13,58±0,24 ^{aB}
C _{14:1}	C	0,98±0,16 ^{aB}	0,43±0,22 ^{aA}	0,80±0,08 ^{aAB}
	W	1,14±0,17 ^{aA}	0,67±0,21 ^{aA}	0,94±0,18 ^{aA}
	RW	1,15±0,01 ^{aA}	0,85±0,49 ^{aA}	0,65±0,26 ^{aA}
	R	0,88±0,02 ^{aA}	0,48±0,21 ^{aA}	0,62±0,30 ^{aA}
C ₁₅	C	1,20±0,02 ^{bA}	1,03±0,26 ^{aA}	1,18±0,00 ^{aA}
	W	1,09±0,00 ^{aA}	1,21±0,02 ^{aB}	1,21±0,01 ^{aB}
	RW	1,19±0,00 ^{bA}	0,79±0,58 ^{aA}	1,20±0,04 ^{aA}
	R	1,16±0,06 ^{abA}	1,23±0,06 ^{aA}	1,24±0,01 ^{aA}
C _{15:1}	C	0,36±0,03 ^{aA}	0,33±0,01 ^{aA}	0,37±0,00 ^{aA}
	W	0,51±0,18 ^{aA}	0,37±0,02 ^{aA}	0,38±0,02 ^{aA}
	RW	0,38±0,01 ^{aA}	0,81±0,56 ^{aA}	0,37±0,01 ^{aA}
	R	0,38±0,01 ^{aA}	0,37±0,23 ^{aA}	0,32±0,06 ^{aA}

Table 1 continue

Fatty Acids	Samples*	Ripening days		
		1	90	180
C ₁₆	C	33,94±0,04 ^{aA}	36,30±3,34 ^{aA}	32,43±0,43 ^{aA}
	W	32,93±1,98 ^{aA}	33,54±0,29 ^{aA}	32,70±1,13 ^{aA}
	RW	33,73±0,27 ^{aA}	32,64±0,61 ^{aA}	32,19±0,94 ^{aA}
	R	33,81±0,62 ^{aA}	32,76±0,46 ^{aA}	33,64±0,12 ^{aA}
C ₁₇	C	0,65±0,18 ^{bA}	0,88±0,04 ^{aA}	0,48±0,11 ^{bA}
	W	0,24±0,04 ^{aA}	0,64±0,38 ^{aA}	0,30±0,4 ^{aA}
	RW	1,11±0,13 ^{cB}	0,74±0,24 ^{aAB}	0,48±0,05 ^{abA}
	R	0,78±0,01 ^{cAB}	0,94±0,23 ^{ab}	0,39±0,01 ^{abA}
C ₁₈	C	11,21±0,01 ^{aB}	8,50±0,38 ^{aA}	9,23±0,31 ^{abA}
	W	10,35±1,26 ^{aA}	8,56±0,32 ^{aA}	10,09±0,05 ^{bA}
	RW	11,89±0,01 ^{aB}	8,74±0,63 ^{aA}	9,33±0,49 ^{aA}
	R	11,89±0,36 ^{aB}	8,41±0,55 ^{aA}	9,14±0,25 ^{aA}
C _{18:1n9c}	C	26,16±0,01 ^{aB}	24,02±0,08 ^{aA}	23,66±0,50 ^{aA}
	W	24,53±0,68 ^{aA}	23,66±0,18 ^{aA}	23,47±1,40 ^{aA}
	RW	25,05±0,93 ^{aA}	24,35±0,64 ^{aA}	23,87±0,24 ^{aA}
	R	25,88±0,34 ^{aB}	23,90±0,28 ^{aA}	23,65±0,11 ^{aA}
C _{18:2n6c}	C	0,40±0,04 ^{aA}	0,34±0,15 ^{aA}	0,46±0,07 ^{aA}
	W	0,47±0,08 ^{aA}	1,48±0,38 ^{bB}	0,47±0,01 ^{aA}
	RW	0,38±0,06 ^{aA}	0,76±0,44 ^{abA}	0,48±0,02 ^{aA}
	R	0,21±0,30 ^{aA}	0,62±0,35 ^{abA}	0,49±0,06 ^{aA}
C _{18:2n6t}	C	1,22±0,06 ^{aA}	1,38±0,51 ^{aA}	1,89±0,2 ^{aA}
	W	1,29±0,02 ^{aA}	1,49±0,05 ^{aA}	2,68±0,89 ^{aA}
	RW	1,36±0,51 ^{aA}	1,44±0,21 ^{aA}	2,40±1,14 ^{aA}
	R	1,14±0,00 ^{aA}	1,56±0,05 ^{ab}	1,78±0,01 ^{aC}
C _{18:3n6}	C	0,59±0,07 ^{aA}	0,75±1,06 ^{aA}	0,00±0,00 ^{aA}
	W	0,60±0,10 ^{ab}	0,52±0,01 ^{ab}	0,00±0,00 ^{aA}
	RW	0,62±0,00 ^{ab}	0,00±0,00 ^{aA}	0,00±0,00 ^{aA}
	R	0,60±0,08 ^{ab}	0,00±0,00 ^{aA}	0,00±0,00 ^{aA}
C _{18:3n3}	C	0,76±0,03 ^{aC}	0,19±0,01 ^{aA}	0,31±0,00 ^{ab}
	W	0,77±0,37 ^{aA}	0,25±0,12 ^{aA}	0,28±0,10 ^{aA}
	RW	0,77±0,04 ^{ab}	0,18±0,07 ^{aA}	0,33±0,03 ^{aA}
	R	0,88±0,16 ^{ab}	0,36±0,22 ^{aAB}	0,212±0,10 ^{aA}
C ₂₃	C	1,19±0,47 ^{aA}	1,27±0,34 ^{aA}	1,40±0,23 ^{aA}
	W	1,33±0,35 ^{aA}	1,11±0,14 ^{aA}	1,26±0,25 ^{aA}
	RW	0,84±0,11 ^{aA}	1,17±0,01 ^{aAB}	1,61±0,35 ^{ab}
	R	0,98±0,35 ^{aA}	1,39±0,06 ^{aA}	1,27±0,07 ^{aA}

Table 1 continue

Fatty Acids	Samples*	Ripening days		
		1	90	180
C ₂₄	C	0,99±0,21 ^{aA}	2,73±1,10 ^{aA}	2,15±0,03 ^{bA}
	W	1,14±0,11 ^{aA}	1,29±0,32 ^{aA}	1,69±0,21 ^{aA}
	RW	0,92±0,02 ^{aA}	1,73±0,56 ^{aA}	1,76±0,01 ^{abA}
	R	0,78±0,22 ^{aA}	2,58±0,98 ^{aA}	1,58±0,24 ^{aA}
SFA (Saturated fatty acids)	C	69,55±0,22 ^{aA}	72,55±0,69 ^{aB}	72,50±0,97 ^{aB}
	W	70,69±0,74 ^{aA}	71,56±0,56 ^{aA}	71,77±2,34 ^{aA}
	RW	70,28±0,41 ^{aA}	71,62±0,31 ^{aA}	71,90±1,10 ^{aA}
	R	70,03±0,19 ^{aA}	72,71±0,79 ^{aB}	72,92±0,30 ^{aB}
USFA (Unsaturated fatty acids)	C	30,45±0,22 ^{aB}	27,45±0,69 ^{aA}	27,50±0,97 ^{aA}
	W	29,31±0,74 ^{aA}	28,44±0,56 ^{aA}	28,23±2,34 ^{aA}
	RW	29,72±0,41 ^{aA}	28,38±0,31 ^{aA}	28,10±1,10 ^{aA}
	R	29,97±0,19 ^{aB}	27,29±0,79 ^{aA}	27,08±0,30 ^{aA}
MUFA (Mono unsaturated fatty acids)	C	27,50±0,18 ^{aB}	24,79±0,30 ^{aA}	24,84±0,58 ^{aA}
	W	26,18±0,33 ^{aA}	24,70±0,36 ^{aA}	24,80±1,55 ^{aA}
	RW	26,59±0,94 ^{aA}	26,01±0,41 ^{bA}	24,89±0,03 ^{aA}
	R	27,14±0,34 ^{aB}	24,75±0,16 ^{aA}	24,58±0,25 ^{aA}
PUFA (Poly unsaturated fatty acids)	C	2,96±0,03 ^{aA}	2,66±0,39 ^{aA}	2,66±0,39 ^{aA}
	W	3,13±0,41 ^{aA}	3,74±0,20 ^{aA}	3,43±0,79 ^{aA}
	RW	3,14±0,52 ^{aA}	2,37±0,72 ^{aA}	3,21±1,13 ^{aA}
	R	2,83±0,53 ^{aA}	2,54±0,62 ^{aA}	2,49±0,05 ^{aA}
SFA/USFA	C	2,28±0,02 ^{aA}	2,64±0,09 ^{aB}	2,64±0,12 ^{aB}
	W	2,41±0,09 ^{aA}	2,52±0,07 ^{aA}	2,55±0,29 ^{aA}
	RW	2,37±0,05 ^{aA}	2,52±0,04 ^{aA}	2,56±0,14 ^{aA}
	R	2,34±0,02 ^{aA}	2,67±0,11 ^{aB}	2,69±0,04 ^{aB}
n6/n3	C	2,13±0,06 ^{aA}	8,75±2,80 ^{aB}	7,68±1,15 ^{aAB}
	W	2,63±1,36 ^{aA}	13,59±7,56 ^{aA}	12,55±7,47 ^{aA}
	RW	2,27±0,86 ^{aA}	12,85±1,66 ^{aB}	8,85±4,28 ^{aAB}
	R	1,54±0,06 ^{aA}	7,13±3,33 ^{aA}	11,57±5,38 ^{aA}
Total ω-3	C	0,76±0,03 ^{aC}	0,19±0,01 ^{aA}	0,31±0,00 ^{aB}
	W	0,77±0,37 ^{aA}	0,25±0,12 ^{aA}	0,28±0,10 ^{aA}
	RW	0,77±0,04 ^{aB}	0,18±0,07 ^{aA}	0,33±0,03 ^{aA}
	R	0,88±0,16 ^{aB}	0,36±0,22 ^{aAB}	0,22±0,10 ^{aA}
Total ω-6	C	1,22±0,06 ^{aA}	1,38±0,51 ^{aA}	1,89±0,32 ^{aA}
	W	1,29±0,02 ^{aA}	1,49±0,05 ^{aA}	2,68±0,89 ^{aA}
	RW	1,36±0,51 ^{aA}	1,44±0,21 ^{aA}	2,40±1,14 ^{aA}
	R	1,14±0,00 ^{aA}	1,56±0,05 ^{aB}	1,78±0,01 ^{aC}

Table 1 continue

Fatty Acids	Samples*	Ripening days		
		1	90	180
Total ω -9	C	26,16±0,01 ^{aB}	24,02±0,08 ^{aA}	23,66±0,50 ^{aA}
	W	24,53±0,68 ^{aA}	23,66±0,18 ^{aA}	23,47±1,40 ^{aA}
	RW	25,05±0,93 ^{aA}	24,35±0,64 ^{aA}	23,87±0,24 ^{aA}
	R	25,88±0,34 ^{aB}	23,90±0,28 ^{aA}	23,65±0,11 ^{aA}

*: C: control group; W: white wine waste; RW: red and white wine waste (1:1 w/w); R: red wine waste

a, b, c, d: There are significant differences between the means marked with different letters in the same column ($p < 0.05$).

A, B, C, D, E: There are significant differences between the averages marked with different letters on the same line ($p < 0.05$)

fatty acids, butyric (C4), caproic (C6), caprylic (C8) and capric (C10) acids were determined in all cheese samples. Long chain fatty acids such as myristic (C14), palmitic (C16) and oleic (C18:1) acids were found in high amounts. Fatty acids are effective in the formation of taste and aroma (Tomar et al., 2020). The formation of short and medium chain fatty acids increases on the 90th day of ripening, but decreases slightly towards the end of ripening. The same findings were obtained in another study with Tulum cheese (Tomar et al., 2020). In addition, the difference between cheese samples in terms of butyric (C4), caproic (C6), caprylic (C8), capric (C10) and lauric (C12) acid contents of short and medium chain fatty acids was statistically insignificant ($p < 0.05$).

During the 180-day ripening period, myristic acid (C14) values of all cheese samples increased compared to the first day. The highest myristic acid value during the ripening period was 13.58% in the R sample on day 180 ($p < 0.05$).

It was observed that the saturated fatty acid ratio of the cheese samples increased during the ripening period and the highest saturated fatty acid ratio was obtained in the R sample on day 180. The difference between the samples was found to be insignificant ($p > 0.05$).

At the end of the ripening period, unsaturated fatty acids values of all cheese samples decreased compared to the beginning. The highest unsaturated fatty acids value was determined in the control group (C) on the first day of ripening and in the white grape pulp (W) sample on the last day. At the end of ripening, monounsaturated fatty acids values of all samples decreased compared to the beginning of ripening. On the 180th day of ripening, monounsaturated fatty acids values of all cheese samples were close to each other ($p > 0.05$). During ripening, polyunsaturated fatty acids values of all samples (except sample C) increased compared to the first day of ripening. However, the difference between the samples was statistically insignificant ($p > 0.05$).

It has been reported that cheese, which is one of the staple foods of humans, is rich in omega-3 and omega-6 polyunsaturated fatty acids in ideal amounts for health. Omega-3, omega-6 and omega-9 fatty acids are known to have positive effects on health such as brain development,

prevention of coronary heart disease and strengthening of the immune system (Kılıçalp and Yücel, 2020). The highest average omega-3 value during the ripening period was found in sample R (0.49%). The highest omega-6 value was detected in the W sample with 2.68% on the 180th day of ripening. It was observed that omega-9 values decreased at the end of ripening compared to the beginning.

In the study conducted by Akarca (2019), according to the fatty acid distribution of Tulum cheese samples, the most abundant fatty acid during the 90-day ripening period was palmitic acid with 28.98%, followed by oleic acid with 25.38% and myristic acid with 13.35%. It was reported that palmitic acid (C16) had the highest value with 29.86-28.11% according to the fatty acid results of Tulum cheeses (Sert et al., 2014). This was followed by oleic acid, stearic acid and myristic acid respectively. The fatty acid values obtained in this study are similar to the literature studies.

Free Fatty Acids of İzmir Tulum Cheeses

The amount of FFAs is an important parameter indicating the degree of lipolysis in cheese. In this context, it is used to determine lipolytic activity (Şengül et al., 2014). FFAs values of the samples and their changes during ripening are given in Table 2. The most abundant FFAs in all samples were oleic acid (C18:1) followed by stearic acid. Butyric (C4) and caproic (C6) acid contents increased during storage and were highest in RW and R samples at the end of storage. Although the caprylic (C8) and capric (C10) acid content of the control group was higher than the other samples, it was statistically similar to R and RW samples ($p > 0.05$). It was observed that the formation of short chain free fatty acids was higher in grape pomace-added samples, especially in red grape pomace-added samples. Butyric and caproic acids are predominantly situated at the sn-3 position and the sn-1 and sn-3 positions, respectively. The outer esters bond of tri- or diacylglycerides are mostly hydrolyzed by the lipases involved in cheese ripening (Atasoy and Türkoğlu 2009). Recent studies have shown that short-chain free fatty acids have anti-inflammatory, antitumor and antimicrobial effects, especially in terms of colon health (Tan et al., 2014, Silva et al., 2020).

Table 2. Free fatty acids of cheese samples, mg/kg

Fatty acids	Samples*	Ripening Days		
		1	90	180
C4	C	59,91±0,67 ^{aA}	113,53±1,32 ^{aAB}	131,88±33,52 ^{abB}
	W	85,16±17,88 ^{aA}	153,86±12,08 ^{ab}	113,12±23,91 ^{aAB}
	RW	82,40±20,70 ^{aA}	116,61±94,36 ^{aA}	194,20±0,04 ^{ba}
	R	68,96±14,43 ^{aA}	156,67±10,06 ^{ab}	157,56±28,15 ^{abB}
C6	C	58,46±0,98 ^{aA}	52,62±28,45 ^{aA}	70,73±26,58 ^{abA}
	W	66,11±11,75 ^{ab}	59,16±25,70 ^{ABa}	27,37±6,41 ^{aA}
	RW	61,67±4,32 ^{aA}	68,49±48,36 ^{aA}	87,74±2,11 ^{ba}
	R	60,97±2,57 ^{abB}	37,36±4,74 ^{aA}	72,79±18,83 ^{abB}
C8	C	22,48±2,25 ^{ca}	46,42±12,22 ^{ab}	43,24±5,82 ^{aA}
	W	19,53±0,37 ^{abA}	33,77±4,96 ^{aCD}	36,09±2,75 ^{aD}
	RW	16,59±1,36 ^{abA}	27,71±12,27 ^{abB}	37,49±6,26 ^{ab}
	R	15,34±0,44 ^{aA}	21,794,70 ^{aA}	35,31±3,68 ^{ab}
C10	C	51,93±0,95 ^{ca}	79,57±9,31 ^{ab}	77,76±4,16 ^{bb}
	W	45,68±1,16 ^{ba}	67,11±4,72 ^{ab}	45,52±8,61 ^{aA}
	RW	36,35±2,93 ^{aA}	52,33±21,29 ^{abB}	69,13±10,97 ^{baB}
	R	34,36±1,67 ^{aA}	45,32±10,92 ^{aA}	68,14±3,12 ^{bb}
C12	C	77,61±5,25 ^{ca}	110,86±7,53 ^{bc}	102,64±5,66 ^{bbC}
	W	66,03±3,97 ^{ba}	91,53±6,18 ^{abC}	68,56±8,77 ^{aA}
	RW	51,20±3,15 ^{aA}	70,73±25,61 ^{abAB}	91,98±12,73 ^{abB}
	R	50,40±2,04 ^{aA}	62,05±8,72 ^{abB}	91,47±2,64 ^{abC}
C14	C	282,25±38,30 ^{ba}	505,20±88,87 ^{bb}	337,87±4,48 ^{aA}
	W	239,41±6,08 ^{abA}	332,89±10,15 ^{aC}	267,65±27,37 ^{abB}
	RW	197,97±7,95 ^{aA}	263,24±59,71 ^{aAB}	299,83±45,13 ^{ab}
	R	198,55±5,40 ^{abA}	263,41±30,83 ^{abC}	306,155±2,45 ^{aC}
C16	C	1196,19±135,55 ^{ba}	2428,92±434,23 ^{bb}	1302,83±41,26 ^{aA}
	W	1047,19±14,50 ^{abA}	1533,60±69,31 ^{ab}	1233,50±164,58 ^{aA}
	RW	917,40±14,88 ^{aA}	1180,54±81,62 ^{ab}	1102,70±114,13 ^{aAB}
	R	927,81±17,04 ^{aA}	1308,36±110,90 ^{aC}	1170,45±90,32 ^{abC}
C18:0	C	293,08±311,41 ^{aA}	718,18±92,23 ^{ba}	457,27±145,81 ^{aA}
	W	476,45±3,33 ^{aA}	734,89±14,66 ^{bc}	603,21±57,18 ^{ab}
	RW	343,25±134,93 ^{abB}	566,50±24,08 ^{aC}	239,42±31,79 ^{aA}
	R	235,65±11,64 ^{aA}	654,54±38,34 ^{abB}	388,10±256,04 ^{abB}
C18:1	C	1884,01±190,18 ^{ba}	2980,17±1454,33 ^{aA}	2045,96±98,17 ^{aA}
	W	1603,96±32,89 ^{aA}	2401,99±24,79 ^{aC}	2062,88±151,60 ^{ab}
	RW	1466,31±18,32 ^{aA}	1926,68±66,35 ^{ab}	1652,37±202,54 ^{aA}
	R	1494,80±7,48 ^{aA}	2147,03±118,80 ^{aC}	1865,16±245,39 ^{abC}
C18:2	C	22,49±1,44 ^{aA}	435,92±59,81 ^{ab}	634,82±9,66 ^{abBC}
	W	27,31±1,09 ^{ba}	732,61±87,53 ^{bb}	690,56±9,35 ^{bb}
	RW	28,24±2,33 ^{ba}	634,34±48,61 ^{bb}	548,25±32,76 ^{ab}
	R	26,96±0,46 ^{ba}	572,23±64,45 ^{abB}	596,52±82,59 ^{abB}

*: C: control group; W: white wine waste; RW: red and white wine waste (1:1 w/w); R: red wine waste

a, b, c, d: There are significant differences between the means marked with different letters in the same column (p<0.05).

A, B, C, D, E: There are significant differences between the averages marked with different letters on the same line (p<0.05)

Myristic (C12), lauric (C14), palmitic (C16) and stearic (C18) acids, which are medium and long chain saturated fatty acids, were found to be higher in the C sample. Although there was no statistically significant difference between the samples, the lower level of saturated fatty acids in the grape pulp-added samples is an important result for healthy nutrition and cardiovascular stiffness. The oleic and linoleic acid contents (long-chain unsaturated fatty acids) in cheeses reached the highest value on the 90th day of storage and were found to be higher than at the beginning, even if their amounts decreased at the end of storage. Despite the quantitative importance of medium and long-chain FFAs, they are not the main contributors to cheese flavour (Atasoy and Türkoğlu, 2008)

Kırdar and Atamer (2021) reported that acetic, butyric, caproic, caproic, caprylic and capric acids showed the fastest increase in ripening time in a study they conducted with Tulum cheese. Short and medium chain fatty acids are generally hydrolyzed by lipoprotein lipase (Karakus et al., 2022). Tekin and Güler (2021) reported that oleic acid was the highest in terms of FFAs in Tulum cheese samples ripened with different packaging materials, followed by stearic, myristic, and capric acid. Arslaner and Bakırcı (2016) determined that myristic acid, palmitic acid, stearic acid and oleic acid were the highest in Tulum cheese samples during the ripening period.

Volatile Components of İzmir Tulum Cheese

Aroma is the most important of factors that contribute to the development of cheese flavor. The formation of volatile compounds in Tulum cheese is a result of microbial and biochemical activities during the storage period. These activities result from various chemical mechanisms such as lactose and citrate metabolism, amino acid catabolism, lipid breakdown and non-enzymatic transformations (Avşar et al. 2011).

A total of 32 volatile components (12 acids, 3 alcohols, 9 esters, 3 terpenes, 2 hydrocarbons, 3 other compounds) were detected in İzmir Tulum cheese during the ripening period. The values of these volatile components and their changes during storage are shown in Table 3. Hexanoic and butanoic acid were the most detected acids in İzmir Tulum cheeses, followed by heptanoic acid. Although a few acids (n-Hekza dekanic acid) decreased as storage progressed, they generally increased. The difference between storage days and samples was significant ($p < 0.05$). Especially in the R sample ripened in brine with the addition of red wine waste, the amount of volatile acids was higher and this difference was statistically significant ($p < 0.05$).

Çakır et al. (2016) reported that acetic, butyric, and pentanoic acids are the primary acids found in Erzincan Tulum cheese samples. During the ripening process of Erzincan Tulum cheese, Çakır et al. (2016) also identified

propanoic, 2-methyl propanoic, hexanoic, heptanoic, octanoic, and decanoic acids. Similar acids were detected in Tulum cheeses by Hayaloğlu et al. (2007). Additionally, hexanoic acid was highlighted as a significant flavor component in Blue-type cheeses. However, these acids not only serve as aroma compounds themselves but also act as precursors for other compounds, including methyl ketones, alcohols, lactones, aldehydes, and esters (Çakmakçı et al. 2013).

The formation of alcohols occurs through lactose metabolism, reduction of methyl ketones, and breakdown of linoleic or linolenic acids (Hayaloğlu et al. 2007). The highest alcohol determined in Tulum cheeses was phenylethyl alcohol in the R sample on Day 180. Again, it was observed that the amount of alcohol in red wine waste and red-white mixture wine waste samples was higher than the other samples ($p < 0.05$). In the study conducted by Hayaloglu and Karabulut (2013), ethanol was found as the main alcohol in Tulum cheese and other cheese samples, followed by 2-butanol and 3-methyl-1-butanol, respectively. Phenylethyl alcohol is associated with floral flavors in cheese and is usually synthesized from styrene, toluene or methyl ethyl acetate (Whetstine et al. 2005).

Another group that affects taste and aroma in cheeses is esters. Esters are obtained from esterification reactions occurring between acids, and primary and secondary alcohols derived from lactose fermentation or from amino acid catabolism (Atasoy et al. 2013).

Methyl and ethyl ester of octanoic acid and ethyl ester of hexanoic acid were detected more in R and WR samples compared to other samples ($p < 0.05$). In a study, Çakır et al. (2016) reported that esters were the most abundant of the volatile compounds determined in Erzincan Tulum cheeses and thirty-one esters were detected during the ripening period of the cheeses. It was stated that methyl esters were the most abundant ester type followed by ethyl esters. Ethyl esters were also found to be the main esters found in Tulum cheeses by other researchers (Hayaloğlu et al. 2007, Hayaloğlu and Karabulut 2013).

Terpenes are one of the most important compounds affecting the quality and volatile components in dairy products. However, it is stated that there is not enough information on whether terpenes have a significant sensory effect on flavor. The formation of terpenes is related to animal nutrition (Bontinis et al. 2012). Their amount increases according to the amount of green feed eaten by the animals. In this study, D-limonene was determined in almost all samples. Their amounts decreased during storage. The D-limonene contents of WR and R samples on Day 1 were different and greater than the control C and W samples ($p < 0.05$). P-cymene and o-cymene were not detected in C, W and R samples, but were detected in WR sample.

Table 3. Volatile compounds of cheese samples ($\mu\text{g/g}$)

Acids	Samples	Ripening days		
		1	90	180
Benzoic acid	C*	0,08±0,00 ^{aA}	0,35±0,09 ^{abB}	0,35±0,09 ^{aB}
	W	0,61±0,74 ^{aA}	0,23±0,03 ^{aA}	0,39±0,18 ^{aA}
	RW	0,28±0,13 ^{aA}	0,25±0,01 ^{abA}	0,83±0,05 ^{bB}
	R	0,09±0,06 ^{aA}	0,48±0,13 ^{bAB}	0,69±0,22 ^{abB}
Butanoic acid	C	0,66±0,06 ^{aA}	ND	9,14±1,38 ^{aB}
	W	2,36±2,03 ^{aA}	ND	ND
	RW	2,13±1,00 ^{aB}	ND	ND
	R	2,26±0,26 ^{aA}	ND	41,67±7,14 ^{bB}
Dodekanoic acid	C	0,11±0,07 ^{abA}	0,38±0,26 ^{aA}	0,90±0,38 ^{aA}
	W	ND	0,47±0,09 ^{aAB}	1,22±0,57 ^{aB}
	RW	0,36±0,17 ^{bA}	0,44±0,05 ^{aA}	1,88±0,32 ^{aB}
	R	0,29±0,06 ^{bA}	0,78±0,14 ^{aA}	1,17±1,66 ^{aA}
Hekzanoic acid	C	1,30±0,48 ^{aA}	6,91±1,09 ^{bAB}	10,46±3,15 ^{abB}
	W	4,74±3,40 ^{aA}	6,86±1,21 ^{bA}	17,12±8,07 ^{bA}
	RW	4,92±2,32 ^{aB}	ND	ND
	R	7,15±0,49 ^{aA}	ND	93,63±4,09 ^{cB}
Heptanoic acid	C	0,06±0,08 ^{aA}	ND	0,18±0,26 ^{aA}
	W	0,02±0,03 ^{aA}	ND	0,51±0,24 ^{aB}
	RW	0,10±0,05 ^{aB}	ND	ND
	R	0,10±0,02 ^{aA}	ND	21,90±29,26 ^{aA}
n-Dekanoic acid	C	0,29±0,14 ^{aA}	1,06±0,10 ^{aB}	1,97±0,36 ^{aC}
	W	1,20±0,81 ^{aA}	1,145±0,20 ^{aA}	6,98±3,29 ^{aA}
	RW	1,11±0,52 ^{aA}	1,59±0,29 ^{aA}	5,70±2,71 ^{aA}
	R	0,81±0,07 ^{aA}	5,30±0,87 ^{bA}	5,95±3,36 ^{aA}
n-Hekza dekanolic acid	C	0,56±0,79 ^{aA}	3,00±2,05 ^{aA}	5,64±6,26 ^{aA}
	W	8,79±12,06 ^{aA}	2,79±0,01 ^{aA}	5,57±2,63 ^{aA}
	RW	4,49±2,11 ^{aA}	2,70±0,25 ^{aA}	7,07±1,68 ^{aA}
	R	1,70±0,36 ^{aA}	6,92±3,75 ^{aA}	7,24±10,24 ^{aA}
Nonanoic acid	C	0,11±0,13 ^{abA}	0,39±0,25 ^{aA}	0,31±0,08 ^{aA}
	W	0,05±0,07 ^{aA}	0,11±0,02 ^{aA}	0,29±0,14 ^{aA}
	RW	ND	0,39±0,23 ^{aA}	0,77±0,67 ^{abA}
	R	0,26±0,01 ^{bA}	0,26±0,21 ^{aA}	1,48±0,26 ^{bB}
Octanoic acid	C	0,478±0,17 ^{aA}	ND	4,30±1,81 ^{bB}
	W	1,12±1,14 ^{aA}	2,34±0,43 ^{bA}	ND
	RW	ND	ND	ND
	R	1,44±0,09 ^{aB}	4,40±0,54 ^{cC}	ND
9-Dekenoic acid	C	0,04±0,06 ^{aAB}	ND	0,15±0,04 ^{aB}
	W	ND	ND	ND
	RW	ND	ND	0,48±0,27 ^{aA}
	R	ND	ND	0,49±0,27 ^{aA}

Table 3 continue

Acids	Samples	Ripening days		
		1	90	180
Palmitoleic acid	C	0,03±0,05 ^{aA}	ND	ND
	W	ND	ND	ND
	RW	ND	ND	ND
	R	ND	ND	ND
Pentanoic acid	C	0,01±0,01 ^{aA}	0,04±0,05 ^{aA}	0,10±0,14 ^{aA}
	W	0,01±0,01 ^{aA}	ND	0,25±0,12 ^{aB}
	RW	ND	ND	ND
	R	ND	ND	ND
Alcohols				
Benzil alcohol	C	ND	ND	ND
	W	0,03±0,04 ^{aAB}	ND	0,21±0,10 ^{aB}
	RW	ND	ND	ND
	R	ND	ND	ND
Feniletil alcohol	C	ND	ND	0,07±0,09 ^{aA}
	W	0,22±0,31 ^{abA}	0,59±0,11 ^{abA}	0,92±0,43 ^{abA}
	RW	1,19±0,56 ^{bA}	1,34±0,54 ^{bA}	3,48±0,62 ^{abB}
	R	2,39±0,47 ^{cA}	3,12±0,56 ^{cA}	6,97±4,58 ^{bA}
Silanediol, dimetil-	C	0,13±0,19 ^{aA}	0,83±1,17 ^{aA}	ND
	W	0,24±0,34 ^{aA}	ND	ND
	RW	1,73±0,81 ^{bB}	ND	ND
	R	1,83±0,28 ^{aB}	ND	ND
Esters				
Butanoic acid, etil ester	C	ND	ND	ND
	W	ND	10,20±1,75 ^{bB}	ND
	RW	0,36±0,17 ^{bA}	15,45±0,97 ^{cB}	33,13±5,7 ^{bC}
	R	0,49±0,03 ^{bA}	19,66±2,99 ^{cB}	ND
Tris(trimethylsilyl) ester	C	ND	ND	ND
	W	ND	ND	0,17±0,08 ^{aB}
	RW	ND	ND	1,30±1,84 ^{aA}
	R	ND	ND	ND
Decanoic acid, etil ester	C	ND	ND	0,37±0,03 ^{aB}
	W	ND	0,92±,12 ^{bB}	ND
	RW	0,24±0,11 ^{aA}	ND	9,25±3,33 ^{abB}
	R	0,22±0,02 ^{aA}	ND	16,77±5,85 ^{bA}
Dekanoic acid , methylester	C	ND	0,27±0,22 ^{aA}	ND
	W	ND	ND	ND
	RW	0,22±0,11 ^{bA}	1,74±0,65 ^{bB}	ND
	R	0,18±0,01 ^{bB}	ND	ND
Heptanoic acid, etil ester	C	ND	ND	ND
	W	ND	0,15±0,02 ^{bAB}	0,51±0,24 ^{bB}
	RW	ND	0,28±0,07 ^{cB}	ND
	R	ND	0,48±0,06 ^{dB}	1,48±0,38 ^{bB}

Table 3 continue

Acids	Samples	Ripening days		
		1	90	180
Hekzanoic acid, etil ester	C	0,06±0,09 ^{aA}	ND	3,37±0,27 ^{aB}
	W	ND	13,40±1,31 ^{bAB}	37,63±17,74 ^{bB}
	RW	2,01±0,95 ^{aA}	36,12±3,73 ^{bB}	ND
	R	ND	ND	ND
Hekzanoic acid, methylester	C	ND	4,11±0,11 ^{bC}	3,13±0,00 ^{bB}
	W	ND	2,58±0,10 ^{abB}	ND
	RW	ND	ND	ND
	R	ND	51,81±2,23 ^{cB}	ND
Octanoic acid, etilester	C	ND	ND	1,16±0,42 ^{aB}
	W	ND	4,59±0,50 ^{bB}	ND
	RW	ND	ND	42,55±15,4 ^{abB}
	R	ND	ND	67,82±28,2 ^{bB}
Octanoic acid, metylester	C	1,16±0,42 ^{aA}	ND	ND
	W	ND	ND	21,80±10,28 ^{bB}
	RW	42,55±15,38 ^{bA}	11,59±3,75 ^{bB}	ND
	R	67,82±28,20 ^{abA}	13,94±0,94 ^{bB}	ND
Terpens				
D-Limonen	C	0,25±0,36 ^{aA}	1,30±0,76 ^{aA}	0,34±0,47 ^{aA}
	W	ND ^{aA}	11,07±6,10 ^{aA}	1,57±0,74 ^{aA}
	RW	37,19±17,53 ^{bA}	15,75±19,57 ^{aA}	14,87±21,1 ^{aA}
	R	34,74±8,29 ^{bB}	0,55±0,78 ^{aA}	1,72±0,39 ^{aA}
p-Cymene	C	ND	ND	ND
	W	ND	ND	ND
	RW	0,57±0,27 ^{bA}	0,39±0,55 ^{aA}	0,72±1,01 ^{aA}
	R	ND	ND	ND
o-Cymene	C	ND	ND	ND
	W	ND	0,49±0,37 ^{aA}	ND
	RW	1,58±0,75 ^{bA}	0,39±0,56 ^{aA}	ND
	R	1,74±0,51 ^{bB}	ND	ND
Hydrocarbons				
Benzen, 1,4-dichloro-	C	0,78±1,05 ^{aA}	2,14±2,85 ^{aA}	0,09±0,13 ^{aA}
	W	0,03±0,00 ^{aA}	3,57±1,48 ^{aB}	ND
	RW	4,57±2,16 ^{abA}	12,33±16,15 ^{aA}	4,56±6,44 ^{aA}
	R	6,93±3,17 ^{bB}	ND	0,15±0,21 ^{aA}
Oksime-, metoksi-fenil-	C	0,80±0,63 ^{aA}	6,70±5,00 ^{aA}	3,50±1,72 ^{abA}
	W	0,42±0,27 ^{aA}	1,47±0,75 ^{aA}	2,60±1,22 ^{abA}
	RW	1,69±0,80 ^{aA}	2,41±1,04 ^{aA}	1,92±2,72 ^{aA}
	R	1,84±0,36 ^{aA}	3,00±0,37 ^{aA}	8,20±2,25 ^{bB}

Table 3 continue

Acids	Samples	Ripening days		
		1	90	180
Others				
p-ksilen	C	0,28±0,40 ^{aA}	ND	0,05±0,07 ^{aA}
	W	ND	9,29±5,58 ^{aA}	ND
	RW	16,51±7,78 ^{aA}	4,52±4,60 ^{aA}	11,75±15,9 ^{aA}
	R	7,41±8,58 ^{aA}	ND	1,52±2,15 ^{aA}
o-ksilen	C	ND	2,13±3,01 ^{aA}	ND
	W	ND	ND	ND
	RW	1,96±0,92 ^{bA}	ND	0,94±1,33 ^{aA}
	R	0,47±0,66 ^{aA}	ND	ND
Fenol	C	0,11±0,12 ^{aA}	0,38±0,43 ^{aA}	0,08±0,12 ^{aA}
	W	0,25±0,35 ^{aA}	ND	ND
	RW	ND	ND	ND
	R	ND	0,20±0,14 ^{aA}	0,15±0,21 ^{aA}

*: C: control group; W: white wine waste; RW: red and white wine waste (1:1 w/w); R: red wine waste

a, b, c, d: There are significant differences between the means marked with different letters in the same column ($p < 0.05$).

A, B, C, D, E: There are significant differences between the averages marked with different letters on the same line ($p < 0.05$)

Hydrocarbons are derived from animal feed and/or from the oxidation of lipids during the maturation process and precursors of some aromatic compounds. Due to their low levels, they have been proven to have a minor contribution to flavor (Kondyli et al. 2016). In all cheeses, it increased at 90 days of storage and decreased slightly at 180 days.

When the results obtained in this study were evaluated, it was observed that the volatile components formed varied according to the grape pomace type used. Especially in red wine (R) and red-white (WR) blend waste samples, the formation of volatile aroma components was higher and this difference was found to be statistically significant. It can be said that red and blended pulps affect the aroma formation in cheese.

CONCLUSION

The use of grape pomace, which is used in winemaking, in many areas shows that it also has the potential to be used in dairy products. With the use of grape pomace, environmental impacts are reduced and the production of products beneficial to health is also possible. In this study, the fatty acids, free fatty acids and volatile components of Izmir Tulum cheese ripened in brine prepared with different grape pomaces, which are winemaking waste, were examined. It has been observed that grape pomace affects the formation of fatty acids and volatile aroma compounds that are effective in taste and aroma, and especially increases the amount of short-chain free fatty acids necessary for colon health. More works needs to be done on this subject to transform waste into value-added products.

ACKNOWLEDGEMENTS

We would like to thank Ege University Scientific Research Projects Coordination Office for providing financial support to our project (Project ID No. 22972)

REFERENCES

- Ackman RG (1998) Remarks on official methods employing boron trifluoride in the preparation of methyl esters of the fatty acids of fish oils. *Journal of the American Oil Chemists' Society*, 75:541–545.
- Akan E (2020) İzmir Tulum peynirinden ekstrakte edilen endojen ve eksojen peptitlerin olası biyolojik etkilerinin değerlendirilmesi. *Doktora Tezi*, Ege Üniversitesi Fen Bilimleri Enstitüsü Süt Teknolojisi Anabilim Dalı, 104, İzmir.
- Akarca G (2019) Lipolysis and aroma occurrence in Erzincan Tulum cheese, which is produced by adding probiotic bacteria and ripened in various packages. *Food Science and Technology*, 40: 102–116.
- Arslaner A, Bakırcı İ (2016) Effect of milk type, pasteurization and packaging materials on some physicochemical properties and free fatty acid profiles of Tulum cheese, *Akademik Gıda*, 14(2): 98–104.
- Augustin MAP, Udabage P, Juliano P., Clarke. T (2013) Towards a more sustainable dairy industry: Integration across the farmfactory interface and the dairy factory of the future. *International Dairy Journal*, 31:2–11.
- Atasoy AF, Hayaloglu AA, Kırmacı HA, Levent O, Türkoglu H (2013) Effects of partial substitution of caprine for ovine milk on the volatile compounds of fresh and mature Urfa cheeses. *Small Ruminant Research*, 115:113–123.
- Atasoy AF, Türkoglu H (2008) Changes of composition and free fatty acid contents of Urfa cheeses (a white-brined Turkish cheese) during ripening: Effects of heat treatments and starter cultures. *Food Chemistry*, 110:598–604.

- Atasoy AF, Türkoglu H (2009) Lipolysis in Urfa cheese produced from raw and pasteurized goats' and cows' milk with mesophilic or thermophilic cultures during ripening. *Food Chemistry*, 115:71–78.
- Avşar YK, Karagül-Yüceer Y, Hayaloğlu, AA (2011) Peynirde Aroma. *Peynir Biliminin Temelleri*. Editörler: Hayaloğlu, A.A., Özer, B. Sidas Medya, İzmir.
- Bennato F, Di Luca A, Martino C, Ianni A, Marone E, Grotta L (2020). Influence of grape pomace intake on nutritional value, lipid oxidation and volatile profile of poultry meat. *Foods*, 9(4):508
- Bordiga M, Travaglia F, Locatelli M (2019). Valorisation of grape pomace: An approach that is increasingly reaching its maturity—a review. *International Journal of Food Science and Technology*, 54(4):933–942.
- Bontinis G, Mallatou H, Pappaa EC, Massouras T, Alichanidis E (2012) Study of proteolysis, lipolysis and volatile profile of a traditional Greek goat cheese (Xinotyri) during ripening. *Small Rumin. Res.* 105:193–201.
- Brockman C, Beeren CJM (2011) Consumer perceptions of additives in dairy products. Pages 41–48 in *Encyclopedia of Dairy Sciences*. 2nd ed. J. W. Fuquay, ed. Elsevier Ltd., Cambridge, UK.
- Cabaroğlu T (2023) Current Situation and Problems of Wine Sector of Türkiye. *Bahçe*, 52: 269–275.
- Crespo JG, Brazinha C (2010) Membrane processing: Natural antioxidants from winemaking by-products, *Filtration and Separation*, 47 (2):32–35.
- Çakmakçı S, Dağdemir E, Hayaloğlu AA, Gurses M, Çetin B, Tahmas-Kahyaoglu D (2013) Effect of *Penicillium roqueforti* and incorporations of whey cheese on volatile profiles and sensory characteristics of mould-ripened Civil cheese. *International Journal Dairy Technology*, 66:512–526.
- Çakır Y, Çakmakçı S, Hayaloğlu AA (2016) The effect of addition of black cumin (*Nigella sativa* L.) and ripening period on proteolysis, sensory properties and volatile profiles of Erzincan Tulum (Savak) cheese made from raw Akkaraman sheep's milk. *Small Ruminant Research*, 134: 65–73.
- Deeth HC, Fitz-Gerald CH, Snow AJ (1983) A gas chromatographic method for the quantitative determination of free fatty acids in milk and milk products. *New Zealand Journal of Dairy Science and Technology*, 18:13–20.
- Demirkol M (2016) Kokulu Üzüm (*Vitis labrusca* L.) Posası Katkılı Yoğurtların Depolama Süresince Bazı Fizikokimyasal Özelliklerinin İncelenmesi, Yüksek Lisans Tezi, Ordu Üniversitesi Fen Bilimleri Enstitüsü, 75s.
- Deolindo CTP, Monteiro PI, Santos JS, Cruz AG, da Silva MC, Granato D (2019). Phenolic-rich Petit Suisse cheese manufactured with organic Bordeaux grape juice, skin, and seed extract: Technological, sensory, and functional properties. *Lebensmittel-Wissenschaft & Technologie*, 115:108493.
- FAO (2023) Food and Agricultural Organization. Statistic. <https://www.fao.org/faostat/en/#home> Available date 10.09.2023.
- Fontana AR, Antonioli A, Bottini R (2013) Grape pomace as a sustainable source of bioactive compounds: Extraction, characterization, and biotechnological applications of phenolics. *J. Agric. Food Chem.*, 61:8987–9003.
- Frühbauerová M, Červenka L, Hájek T, Salek RN, Velichová H, Buňka F (2020) Antioxidant properties of processed cheese spread after freeze-dried and oven-dried grape skin powder addition. *Potravinarstvo*, 14.
- García-Lomillo J, Gonzalez-SanJose ML (2017) Applications of wine pomace in the food industry: Approaches and functions. *Comprehensive Reviews in Food Science and Food Safety*, 16(1): 3–22.
- Guler Z (2008) Evaluation of lipolysis in set-type fermented milks made with different commercial yogurt starter cultures during storage. *Milchwissenschaft*, 33:73–77.
- Hayaloğlu AA, Çakmakçı S, Brechany EY, Deegan KC, McSweeney PL (2007) Microbiology, biochemistry, and volatile composition of Tulum cheese ripened in goat's skin or plastic bags. *Journal of Dairy Science*, 90: 1102–1121.
- Hayaloglu AA, Karabulut I (2013) SPME/GC-MS characterization and comparison of volatiles of eleven varieties of Turkish cheeses. *International Journal of Food Properties*, 16(7):1630-1653.
- Ianni A, Martino G (2020) Dietary grape pomace supplementation in dairy cows: Effect on nutritional quality of milk and its derived dairy products. *Foods*, 9(2):168.
- Karakus M S, Yildiz-Akgul F, Korkmaz A, Atasoy AF (2022) Evaluation of fatty acids, free fatty acids and textural properties of butter and sadeyag (anhydrous butter fat) produced from ovine and bovine cream and yoghurt. *International Dairy Journal*, 126:105229.
- Kalli E, Lappa I, Bouchagier P, Tarantilis PA, Skotti E (2018) Novel application and industrial exploitation of winery by-products. *Bioresources and Bioprocessing*, 5(1):1–21.
- Kamber U (2007) The traditional cheeses of Turkey: the Aegean region. *Food Reviews International*, 24(1):39–61.
- Kandyliis P, Dimitrellou D, Moschakis T (2021) Recent applications of grapes and their derivatives in dairy products. *Trends in Food Science & Technology*, 114:696–711.

- Kılıçalp N, Yücel C (2020) Effect of dietary omega-3 polyunsaturated fatty acids during the flushing period on Kirdar SS, Atamer M (2021) Quality criteria of Tulum cheese produced from cow's milk preserved by activation of lactoperoxidase system. *Journal of Food Processing and Preservation*, 45(4):15210.
- Kondyli E, Pappa EC, Svarnas C (2016) Ripening changes of the chemical composition, proteolysis, volatile fraction and organoleptic characteristics of a white-brined goat milk cheese. *Small Ruminant Research*, 145:1–6.
- Makris DP, Boskou G, Andrikopoulos NK (2007) Polyphenolic content and in vitro antioxidant characteristics of wine industry and other agri-food solid waste extracts. *Journal of Food Composition and Analysis*, 20:125–132.
- Mollet B, Rowland I (2002) Functional foods: At the frontier between food and pharma. *Curr. Opin. Biotechnol.* 13:483–485.
- OIV (2019). Statistical report on world vitiviniculture 2019. International Organization of Vine and Wine. <http://www.oiv.int/public/medias/6782/oiv-2019-statistical-report-onworld-vitiviniculture.pdf/>. (Accessed 15 June 2023)
- Sert D, Akin N, Aktumsek A (2014) Lipolysis in Tulum cheese produced from raw and pasteurized goats' milk during ripening. *Small Ruminant Research*, 121:351–360.
- Silva YP, Bernardi A, Frozza RL (2020) The role of short-chain fatty acids from gut microbiota in gut-brain communication. *Frontiers in Endocrinology*, 11: 25.
- Stashenko EE, Martínez JR (2007) Sampling volatile compounds from natural products with headspace/solid-phase micro-extraction. *Journal of Biochemical Biophysical Methods*, 70 (2): 235–242.
- reproductive performance of Karayaka ewes. *Indian J Anim Res*, 54:869–873.
- Şengül M, Erkaya T, Dervişoğlu M, Aydemir O, Gül O (2014) Compositional, biochemical and textural changes during ripening of Tulum cheese made with different coagulants. *International Journal of Dairy Technology*, 67(3):373–383.
- Tan J, McKenzie C, Potamitis M, Thorburn AN, Mackay CR, Macia L (2014) The role of short-chain fatty acids in health and disease. *Advances in Immunology*, 121:91–119.
- Tekin A, Güler Z (2021) The effect of ripening medium (goat skin bag or plastic barrel) on the volatile profile, color parameter and sensory characteristics of Tulum cheese. *Journal of Central European Agriculture*, 22(1):19–38.
- Tomar O, Akarca G, Gök V, Çağlar M Y (2020) The effects of packaging materials on the fatty acid composition, organic acid content, and texture profiles of Tulum cheese. *Journal of Food Science*, 85:3134–3140.
- Whetstone CME, Cadwallader KR, Drake M (2005) Characterization of aroma compounds responsible for the rosy/floral flavor in Cheddar cheese. *Journal of Agricultural and Food Chemistry*, 53(8):3126–3132.
- Yu J, Ahmedna M (2013) Functional components of grape pomace: their composition, biological properties and potential applications. *Int. J. Food Sci. Technol.* 48:221–237.
- Zhu F, Du B, Zheng L, Li J (2015) Advance on the bioactivity and potential applications of dietary fibre from grape pomace. *Food Chemistry*. <http://dx.doi.org/10.1016/j.foodchem.2014.07.057>.

