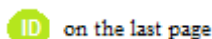




Evaluation of fatty acid compositions and physicochemical quality parameters of ancient and recent olive (*Olea europaea* L.) oil varieties of Southeast Anatolia

Halim AVCI^{1,*}, Yilmaz UGUR², Selim ERDOGAN³



¹Kilis 7 Aralık University, Faculty of Science and Art, Department of Chemistry, Kilis, Turkey

²Apricot Research Institute, Malatya, Turkey

³Inonu University, Faculty of Pharmacy, Department of Analytical Chemistry, Malatya, Turkey

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*Corresponding author e-mail: halimavci77@gmail.com

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ABSTRACT

Upper Mesopotamia is known to be the homeland of olive, spanning over Mardin, Hatay, Kilis and the western shores of Syria and Palestine. Having earliest habitation in the Southeastern Anatolia region, olive lies down along the Western Anatolia. The goal of the present study is to evaluate the fatty acid compositions of the olive oils obtained from the olive trees which are the kinds of Kilis yağlık (KY), Halhalı (H), Gemlik-Kilis (GK), and Gemlik-Gemlik (GG). While GG variety has the lowest total saturated fatty acids (SFAs), KY, GK and H varieties show similarity in respect to the total SFAs levels, and these have the higher SFAs levels. While GG variety has the lowest total SFAs and polyunsaturated fatty acids (PUFAs), KY contains the highest total SFAs and PUFAs. While mean docosahexaenoic acid (DHA) values in KY and GK varieties are minor levels, it is found below the detection limit of GC-FID method for all of H and GG samples.

Keywords: Olive oil, fatty acid composition, genotype, physicochemical quality parameters.

Güneydoğu Anadolu'nun eski ve yeni zeytin (*Olea europaea* L.) yağı çeşitlerinin yağ asiti bileşimleri ve fizikokimyasal kalite parametrelerinin değerlendirilmesi

ÖZ

Zeytinin anavatanının Mardin, Hatay, Suriye'nin ve Filistin'in batı kıyılarını içerisine alan yukarı Mezopotamya olduğu bilinmektedir. Güneydoğu Anadolu bölgesinde ilk yerleşime sahip olan zeytin Batı Anadolu'ya kadar uzanmıştır. Bu çalışmanın amacı, Kilis yağlık (KY), Halhalı (H), Gemlik-Kilis (GK) ve Gemlik-Gemlik (GG) çeşitleri olan zeytin ağaçlarından elde edilen zeytinyağlarının yağ asidi bileşimlerini değerlendirmektir. GG türü en düşük doymuş yağ asidine (SFAs) sahip iken, KY, GK and H türleri toplam SFAs seviyelerine benzerlik gösterir ve bunlar daha yüksek SFAs seviyelerine sahiptir. GG türü en düşük toplam SFAs ve çoklu doymamış yağ asitlerine (PUFAs) sahip iken KY en yüksek toplam SFAs ve PUFAs içermektedir. KY ve GG türlerinde ortalama docosahexaenoic asit (DHA) düşük seviyelerde iken, H ve GG örneklerinin hepsi için GC-FID metodunun gözlenebilir sınırdan altındadır.

Anahtar Kelimeler: Zeytinyağı, yağ asidi bileşimi, genotip, fizikokimyasal kalite parametreleri.

1. INTRODUCTION

Olive farming has a history and may be as old as human kind itself, and on the other hand olive is said to be “the father of all trees”. The importance of olive is cited in all religious texts and in myths of creation and origin. The Archeological and geological finds also reveal that olive has been used since 6000 BC.^{1,2} Upper

Mesopotamia is known to be the homeland of olive, spanning Mardin, Hatay, Kilis and the western shores of Syria and Palestine. This judgment is reached by means of the subspecies of olive tree seen on the across strips of these provinces, recently. Olive, having earliest habitation in the Southeastern Anatolia region, is lies down along the Western Anatolia, and continuous from there to Greece, Italy, France and Spain through the

Aegean islands. At the same time, it reaches North Africa through Sicily, and merges with the second stripe that comes from the Southeastern Anatolia region and moves through Syria and Egypt and finally spread over all southern coasts of the Mediterranean.^{2,3}

Olive farming is carried out along the shores in Mediterranean, Southeastern Anatolia, Marmara, Aegean and Black Sea regions of Turkey and in most provinces reaching up to Mardin in the Southeastern Anatolia. Within this ecological environment, olive has various species in Turkey, its homeland.^{2,3} The Turkish Agriculture Ministry has recorded total 88 assortment of native olives (include 26 foreign olive varieties) derived from both pomological and morphological variables in 1990 year.⁴ Gemlik variety, the dominant olive variety in the Marmara Region, particularly in the Gemlik Gulf, has also been grown in other parts of Turkey since 40 years. On the other hand, the most widely known and grown olive varieties in South Anatolia Region are Kilis Yağlık (KY) and Halhalı (H).^{5,6}

The most important factor that affects the chemical composition of olive is its variety. Other factors are the environmental conditions that the olive trees are grown. In determination of the quality of the olive, the big difficulty is encountered due to olive variety grown in different geographic region, and environment. While the environmental factors can be said as soil and climate, the agronomic factors are mentioned as irrigation and fertilization. Harvesting and ripeness which form cultivation factors have important effects as well as the technological factors such as post-harvest storage and extraction system. Geographical indication about olive variety informs about the authenticity of the extra-virgin olive oils in Europe.⁷⁻¹⁰

In this study, olive oil samples of ancient (native) and recent varieties obtained from the local producers were extracted by three-phase continuous centrifugation process. The chemical compositions of all extracted olive oil samples (Kilis Yağlık (KY), Gemlik-Kilis (GK), Gemlik-Gemlik (GG) and Halhalı (H)) were determined. Moreover, the physicochemical quality parameters including peroxide value (PV), free fatty acid (FFA) content, UV-spectrophotometric characteristics, refractive index and the viscosity were also determined in all the olive oil samples.

2. MATERIALS AND METHODS

2.1. Study area

Saline-sodic soils are known to covering 1.5 million ha in Turkey. The soils in Southeastern Anatolia are highly calcareous with high pH. Calcium carbonate is abundant in regional soils.¹¹ The soil in Kilis is generally geologically with limestone, and it contains high

amounts of lime, generally in the range of 30-60 % CaCO₃.

The olive (*Olea europaea* L.) oil samples of KY, GK, GG, and H (İskenderun-Hatay) were obtained from the local producers in Kilis, Gemlik and İskenderun, respectively (Figure 1).



Figure 1. The map showing three regions where the olive tree varieties grown in Gemlik (G), İskenderun (I) and Kilis (K).

2.2. Plant material

The most significant type of olive oil in the Southeastern Anatolia is KY olive oil. This type is widespread in Gaziantep, Kilis, Şanlıurfa, Kahramanmaraş, and Mardin provinces. Its fruits are very small compared to its bigger seeds. The fruits of KY olive contain oil up to 31.8%. When considered as one of the most known olive types in terms of oil containment, the KY olive is a significant olive type of the Southeastern Anatolia region with its strength, fertility and adaptable nature to the region.¹² The GG olive cultivar is originated from Gemlik Gulf at Marmara region and it is the major olive variety of the Marmara and Aegean region. Gemlik is a typical olive variety in the Marmara Region. More than 80% of the olives in the region are composed of Gemlik variety. The shell of Gemlik olive is thin and attached to the flesh, the kernel is small, round, and smooth. An important feature of the Gemlik variety is the being very aromatic. Its oil ratio is about 25-28%. Although it is generally regarded as black table olive, it can sometimes be processed as fat. Gemlik, in Turkey's various regions, has been grown intensively since the last 30-40 years.¹³ Halhalı olive variety which is belong to Eastern Mediterranean and Southeastern Anatolia Regions widely grows in Hatay, Gaziantep, Kahramanmaraş, Kilis, Mardin, and their surroundings. In addition to being considered more fat, it is also regarded as green or black table olive. Its fruit is small and round. The Fat percentage is very high, about 30-32%.¹³

2.3. Olive oils

Monovarietal virgin olive oils of Kilis yağlık (KY), Gemlik-Kilis (GK) and Halhalı (H) samples used in this study were collected from the commercial small or medium-scale olive oil producers in 2014 year. 100 ml of olive oil samples were placed in dark glass bottles and were stored in a refrigerator at 4°C until the samples were brought to analyze. The dominant olive variety grown in Kilis is Kilis yağlık, but Gemlik variety has been also cultivated in Kilis region for 20-30 years. The original growing area of Gemlik variety is the North Marmara Region. Growing area of Halhalı is East Mediterranean and Southeast Anatolia. Growing area of Kilis yağlık is Southeast Anatolia.

The olive (*Olea europaea* L.) oil of KY (n = 18), GK (n = 10), GG (n = 10), and H (İskenderun) (n = 18) samples were obtained from the local producers in Kilis, Gemlik, and İskenderun, respectively and all samples were extracted by three-phase continuous centrifugation process. The temperature was less than 40°C in all processing stages. The olive cultivars were identified according to the morphological properties.

Oil Extraction Systems: The centrifugation system, called “the three-phase system decanter”, is the most popular extraction system in Turkey. In the three-phase system decanter, water was added to the system. As the centrifuge rotates at a high speed (3500-3600 rpm), the non-miscible liquids (olive oil and vegetation water) were separated by proper nozzles from oil pomace due to specific weight differences. This liquid is then taken to a vertical centrifuge where the olive oil is separated from the fruit’s vegetable water.¹⁴

2.4 Sample preparation and analysis of fatty acids

The olive oil samples were prepared as described in the official journal of the European Official Method. The vials were stored at -20 °C in a deep freeze until the moment of chromatographic analysis. 37 fatty acid methyl esters (FAME), mix certified reference material (18919-1 AMP SUPELCO) containing C4–C24 (1.343–6.640 wt. % relative concentration) was used as a reference, and the results were expressed in relative percentages of each fatty acid. Three replicate measurements were performed for each sample.

Chromatographic analyses were achieved on a Shimadzu 2010 gas chromatography (GC) (Shimadzu Technologies, Kyoto, Japan) consisting of autosampler, in-line degasser and a flame ionization detector (FID). The instrumental configuration and analytical conditions were summarized in the following; Shimadzu 2010 GC-FID instrument equipped with a RT-2560 capillary column (100 m x 0.25 mm x 0.20 µm, RESTEK Scientific) under the following temperature program: 140 °C for 5 min followed by an increase to 240 °C at a rate of 20 °C/min for 45 min. The injector and flame ionization detector temperatures were set at 240 °C. The

instrumental configuration for analysis of fatty acid methyl esters from all samples are summarized below.

Column: Rt-2560, 100 m x 0.25 mm I.D., 0.20 µm, **Oven:** 140 °C (5 min), 4 °C/min to 240 °C (at 4 °C min), hold 15 min, **Injection:** 1 µl, split 1:80, **Inj. Temp.:** 240 °C, **Carrier Gas:** Helium, 20 cm/sec., 150 °C, **Detector:** FID, 240 °C. **Liner:** 4 mm I.D split, cup design.

The fatty acids mentioned in section 2.3 were analyzed by GC-FID after the olive oil samples were digested (extracted). The fatty acid composition is a quality parameter and authenticity indicator for virgin olive oils. Typical chromatograms are shown in Figure 2.

The determinations of fatty acid methyl esters (FAME) was carried out by comparing the retention time with those of the 37 components’ reference standard mixture (37-Component FAME Mix, Supelco) analyzed under the same analytical conditions.

2.4. Physicochemical parameters of olive oils

Analysis of peroxide values (PV), fatty acids (FAs) and UV absorption characteristics were performed according European Community (EC) methods (European Economic Community, EEC/2568/91 and European Economic, EE/1429/92).¹⁵

The PV (meq O₂/kg oil) was determined by reacting oil and 3:2 chloroform/acetic acid with potassium iodide in darkness. Free fatty acid (FFA; as oleic acid %) was determined by volumetric method. K232 and K270 extinction coefficients were calculated by means of UV-Vis spectrophotometer.¹⁶ In addition, the refractive index and the viscosity of the oil samples were also determined by a refractometer and a viscosimeter, respectively. The results of FFA, PV, and UV absorption characteristics (K232, K270), the refractive index, and the viscosity are shown in Table 1.

3. RESULTS AND DISCUSSION

3.1. Fatty acids composition of olive oils

A proportionally small amount of polyunsaturated fatty acids (PUFAs) and large amount of monounsaturated fatty acids (MUFAs) determine fatty acid components which have a total up to 98% in oil constituents. The largest amount (98% to 99%) of the fatty acid structure composes of triglycerides, whereas diglycerides and monoglycerides constitute very small amounts; 1% to 1.5% and below 1%, respectively. The different amounts of saturated palmitic (C16:0; 7.5–20%) and stearic fatty acids (C18:0; 0.5–3.5%) as well as monounsaturated palmitoleic (C16:1; 0.3–3.5%) and oleic fatty acids (C18:1; 56.0–85.0%); with polyunsaturated linoleic (C18:2; 3.5–20%) and linolenic fatty acids (C18:3; 0–1.5%), an acid that humans do not amalgamate, constitute nearly all of fatty acid component.¹⁷

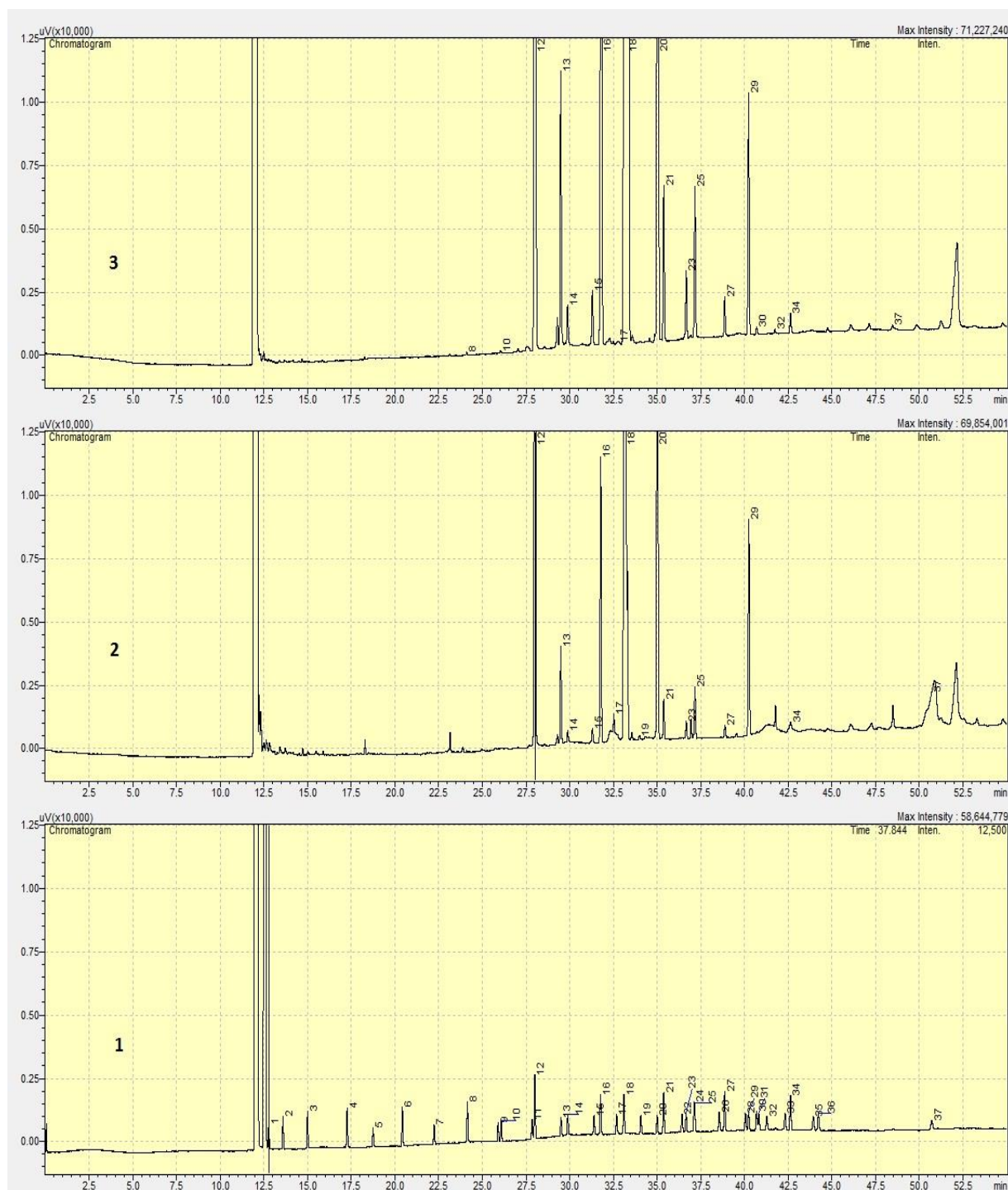


Figure 2. Typical GC-FID chromatograms for (1) composition of 37 components FAME mix standart, (2) GK and (3) KY olive oil.

1: C4:0 (Butyric), **2:** C6:0 (Caproic), **3:** C8:0 (Caprylic), **4:** C10:0 (Capric), **5:** C11:0 (Undecanoic), **6:** C12:0 (Lauric), **7:** C13:0 (Tridecanoic), **8:** Myristic acid C14:0, **9:** Myristoleic acid C14:1, **10:** Pentadecanoic acid C15:0, **11:** cis-10-pentadecanoic acid C15:1, **12:** Palmitic acid C16:0, **13:** Palmitoleic acid C16:1, **14:** Heptadecanoic acid C17:0, **15:** cis-10 heptadecanoic acid C17:1, **16:** Stearic acid C18:0, **17:** Elaidic acid C18:1n9t, **18:** Oleic acid C18:1n9c, **19:** Linolelaidic acid C18:2n6t, **20:** Linoleic acid C18:2n6c, **21:** Arachidic acid C20:0, **22:** gamma-linolenic acid C18:3n6, **23:** cis-11-eicosenoic acid C20:1, **24:** Linolenic acid C18:3n3, **25:** Henoicosanoic acid C21:0, **26:** cis-11,14-eicosadienoic acid C20:2, **27:** Behenic acid C22:0, **28:** cis-8,11,14-eicosatrienoic acid C20:3n6, **29:** Erucic acid C22:1n9, **30:** cis-11,14,17-eicosatrienoic acid C20:3n6, **33:** cis-13,16-docosadienoic acid C22:2, **34:** Lignoceric acid C24:0, **35:** cis-5,8,11,14,17- eicosapentaenoic C20:5n3 (EPA), **36:** Nervonic acid C24:1, **37:** cis-4,7,10,13,16,19-docosahexaenoic acid C22:6n3 (DHA).

The big difficulty is encountered in qualifying the olive oil in terms of varieties and geographic derivation due to how it is composed of different factors. While the environmental factors are soil and climate, irrigation and fertilization form the agronomic factors. Harvesting and ripeness which form cultivation factors have effects as well as the technological factors do, such as post-harvest

storage and extraction system. How chemically vegetable oils are formed is mostly affected by climate the impact of which on monovarietal features is a widely examined subject in research.⁹ Several studies are conducted on fatty acid compositions of the virgin olive oils of KY, H, and Gemlik varieties (Table 2).

Table 1. The determined physicochemical quality parameters of olive oils and the European Regulation Standard limit values for olive oil quality parameters¹⁵

Olive Oil Species and Quality Indexes	Free acidity (oleic acid; %)		Peroxide value (meq O ₂ /kg oil)		K232		K270		Refractive index		Viscosity (cp)	
	Range	Mean± SD	Range	Mean± SD	Range	Mean± SD	Range	Mean± SD	Range	Mean± SD	Range	Mean± SD
Kilis yağlık (Kilis)	1.0-7.8	3.9±1.9	9.6-46.6	27.8±10.5	1.9-3.3	2.6±0.4	0.09-0.41	0.25±0.1	1.467-1.469	1.468±0.00	74.6-81.4	79.0±1.5
Gemlik (Kilis)	0.4-5.9	2.5±1.9	8.6-28.8	18.0±7.3	1.5-2.0	1.8±0.1	0.17-0.32	0.22±0.0	1.468-1.470	1.469±0.00	77.0-80.4	78.1±1.1
Gemlik (Gemlik)	0.3-3.7	1.4±1.1	2.0-27.3	14.8±7.1	1.4-2.0	1.8±0.2	0.15-0.27	0.20±0.0	1.468-1.469	1.469±0.00	76.1-79.3	78.1±0.9
Halhalı (Iskenderun)	0.3-4.8	1.7±1.1	3.3-29.2	15.6±7.2	1.8-2.1	2.0±0.1	0.15-0.76	0.25±0.13	1.467-1.469	1.467±0.00	74.7-82.6	79.0±1.9
Extra virgin OO	≤ 1.0		≤ 20		≤ 2.5		≤ 0.20					
Virgin OO	≤ 2.0		≤ 20		≤ 2.6		≤ 0.25					
Ordinary virgin OO	≤ 3.3		≤ 20		≤ 2.6		≤ 0.25					
Lampante OO	> 3.3		> 20		≤ 3.7		> 0.25					
Refined OO	≤ 0.5		≤ 5		≤ 3.4		≤ 1.20					

Arslan and Ozcan¹⁰ studied on the monovarietal virgin olive oils of four Turkish varieties which are quite common in Turkey: Ayvalık, Gemlik, Kilis yağlık, and Sarulak from Mediterranean and the Southeastern Anatolia Regions of Turkey. In Gemlik and KY olive oils among these varieties, high oleic acid levels were observed up to 70%. Çolakoğlu¹⁸ researched fatty acid levels in the various olive oils collected from the different parts of West and Southeast Anatolia, Mediterranean, Aegean, and Marmara. These varieties were reported according to the oleic, palmitic, linoleic, stearic, palmitoleic, and linolenic acid levels in ranges of 61.0-79.3, 9.0-19.7, 4.7-16.5, 1.4-4.2, 0.3-1.5, and 0.5-1.2%, respectively.

In this study, olive (*Olea europaea* L.) oil samples of KY, GK, GG, and H (Iskenderun-Hatay) (Figure 1) were collected from commercial small or medium-scale olive oil producers. The dominant olive variety grown in Kilis is KY, but Gemlik variety has also been cultivated in Kilis region for 30-40 years. The fatty acid profiles of the olive oils are submitted in Table 3. As can be seen in Table 3, the range for individual fatty acid determined for different varieties virtually are covered the full range of International Olive Council (IOC)¹⁹ and

the Turkish Food Codex standards.²⁰ Generally, the fatty acids levels in each variety of olive oil samples are similar to the same variety samples collected from different regions.

Olive oils are divided into two types according to their fatty acid composition. The first type of olive oil has low linoleic and palmitic content but also high oleic acid content. In the second type of olive oil, it has high linoleic and palmitic content and low oleic acid content. According to this group; Turkish virgin olive oil (such as Spanish, Italian and Greek) is the first type, and Tunisian oils are the second type.⁸ The most important major fatty acid in olive oils is oleic acid. The highest average level of oleic acid (72.6%) is determined for GG in Marmara Region. The oleic acid levels in all varieties are different with a confidence level of 0.05, but at higher confidence level (lower α values), significant differences is not found between KY and GK, and between KY and H ($p < 0.1$). At a confidence level of 0.2, there are three groups according to oleic acid levels; (a) KY and H, (b) GK, and (c) GG. Arslan and Ozcan¹⁰ studied on the monovarietal virgin olive oils of four Turkish varieties: Ayvalık, Gemlik, Kilis yağlık, and Sarulak from Mediterranean and the Southeastern Anatolia Regions of Turkey.

Table 2. Reported fatty acids contents (m/m %) of virgin olive oil of KY, GK, GG, and H (İskenderun), and some varieties of different parts of Turkey
¹Gemlik variety olives grown in three different districts (Gemlik, Nilüfer and Orhangazi) of Bursa. ²Turkish virgin olive samples of five groups from the Aegean Region (n = 64)

Variety	Location	C16:0	C16:1	C17:0	C17:1	C18:0	C18:1n9c	C18:2n6c	C18:3	C20:0	C20:1	C18:3n3	C21:0	C22:0	C24:0	Literature
Kilis yağlık	Kilis	13.4-15.0	0.9-1.7			2.7-3.6	66.8-70.6	7.6-11.2		0.40-0.70		1.0-1.60		0.30-0.50		10
Kilis yağlık	Kilis-Agdere	14.9	1.0	0.12	0.17	3.1	70.1	9.1		0.46	0.23	0.65		0.11	0.06	22
Kilis yağlık	Kilis-Besenli	15	1.1	0.13	0.18	3.2	70.4	8.3		0.53	0.26	0.66		0.14	0.07	22
Nizip yağlık	Nizip	14.2-16.8	1.2-1.3	0.12-0.16	0.17-0.23	3.1-3.2	66.4-73.4	6.3- 10.5		0.44-0.50	0.20-0.21	0.63- 0.68		0.09-0.12	0.04-0.06	22
Nizip yağlık	Nizip	15.4	1.2	0.12	0.16	3.7	68.8	9		0.59	0.24	0.58		0.15	0.09	32
Gemlik	Kilis	14.7	1.6	0.12	0.22	2.9	70.7	8.2		0.40	0.21	0.78		0.09	0.04	22
Gemlik	Gemlik	14	0.9	-	-	2.9	70.7	10.1		0.78	-	0.39		0.15	-	9
Gemlik	İzmir	8.6-16.8	0.9-1.4	0.01-0.13	nd	2.3-3.3	61.9- 65.3	12.8-16.0		0.33-0.56	0.51-0.60	0.19-0.27		0.00-0.12	0.00- 0.10	31
Gemlik ¹	Bursa	12.8-16.9	1.1- 1.3		0.10- 0.16	3.1 -4.0	68.6- 74.4	6.8- 8.7		0.36- 0.50	0.20- 0.25	0.54- 0.60		0.05- 0.13	0.03-1.18	21
Gemlik	Bursa	12.5	1.2	-	-	2.3	72.3	7.9		0.36	0.25	-		0.04	-	23
Gemlik	İzmit	13.9	1.6	0.1	0.2	2.2	72.6	8.1		0.32	0.22	0.6		0.13	0.02	22
Gemlik	Edremit	13.7	1.1	-	-	3.2	72.1	8.9		0.59	-	0.37		0.14	-	9
Gemlik	Mediterranean	14.1-15.8	1.2-1.6			2.3-3.1	67.2-70.2	6.4-10.3		0.50-0.50		0.6-1.5		0.30-0.50		10
Gemlik	İzmir-Torbalı	12.8	1.1	0.12	0.19	2.9	73.4	8.1	0.67	0.38	0.24	0.75		0.09	0.06	22
Gemlik	Karlıdağ	13.9	1.6	0.11	0.23	2.8	69.8	10.0	0.91	0.38	0.23	0.67		0.08	0.06	22
Gemlik	Hatay	9.05	1.9	0.07	0.22	1.3	75.4	10.8		0.17	0.24	0.8		0.05	0.01	22
Gemlik	Antakya	15.6-16.3	1.5-2.2	0.11-0.13	0.21-0.23	2.8-3.0	69.8-71.8	5.6-7.1		0.42-0.46		0.21-0.62	0.14-0.28	0.11-0.11	0.06-0.10	13
Halhali	Antakya	14.4-16.4	0.9-1.3	0.11-0.14	0.15-0.17	3.2-4.2	70.9-71.6	5.8-6.8		0.58-0.69		0.53-0.59	0.18-0.32	0.10-0.19	0.10-0.11	13
Halhali	Reyhanlı. Kırkkhan	11.7-15.4	0.8-1.11	0.10-0.15	0.17-0.18	2.2-4.2	71.1-75.0	6.3-8.3	0.55-0.56	0.61-0.69	0.21-0.23	0.55		0.08-0.16	0.04-0.09	22
Halhali	Nizip	15.1	0.9	0.06	0.05	3.9	63.4	15.3		0.43	0.2	0.59		0.09	0.03	22
Five groups ²	Aegean	12.1	0.78	0.1	0.16	2.5	73.1	9.7		0.40	0.31	0.56		0.12	0.05	29
Different varieties	N.Aegean	12.9	1.04	0.11	0.19	0.2	71.5	10.6	0.69	0.36	0.26			0.10	0.04	22
Different varieties	S.Aegean	12.7	1.06	0.06	0.11	2.1	73.8	8.7	0.74	0.33	0.26			0.09	0.03	22
Different varieties	Mediterranean	13.1	1.77	0.13	0.23	2.5	69.3	11.	0.72	0.38	0.25			0.11	0.04	22
Different varieties	S.Anatolia	15.2	1.12	0.13	0.18	3.2	70.1	8.6	0.65	0.48	0.22			0.11	0.05	22
Different varieties	Bursa. Akhisar	9.0-13.9	0.73-	0.03-0.20	0.04-0.29	1.6-3.6	68.6-76.4	6.9-11.4		0.24-0.57		0.48-0.71		0.06-0.16	0.03-0.07	6
Different varieties	N.Aegean	11.6-13.9	0.7-1.3	0.05-0.17		1.6-3.5	70.0-77.1	7.47-11.0		0.24-0.47	0.22-0.37	0.45-0.71		0.09-0.13	0.03-0.06	32
Different varieties	Different regions	13.7-18.2	1.2-1.7	0.1-0.1	0.16-0.21	2.6-4.3	64.9-72.9	6.8-11.0		0.4-0.6	0.21-0.27	0.53-0.77		0.10-0.15	0.06-0.1	6

Table 3. Fatty acids (FAs) contents (m/m %) of virgin olive oil varieties from different geographic regions of Turkey*. Suggested or regulatory levels thresholds from relevant sources are also provided

Types of FAs	Formula of FAs	IUPAC name of FAs	Kilis Yağlık (KY) n= 18		Gemlik-Kilis, (GK) n = 10		Gemlik-Gemlik (GG) n = 10		Halhali (H) (İskenderun) n = 18		Turkish Food Codex, IOC
			Range	Mean±SD	Range	Mean±SD	Range	Mean±SD	Range	Mean±SD	
SFAs	C 14:0	Myristic	0.000-0.05	0.012±0.014	0.000-0.013	0.006±0.006	0.000-0.014	0.007±0.005	0.01-0.016	0.012±0.001	≤ 0.05
	C15:0	Pentadecanoic	0.000-0.016	(0.005±0.005)ab	0.000-0.017	(0.004±0.06)ab	0.000-0.017	(0.005±0.005)ab	0.006-0.009	(0.008±0.001)b	
	C16:0	Palmitic	12.93-14.98	(13.946±0.511)a	12.41-16.56	(13.740±1.373)a	10.9-13.081	(11.820±0.78)b	12.160-15.00	(14.040±0.810)a	7.5-20
	C17:0	Heptadecanoic	0.11-0.141	(0.126±0.009)a	0.113-0.268	(0.144±0.047)a	0.094-0.18	(0.138±0.025)a	0.114-0.168	(0.141±0.013)a	≤ 0.3
	C18:0	Stearic	3.409-4.08	(3.716±0.181)b	3.133-3.851	(3.507±0.242)a	2.873-3.909	(3.402±0.310)a	2.995-3.815	(3.516±0.241)a	0.5-5.0
	C20:0	Arachidic	0.401-0.58	(0.568±0.049)c	0.401-0.594	(0.496±0.059)b	0.415-0.546	(0.457±0.037)a	0.462-0.634	(0.543±0.043)c	≤ 0.6
	C21:0	Henoicosanoic	0.546-0.637	(0.590±0.049)a	0.561-0.776	(0.631±0.059)b	0.447-0.660	(0.557±0.070)a	0.561-0.735	(0.632±0.046)b	
	C22:0	Behenic	0.127-0.178	(0.152±0.025)b	0.091-0.233	(0.152±0.041)b	0.106-0.150	(0.127±0.014)a	0.120-0.176	(0.141±0.016)ab	≤ 0.2
	C24:0	Lignoceric	0.069-0.409	(0.150±0.0130)a	0.00-0.989	(0.234±0.298)a	0.000-1.168	(0.270±0.465)a	0.063-0.117	(0.081±0.014)a	≤ 0.2
	ΣSFA (%)		19.256		18.904		16.787		19.114		
MUFAs	C14:1	Myristoleic	0.000-0.000	-	0.000-0.000	-	0.000-0.000	-	0.000-0.000	-	
	C15:1	cis-10-pendacanoic	0.000-0.458	(0.0459±0.134)a	0.000-0.000	0.000a	0.000-0.000	0.000 a	0.006-0.013	(0.009±0.002)a	
	C16:1	Palmitoleic	0.799-1.060	(0.913±0.090)ab	0.940-1.193	(1.057±0.095)a	0.601-1.176	(0.826±0.169)ab	0.515-1.276	(0.988±0.252)b	0.3-3.5
	C17:1	cis-10-pendacanoic	0.161-0.213	(0.187±0.012)a	0.165-0.240	(0.192±0.020)ab	0.147-0.275	(0.230±0.037)c	0.165-0.238	(0.206±0.020)b	≤ 0.3
	C18:1n9c	Oleic	64.40-70.25	(68.436±1.507)ab	60.35-72.96	(67.059±4.971)a	70.37-74.73	(72.564±1.655)c	64.596-72.07	(69.426±1.760)b	55.0-83.0
	C20:1	cis-11-eicosenoic	0.209-0.274	(0.258±0.015)a	0.160-0.266	(0.224±0.036)a	0.229-0.306	(0.250±0.022)a	0.251-0.340	(0.269±0.028)a	≤ 0.4
	C22:1n9	Erucic	0.713-5.453	(1.686±1.198)ab	0.694-3.990	(2.029±1.340)b	0.694-2.269	(1.189±0.571)a	0.672-1.421	(0.950±0.212)a	
		ΣMUFA (%)		71.523		70.561		73.870		70.897	
	MUFA/PUFA		6.92		6.98		8.65		7.77		

*Results of statistical analyses (Duncan Multiple Test) of fatty acid levels in olive oil varieties are indicated in the "Mean" columns of this table. Values with the same letter are not statistically different ($p < 0.05$) for that element across the species.

SD: standard deviation; **FA:** fatty acid, **SFA:** saturated fatty acid, **MUFA:** monounsaturated fatty acid, **PUFA:** polyunsaturated fatty acid, **USFA:** unsaturated fatty acid, **O/L:** oleic/linoleic, **PA/L:** palmitic/linoleic; **DHA:** Cis-4,7,10,13,16,19-docosahexaenoic acid.

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Table 3's continuation

PUFAs	C18:2n6c	Linoleic**	7.372-9.879	(8.755±0.837)b	6.304-9.838	(8.511±1.131)ab	6.575-10.58	(7.915±1.284)a	6.449-10.930	(8.557±1.313)ab	3.5-21.0
	C20:3n6	cis-8.11.14- eicosatrienoic	0.000-0.012	(0.001±0.0030)a	0.000-0.000	0.000a	0.000-0.027	(0.016±0.009)b	0.000-0.000	0000a	
	C20:3n6	cis-11.14.17- eicosatrienoic	0.021-0.209	(0.034±0.044)a	0.000-0.028	(0.013±0.013)a	0.047-0.327	(0.096±0.085)b	0.023-0.032	(0.026±0.003) a	
	C22:6n3	DHA	0.000-1.253	(0.287±0.368)a	0.000-3.140	(0.937±1.200)b	0.000-0.00	0.000a	0.006-0.061	(0.020±0.016)a	
		∑PUFA (%)		10.332		10.101		8.542		9.122	
	∑USFA (%)		81.855		80.662		82.412		80.019		
	∑(MUFA+PUFA)										
	O/L		6.73-9.47	(7.89±0.83)a	6.180-10.03	(8.073±1.388)a	7.116-1.340	(9.321±1.426)b	6.206-11.176	(8.279±1.485)a	

*Results of statistical analyses (Duncan Multiple Test) of fatty acid levels in olive oil varieties are indicated in the "Mean" columns of this table. Values with the same letter are not statistically different ($p < 0.05$) for that element across the species.

** $p < 0.1$.

SD: standard deviation; **FA**: fatty acid, **SFA**: saturated fatty acid, **MUFA**: monounsaturated fatty acid, **PUFA**: polyunsaturated fatty acid, **USFA**: unsaturated fatty acid, **O/L**: oleic/linoleic, **PA/L**: palmitic/linoleic; **DHA**: Cis-4,7,10,13,16,19-docosahexaenoic acid.

They reported that Gemlik and KY olive oil samples had high oleic acid levels rising to 70%, but KY had low linoleic and palmitic acids levels which made it superior to Gemlik.

The other major fatty acids (FAs) are palmitic, stearic, and linoleic acids. The mean levels of palmitic and linoleic acids in KY, GK, and H varieties are similar and higher than those in GG (Table 3). In our study, olive oil of GG variety has the lowest mean linoleic and palmitic acid levels. However, Arslan and Ozcan¹⁰ reported lower linoleic and palmitic acids levels in olive oil samples of Kilis yağlık than those reported in Gemlik variety from Mediterranean Region.

In this study, mean oleic and linoleic acids in KY, Gemlik, and H varieties are generally similar to the mean levels of FAs in the same varieties reported by some authors (Table 3).^{9,13,21-23} The levels of palmitic and oleic acids in GK and GG olive oil samples cultivated in different regions show significant differences at a confidence level of 0.05, but significant differences are found for stearic and linoleic acids levels in the same varieties and at the same confidence level. At lower confidence levels (higher α values), significant differences are found for palmitic ($p < 0.1$) and oleic acids ($p < 0.1$) (Table 3). The ratios of oleic to linoleic acid and MUFAs to PUFAs are important indicators of oxidative stability. KY, GK, and H varieties had similar oleic/linoleic acid ratios (O/L) while GG variety had higher O/L ratio. The O/L ratios did not show a significant difference at a confidence level of 0.05 among KY, GK, and H varieties, but differences appeared at confidence levels of 0.1 and 0.2. These O/L ratios are in agreement with the findings of Arslan and Ozcan¹⁰ and Pinelli et al.²⁴

Negative relationships between oleic and linoleic acid levels in the South Anatolian Region varieties were determined from coefficient of determination, H ($R^2 = -0.66$), GK ($R^2 = -0.65$), and KY ($R^2 < -0.30$). Oleic acid may have been transformed into linoleic acid especially in olive oils produced from these varieties.

Brescia et al.²⁵ reported that fatty acid content did not present any particular trend with ripening, however, the oil fraction sensibly enriched in longer-chain fatty acids with ripening. It has been reported that the contents of mono- and polyunsaturated fatty acids are reduced with olive ripening and this can be expressed by the conversion of enzyme activity to oleic acid and linoleic acids. Baccouri et al.²⁶ reported that the oleic acid level decreased with ripeness or harvest time. However, some authors^{27,28} reported that the increase in linoleic acid content was due to the fact that besides the continuing biosynthesis of triglycerides, with the formation of oleic acid, the enzyme oleate desaturase was active, transforming oleic acid into linoleic acid.

The mean levels of palmitoleic, arachidic, henoicosanoic, behenic and erucic acid being minor FAs in KY, GK, and H are similar, while the mean levels of the FAs in GG is lower. Hepta decanoic acid mean levels in four varieties are near to each other. However, mean levels of cis-10-pendacanoic and cis-11,14,17-eicosatrienoic acid considered as minor fatty acids in GG are higher than the those of the other varieties. On the other hand, it cannot be made generalizations among four varieties in terms of the mean levels of the other minor fatty acids such as heptadecanoic, cis-10-pendacanoic, cis-11-eicosenoic, linolenic, cis-8.11.14-eicosatrienoic acid, and docosahexaenoic acid (DHA) (Table 3).

Some minor FAs, heptadecanoic, lignoceric, cis-11-eicosenoic acids levels in GK and GG olive oil samples as being the same varieties but cultivated in different regions did not show significant differences at a confidence level of 0.05, but significant differences were found for arachidic, henoicosanoic, behenic palmitoleic, and linolenic acids levels at the same confidence level in the same samples (Table 3).

Minor FAs, heptadecanoic, lignoceric, and cis-11-eicosenoic acids levels were not different at a confidence level of 0.05, but significant differences were found at a confidence level of 0.05 for arachidic, henoicosanoic, beheni, palmitoleic, cis-10-pendacanoic, erucic, and linolenic acids levels between olive oil samples of KY and GK varieties cultivated in the same region.

Significant differences ($p < 0.05$) are present between KY and GK varieties cultivated in the same region, minor SFAs such as arachidic and henoicosanoic, minor MUFAs such as palmitoleic, cis-10-pendacanoic, and erucic, and minor PUFA and DHA. However, significant differences ($p < 0.05$) were not found for the levels of some minor SFAs such as heptadecanoic, behenic, and lignoceric acids and a minor MUFA, cis-11-eicosenoic acids. The differences between levels of major FAs in KY and GK varieties cultivated in the same region can be primarily attributed to genotype. The differences and/or similarities between levels of FAs between GK and GG olive oil samples being the same varieties but cultivated in different regions can be attributed to the physicochemical properties of soils and the other ecological factors. Genotype, clearly affects the composition of the fatty acid. The fatty acid profile of an olive oil is genotype dependent in terms of unsaturated/saturated fatty acids ratio.⁷

KY variety contains the highest percentage of total SFAs (19.3%) and PUFAs (10.3%) while GG variety has the lowest total SFAs (16.8%) and PUFAs (8.5%). Because linoleic acid includes the major fatty acid in the fraction, generally, the varieties including higher linoleic acid levels have high total PUFA. GG variety has the lowest total SFAs (16.8%) essentially due to the lowest palmitic acid content, which represents the major fatty

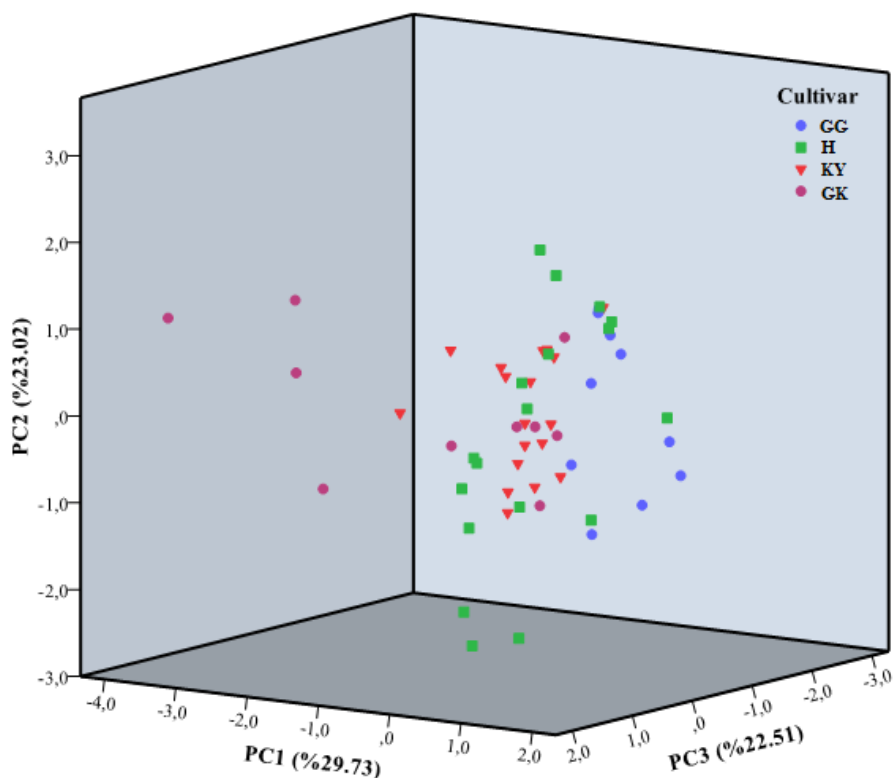


Figure 3. Principal component analyses showing the natural interposition of olive oils of Gemlik-Gemlik (GG), Halhalı (H), Kilis yağlık (KY), and Gemlik-Kilis (GK) varieties.

acid in the SFA fraction. KY, GK, and H varieties may accept similar in respect to the total SFAs levels as 19.3, 18.9 and 19.1%, respectively. SFAs levels between GK and GG olive oil samples as being the same varieties show significant differences at a confidence level of 0.05, except for minor SFAs, heptadecanoic, and lignoceric acids levels among all varieties (Table 3).

In general, SFAs are found at greater levels in the varieties cultivated in the Southeast and Mediterranean Regions. However, significant differences are not seen clearly among mean SFAs levels reported in Gemlik varieties cultivated in Marmara^{9,21-23}, in Aegean^{9,22,29}, in Mediterranean^{13,22} and in Southeastern Anatolia²² Regions (Table 3). GG variety contains the highest percentage (73.9%) of total MUFAs due to the highest oleic acid (C18:1n9c) content (72.6%), which represents the major fatty acid in the MUFA fraction. The total MUFAs of KY, GK, and H are 71.5, 70.6, and 70.9%, respectively. While the mean levels of DHA in KY and GK varieties are minor, it is at trace levels in all of H variety and below the detection limit of GC-FID method in all samples of GG variety. In vitro studies have suggested that PUFAs are more proinflammatory than

monounsaturated FAs (MUFAs) and saturated FAs (SFAs). In fact, linoleic acid has more oxidative and inflammatory stress induction capacity than other fatty acids.³⁰

The levels of minor FAs such as arachidic, henoicosanoic, beheni, palmitoleic, cis-10-pendacanoic, and erucic acids are significantly ($p < 0.05$) different among olive oil samples of four varieties, but the levels of heptadecanoic, lignoceric, and cis-11-eicosenoic are not different among the same samples at the same confidence level. The variations in FAs composition observed in olive oils analyzed (Table 3) are probably related to both genetic factors and environmental conditions during the development and the progress in maturity of the fruit.

3.2. Principal component analyses

There are formed 8 factors, and the total variance ratio is 84.796% when all components are examined together (Figure 3). Fatty acids analyzed in olive oil samples of GG, H, KY, and GK were separated into three components;

Major components (> 1.0%): Major fatty acids are oleic (C18:1n9c), linoleic (C18:2n6c), palmitic (C 16:0), stearic (C18:0), and erucic (C22:1n9). Varimax vertical rotation method is applied after factor loadings are calculated for major components. Eigenvalue have two factors greater than 1. The explained variance rate is 65.732%. The factor loadings of each component are as follows. The variance ratio for PC1 explained by the first component is 34.15% while the variance ratio for PC2 explained by the second component is 31.58%. C18:1n9c, and C22:1n9 acids were the most important factors for PC1 whereas C18:2n6c, C16:0, and C18:0 were the most important factors for PC2 (Figure 3). According to PCA values, GG, H, and GK species seem to be clearly separated. KY olive oil type is mixed partially with GG and H.

Minor components (0.01-1%): Minor components of fatty acids in olive oils are arachidic (C20:0), cis-10-pendacanoic (C17:1), behenic (C22:0), cis-11-eicosenoic (C20:1), palmitoleic (C16:1), henoicosanoic (C21:0), lignoceric (C24:0), linolenic (C18:3n3), and heptadecanoic (C17:0). There are formed 4 factors above Eigen Value 1. The total variance ratio explained is 72.74%. The explained variances for the first three components (PC1, PC2, and PC3) are 22.19%, 17.48%, and 17.26%, respectively. The most important factors are C20:0, C22:0, C20:1, and C17:1 for PC1, C16:1 and C21:0 for PC2, and C24:0 and C18:3n3 for PC3.

Trace components (< 0.01%): Trace components of fatty acids in olive oils are pentadecanoic (C15:0), lignoceric acid (C24:0), cis-10-pendacanoic (C15:1), cis-11.14.17-eicosatrienoic (C20:3n6), and DHA (C22:6n3). There are formed 3 factors above Eigen Value 1. The explained variances for the first three components (PC1, PC2, and PC3) are obtained as 29.73%, 23.02%, and 22.51%, respectively. The most important factors are C15: 0 and C24: 0 for PC1, C15: 1 and C20: 3n6. for PC2.

4. CONCLUSIONS

The present study provide information regarding the fatty acid composition of Kilis yağlık (KY), Gemlik-Kilis (GK), Gemlik-Gemlik (GG), and Halhalı (H) (İskenderun) varieties that are collected from commercial olive oil producers. Individual fatty acid determined for different varieties has covered with the range of the IOC and the Turkish Food Codex standards.

The levels of oleic and palmitic acids in GK and GG showed significant differences at a confidence level of 0.05, but significant differences were found for stearic and linoleic acids levels in the same varieties and at the same confidence level. However, at a confidence level of 0.2, there were three groups according to oleic acid levels; (a) KY and H, (b) GK and (c) GG. Also, KY, GK, and H varieties had similar oleic/linoleic acid ratios (O/L) while GG variety had higher O/L ratio than the

other varieties' O/L ratios. The mean levels of other major FAs such as palmitic, stearic, and linoleic acids in KY, GK, and H varieties were similar, but higher than those in GG. GG variety had both the lowest mean of linoleic and palmitic acid levels. Negative relationships between oleic and linoleic acid levels in the South Anatolian Region varieties were determined from coefficient of determination, H ($R^2 = -0.66$), GK ($R^2 = -0.65$), and KY ($R^2 < -0.30$). The results could be attributed to oleic acid having been transformed into linoleic acid in olive oils produced from these varieties.

The mean levels of palmitoleic, arachidic, henoicosanoic, behenic, and erucic acids being minor FAs in KY, GK, and H were similar, while the mean levels of the FAs in GG was lower. Also, the mean levels of cis-10-pendacanoic and cis-11,14,17-eicosatrienoic acids as minor fatty acids in GG were higher than of other varieties. On the other hand, it cannot be made generalizations among four varieties in terms of the mean levels of the other minor fatty acids such as heptadecanoic, cis-10-pendacanoic, cis-11-eicosenoic, linolenic, cis-8.11.14-eicosatrienoic acid, and DHA.

There were significant differences ($p < 0.05$) between KY and GK in terms of minor SFAs such as arachidic and henoicosanoic, minor MUFAs such as palmitoleic, cis-10-pendacanoic, erucic, and minor PUFA and DHA. However, significant differences were not found for the levels of some minor SFAs such as heptadecanoic, behenic, and lignoceric acids, and a minor MUFA, cis-11-eicosenoic acids. The differences between levels of major FAs in KY and GK varieties cultivated in the same region can be primarily attributed to genotype. It was seen that KY variety contained the highest percentage of total SFAs (19.3%) and PUFAs (10.3%), while GG variety had the lowest total SFAs (16.8%) and PUFAs (8.5%). Because linoleic acid had the major fatty acid in fraction, generally the varieties having higher linoleic acid levels had high total PUFAs. While DHA mean levels in KY and GK were minor, it was at trace levels in all of H and below the detection limit of GC-FID method in all samples of GG variety.

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Conflict of interest

Authors declare that there is no a conflict of interest with any person, institute, company, etc.

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ORCID

 ID 0000-0002-6252-1819 (H. Avci)

 ID 0000-0002-9040-4249 (Y. Uğur)

 ID 0000-0002-9169-9771 (S. Erdoğan)