

## Effects of different holding types and times on quality attributes of oil obtained from olives belonging to Akhisar Region, Turkey

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### Abstract

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**Introduction.** In this study, it was aimed to determine the effects of different holding types and times on quality attributes of oil obtained by extraction using Abencor system.

**Materials and methods.** Edremit and Uslu types of olives which are widely grown in the region of Akhisar, constitute the main material of the study. These two kinds of olives were filled into plastic boxes or nylon sacks. After waiting for 0, 7, 14, 21 days, oils were extracted by using Abencor system. Free fatty acid, peroxide value, UV absorption values (at 232 and 270 nm), the amount of total phenol, refractive index, the amount of total chlorophyll and carotenoid contents, fatty acid composition and color values were determined in olive oil samples. Oxidative stabilities and sensorial properties of olive oils were also examined.

**Results and discussion.** The results indicated that loss in quality of the samples, especially the Uslu variety, held in sacks. Total phenol content of olive oils were decreased excessively during the holding time for both types of samples. Although the majority of the chemical parameters fell within the limits established by the Legislation. Chlorophyll and carotenoid contents ranged, respectively from 0.7 to 8.69 mg/kg and from 0.7 to 3.44 mg/kg for Edremit olive oil and from 0.93 to 2.17 mg/kg and from 0.96 to 1.49 mg/kg. Oleic (C18:1), linoleic (C18:2), palmitic (C16:0) and stearic (C18:0) acids found as predominant fatty acids in all samples. Oil samples obtained from Edremit variety on the first day of holding and the 7th day of the holding period in sack were classified as extra virgin olive oil. Also oil obtained from Uslu variety on the first day of the holding period showed the characteristics of extra virgin olive oil according to the sensorial properties. Whereas other samples were classified as virgin olive oil. The initial Induction Period (IP) of Edremit and Uslu olive oils were 3.9 and 3.8 h. For Uslu variety, it was observed a decrease during holding period at each holding types until 21 th day. For Edremit variety, it was not observed a significant decrease during holding period.

**Conclusion.** According to the results, the chemical parameters analysed in the different samples are within the limits established by the legislation, but vary during the holding period and according to the variety.

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## Introduction

Olive is a typical plant of Mediterranean countries grown in Aegean, Marmara, Mediterranean and South East Anatolian regions of Turkey. Spain is leading the field in olive oil production and olive growing. Italy, Greece and Turkey follow Spain in olive oil production, respectively. The share of our country in world olive oil production is 5,8 %. With mentioned the positive effects of olive oil on health by the experts, its consumption gradually increase in countries with high level of income. Greece continues to lead the ranking with 12.8 kg per capita per year. It is followed by Spain (11.3 kg), Italy (11.3 kg), Portugal (10.5 kg), Cyprus (5.5 kg), Luxemburg (3.2 kg), Malta (3 kg), France (1.7 kg) and Turkey (1.4 kg) [IOOC, 2015]. Period among the planting of olive tree and serving the olive and olive oil to consumer includes extensive processes such as cultural applications like using appropriate variety, planting and pruning and the other processes like harvesting, transportation, pressing and holding. These processes directly affect both olive and olive oil quality and the amount of product which might be obtained in the future.

In this study, it was aimed to determine the effects of different holding types and times on quality attribute of oil obtained after the pressing. Edremit and Uslu type olive cultivars which are widely grown in the region of Akhisar, Turkey that constitute the main material of the study are harvested in 2012-2013 crop season. Their maturation index was indicated. Then these two kinds of olives filled into boxes and nylon sacks and after waiting for 0, 7, 14, 21 days, oils were extracted by using Abencor system. Free fatty acid, peroxide value, UV absorption values (232 and 270), the amount of total phenol, refractive index, color values, the amount of total chlorophyll and carotenoid contents, fatty acid composition were determined as analytical criteria in olive oil samples. The results of the analyses were compared with the criteria given in the Communication for Edible Olive Oil and Olive Pomace of Turkish Food Codex and International Olive Council Legislation and scientific researches [Anon 2014; IOC 2015]. In addition, sensory properties and oxidative stability were evaluated as quality criteria. Up to our knowledge there are rather limited studies about determination the effects of holding times and types on olive oil quality so it's invented that these results elucidate to researchers.

## Materials and Methods

### Materials

Edremit and Uslu varieties of olive which are widely cultivated in Akhisar Region of Manisa/Turkey were used. Olives were manually harvested randomly as 10 kg for each variety at 2012 and 2013 harvest season. Harvested samples for each variety were put into plastic boxes (53x37x31 cm) and nylon sacks (60x90 cm) and were kept inside of them for 0, 7, 14 and 21 days (Figure 1 and Figure 2). Samples were randomly taken from the plastic boxes or nylon sacks for each analysis period. Then they were crushed by Abencor system at Institute of Olive Researches, Bornova/Izmir. They were filtered and filled into the dark-bottles and kept at 4°C until being analyzed. Each of the analyses were repeated three times for each sample.



**Figure 1. Olive samples held in plastic boxes**



**Figure 2. Olive samples held in nylon sacks**

### **Maturation index**

The MI of olive fruits was determined according to the method given in by Vinha *et al.* [2005]. MI was calculated after visual colour inspection over a hundred randomly chosen olives, according to the following formula:  $MI=(a \times 0)+(b \times 1)+(c \times 2)+(d \times 3)+(e \times 4)+(f \times 5)+(g \times 6)+(h \times 7)/100$ . Where a, b, etc. are the number of olives in each of the seven colour classes from dark green to dark black.

### **Olive oil extraction by Abencor system**

Oil was extracted using an Abencor laboratory mill (Polat Machinery Inc.), simulating commercial oil extraction systems. Extraction process consisted of the following steps: fruit

crushing and malaxation for 60 min at  $35 \pm 1^\circ\text{C}$ , two rounds of centrifugation, 60 s each at 3000 rpm, with 100 mL water added between rounds. After centrifugation, oils were decanted, filtered, transferred into amber glass bottles and held at  $4^\circ\text{C}$  until analysed.

### **Determination of physical characteristics**

Color values of the olive oil samples were determined in terms of  $L^*$ ,  $a^*$ , and  $b^*$  criteria using Lovibond PFX880-Tintometer [AOCS 1993a]. The optical path length of the glass cell was 1" for extra virgin olive oil. The refractive index values of the samples were carried out using method of Turkish Standard 4960 EN ISO 6320 by Abbe Refractometer at  $20^\circ\text{C}$  [Anon, 2010].

### **Determination of quality criteria**

The samples were analyzed after each holding times and types. Acidity (% as oleic acid) and peroxide value (meq  $\text{O}_2/\text{kg}$ ) of the virgin olive oil samples were carried out according to the method reported by AOCS [1993b]. and Salvador et al. [2000]. UV specific extinctions of K232 and K270 were determined by Anon [2014]. Chlorophylls and carotenoids were determined colorimetrically following the method [Morello *et al.* 2004; Beltran *et al.* 2005]. The maximum absorption at 670 nm is related to the chlorophyll fraction and at 470 nm is related to carotenoid fraction.

### **Total phenol content**

Total polyphenol content was analyzed as described [Gutfinger, 1981]. The phenolic compounds were isolated from a solution of oil in hexane by extraction with a water/methanol mixture (60:40, v/v). The Folin–Ciocalteu reagent was added to a suitable aliquot of the combined extracts, and the absorption of the solution was measured at 725 nm. The results were evaluated in terms of gallic acid as mg GA/kg oil.

### **Fatty acid composition**

Fatty acid compositions of the oil samples were determined by GC [AOCS, 1997]. Methyl esters were prepared by vigorous shaking of a solution of each olive oil sample in n-hexane (0.2 g in 5 mL) with 0.5 mL 2 N methanolic potassium hydroxide solution. Chromatographic analysis was performed on a Agilent Technologies 6890N Gas Chromatograph (Hewlett-Packard Company, Wilmington, DE, USA), equipped with a capillary column (Agilent 122-2362: 60 m x 0.25 mm x 0.25  $\mu\text{m}$ ), an injector split-splitless and a FID detector. The carrier gas was nitrogen, with a flow rate of 25 ml/min. Initial oven temperature was  $170^\circ\text{C}$ , raised at  $2^\circ\text{C}/\text{min}$  to  $210^\circ\text{C}$  and hold for 15 min. The temperatures of the injector and the detector were held at  $250^\circ\text{C}$ . Fatty acids were identified by comparing retention times with those of standard compounds.

### **Measurement of oxidative stability**

An automated Metrohm Rancimat apparatus model 743 (Metrohm, Switzerland) capable of operating over a temperature range of  $50\text{--}200^\circ\text{C}$  was used for induction period determination of oil samples. Fourteen oil samples were analyzed in the equipment at the same time. For oxidative stability measurement, each oil sample ( $3.0 \pm 0.1$  g) was weighted

into the reaction vessel glassware. The conductimetry cells were filled with deionized water up to 90 ml. Air was flown through the heated oil at different rates, depending on the temperature. The flow rate of air was 20 l/h used for 120°C. Although this equipment (Model 743 Metrohm) presents a cleaner system of procedure, the glassware was rigorously cleaned between each run to avoid any contamination that would catalyze the peroxidation [Anwar *et al.* 2003; Mateos *et al.* 2006].

### **Sensory profile analysis**

Sensory analysis was carried out by a panel of 10 selected trained judges of the Olive Oil Research Institute, İzmir, Turkey, coordinated by a panel head [IOOC, 2010]. The sensory sessions were conducted in a sensory room equipped with 10 booths. Panellists evaluated olive oil samples as their negative and positive properties on a scale from 0 to 5.

### **Statistical Analysis**

The data was analyzed utilizing the PROC MIXED procedure of SAS [SAS, 2000]. To find the treatment combination, means including all treatment combinations (holding time, holding types) for each olive variety were generated using the PROC MIXED procedure and these were compared to a control.

## **Results and Discussion**

### **Physical characteristics of olive oil samples**

According to the different holding types and times, physical characteristics of olive oils obtained from Edremit and Uslu varieties which were harvested at the maturation index of 3.75 and 4.5 respectively, are given in Table 1 and 2.

L\*, a\* and b\* values of olive oils were ranged between 19.9-85.81, 2.53-12.64 and 30.33-92.8, respectively. After holding period of the olive samples, L\*, a\* and b\* values of the olive oils increased at both type (Table 1-2). As regards to the statistical results, the interaction of holding types and times was found significantly important for L\*, a\* and b\* values at Edremit variety while it was found significantly important only for L\* and b\* values at Uslu variety ( $p < 0,0001$ ). The holding conditions and holding times of olives after harvest, cultivar, soil, climate conditions, degree of ripeness and irrigation can affect color, smell, taste and stability of oils [Kritsakis, 1998]. So, there are differences between initial and subsequent color values of olive oil samples.

One of the physical purity criteria is refractive index values for natural olive oils. Refractive index values of the samples were among 1,4696-1,4708. When compared with initial values, there were a slight decrease for each variety due to the holding types and times (Table 1-2).

**Table 1**  
Least squares means of holding times and holding types on color values (L, a\*, b\*) and refractive index (RI) value for Edremit variety olive oil<sup>a</sup>

Source of variation	L	a*	b*	RI (nD 20° C)
<b>Holding times, week</b>				
0	85,79	12,09	80,09	1,4704a
7	82,49	6,56	50,02	1,4702b
14	37,49	5,2	50,73	1,4698c
21	45,53	6,07	60,81	1,4697c
P-value	<0,001	<0,0001	<0,0001	0,0002
<b>holding types</b>				
box	66,33	9,44	76,21	1,4699
sack	59,32	5,52	44,61	1,47
P-value	<0,0001	<0,0001	<0,0001	0,24
<b>holding timesxholding types</b>				
0/b	85,77f	12,12g	80,07d	1,4704
0/s	85,81f	12,06g	80,1d	1,4703
7/b	80,86e	11,61f	92,8e	1,47
7/s	84,12f	1,52a	71,24c	1,4704
14/b	55,09d	7,87e	71,13c	1,4698
14/s	19,9a	2,53b	30,33a	1,4698
21/b	43,62b	6,16d	60,84b	1,4696
21/s	47,44c	5,98c	60,77b	1,4697
P-value	<0,0001	<0,0001	<0,0001	0,06

Within columns, means with different letters differ ( $P < 0.05$ ).

<sup>a</sup> Letters were not assigned to main effect if means when interaction was significant

**Table 2**

Least squares means of holding times and holding types on color values (L, a\*, b\*) and refractive index (RI) value for Uslu variety olive oil <sup>a</sup>

Source of variation	L	a*	b*	RI (nD 20° C)
<b>Holding times, week</b>				
0	85,29	12,6a	82,19	1,4705
7	77,13	10,19b	73,63	1,4707
14	80,19	10,02b	74,81	1,4703
21	80,54	10,26b	74,68	1,4701
<i>P-value</i>	0,0006	0,0004	<0,0001	0,11
<b>Holding types</b>				
box	83,24	10,91	78,22	1,4704
sack	78,33	10,63	74,43	1,4703
<i>P-value</i>	0,0001	0,16	0,0001	0,0002
<b>Holding timesxholding types</b>				
0/b	85,35e	12,64	82,2f	1,4706d
0/s	85,22e	12,56	82,17f	1,4704c
7/b	84,4e	10,72	74,65c	1,4706d
7/s	69,86a	9,67	72,61b	1,4708e
14/b	83,64de	10,02	78,95e	1,4702b
14/s	76,73b	10,03	70,68a	1,4704c
21/b	79,55c	10,26	77,1d	1,4704c
21/s	81,53cd	10,26	72,25ab	1,4698a
<i>P-value</i>	0,0001	0,15	0,003	0,001

Within columns, means with different letters differ ( $P < 0.05$ ).

<sup>a</sup> Letters were not assigned to main effect if means when interaction was significant

### Quality criteria of olive oil samples

According to the quality indices (free fatty acids, peroxide value and UV absorption parameters, K232 and K270), all samples were in line with the limit values for EVOO, OO and RFOO set by the Turkish Food Codex on Olive Oils and Olive Pomace Oils [Anon, 2014].

As seen in Table 3 and Table 4, FFA of Edremit and Uslu type olive oils increased constantly during holding time. In Edremit variety, FFA increased to 2.7 % from 0.62 % and for Uslu variety it reached upto 1.33% from 0.25% at the end of holding period in sacks. Regarding the influence of holding conditions, interaction of holding types and

holding times affect FFA content of Edremit olive oil variety ( $p < 0.0001$ ) whereas in Uslu olive oil variety only holding times affect FFA content ( $p < 0.0001$ ). It is thought that the reason of the high free fatty acidity of the oil was arisen from the mechanical injury of the olives during harvest and the harms occurred during 21-days holding period.

**Table 3**  
Least squares means of holding times and holding types on some analytical criteria and total phenol content for Edremit variety olive oil <sup>a</sup>

Source of variation	FFA (% as oleic acid)	PV (meqO <sub>2</sub> /kg)	K232 (E% <sup>1</sup> 1cm)	K270 (E% <sup>1</sup> 1cm)	Total chlorophyll (mg/kg)	Total carotenoid (mg/kg)	Total phenol (mg/kg GA)
<b>Holding times, week</b>							
0	0,62	4,88	1,69	0,06a	0,61	0,67	251,43
7	0,74	4,94	1,3	0,06a	6,59	2,77	87,28
14	1,6	3,4	1,7	0,18b	4,37	2,22	62,77
21	2,05	3,81	1,85	0,15c	4,21	2,25	44,27
P-value	<0,0001	<0,0001	0,16	<0,0001	<0,0001	<0,0001	<0,0001
<b>Holding types</b>							
box	1,06	3,98	1,48	0,1a	3,48	1,85	113,22
sack	1,44	4,54	1,78	0,12b	4,4	2,1	109,64
P-value	<0,0001	0,0007	0,13	0,03	0,001	0,015	0,21
<b>Holding timesxholding types</b>							
0/b	0,61a	4,83a	1,68	0,06	0,7a	0,7a	235,03f
0/s	0,62a	4,93a	1,7	0,06	0,52a	0,64a	267,83g
7/b	0,75c	4,9a	0,9	0,41	4,5b	2,11b	100,13e
7/s	0,73b	4,99a	1,7	0,82	8,69c	3,44c	74,39d
14/b	1,47e	3,11d	1,7	0,18	4,43b	2,34b	64,48c
14/s	1,73f	3,69c	1,7	0,18	4,3b	2,1b	61,06bc
21/b	1,41d	3,08d	1,67	0,13	4,31b	2,28b	53,26b
21/s	2,7g	4,55b	2,03	0,17	4,1b	2,22b	35,28a
P-value	<0,0001	0,0034	0,33	0,18	0,001	0,0005	0,001

Within columns, means with different letters differ ( $P < 0.05$ ).

<sup>a</sup> Letters were not assigned to main effect lsmeans when interaction was significant

FFA: Free fatty acid, PV: Peroxide value

It is thought that free fatty acid values of the sample increased due to the activity of lipolytic enzymes in the olive fruit. The activity of this enzyme is influenced from fruit quality, climate conditions, maintenance requirements olives and process conditions [Mateos *et al.* 2006].



The effect of holding time and container type on quality of extra virgin olive oil have reported by Mendez and Falqu  [2007]. According to their findings, the acidity of olive oil samples increased to 0.38% from 0.25% at the end of 6-month holding period in plastic containers. It is thought that the differences among the results of the researches with literature data depend to a number of factors as olive harvest time, applied technology, preservation and holding conditions of olives.

**Table 4**  
**Least squares means of holding times and holding types on some analytical criteria and total phenol content for Uslu variety olive oil <sup>a</sup>**

Source of variation	FFA (% as oleic acid)	PV (meqO <sub>2</sub> /kg)	K232 (E% <sup>1</sup> 1cm)	K270 (E% <sup>1</sup> 1cm)	Total chlorophyll (mg/kg)	Total carotenoid (mg/kg)	Total phenol (mg/kg GA)
<b>Holding times, week</b>							
0	0,25a	6,09c	2,06	0,13a	1,04a	1	174,8a
7	0,47b	9,08a	2,27	0,14a	1,97b	1,22	69,82b
14	1,11c	8,28ab	1,9	0,17ab	1,58ab	1,02	59,37c
21	1,31d	8b	2,62	0,23b	2,1b	1,37	20,22d
P-value	<0,0001	0,0015	0,12	0,04	0,03	0,007	<0,0001
<b>Holding types</b>							
box	0,78	8,19b	2,38	0,17	1,54	1,02	81,74
sack	0,79	7,53a	2,05	0,16	1,8	1,29	80,37
P-value	0,46	0,04	0,15	0,48	0,18	0,003	0,64
<b>Holding times-holding types</b>							
0/b	0,28	6,39	2,1	0,13	0,93	0,96b	177,33
0/s	0,22	5,79	2,02	0,12	1,14	1,04b	172,26
7/b	0,48	9,49	2,37	0,15	1,76	1,13bc	71,48
7/s	0,47	8,68	2,18	0,14	2,17	1,3cd	68,16
14/b	1,09	8,66	2,37	0,2	1,41	0,72a	60,87
14/s	1,14	7,9	1,42	0,14	1,76	1,33cd	57,88
21/b	1,28	8,24	2,67	0,22	2,1	1,26c	17,26
21/s	1,33	7,76	2,57	0,24	2,14	1,49d	23,18
P-value	0,43	0,95	0,36	0,56	0,89	0,05	0,5

Within columns, means with different letters differ ( $P < 0.05$ ).

<sup>a</sup> Letters were not assigned to main effect lsmeans when interaction was significant

The peroxide value underwent a significant increase up to the 7th day of holding for both variety, then slowly decreased up to the 21th day in box and sack, probably due to the fact that the newly formed oxidation products were further converted to secondary ones (Table 3-4). But nevertheless, they did not exceed the legal limit of  $\leq 20$  meqO<sub>2</sub>/kg for extra virgin olive oil set by the Anon [2014] and IOC [2015]. According to the statistical results, the interaction of holding types and holding times was found important for Edremit olive oil variety ( $p < 0.05$ , Table 3), whereas for Uslu olive oil variety, the effect of holding times and holding types were found significantly important ( $p < 0.05$ , Table 4). The peroxide value of Edremit and Uslu type olive oils from different regions and different harvest period have identified. Their peroxide value was changed between 3,01–8,05 meq O<sub>2</sub>/kg and 4,52–8,16 meq O<sub>2</sub>/kg, respectively [Yavuz, 2008]. It was observed that these values are similar to the values in our study. It is known that oxygen in the air causes a rise in peroxide value. Heat and light accelerate these oxidative chemical reactions. Therefore, olives and olive oils should not be exposed to the oxygen for a long time and should be kept in dark places at 10–15 °C [Allalout *et al.* 2009].

The initial values of the coefficients K232 and K270 are between 1.68 and 0.06 for Edremit variety and 2.12 and 0.12 for Uslu variety, within the limits permitted by the International Olive Council Legislation [IOC, 2015]. In Table 3 and 4, it can be seen that the effect of different holding times and holding types did not significantly affect the changes in K232 value of both type ( $p > 0.05$ ). But it was found that holding types and holding times had separately significant importance on the changes in K270 value of Edremit olive oil, whereas only the effect of holding times on K270 value was found significantly important for Uslu olive oil ( $p < 0.05$ ). K270 value of Uslu olive oil was slightly exceeded the legal limit of  $\leq 2.60$  for virgin olive oil as required by IOC at 21th day of holding [IOC, 2015] (Table 4). This occur due to the increase in the number of compounds resulting from the degradation of the hydroperoxides, which is confirmed by an increase of K270 value [Yavuz, 2008].

The natural pigments contents of the oils are important for the quality parameters because they correlate with colour and play a key role as a factor of sensorial acceptability among consumers. Virgin olive oil has a color changing among green-yellow to gold, depending on the variety and the stage of maturity [Salvador *et al.* 2000]. They undergo during fruit ripening and oil holding so they could be considered as a product freshness indicator [Allalout *et al.* 2009]. As shown in Table 3 and Table 4, Chlorophyll and carotenoid contents ranged, respectively from 0.7 to 8.69 mg/kg and from 0.7 to 3.44 mg/kg for Edremit olive oil and from 0.93 to 2.17 mg/kg and from 0.96 to 1.49 mg/kg.

These results are in agreement with the findings of which reported that the chlorophylls and carotenoids contents ranged, respectively from 0.05 to 1.52 mg/kg and 2.03 mg/kg for Moroccan cultivars [Tanouti *et al.* 2011]. It should be noted that the values found for the two pigments were low. This can be attributed to the geographical origin, olive cultivar, fruit ripeness, soil and climatic condition and the processing procedures [Psomiadou and Tsimidou, 2001]. Generally, oils obtained in irrigation regime had higher levels of chlorophyll pigments than non-irrigated one. However, the content of carotenoids was less affected by water supply [Baccouri *et al.* 2008]. Chlorophyll and carotenoid contents of Edremit olive oils have been identified as 0.95–6.73 mg/kg and 1.51–5.13 mg/kg, respectively. They reported that their contents decreased as maturation index increased [Yorulmaz *et al.* 2013].

The effect of interaction of the type of holding types and holding times significantly affected the changes in chlorophyll and carotenoid contents of Edremit variety ( $p < 0.001$ , Table 3), whereas for Uslu variety while the interaction of holding types and holding times

affected the carotenoid content ( $p < 0.05$ , Table 4). For chlorophyll content, only the effect of holding times was found statistically important ( $p < 0.05$ , Table 4).

### **Total Phenol content of olive oil samples**

The amount of total phenolic compounds in virgin olive oil is an important factor when evaluating its quality, given that the natural phenols improve its resistance to oxidation, and to certain extent, are responsible for its bitter taste [Baccouri *et al.* 2008]. The concentration of total phenols ranged between 35.28–267.83 mg/kg and 17.26–177.33 mg/kg for Edremit and Uslu olive oil varieties, respectively (Table 3–4). The higher concentrations of total phenol were found at the beginning of holding for both types. Then total phenols underwent a decrease during holding period. The literature reports that their decrease is due to the decomposition process and the oxidative activity of these compounds [Morello *et al.* 2004]. The most significant decrease of total phenol content occurred during holding in plastic boxes for both types.

Generally, total phenol content of olive oils was lower according to the literature reports. The characteristics of olive and olive oils during ripening have been examined and they found total phenol content between 550.33–810.98 mg/kg in Edremit olive oil [Yorulmaz *et al.* 2013]. In another study, total phenol content of “Bosana” extra virgin olive oil was found between 248–409 mg/kg oil during 16 months of holding under the light and dark [Del Caro *et al.* 2006]. Total phenol content of Edremit virgin olive oils were found between 89.2–128.7 mg/kg harvested in 2005 [Andjelkovic *et al.* 2009]. This results showed similarity with our results. These differences of total phenol content of virgin olive oils are depending to the variety, climatic conditions, maturity index, irrigation, time and type of harvesting, holding and processing technology [Boskou, 1996].

According to the statistical analyses results, it was determined that the interaction of holding types and holding times affected the changes in total phenol content of Edremit olive oil ( $p < 0.001$ , Table 3). On the other hand, only holding time was found statistically important on the changes in total phenol content of Uslu olive oil ( $p < 0.0001$ , Table 4).

### **Fatty acid composition of olive oil samples**

The major fatty acids found in Edremit and Uslu olive oils had oleic (C18:1), linoleic (C18:2), palmitic (C16:0) and stearic (C18:0) acids. These predominant fatty acids were ranged in all samples between 70.05–71.97 %, 10.22–12.13 %, 12.40–13.87 %, 2.01–2.62 %, respectively. The distribution of fatty acids in the samples was in agreement with the Turkish Food Codex on Olive Oils and Olive Pomace Oils [TFC, 2014/53]. Palmitoleic (C16:1), heptadecanoic (C17:0), heptadecenoic (C17:1), linolenic (C18:3), arachidic (C20:0) and gadoleic (C20:1) acids were determined in low amounts.

In Edremit and Uslu virgin olive oils, total SFA, MUFA and PUFA were almost stable due to the different holding types and times as seen in Table 5 and Table 6. The samples showed the lowest total PUFA (10.7–12.05 % for Edremit, 12.16–12.67 % for Uslu) and the highest total MUFA (71.80–73.32 % for Edremit, 71.31–71.99 % for Uslu). As for that total SFA, it varied between 15.77–16.42 % for Edremit and 15.34–16.26 % for Uslu (Table 5–6). Fluctuations in total SFA, MUFA and PUFA contents observed in olive oil samples are related to holding conditions (Table 5–6). The holding times did not affect their contents in both types. The effect of the interaction of holding time and type was found statistically important on the changes of total MUFA and PUFA contents of Edremit olive oil ( $p < 0.05$ , Table 5), whereas holding time caused significant effect for SFA and PUFA

contents and also interaction of holding time and type was found important on the changes of total MUFA contents of Uslu olive oil ( $p < 0.05$ , Table 6).

The fatty acid profiles of our samples were also in good agreement with values obtained for the same varieties in some other studies [Allalout *et al.* 2009; Yorulmaz *et al.* 2014]. Some characterization of Turkish olive oils have been investigated by Yorulmaz *et al.* [2014]. As they reported, average oleic, linoleic, palmitic and stearic acids of Uslu and Edremit olive oils were 72.34–71.57%, 10.18–10.37%, 12.65–12.98% and 1.70–2.25%, respectively. the total SFA, MUFA and PUFA contents in different cultivars of Spanish and Greece olive oils have been identified as 14.22–20.18%, 61.24–76.61% and 8.82–13.56%, respectively [Allalout *et al.* 2009]. Total SFA, MUFA and PUFA contents were 15.5 %, 73.3 % and 8.6 % in different cultivars [Mendez and Falqu , 2007]. Total SFA, MUFA and PUFA contents of Edremit olive oil were 12.59 %, 74.29 % and 8.93 %. They also indicated the major fatty acid as oleic acid (72.78 %) in Edremit olive oil [Matthaus and I zcan, 2011]. The fatty acid composition of olive oil varies widely depending on the cultivar, maturity of the fruit, altitude climate and several other factors [Matthaus and I zcan, 2011]. Various factors such as harvest period, cultivar and origin affect the formation of the main fatty acids of olive oil in different ways [Bruni *et al.* 1994].

**Table 5**  
Least squares means of holding times and holding types on total fatty acids composition (Totals SFA, MUFA and PUFA) for Edremit variety olive oil <sup>a</sup>

Source of variation	Totals		
	SFA (%)	MUFA (%)	PUFA (%)
<b>Holding times, week</b>			
0	16,38	71,86	11,81
7	15,91	72,96	11,02
14	16	72,07	11,88
21	16,1	72,19	11,62
P-value	0,18	0,0003	<0,0001
<b>Holding types</b>			
box	16,15	72,41	11,43
sack	16,1	72,13	11,73
P-value	0,44	0,01	0,0005
<b>Holding timesxholding types</b>			
0/b	16,42	71,8a	11,9de
0/s	16,34	71,92a	11,72c
7/b	15,93	73,32d	10,7a
7/s	15,89	72,59c	11,33b
14/b	15,81	72,16b	11,94de
14/s	16,2	71,97a	11,82cd
21/b	16,43	72,34b	11,19b
21/s	15,77	72,04ab	12,05e
P-value	0,07	0,05	0,0003

Within columns, means with different letters differ ( $P < 0.05$ ).

<sup>a</sup> Letters were not assigned to main effect lsmeans when interaction was significant

SFA: Saturated fatty acids, MUFA: Monounsaturated fatty acids, PUFA: Polyunsaturated fatty acids

**Table 6**  
**Least squares means of holding times and holding types on total fatty acids composition (Totals SFA, MUFA and PUFA) for Uslu variety olive oil <sup>a</sup>**

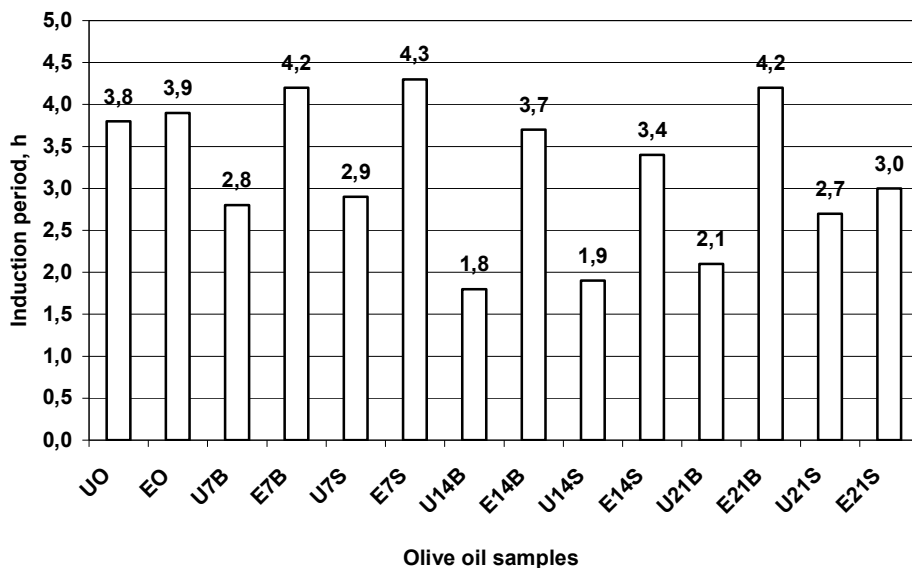
Source of variation	Totals		
	SFA (%)	MUFA (%)	PUFA (%)
<b>Holding times, week</b>			
0	15,77ab	71,84	12,48ab
7	16,21a	71,47	12,29b
14	15,43b	71,81	12,61a
21	15,44b	71,9	12,62a
P-value	0,03	0,004	0,02
<b>Holding types</b>			
box	15,69	71,8	12,55
sack	15,73	71,7	12,48
P-value	0,83	0,14	0,15
<b>Storage times x holding types</b>			
0/b	15,68	71,93cd	12,6
0/s	15,86	71,75bc	12,36
7/b	16,26	71,31a	12,41
7/s	16,15	71,62b	12,16
14/b	15,34	71,97cd	12,62
14/s	15,51	71,64b	12,6
21/b	15,49	71,99d	12,57
21/s	15,39	71,8bcd	12,67
P-value	0,84	0,02	0,2

Within columns, means with different letters differ ( $P < 0.05$ ).

<sup>a</sup> Letters were not assigned to main effect means when interaction was significant

### Oxidative Stability of olive oil samples

The Rancimat method is an accelerated stability test that provides very useful information about the resistance of oil to oxidation [Beltran *et al.* 2005]. In Figure 3 is given the changes of IP values at 120°C for Edremit and Uslu olive oils obtained according to the different holding types and holding times. The initial IP of Edremit and Uslu olive oils were 3.9 and 3.8 h. For Uslu variety, it was observed a decrease during holding period at each holding types until 21 th day. At 21th day, there was a slight increase from 1.8 h to 2.1 h in box and 1.9 h to 2.7 h in sack. This caused that nonhomogenous sampling. For Edremit variety, it was not observed a significant decrease during holding period.



**Figure 3. Changes of induction period (hour) of Edremit and Uslu Olive Oil Varieties according to the different storage types and times**

- UO – Uslu variety first day of harvest
- EO – Edremit variety at first day of harvest
- U7B – Uslu variety holded at 7 days in box,
- E7B – Edremit variety holded at 7 days in box
- U7S – Uslu variety holded at 7 days in sack
- E7S – Edremit variety holded at 7 days in sack
- U14B – Uslu variety holded at 14 days in box
- E14B – Edremit variety holded at 14 days in box
- U14S – Uslu variety holded at 14 days in sack
- E14S – Edremit variety holded at 14 days in sack
- U21B – Uslu variety holded at 21 days in box
- E21B – Edremit variety holded at 21 days in box
- U21S – Uslu variety holded at 21 days in sack
- E21S – Edremit variety holded at 21 days in sack

IP of Frantoio cultivar olive oil was determined 3.7 h at 120°C [Mateos *et al*, 2006]. This finding is similar with our initial IP results. As the ripening occurs, the oil stability decreases according to the results described previously [Gutierrez *et al*, 1999].

### Sensory Profile of olive oil samples

According to the results of sensory evaluation, two positive attributes of Uslu olive oil as fruitiness and pungency was found higher at 0th day. On the other hand, negative attributes as fusty-humid and winy of Uslu olive oil were found higher at 14th day holding in box (Table 7). All olive oil samples were located at mature index.

**Table 7**  
**Negative and Positive Sensorial Properties of Edremit and Uslu olive oil varieties according to the different holding times and holding types**

Variety	Holding Times (week)	Holding Types	Negative properties					
			Muddy Sediment	Fusty-humid	Winey	Wet Wood	Rancid	Others
<b>Uslu</b>	0	-	-	-	-	-	-	-
	7	box	-	2	-	-	1,9	-
	14	box	0,75	2,5	3,2	-	-	-
	21	box	3,3	2	-	-	-	-
	7	sack	-	-	2,5	-	-	-
	14	sack	3	-	1	-	-	-
	21	sack	2,7	1,25	2,5	-	-	-
<b>Edremit</b>	0	-	-	-	-	-	-	-
	7	box	-	1,5	-	-	-	-
	14	box	-	2,4	-	-	-	-
	21	box	2	-	-	-	-	-
	7	sack	-	-	-	-	-	-
	14	sack	2,25	-	2	-	-	-
	21	sack	-	2,5	3	-	-	-

Variety	Holding Times (week)	Holding Types	Positive properties			
			Fruity	Bitter	Pungent	Maturity
<b>Uslu</b>	0	-	3,5	0,5	2	mature
	7	box	2,15	-	1,3	mature
	14	box	1,5	-	-	mature
	21	box	1,4	-	-	mature
	7	sack	2	-	1,3	mature
	14	sack	1,4	-	1,6	mature
	21	sack	1,85	-	-	mature
<b>Edremit</b>	0	-	2,75	-	1,5	mature
	7	box	2,45	-	1,5	mature
	14	box	2	0,5	-	mature
	21	box	2	-	-	mature
	7	sack	3,1	1	1,5	mature
	14	sack	2	-	-	mature
	21	sack	1,5	-	-	mature

According to the Table 8, while Edremit olive oil at initial day of holding, 7th day in sack and Uslu olive oil at initial day of holding were taken a part of extra virgin olive oil, other samples were classified as virgin olive oil according to the IOOC Method [IOOC, 2010]. Fruity score ranged from 1.4 to 3.5 for both olive oil samples. Fruit score were determined score as from 1.8 to 3.2 in Cornicabra virgin olive oils [Salvador *et al.* 2000]. Generally, muddy-sediment occurred at the latest holding times as 14th and 21th day for

both types. One of the negative attributes wet wood was not perceived in any sample (Table 7). Additionally, intensity of rancid was only determined in Uslu olive oil on 7th day of holding at box.

**Table 8**

**Limits established for Median of defect and fruity attribute (IOC, 2015)**

	Median of defect	Median of the fruity attribute
<b>Extra virgin olive oil</b>	Me=0	Me>0
<b>Virgin olive oil</b>	0<Me≤3.5	Me>0
<b>Ordinary virgin olive oil</b>	3.5<Me≤6.0	
<b>Lampante virgin olive oil</b>	Me>6	

The sensory characteristics of bitterness and pungency are due to the activation of taste receptors and trigeminal nerve endings associated with taste buds in fungiform papillae, sensitive to chemical stimuli. In virgin olive oils, these sensations are related to the presence of phenolic compounds and can persist for rather long times after deglutition, showing a clear after-effect that can greatly vary among olive oils in intensity and duration and might affect consumer acceptance [Nieto *et al.* 2010].

Researches showed that the main reason of quality reduction in the olives which are holded before being processed is the increase in acidity and peroxide number [Cimato, 1990; Garcia *et al.* 1996].

## Conclusion

The evaluation of the influence of holding types and times of olives on quality parameters and oxidative stability of olive oils is important for exhibit the requirement to not holding olives long time at inconvenient conditions. This study is also of great interest to the local industrial sector and consumers. Quality criteria of olive oils becomes much closer or exceeds the upper legal limits due to negative factors such as harvesting olives at inappropriate maturity index, the long holding period until processing, transport stage after harvesting. In addition to these, producer awareness has also effect on changes of olives quality criteria. Therefore, harvest should be made with appropriate methods and then olives transport to the place of processing and be holded in properly packaged without losing time. At this point, it is not the right approach to say that olive producers has the same consciousness in each region.

Post-harvest transport stage is the stage that showed the most increase the amount of free fatty acidity and oxidation as a result of the continued respiration in fruit and hydrolytic degradation.

According to the results, the chemical parameters analysed in the different samples are within the limits established by the legislation, but vary during the holding period and according to the variety. With respect to holding type, the acidification and oxidation of the olive oil increase during holding in sack. When compared two types of olives, Edremit variety has higher quality than Uslu variety in terms of phenolic compounds, peroxide value and thereby oxidative stability. The scores of positive sensorial properties of Edremit variety are higher, as well.



It is seen that the traditional olives holding methods using generally at villages and towns are not suitable for stability and quality of olive oils obtained from these olives. As a consequence, oxygen and light induce a rapid deterioration in sacks and boxes. Likewise, acidification, loss of phenolic compounds, oxidative rancidity and sensorial quality is favoured. However, olives should be holded at concrete and smooth surfaced panes or under plastic bedstead for not more high than 10-12 cm, after harvesting up to one or two months at maximum. The most important issue for the holding of olive is to provide getting the air and not be crushed.

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