



Evaluation of Marketed Almond Oils [*Prunus dulcis* (Mill.) D.A. Webb] in Terms of European Pharmacopoeia Criteria

Aysel BERKKAN¹, Berra Nur DEDE TÜRK², Sultan PEKACAR², Onur Kenan ULUTAŞ³, Didem DELİORMAN ORHAN^{2*}

¹Gazi University, Faculty of Pharmacy, Department of Analytical Chemistry, Ankara, Türkiye

²Gazi University, Faculty of Pharmacy, Department of Pharmacognosy, Ankara, Türkiye

³Gazi University, Faculty of Pharmacy, Department of Toxicology, Ankara, Türkiye

ABSTRACT

Objectives: Almond oil marketed for health benefits and cosmetic purposes should be in compliance with the European Pharmacopoeia (EP) criteria. Therefore, in this study, 17 almond oil samples sold in pharmacies, herbal shops, online, and cosmetics stores were analyzed in terms of "almond oil" monograph criteria, which have been mentioned in EP 7.0.

Materials and Methods: In this study, 17 almond oil samples sold in pharmacies, herbal, online, and cosmetics stores were analyzed in terms of "almond oil" monograph criteria, which have been mentioned in EP 7.0. Appearance, acidity value, and peroxide value of each sample were determined and the ingredients were identified by thin layer chromatography. Fatty acids were analyzed by gas chromatographic method using flame ionization detector.

Results: It was determined that two of the 17 samples complied with EP 7.0 criteria.

Conclusion: Almond oil, which is currently marketed according to the manufacturer's own marketing and quality criteria, is excluded from the Turkish Food Codex Standards. Our research has shown that most of the products do not comply with the EP standards. For this reason, it should be ensured that almond oil is listed in this codex and urgent arrangements should be made for quality control analysis.

Key words: Fatty acids, gas chromatography-mass spectrometry, *Prunus dulcis*, quality control

INTRODUCTION

Türkiye, with its diversity of plant species, is one of the world's most important gene sources. The almond is one among the important species in this gene source, and is grown or cultivated across widespread in Türkiye. Almond [*Prunus dulcis* (Mill.) D.A. Webb, syn. *P. amygdalus* Batsch, and *P. communis* (L.)] are divided into two varieties,¹ pomologically, sweet almond (*P. dulcis* var. *dulcis*) and bitter almond (*P. dulcis* var. *amara*) (*P. amygdalus* Batsch), where all belongs to the family Rosaceae, along with raspberries, peaches, apples, and pears. About of 20 species have been reported to grow in Iran, while there have been more than 30 wild species in the world.²

Almonds, which are typically used as snack foods or found as an ingredient in many products,^{3,4} have a considerable

economic value with their different usage areas (gastronomy, confectionery etc.).⁵⁻⁹ They are commonly cultivated for their fruits¹⁰ and place number one among the products of tree nuts.¹¹ Due to their high nutritional content and their promising effects on human health with their high levels of monounsaturated fatty acid and polyunsaturated fatty acid content, consumption of and demand for almonds remains high.⁵⁻⁷ The almond fixed oil obtained from almond seeds is used as medicine, pharmaceutical, and cosmetic products to treat dry skin disorders such as psoriasis.¹² Many studies on the biological value and chemical properties of nut proteins and oils have been reported.¹³ *In vivo* studies have reported that almond seeds and oils possess hepatoprotective, anti-inflammatory, anticancer, and immunestimulant effects. Among tocopherols, phytosterols, and many other health-promoting micronutrients,¹⁴ almond with

*Correspondence: didemdeliorman@gmail.com; didem@gazi.edu.tr, Phone: +90 533 745 96 68, ORCID-ID: orcid.org/0000-0003-3916-4048

Received: 13.05.2021, Accepted: 06.09.2021

©Turk J Pharm Sci, Published by Galenos Publishing House.

its high content of mono-unsaturated fatty acids, can reduce gastric carbohydrate absorption rate and increase insulin sensitivity,¹⁵ while also helpful for constipation and restless bowel syndrome.¹⁶⁻¹⁸ The importance of almond fruit with rich oleic, linoleic, and linolenic acid content has increased due to the positive effects on cholesterol and cardiovascular disease in human,¹² while the observed blood cholesterol-lowering effects of nuts were far better than what was predicted according to their dietary fatty acid profiles.^{19,20}

The almond oil market was valued at \$1118 million in 2016 and is expected to reach up to \$2680 million by 2023, while the growth of almond oil market is driven by the rise in production of aromatherapy products, increase in preference of customers toward cosmetic products containing natural ingredients, rapid urbanization, and growth in applications of almond oil in pharmaceutical industry.²¹

Differences in the major and minor components of the medicinal oils significantly affect their nutritional, health-promoting activities, and their organoleptic properties. Therefore, in this study, 17 almond oil samples sold in pharmacies, herbal shops, online, and cosmetics stores were analyzed in terms of the criteria specified in the "almond oil" monograph in European Pharmacopoeia (EP) 7.0.²²

MATERIALS AND METHODS

Materials

Seventeen different brands of almond oils were purchased from pharmacies, herbal, online, and cosmetics stores in Ankara, Türkiye between 2017 and 2018. All chemicals used were of analytical reagent grade. Oksan Co., Ltd. (Ankara, Türkiye) provided helium, hydrogen, and dried air gases for gas chromatography (GC) with 99.99% purity. Deionized water was obtained from a Milli-Q water purification system (Millipore, Bedford, MA, USA).

GC (7890A GC System, Agilent Technologies Inc, US), a capillary column Rt-2560 (100 m, 0.25 mm ID, 0.2 μ m) (Restek Corporation Bellefonte, US), vial insert, 250 μ L, glass with polymer feet, vial, screw top, 2 mL, amber and cap, screw, blue, PTFE/red silicone septa (Agilent Technologies Inc., US) were used.

Methods

Seventeen brands of almond oils were analyzed according to the criteria of EP 7.0²² (appearance, identification, acid value, peroxide value, and composition of fatty acids) and the results were compared with the pharmacopeia quality of reference standard almond oil from *P. dulcis* (63445-250 mL, Sigma Aldrich, Lot BCBV9057).

Statistical analysis

All the results are given for at least in triplicate and the values are given as mean \pm standard deviation. Calculations made in the statistical analysis were made using Microsoft Excel software.

RESULTS

Appearance

All oil samples were placed into glass droppers and their colors were compared with the pharmacopeia quality of reference almond oil (Table 1).²²

Identification

Thin layer chromatography (TLC) was used to identify the almond oils.²³ C-18 silica TLC plate (Supelco 10 x 10 cm, 0.2 mm) was used as the stationary phase. An approximately 20 mg of oil sample, and a reference solution of almond oil complying with "Almond Oil European Pharmacopoeia" standards were dissolved in 3 mL of CH_2Cl_2 (for GC MSSupraSolv[®]). TLC plate was first eluted with ether (Sigma Aldrich, Germany) mobile phase up to 0.5 cm. Then, the plate was then removed from the tank and immersed in the other tank containing CH_2Cl_2 : glacial acetic acid (Sigma Aldrich, Germany): Acetone (Sigma Aldrich, Germany) (2:4:5, by volume) mobile phase, and eluted 8 cm. The plate removed from the tank and dried in air, and then 100 g L^{-1} solution of phosphomolybdic acid (Sigma Aldrich, Germany) in alcohol as revelator was sprayed. TLC plate was heated at 120°C for 3 min. Then, retention times and stain of the commercial almond oils were compared with the retention times and stains of reference almond oil from *P. dulcis* (Figure 1). A thin layer chromatogram of the samples and reference almond oil (Figure 1) was also compared with the reference chromatogram in EP 8.0 (Figure 2).²³

Acid (I_A) and peroxide (I_P) values

Acid value is expressed as milligrams of KOH (Sigma Aldrich, Germany) required to neutralize the free acids in 1 g of the oil. Therefore, about 10 g of each oil sample was dissolved in 50 mL of 96% methanol (Merck, Germany) and peroxide-free ether (Merck, Germany, Germany) mixture (1:1, by volume), then titrated with 0.1 M KOH in the presence of phenolphthalein (Sigma Aldrich, Germany) indicator until the pink remained stable for at least 15 s acid values of samples were compared with the value of maximum of 2.0 in a 5.0 g oil sample (Table 1).^{22,24}

The peroxide value is expressed as milliequivalent of active oxygen, the quantity of peroxide contained in 1000 g of the substance. So, about 5 grams of oil was placed in a 250 mL conical flask fitted with a ground-glass stopper. 30 mL of a mixture of chloroform (Merck, Germany) and glacial acetic acid (2:3, by volume) was added. After the oil dissolved, 0.5 mL of saturated potassium iodide (Merck, Germany) solution was added and shaken for exactly 1 min, then 30 mL water was added. It was titrated with 0.01 M sodium thiosulfate (Sigma Aldrich, Germany) until the yellow was almost discharged. 5 mL of starch solution was added and continued the titration, until the color was discharged. It was carried out with a blank test under the same conditions. Peroxide values were compared the value of maximum 15.0 in 5 g oil (Table 1).^{22,24}

Preparation of fatty acid methyl esters (FAMES) standard

All standard solutions were prepared in an ice bath and stored at -20°C. 0.1 g of FAME37, C4-24 (Sigma Aldrich, Germany)

reference standard was dissolved in 250 μL of CH_2Cl_2 (400 mg mL^{-1} FAME37, C4-24), and then, 75 μL of 400 mg mL^{-1} FAME37, C4-24 was diluted to 1.0 mL with CH_2Cl_2 (30 mg mL^{-1} FAME37).

Preparation of FAMES in almond oils

Each oil was dried at 100-105°C. The oils (1.0 g for each) were weighed into a 25 mL round-bottomed flask with a ground-glass neck fitted with a reflux condenser and a gas port into the flask. Anhydrous methanol (10 mL) and 60 g L^{-1} KOH (0.2 mL) in methanol were added. The reflux condenser was attached, passed nitrogen through the mixture at a rate of about 50 mL

min^{-1} shacked, and heated to boiling. When the solution was clear, it was continued heating for a further 5 min and cooled the flask and transferred the contents to a separating funnel. The flask was rinsed with 5 mL of anhydrous chromatographic quality of heptane (99%, Sigma Aldrich, Germany), then, the rinsing was transferred to a separating funnel and stacked. 10 mL of a 200 g L^{-1} NaCl (Sigma Aldrich, Germany) solution was added and stacked vigorously. It was allowed to form two separate phases and transferred the upper organic layer to a vial containing anhydrous Na_2SO_4 (Sigma Aldrich, Germany), allowed to stand, then filtered.

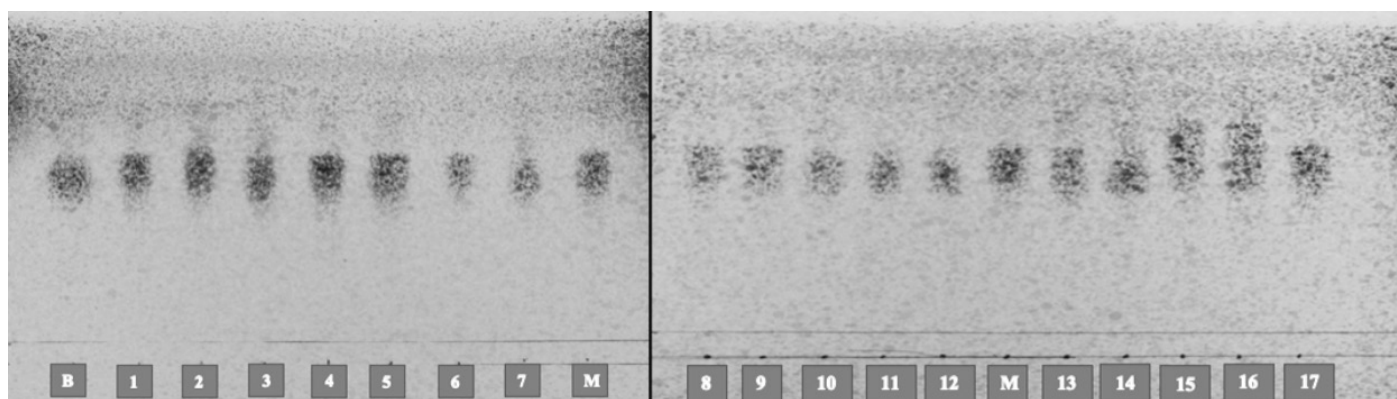


Figure 1. Thin layer chromatography of almond oil samples (s1 to s17), corn oil (M), and reference almond oil obtained from *Prunus dulcis* (B)

Table 1. Appearance, acid value and, peroxide value of almond oils

Sample no	Appearance	$I_A^a \pm \text{SD}^b$ mg KOH g^{-1} oil	$I_P^c \pm \text{SD}^b$ mL g^{-1}
s1	Clear liquid	0.34 ± 0.04	10.61 ± 0.86
s2	Pale yellow	0.48 ± 0.01	17.30 ± 0.87
s3	Pale yellow	3.30 ± 0.01	4.85 ± 0.99
s4	Clear liquid	0.36 ± 0.04	10.94 ± 1.60
s5	Clear liquid	0.80 ± 0.03	9.40 ± 0.82
s6	Clear liquid	0.32 ± 0.001	8.16 ± 0.77
s7	Dark yellow	9.18 ± 0.09	3.06 ± 0.77
s8	Clear liquid	0.36 ± 0.01	3.76 ± 0.14
s9	Pale green	1.22 ± 0.02	5.40 ± 0.18
s10	Yellowish green	8.65 ± 0.32	4.46 ± 0.32
s11	Clear liquid	0.18 ± 0.01	7.14 ± 0.07
s12	Pale yellow	1.24 ± 0.06	7.68 ± 0.28
s13	Clear liquid	0.35 ± 0.04	9.52 ± 3.30
s14	Dark yellow	6.04 ± 0.16	6.10 ± 0.28
s15	Pale yellow	0.32 ± 0.02	2.43 ± 0.10
s16	Clear liquid	0.35 ± 0.01	19.59 ± 0.41
s17	Dark yellow	0.38 ± 0.03	8.98 ± 0.09
Almond oil (pharmacopoeia quality)	Pale yellow	0.44 ± 0.02	3.46 ± 0.29

^aReference acid value is maximum 2.0, determined on 5.0 g, ^bSD: Standard deviation, n: 3, ^cReference peroxide value is maximum 15.0

Analysis of FAMES with GC- flame ionization detector (FID)

FAME37, C4-24 standard, and fatty acids in almond oil were analyzed by GC equipped with an auto sampler model Agilent 7693, and FID.²⁵ A capillary column Rt-2560 (100 m, 0.25 mm ID, 0.2 μ m), vial insert, 250 μ L, glass with polymer feet, vial, screw top, 2 mL, amber and cap, screw, blue, and PTFE/red silicone septa were used as the column and sample vial, respectively. GC oven was programmed to 100°C, held for 4 min, and then increased by 3°C min ramp to 240°C, held for 10 min. The injector and FID detector temperatures were 225°C and 250°C, respectively. Injection volume was 2 μ L with a split ratio 200:1. Helium was used as the carrier gas at 1.2 mL min⁻¹, 20 cm s⁻¹ at 175°C. FAMES in the oil samples were identified from the chromatogram by comparing their retention times with standard FAME37, C4-24, and the number of FAMES in the oil samples was expressed as a percentage by weight of all FAMES

from the total detected fatty acids (Figures 3, 4). Peak area was used for quantitative analysis of FAMES, where their content in almond oil samples were listed in Tables 2a and b.

DISCUSSION

EP 7.0 states that the appearance of almond oil should be clear or pale yellow. The appearance, acid values, and peroxide values of almond oil samples are given in Table 1. In the evaluations, it was observed that the samples coded s7, s9, s10, s14, and s17 do not have this appearance and their colors are pale green or dark yellow (Table 1).

Calculation of the acid value in fixed oils is an important quality criterion. It also gives an idea of whether the oil has exceeded its shelf life. The increase in the amount of free fatty acids is an indication that there will be a decrease in the stability of the oil

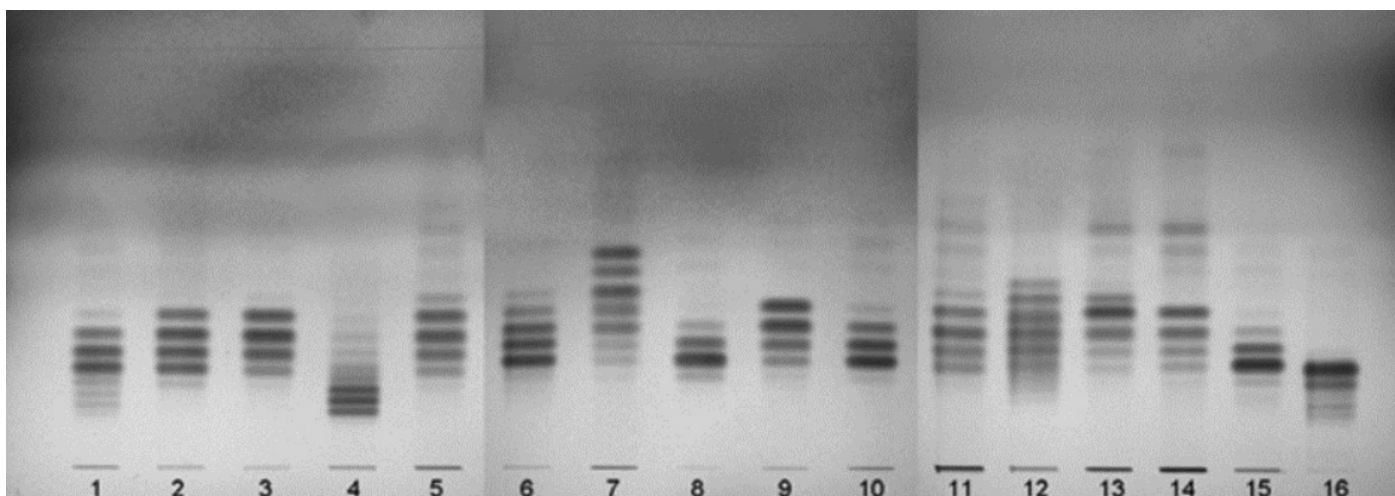


Figure 2. Thin layer chromatograms for the identification of fatty acids in European Pharmacopoeia 8.0²³ [1: Arachis oil, 2: Sesame oil, 3: Corn oil, 4: Rapeseed oil, 5: Soya-bean oil, 6: Rapeseed oil (erucic acid-free) 7: Linseed oil, 8: Olive oil, 9: Sunflower oil 10: Almond oil, 11: Wheat-germ oil, 12: Borage oil, 13: Evening primrose oil, 14: Safflower oil (type I), 15: Safflower oil (type II), 16: Hydrogenated arachis oil]

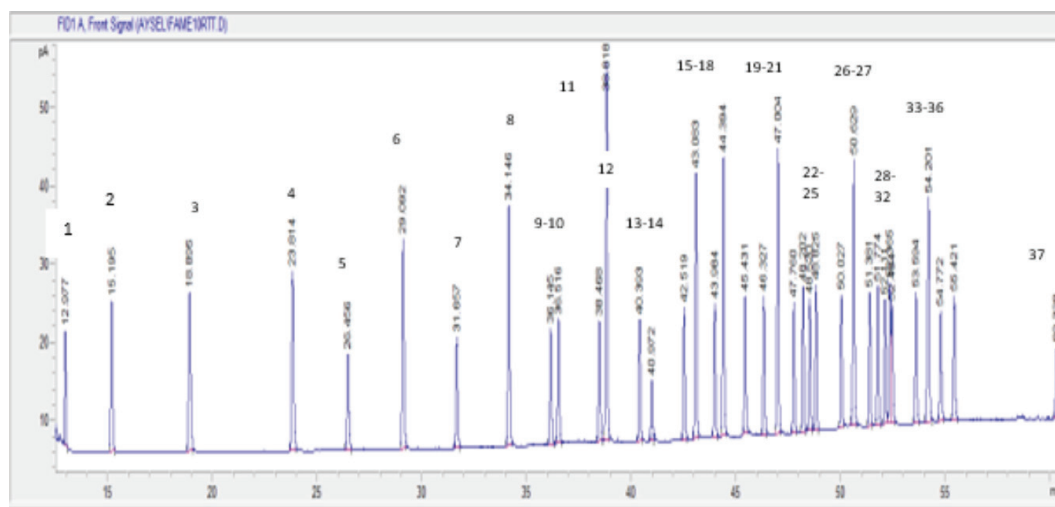


Figure 3. GC-FID chromatogram of FAME37 mix, C4-C24 reference standard material (1- C4:0, 2- C6:0, 3- C8:0, 4- C10:0, 5- C11:0, 6- C12:0, 7- C13:0, 8- C14:0, 9- C14:1 (*cis*-9), 10- C15:0, 11- C15:1 (*cis*-10), 12- C16:0, 13- C16:1 (*cis*-9), 14- C17:0, 15- C17:1 (*cis*-10), 16- C18:0, 17- C18:1 (*trans*-9), 18- C18:1 (*cis*-9), 19- C18:2 (*trans*-9,12), 20- C18:2 (*cis*-9,12), 21- C20:0, 22- C18:3 (*cis*-6,9,12), 23- C20:1 (*cis*-11), 24- C18:3 (*cis*-9,12,15), 25- C21:0, 26- C20:2 (*cis*-11,14), 27- C22:0, 28- C20:3 (*cis*-8,11,14), 29- C22:1 (*cis*-13), 30- C20:3 (*cis*-11,14,17), 31- C20:4 (*cis*-5,8,11,14), 32- C23:0, 33- C22:2 (*cis*-13,16), 34- C24:0, 35- C20:5 (*cis*-5,8,11,14,17), 36- C24:1 (*cis*-15), 37- C22:6 (*cis*-4,7,10,13,16,19))

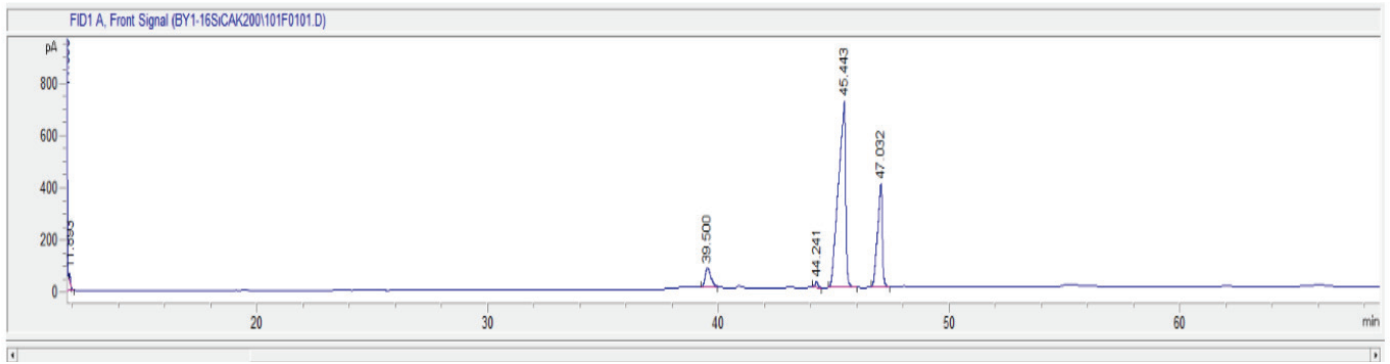


Figure 4. GC-FID chromatogram of reference almond oil (from *Prunus dulcis*)

Table 2a. Content of FAMES in almond oil samples 1-9

FAME	Content, percent in almond oil \pm SD ^a								
	s1	s2	s3	s4	s5	s6	s7	s8	s9
C14:0	0.1 \pm 0.0	0.1 \pm 0.0	-	-	-	-	0.1 \pm 0.0	-	-
C16:0	6.2 \pm 0.5	10.2 \pm 0.3 ^b	4.9 \pm 0.3	5.7 \pm 0.3	5.5 \pm 0.3	5.7 \pm 0.3	6.5 \pm 0.3	5.5 \pm 0.3	5.7 \pm 0.3
C16:1	-	-	0.3 \pm 0.0	-	-	-	0.4 \pm 0.1	0.2 \pm 0.0	-
C18:0	3.4 \pm 0.1 ^b	4.6 \pm 0.1 ^b	1.2 \pm 0.2	3.8 \pm 0.1 ^b	3.9 \pm 0.1 ^b	3.6 \pm 0.1 ^b	1.2 \pm 0.2	3.5 \pm 0.2 ^b	2.8 \pm 0.2
C18:1	29.3 \pm 0.4 ^b	25.9 \pm 0.5 ^b	66.1 \pm 0.6	30.5 \pm 0.6 ^b	29.9 \pm 0.5 ^b	30.4 \pm 0.5 ^b	64.2 \pm 1.0	33.8 \pm 0.3 ^b	36.4 \pm 0.5 ^b
C18:2	57.8 \pm 1.3 ^b	51.4 \pm 0.3 ^b	20.6 \pm 0.1	57.6 \pm 0.6 ^b	58.3 \pm 0.6 ^b	57.9 \pm 0.6	24.9 \pm 1.5	54.3 \pm 1.2 ^b	52.9 \pm 1.2 ^b
C20:0	0.3 \pm 0.0 ^b	0.6 \pm 0.1 ^b	0.4 \pm 0.0 ^b	0.3 \pm 0.0 ^b	0.3 \pm 0.0 ^b	0.3 \pm 0.0 ^b	-	0.3 \pm 0.0 ^b	-
C18:3	0.3 \pm 0.0	0.6 \pm 0.1	0.3 \pm 0.0	0.2 \pm 0.0	0.3 \pm 0.0	0.2 \pm 0.0	0.3 \pm 0.0	0.2 \pm 0.0	0.3 \pm 0.0
C20:1	-	5.2 \pm 0.3 ^b	5.5 \pm 0.3 ^b	0.1 \pm 0.0	-	-	-	-	-
C18:3	0.3 \pm 0.0	0.1 \pm 0.0	-	-	0.1 \pm 0.0	0.1 \pm 0.0	-	-	-
C20:2	0.9 \pm 0.2	0.6 \pm 0.1	0.3 \pm 0.0	0.9 \pm 0.1	0.9 \pm 0.1	0.9 \pm 0.1	-	0.9 \pm 0.1	0.8 \pm 0.1
C24:0	-	-	-	0.4 \pm 0.0	-	-	-	-	-
C20:5	0.4 \pm 0.2	-	-	-	0.4 \pm 0.0	0.4 \pm 0.0	-	0.5 \pm 0.0	-

^aSD: Standard deviation, n= 3, ^bOut of limit, FAME: Fatty acid methyl ester

Table 2b. The content of FAMES in almond oils samples 10-17 and reference almond oil

FAME	Content, % in almond oil \pm SD ^a								
	s10	s11	s12	s13	s14	s15	s16	s17	Reference almond oil
C16:0	5.8 \pm 0.0	5.7 \pm 0.2	5.5 \pm 0.2	3.9 \pm 0.1 ^b	6.3 \pm 0.3	6.9 \pm 0.3	5.7 \pm 0.2	5.7 \pm 0.1	5.5 \pm 0.2
C16:1	-	0.4 \pm 0.1	0.4 \pm 0.1	-	0.4 \pm 0.1	-	0.4 \pm 0.1	0.2 \pm 0.0	-
C18:0	1.2 \pm 0.0	0.8 \pm 0.2	0.5 \pm 0.2	2.5 \pm 0.5	1.0 \pm 0.2	4.3 \pm 0.4 ^b	3.9 \pm 0.2 ^b	1.9 \pm 0.3	0.9 \pm 0.1
C18:1	69.8 \pm 1.5	67.6 \pm 0.5	68.8 \pm 0.9	67.8 \pm 1.3	65.6 \pm 0.6	23.2 \pm 0.6 ^b	31.3 \pm 0.5 ^b	16.7 \pm 0.7 ^b	68.3 \pm 0.1
C18:2	23.2 \pm 0.3	24.5 \pm 0.7	23.3 \pm 0.6	22.7 \pm 0.9	25.3 \pm 0.6	28.5 \pm 0.2	20.2 \pm 0.2	74.0 \pm 1.6 ^b	24.7 \pm 0.4
C20:0	-	-	-	0.3 \pm 0.0 ^b	-	3.7 \pm 0.9 ^b	4.2 \pm 0.3 ^b	0.5 \pm 0.0 ^b	-
C18:3	-	-	-	0.3 \pm 0.0	0.3 \pm 0.0	2.7 \pm 0.0	2.8 \pm 0.0	0.3 \pm 0.1	-
C20:1	-	-	-	-	-	31.3 \pm 0.1 ^b	32.9 \pm 0.4 ^b	-	-
C20:2	-	-	-	1.6 \pm 0.1	-	-	-	0.6 \pm 0.0	-
C22:0	-	-	-	-	-	0.2 \pm 0.1	0.2 \pm 0.1	-	-
C24:0	-	-	-	0.9 \pm 0.0	-	-	-	-	-

^aSD: Standard deviation, n= 3, ^bOut of limit, FAME: Fatty acid methyl ester

against oxidation. This situation is defined as the oil becoming bitter. According to EP 7.0, it has been reported that acid value in the almond oil should be “maximum 2.0 in 5 g oil”. When evaluated in terms of this criterion; acid values of the samples coded s3, s7, s10, and s14 ($I_A= 3.30$ to 9.18) were found to be well above the criteria reported (Table 1).

Peroxide content of the oil samples indicates that the oil has started to oxidize. Peroxidation process occurs due to high temperature and light exposure. Contact with metal surfaces can also cause the oil to oxidize faster. Additionally, oxygen breaks down unsaturated fatty acids, resulting in smaller aldehyde molecules such as malondialdehyde. The lower peroxide values indicate the longer shelf life of the oil. A high peroxide value usually indicates poor processing and poor oil quality. According to EP 7.0, it has been reported that the peroxide values in almond oil should be “maximum 15.0 in 5 g oil”. When the results were investigated, the samples, s2 ($I_p= 17.30$) and s17 ($I_p= 19.59$) were found to have high peroxide values compared to the maximum values (Table 1).

The fatty acids in commercial almond oils were identified by comparing their TLC profiles with that of the reference almond oil from *P. dulcis* under the same conditions (Figure 1). Results of the fatty oils reported in the EP 8.0 (Figure 2)²³ were also used for identification of fatty acids of the samples. TLCs obtained from samples s5, s6, s8, s9, s11, s12, and s13 were similar to the reference almond oil chromatogram and the corresponding pharmacopeia chromatogram as shown in Figure 2. Nevertheless, the general profile of samples s1, s2, s3, s4, s7, s10, s14, s15, and s16 coded expressions are not suitable. Quality of the oil is related to its contents, types, and number of fatty acids. In the almond oil monograph in EP 7.0, defined as “*Amygdalae oleum raffinatum*” and its fatty oil was obtained from the ripe seeds of *P. dulcis* var. *dulcis* or *P. dulcis* var. *amara* or a mixture of both varieties by cold expression. Then, it is refined. A suitable antioxidants may be added. Another almond oil registered in the EP 7.0 is “almond oil, virgin, “*Amygdalae oleum virginale*,” and its fatty oils were obtained from cold

expression from the ripe seeds of *P. dulcis* var. *dulcis* or *P. dulcis* var. *amara*, or a mixture of both varieties.

According to EP 7.0, fatty acid compositions of refined almond oil and virgin almond oil are given as; palmitic acid (4.0-9.0%), palmitoleic acid (0.8% max), margaric acid (not more than 0.2%), stearic acid (3.0% max), oleic acid (62-86%), linoleic acid (20-30%), linolenic acid (0.4% max), arachidic acid (not more than 0.2%), eicosenoic acid (0.3% max), behenic acid (not more than 0.2%), and erucic acid (0.1% max).²² Supelco FAME37 mix, C4-24 was used as a standard reference material to detect the fatty acids in almond oil with GC-FID.

Fatty acids are defined as the organic compounds formed by a hydrocarbonated chain and a carboxylic acid group, which are normally bound with glycerol-forming acylglycerides (mono-, di- or triglycerides).²⁶ While α -linolenic acid and linoleic acids are the essential fatty acids, which the human body cannot produce, the unsaturated fatty acids in almond oil (50-81% oleic and 6-37% linoleic acid) become more desirable for the physicochemical and nutritional properties and its significant role in human diet.²⁷⁻³¹ Their fatty acid profiles depend on the oil's variety and origin, which affects their stability against rancidity during transport, storage, and directly affecting their products and influencing their price.³² The oil content and composition of almond seed and the fatty acids are usually referred to as the quality characteristic of almond conditions,³² while it depends on the genotype, climatic conditions, agriculture, and harvest. The amounts of oleic acid, oleic/linoleic acid ratio and tocopherol concentration are used as quality indicators, while the oleic/linoleic acid ratio is significant in determining the quality of the kernel due to its preventive effect on lipid oxidation.³² When fatty acid contents of the almond oil samples analyzed are evaluated according to the EP 8.0,²⁵ it is seen that oleic acid (C18:1) amounts of samples with s1, s2, s4, s5, s6, s8, and s9 codes are lower than the reference value and linoleic acid (C18:2) amounts are higher than the reference value (Table 3). This difference may also be due to production of gums, as well as possibility of oxidation of oleic acid ($C_{18}H_{34}O_2$) to linoleic

Table 3. Comparison of FAMES in almond oils according to the EP 7.0

FAMES	s1	s2	s3	s4	s5	s6	s7	s8	s9	s10	s11	s12	s13	s14	s15	s16	s17
C16:0	R	↑	R	R	R	R	R	R	R	R	R	R	↓	R	R	R	R
C16:1	-	-	R	-	-	-	R	R	-	-	R	R	-	R	-	R	R
C17:0	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
C18:0	↑	↑	R	↑	↑	↑	R	↑	R	R	R	R	R	R	↑	↑	R
C18:1	↓	↓	R	↓	↓	↓	R	↓	↓	R	R	R	R	↓	↓	↓	R
C18:2	↑	↑	R	↑	↑	↑	R	↑	↑	R	R	R	R	R	R	R	↑
C18:3	R	R	-	-	R	R	-	-	-	-	-	-	-	-	-	-	-
C20:0	↑	↑	↑	↑	↑	↑	-	↑	-	-	-	-	↑	-	↑	↑	↑
C20:1	-	↑	↑	R	-	-	-	-	-	-	-	-	-	-	↑	↑	-
C22:0	-	-	-	-	-	-	-	-	-	-	-	-	-	-	↑	↑	-
C22:1	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-

R: Between reference range, ↑: More than reference value, ↓: Less than reference value, -: Not found, FAME: Fatty acid methyl ester, EP: European Pharmacopoeia

acid ($C_{18}H_{32}O_2$) depending on the production methods (refined, infiltration, cold press *etc.*), storage conditions (heat, light, *etc.*), antioxidant addition and amount. However, the samples with s7, s10, s11, and s12 codes were found to meet the criteria required by the EP 7.0.²² When all the results are evaluated; among the 17 almond oil samples sold in the pharmacy, only 2 (s11 and s12) of them were found to meet the EP 7.0²² criteria for quality (Table 4).

CONCLUSION

Many vegetable oils are sold with health-promoting claims or statements that they are beneficial against diseases, while almond oils are one of them, as they are sold in “natural”, “organic products”, “local products” shops, cosmetics store chains, and pharmacies. Almond oil, which is marketed for health benefits and cosmetics, must meet the EP criteria. Currently, these almond oils are marketed as fixed oils with the producer’s own marketing and quality criteria, while our study shows that most of the products are off-limits of pharmacopeia. A pharmacopeia’s core mission is to protect public health by creating and making available public standards to help ensure the quality of products, while the user or procurer can make an independent judgment regarding quality, thus safeguarding the health of the public. To establish the necessary quality criteria and show no harm to the user, the almond oil (if for human use) needs to be encouraged to be pharmacopeia compliance. Additionally, currently, almond oil is excluded from the Turkish Food Codex Standards. For this reason, it should be ensured

Table 4. Comparison with criteria according to EP 7.0

Sample no	Appearance	TLC	FAMES	I_A	I_P
s1	+	-	-	+	+
s2	+	-	-	+	-
s3	+	-	-	-	+
s4	+	-	-	+	+
s5	+	+	-	+	+
s6	+	+	-	+	+
s7	-	-	+	-	+
s8	+	+	-	+	+
s9	-	+	-	+	+
s10	-	-	+	-	+
s11	+	+	+	+	+
s12	+	+	+	+	+
s13	+	+	-	+	+
s14	-	-	-	-	+
s15	+	-	-	+	+
s16	+	-	-	+	-
s17	-	+	-	+	+

+: Appropriate -: Not appropriate

that almond oil is listed in this codex and urgent arrangements should be made for quality control analysis.

Ethics

Ethics Committee Approval: There is no requirement for ethical approval.

Informed Consent: Not applicable.

Peer-review: Externally peer-reviewed.

Authorship Contributions

Concept: D.D.O., Design: D.D.O., Data Collection or Processing: D.D.O., O.K.U., A.B., S.P., B.N.D.T., Analysis or Interpretation: D.D.O., O.K.U., A.B., S.P., B.N.D.T., Literature Search: D.D.O., O.K.U., B.N.D.T., Writing: D.D.O., O.K.U.

Conflict of Interest: No conflict of interest was declared by the authors.

Financial Disclosure: Gazi University Scientific Research Projects Unit for supporting the economic support necessary for the realization of this study with the project code 02/2018-01 within the scope of a graduate scientific research project.

REFERENCES

- Syngé PMM, Phychorrhaphis J. Tropical crops, fruits and nuts. The Royal Horticultural Society Dictionary of Gardening. London, United Kingdom: Oxford At The Clarendon Press; 1977. p. 217.
- Kermanshah A, Ziarati P, Asgarpanah J, Qomi M. Food values of two endemic wild almond species from Iran. IJPAES. 2014;4:380-388.
- Sathe SK, Wolf WJ, Roux KH, Teuber SS, Venkatachalam M, Sze-Tao KW. Biochemical characterization of amandin, the major storage protein in almond (*Prunus dulcis* L.). J Agric Food Chem. 2002;50:4333-4341.
- Safarian S, Azarmi Y, Jahanban-Esfahlan A, Jahanban-Esfahlan H. The beneficial effects of almond (*Prunus amygdalus* Batsch) hull on serum lipid profile and antioxidant capacity in male rats. Turk J Med Sci. 2016;46:1223-1232.
- Sathe SK, Sze KWC. Thermal aggregation of almond protein isolate. Food Chem. 1997;59:95-99.
- Mexis SF, Riganakos KA, Kontominas MG. Effect of irradiation, active and modified atmosphere packaging, container oxygen barrier and storage conditions on the physicochemical and sensory properties of raw unpeeled almond kernels (*Prunus dulcis*). J Sci Food Agric. 2011;91:634-649.
- Tiwari RS, Venkatachalam M, Sharma GM, Su M, Roux KH, Sathe SK. Effect of food matrix on amandin, almond (*Prunus dulcis* L.) major protein, immunorecognition and recovery. LWT-Food Sci Technol. 2010;43:675-683.
- Raisi M, Ghorbani M, Mahoonak AS, Kashaninejad M, Hosseini H. Effect of storage atmosphere and temperature on the oxidative stability of almond kernels during long term storage. J Stored Prod Res. 2015;62:16-21.
- Sweazea KL, Johnston CS, Ricklefs KD, Petersen KN. Almond supplementation in the absence of dietary advice significantly reduces C-reactive protein in subjects with type 2 diabetes. J Funct Foods. 2014;10:252-259.
- Karatay H, Sahin A, Yilmaz O, Aslan A. Major fatty acids composition of 32 almond (*Prunus dulcis* [Mill.] D.A. Webb) genotypes distributed in East and Southeast of Anatolia. Turk J Biochem. 2014;39:307-316.

11. Sfahlan AJ, Mahmoodzadeh A, Hasanzadeh A, Heidari R, Jamei R. Antioxidants and antiradicals in almond hull and shell (*Amygdalus communis* L.) as a function of genotype. *Food Chem.* 2009;115:529-533.
12. Ahrens S, Venkatachalam M, Mistry AM, Lapsley K, Sathe SK. Almond (*Prunus dulcis* L.) protein quality. *Plant Foods Hum Nutr.* 2005;60:123-128.
13. Morgan AF, Strauch CM, Blume F. The nature and biological availability of almond carbohydrates. *JBC.* 1930;85:385-404.
14. Yada S, Lapsley K, Huang GW. A review of composition studies of cultivated almonds: macronutrients and micronutrients. *J Food Comp Anal.* 2011;24:469-480.
15. Hou YY, Ojo O, Wang LL, Wang Q, Jiang Q, Shao XY, Wang XH. A randomized controlled trial to compare the effect of peanuts and almonds on the cardio-metabolic and inflammatory parameters in patients with type 2 diabetes mellitus. *Nutrients.* 2018;10:1565.
16. Jenkins DJ, Kendall CW, Josse AR, Salvatore S, Brighenti F, Augustin LS, Ellis PR, Vidgen E, Rao AV. Almonds decrease postprandial glycemia, insulinemia, and oxidative damage in healthy individuals. *J Nutr.* 2006;136:2987-2992.
17. Chen CY, Lapsley K, Blumberg J. A nutrition and health perspective on almonds. *J Sci Food Agr.* 2006;86:2245-2250.
18. Mericli F, Becer E, Kabadayi H, Hanoglu A, Yigit Hanoglu D, Ozkum Yavuz D, Ozek T, Vatanserver S. Fatty acid composition and anticancer activity in colon carcinoma cell lines of *Prunus dulcis* seed oil. *Pharm Biol.* 2017;55:1239-1248.
19. Jenkins DJ, Kendall CW, Marchie A, Parker TL, Connelly PW, Qian W, Haight JS, Faulkner D, Vidgen E, Lapsley KG, Spiller GA. Dose response of almonds on coronary heart disease risk factors: blood lipids, oxidized low-density lipoproteins, lipoprotein(a), homocysteine, and pulmonary nitric oxide: a randomized, controlled, crossover trial. *Circulation.* 2002;106:1327-1332.
20. Kris-Etherton PM, Pearson TA, Wan Y, Hargrove RL, Moriarty K, Fishell V, Etherton TD. High-monounsaturated fatty acid diets lower both plasma cholesterol and triacylglycerol concentrations. *Am J Clin Nutr.* 1999;70:1009-1015.
21. Prasannan A. Almond oil market by type (sweet almond oil and bitter almond oil), application (food preparation, cosmetic, pharmaceutical, and others), and distribution channel (hypermarkets & supermarkets, food specialty stores, pharmacy, cosmetic discounters, and others) - Global Opportunity Analysis and Industry Forecast, 2017-2023. 2020.
22. EU. European Pharmacopeia 7th Edition. In: Healthcare EDftQoMa (ed). Vol 1. Germany, Druckerei C.H. Beck Press, 2011;1346.
23. EU. European Pharmacopeia 8th Edition. In: Healthcare EDftQoMa (ed). Germany, Druckerei C.H. Beck Press, 2014;122.
24. EU. European Pharmacopeia 8th Edition. In: Healthcare EDftQoMa (ed). Vol a. Germany, Druckerei C.H. Beck Press, 2014;155.
25. EU. European Pharmacopeia 8th Edition. In: Healthcare EDftQoMa (ed). Vol 1. Germany, Druckerei C.H. Beck Press, 2014;136.
26. Khodadoust S, Mohammadzadeh A, Mohammadi J, Irajie C, Ramezani M. Identification and determination of the fatty acid composition of *Quercus brantii* growing in southwestern Iran by GC-MS. *Nat Prod Res.* 2014;28:573-576.
27. Farhoosh R, Tavakoli J. Physicochemical properties of kernel oil from *Amygdalus scoparia* growing wild in Iran. *J Food Lipids.* 2008;15:433-443.
28. Moayedi A, Rezaei K, Moini S, Keshavarz B. Chemical compositions of oils from several wild almond species. *J Am Oil Chem Soc.* 2010;88:503-508.
29. Moayedi H, Huat BBK, Ali TAM, Asadi A, Moayedi F, Mokhberi M. Preventing landslides in times of rainfall: case study and FEM analyses. *Disaster Prev Manag.* 2011;20:115-124.
30. Sorkheh K, Kiani S, Sofo A. Wild almond (*Prunus scoparia* L.) as potential oilseed resource for the future: studies on the variability of its oil content and composition. *Food Chem.* 2016;212:58-64.
31. Mirzaei H, Rezaei K. Amygdalin contents of oil and meal from wild almond: effect of different heat pretreatment and extraction methods. *J Am Oil Chem Soc.* 2019;96:1163-1171.
32. Kodad O, Socias I Company R. Variability of oil content and of major fatty acid composition in almond (*Prunus amygdalus* Batsch) and its relationship with kernel quality. *J Agric Food Chem.* 2008;56:4096-4101.