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Variation in Fatty Acid Composition of Four Turkish Registered Poppy (*Papaver somniferum* L.) Seeds in two Locations (Ankara and Boldavin) of Turkey

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Abstract

Opium poppy (*Papaver somniferum* L.) has two major products: alkaloids in the capsules and the seeds. Poppy seed oil is a rich source of polyunsaturated fatty acids. It is known that polyunsaturated fatty acids present not only basic nutriments for human body, but protects against cardiovascular diseases, heart attacks and many inflammatory diseases. The aim of this study was to determine oil content and fatty acids composition in four Turkish registered poppy cultivars grown in experimental fields of the Agronomy Department, of Ankara University and Bolvadin Factory of Alcaloids at the Afyon province, Turkey. All seeds were sown in 15 October 2009 and harvested in 20 July 2010. The oil was extracted and analyzed with hexane by foss soxtec 2055 apparatus and fatty acids were analyzed by gas chromatography. Seed oil and fatty acids percentage of four different cultivars in two locations were determined. Oil contents of seed varieties ranged 40.20% - 47.95%. The major fatty acid in seed oils was linoleic acid (68.16 - 74.15%) whereas oleic and palmitic acid contents of seed oils ranged 14.22 - 16.47% and 7.96 - 12.87% respectively. In terms of oil content and unsaturated fatty acids concentration Bolvadin location is better compared to Ankara location.

Keywords: Poppy, Unsaturated fatty acid, oil content, Alkaloid, Capsule

Introduction

The opium poppy (*Papaver somniferum* L.) a member of *Papaveracae* family is used by humans since thousands of years. It is a multipurpose medicinal or ornamental plant, and a source for seed oil (Dewick, 2002). The experiments about medicinal use of opium poppy as a sedative, anesthetic and anti-diarrheal matter can be seen in the ancient documents. It is known that opium poppy today contains alkaloid (morphine, codein, tebaine, noscapine, papaverine) in great quantities. Opiate alkaloids and their synthetic derivatives (oxycodone, hydrocodone, pholcodine) are widely used in medicine and these compounds are produced in hundreds of tons for the medicine industry (Holzer, 2009; Francisa and *et al*., 2008). When the plant reache complete maturity, the leaves are dry and the seeds contain a maximum of poppy seed oil (an unsaturated fixed oil). Poppy seed oil is used as a culinary salad oil**,** cooking oil, drying oil for use in art, and as a vehicle for various parenteral formulations (Bruneton, 1995; Trease and Evans, 1983; Morton, 1977). Plants are important renewable sources of fatty acids because many species accumulate the fatty acids in the form of triglycerides as the main substances in seeds. Therefore it is also very important to understand which factors limit the accumulation of fatty acid structures in seeds (Thelen and

Ohlrogge, 2002). Quality and nutritive value of poppy seeds is based on oil content and mainly polyunsaturated fatty acids. Generally, poppy seeds contain 50% of edible oil (Singh *et al*., 1990). Polyunsaturated fatty acids (PUFA) are considered to be necessary components of cell membranes and play a key role in many cell processes (Abbate *et al*., 1996). Poppy oil is also suitable alternative for sunflower oil in dietary products (Kusmenoglu *et al*., 2002).

A need arise for new and cheaper proteins as protein sources for animal diets are very expensive worldwide. Poppy seed meal is a high-level protein source produced in many region of the world. Poppy seed meal containing about 30-36% crude protein and 1-15% crude oil is a good and cheap alternative for common protein sources for farm animals (Kutlu, 2002). There are some minerals in poppy seed oil; important mineral matters in the opium poppy oil are K: 5906 (mg/kg), P: 5795 (mg/kg), Mg: 4256(mg/kg), S: 2113(mg/kg), Ba: 118(mg/kg), Fe: 91(mg/kg), Sr: 71(mg/kg), Mn: 56.1(mg/kg), Zn: 43(mg/kg), B: 30.3(mg/kg), Al: 19.6(mg/kg) and V: 13.9(mg/kg) (Ozcan, 2004). The production of poppy either is done for opium, for dry capsule or nutritional purposes, which needs some kind of narcotic control measures. As a consequence, all countries interested in the production of poppy have to take into consideration the UN Convention on Narcotics, signed in 1988. This Convention forced the countries not only for controlling their cultivation methods and patrolling cultivation areas, but to build up a new strategy for developing special varieties (Bernáth and Németh, 1999; Németh *et al*., 2002). Turkey such as some countries is a legal opium poppy producer and UN recognized Turkey and India as traditional poppy producing countries. The plant production is performed in fifteen provinces of Turkey. (Kutlu, 2002; Rahimi *et al*., 2011). The increasing demand for poppy alkaloids is the consequence of the widening of the medicinal applications of morphine and its related compounds (Anonyms, 1998, 1999). According to FAO STAT (2009) database, areas of poppy seeds cultivated in the world achieved in 2009 year 140 534 ha, with total production quantity of 98 835 tons. poppy seeds with no narcotic effect are highly valuable due to high nutritive value having protein up to 24% and high amounts of linoleic acid (up to 68%) in seed oil, which helps in lowering the blood cholesterol in the human system (Singh *et al*., 1990; Singh *et al*.,1995). A few samples were chosen and analyzed to determine oil content and fatty acid composition of poppy seeds. The objective of our study was to determine oil content and fatty acids composition in poppy seed samples grown in Turkey and to evaluate the effect of cultivar and location on their amounts.

Materials and Methods

Materials

Four registered Turkish poppy (*Papaver somniferum* L.) cultivars were used in this study. Some characteristics of cultivars are summarized in Table-1 .The seeds of cultivars were obtained from Turkish Grain Board (TMO). These cultivars were sown in the experimental fields of the Agronomy Department, Faculty of Agriculture of Ankara University (32:52 E; 39:56 N) and Bolvadin Alkaloid Factory (30:33 E; 38:45 N) in 15 October 2009 and harvested in 20 July 2010.

Table-1: Some identification charachterisitics of poppy seeds

Cultivar	Seed Color	Flower Color	Association
Ofis 96	Yellow	White	TMO*
TMO 1	Yellow	White	TMO
Afyon	Yellow	White	TMO
Ofis 95	Yellow	White	TMO

*Turkish Grain Board (TMO)

Table- 2: Oil content (%) of poppy seeds in different locations and cultivars

Location	Cultivar	Oil (%)
Ankara	Ofis96	40,77
Ankara	TMO1	40,20
Ankara	Afyon95	40,59
Ankara	Ofis95	40,92
Average of Ankara location		**40,62**
Bolvadin	Ofis96	43,73
Bolvadin	TMO1	46,14
Bolvadin	Afyon95	47,95
Bolvadin	Ofis95	45,43
Average of Bolvadin location		**45,81**

The climatic data (Table-4) and soil analysis (Table-5) results of the experimental locations are shown.

Oil and Fatty Acids Analyze

The seeds (~1 g) were ground and extracted with hexane by foss soxtec 2055 apparatus. Fatty acid methyl esters were prepared according the AOAC method (AOAC, 1990) and analyzed by Shimadzu (Kyoto, Japan) gas chromatography equipped with db 23 capillary column (30mx0.25mm film thickness 0.25µm) and fid (flame ionization detector). Helium at a flow rate of 1.0ml/min was used as a carrier gas. Injector and detector temperatures were 230 and 240°c, respectively. Column temperature was kept at 190°c for 30 min. A sample of 1 µl was injected by the autosampler with a split mode (split ratio of 1:80). The fatty acid identification was based on the comparison of their relative retention times with the corresponding fatty acid methyl ester standards. Individual reference methyl ester standards (myristic acid (C14:0), palmitic acid (C16:0), stearic acid (C18:0), oleic acid (C18:1), linoleic acid (C18:2), arachidic acid (C20:0), gadoleic acid (C20:1), behenic acid (C22:0) and lignoceric acid (C24:0) and as well as fatty acid methyl ester mix (37 components fame mix) were purchased from sigma chemical co. (Sigma-Aldrıch Gmbh, Sternheım, Germany).

Results

The oil contents in evaluated poppy cultivars, grown in two locations are shown in Table-3. In given location of cultivation, any of cultivars in Bolvadin contained higher oil content compared to oil content of any cultivar at Ankara location. Total oil content ranged 40.20% (cv. TMO1 in Ankara location) to 47.95% (cv. Afyon 95 in Bolvadin location). The average of oil content at Bolvain and Ankara was 45.81% and 40.62% respectively. In Bolvadin the oil concentration was the highest (47.95%) in cv. Afyon 95 whereas cv. Ofis 96 had the lowest oil yield (43.73%). At Ankara the difference among cultivars was not very much thus the oil concentration was the highest (40.92%) in cv. Ofis 95 whereas TMO1 sample had the lowest oil yield (40.20%). Hlinková and *et al.* (2012) reported that oil content during 2007 ranged 43.3 – 49.9% and in 2009 ranged 40.8 - 50.1%. Ozcan and Atalay (2006) in a study under 6 Turkish cultivars and 1 line, showed that the oil content varied between 32.4% (cv. Ankara 94) and 45.5% (cv. Şuhut 94). Sener *et al*. (1999) found that Turkish poopy seeds contained oil in range of 45-50%; whereas Arsalan *et al.* 2000 found that it ranged 38.86 - 53.39%.

Table-3: Fatty acid composition (%) of poppy seeds in different locations and cultivars

Location	Cultivar	C14:0	C16:0	C17:0	C18:0	C20:0	S.*	C16:1	C17:1	C18:1	C18:2	C18 :3	C20: 1	U.S.*
Ankara	Ofis96	0,05	9,06	0,04	2,29	0,08	11,53	0,16	0,02	15,19	72,46	0,59	0,06	88,47
Ankara	TMO1	0,05	8,96	0,05	2,20	0,08	11,33	0,15	0,03	15,42	72,37	0,65	0,05	88,67
Ankara	Afyon95	0,05	8,42	0,05	2,24	0,08	10,84	0,12	0,02	15,80	72,63	0,52	0,06	89,16
Ankara	Ofis95	0,04	12,87	0,05	3,86	0,16	16,98	0,11	0,02	14,22	68,16	0,46	0,06	83,02
Average of Ankara		**0,05**	**9,83**	**0,05**	**2,65**	**0,10**	**12,67**	**0,14**	**0,02**	**15,16**	**71,40**	**0,55**	**0,06**	**87,33**
Bolvadin	Ofis96	0,05	8,39	0,06	2,27	0,09	10,85	0,13	0,02	15,52	72,73	0,68	0,06	89,15
Bolvadin	TMO1	0,04	8,44	0,05	2,19	0,09	10,81	0,17	0,03	14,26	74,05	0,62	0,06	89,19
Bolvadin	Afyon95	0,05	8,29	0,04	2,31	0,10	10,79	0,13	0,03	16,47	71,98	0,55	0,06	89,21
Bolvadin	Ofis95	0,04	7,96	0,04	2,09	0,09	10,22	0,13	0,02	14,84	74,15	0,58	0,06	89,78
Average of Bolvadin		**0,05**	**8,27**	**0,05**	**2,21**	**0,09**	**10,67**	**0,14**	**0,02**	**15,27**	**73,23**	**0,61**	**0,06**	**89,33**

*S.: saturated fatty acids and *U.S.: unsaturated fatty acid

Fatty acid composition of poppy seed oils is presented in Table-4. Major fatty acids in all analyzed oil samples were linoleic (C 18:2), oleic (C 18:1) and palmitic acids (C 16:0). As minority fatty acids were characterized as stearic (C 18:0) and linolenic acids (C 18:3). Minor fatty acids in all oil samples were palmitoleic (C 16:1), arachidic (C 20:0), myristic (C 14:0), gadoleic (C 20:1), heptadecanoic (17:0), heptadecenoic acids (17:1). Similarly Some researchers have reported that the linoleic (C 18:2), oleic (C18:1) and palmitic acids (C16:0) are major fatty acids in the poppy seed oil (Erinc *et al*., 2009; Singh and *et al*., 1990; Sener and *et al*., 1999; Bezáková *et al*., 1994; Bajpai, 1999; Bozan and Temellı, 2008 and Luthra and Singh, 1989). Poppy seed oils contained 83.02% (Ofis 95 in Ankara location) to 89.78% (Ofis 95 in Bolvadin location) unsaturated fatty acids made up mainly linoleic acid (C18:2). The difference of Ofis 95 unsaturated fatty acids percentage between two locations is strong. Probably this characteristic of the cultivar is affected by the condition of locations. The average of unsaturated fatty acids in four cultivars related to Ankara and Bolvadin locations were 87.33% and 89.33% respectively. Major fatty acid linoleic acid (C18:2) the first most abundant unsaturated fatty acid and also major PUFA in poppy seed oils ranged 68.16% (cv. Ofis 95 in Ankara location) - 74.15% (cv. Ofis 95 in Bolvadin location). The average of linoleic acid (C18:2) in four cultivars related to Ankara and Bolvadin locations was 71.40% and 73.23% respectively. The levels of oleic acid (18:1) ranged 14.22% (Ofis 95 in Ankara location) - 16.47% (Afyon 95 in Bolvadin location) is the second most abundant unsaturated fatty acid and also major MUFA in poppy seed oils. The average of oleic acid (C18:1) in four cultivars related to Ankara and Bolvadin locations were 15.16% and 15.27% respectively. Linolenic acid (C18:3) is the third fatty acid and the second PUFA in poppy seed oils and its content ranged 0.46% (Ofis 95 in Ankara location) - 0.68% (Ofis 96 in Bolvadin location). The average of linolenic acid (C18:3) in four cultivars related to Ankara and Bolvadin locations were 0.55% and 0.61% respectively. Poppy seed is very suitable crop for food industry due to high level of linoleic (C18:2) and low level of linolenic acid (C18:3) (Singh *et al*., 1998). High level of linolenic acid (C18:3) is unsuitable for food industry because of its instability and modifications associated with autooxidation (Green, 1986). The

amounts of the other unsaturated fatty acids were below 0.18%. Palmitoleic (C16:1, 0.11-0.17%), heptadecenoic (C17:1, 0.02-0.03%) and gadoleic acid (C20:1, 0.05-0.06%) were MUFA found in trace amounts in all oil samples. Palmitic (C16:0) and stearic acid (C18:0) were found as major saturated fatty acids in all seed oils. The percentage of palmitic acid (C16:0) in seed oils ranged 7.96 (Ofis 95 in Bolvadin location) - 12.87% (Ofis 95 in Ankara location). The average of palmitic acid (C16:0) in four cultivars related to Ankara and Bolvadin locations were 9.83% and 8.27% respectively. Stearic acid (C18:0) varied between 2.09 (Ofis 95 in Bolvadin location) and 3.86% (Ofis 95 in Ankara location). The average of stearic acid (C18:0) in four cultivars related to Ankara and Bolvadin locations were 2.65% and 2.21% respectively. The other three saturated fatty acid in poppy seed oils, myristic (C14:0), heptadecanoic (C17:0) and arachidic acid (C20:0) were also present in small concentrations usually accounting for less than 0.17% of the oil composition.

Table-4. The long term and (2009-2010) outdoors climatic data of the experimental cities*

Months	**Rainfal (mm)**		**Temperature (C°)**		**Humidity (%)**	
	(2009-2010)		**(2009-2010)**		**(2009-2010)**	
	Ankara	**Bolvadin**	**Ankara**	**Bolvadin**	**Ankara**	**Bolvadin**
September	10.3	21.6	18.3	17.1	49.5	55.3
October	13.7	13.1	16.7	15.3	49.8	55.9
November	43.1	28.9	7.3	7.1	75.0	73.0
December	68.0	71.6	5.4	5.6	79.6	80.6
January	63.0	49.1	3.1	3.6	78.3	78.4
Fabruary	65.1	73.4	6.5	6.1	70.8	71.5
March	44.6	38.2	8.3	8.1	60.1	62.2
April	37.5	72.9	12.0	10.7	55.8	66.5
May	31.0	15.2	17.8	16.3	47.1	53.2
June	57.8	51.1	21.3	19.3	56.2	61.6
July	25.7	4.9	25.7	24.2	46.6	48.0
August	0.4	1.3	28.1	26.1	32.8	36.6

* The government meteorological association of Turkey

Table-5. Soil analyses results of the experimental soil samples in two locations*

Soil characteristics	**Ankara**	**Bolvadin**
pH	7.57	8.04
CaCo3 (%)	11.07	23.53
Organic Material (%)	1.10	2.47
N (%)	0.13	0.12
P_2O_5 (ppm)	8.20	10.58
K_2o (ppm)	612.5	920.3
Salt (%)	0.062	0.094
Texture	Clay-Loam	Clay-Loam

*Ercies university, soil department laboratory, Turkey

Discussion

Modification in fatty acids composition is probably caused by environmental effects. Many researchers confirmed that plants produce polyunsaturated fatty acids during cold stress. To other

factors, which influence the fatty acids composition, belonging to salt and drought (Hlinková and *et al*., 2012). Mikami and Murata (2003) demonstrated that tolerance of plants to salt and drought is strongly dependent on the inheritance of fatty acids levels. Matos and *et al*. (2001) showed that water stress causes the inhibition of oil biosynthesis. Jemal *et al*. (2000) confirmed the decrease of unsaturation in leaves with plants exposure by heavy metals. According to Singh *et al*. (1998), significant negative correlation was found between oleic (C18:1) and linoleic (C18:2) acids. Luthra and Singh (1989) showed that palmitic acid (C16:0) positively correlated with stearic (C18:0) and oleic (C18:1) acids. Correlations between fatty acids are interpreted on the basis of the biosynthetic pathway. Sener and *et al*. (1999) reported that linoleic (C18:2) and oleic acid (C18:1) content of poppy seeds from different locations changed between 32.63 - 74.31% and 10.38 - 27.04%, respectively. Most of the results with respect to linoleic (C18:2) and oleic acid (C18:1) content are consistent with (Sener *et al*., 1999). These samples were obtained from different locations in Turkey so these differences may be due to samples obtained from different locations. Azcan *et al*. (2004) found that depending on the color of the seeds the amount of linoleic acid (C18:2) was at maximum level in range of 56.4-69.2%. Other quantitatively dominant fatty acids were oleic acid (C18:1) (16.1-24.7%) and palmitic acid (C16:0) (10-13%). According to Nergiz and Otles (1994), poppy oil contained 50-60% of linoleic acid (C18:2) , 30% of oleic acid (C18:1) and 6-9% of palmitic acid (C16:0). Marin and *et al*. (1989) reported that linoleic (C18:2) and oleic acid (C18:1) contents were about 79.3% and 9.0% in *Papaver somniferum* L. oil, respectively. Linoleic (C18:2) and oleic acid (C18:1) contents of Indian poppy seed oils were reported as 41.0-68.0 and 13.22-36.79% respectively (Singh and *et al*., 1990). Linoleic acid (C18:2) contents in most of samples was higher compared to data of (Azcan *et al*., 2004; Sener *et al*., 1999) and lower than (Marin *et al*., 1989). Oleic acid (C18:1) contents were similar to the data of Azcan *et al*., (2004) and Singh and *et al*., (1990) and lower than results of Marin *et al*., (1989). Palmitoleic (C16:1), heptadecenoic (C17:1) and gadoleic acids (C20:1) were not identified by Azcan *et al*. (2004), Singh and *et al*. (1990), Sener *et al*. (1999) and Marin *et al*. (1989). Palmitic acid (C16:0) and stearic acids (C18:0) were found in Indian poppy seed oils as 8.90- 21.48% and 1.40-10.80%, respectively (Singh and *et al*., 1990). Azcan et al. (2004) reported that palmitic (C16:0) and stearic acids (C18:0) changed depending on the color of seeds and their percentage ranged 10.0-13.0% and 2.5-3.2%, respectively. Sener *et al*. (1999) determined palmitic (C16:0) and stearic acid (C18:0) contents as 8.33-23.00% and 0-4.30%, respectively. Palmitic (C16:0) and stearic acids (C18:0) were found as 9.6 and 1.9% by Marin *et al*. (1989).

In this study, four selected poppy cultivars were analyzed for oil content and fatty acids composition and evaluated for the effect of location. In terms of oil content and unsaturated fatty acids consenteration Bolvadin is better compared to Ankara. In Ofis 95 cultivar in term of unsaturated fatty acids there is strong difference (6.76%) between Bolvadin and Ankara locations; in this affair probably the cultivar is effected by location conditions. As results the poppy seed oils had a balanced fatty acid distribution having a high content of unsaturated fatty acids. The major fatty acids in the seed oil were linoleic (C18:2), oleic (C18:1) and palmitic acids (C16:0). Linoleic (C18:2) and oleic acids (C18:1) are two important unsaturated fatty acids in human diet. Increase in their concentration relates to the quality of related oil. With a balanced fatty acid composition, the seeds could be used in some foods to improve their nutritional value. In conclusion, opium poppy seeds are major source of raw materials such as oil with potential application as nutraceuticals and functional foods; where as the number of researches about poppy seed oil content and its quality and the conditions that affect on them are little.

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