



Effects on Fatty Acid Compositions of Müsküle Grape Leaves (*Vitis vinifera* L.) of Different Harvest Times

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The present study, deals with the fatty acid profile in six different harvest times of Müsküle grape leaves grown in Turkey. The fatty acid profile was determined using gas chromatography. Twenty eight fatty acids were detected in grape leaf oils. Generally, C 18:1 oleic acid, C 14:0 myristic acid, C 18:0 stearic acid, C 18:2 ω6 linoleic acid, C 16:0 palmitic acid and C 18:3 ω3 linolenic acid were found to be the major fatty acids in all dates. The monounsaturated fatty acids were determined at 37.43, 41.43, 45.18, 50.05, 51.75 and 54.96 % in 16.05.2010, 31.05.2010, 16.06.2010, 03.07.2010, 20.07.2010, 07.08.2010, respectively. Monounsaturated fatty acids content was found to be more than the polyunsaturated fatty acid content in all times. Despite of the saturated fatty acid showed a decrease against time; there was an increasement with the monounsaturated fatty acids.

Key Words: Müsküle grape leaves (*Vitis vinifera* L.); Fatty acid profile; Harvest time.

INTRODUCTION

Leave of Müsküle grape variety is considered as picked in Turkey. Harvest times of leaves of Müsküle grape variety is begin in May, continues until July. Grape leaves are used in the cuisines of a number of cultures, including Turkish cuisine, Greek cuisine, Bulgarian cuisine, Arab cuisine, Romanian cuisine and Vietnamese cuisine. They are most often picked fresh from the vine and stuffed with a mixture of rice, meat and spices and then cooked by boiling or steaming.

It has been reported in the literature in 100 g of leaf nutrient components as follows. 2.12 g total fat, 1.06 g polyunsaturated fat, 0.08 g monounsaturated fat, 5.6 g protein, 17.31 g carbohydrates, 11.00 g dietary fiber, 6.3 g sugars, 9 mg sodium, 363.08 mg of calcium, 2.63 g of iron, 91.02 mg of phosphorus, As 1376 IU of vitamin A, 120 mg/100 g vitamin C¹, 2.9 kcal/100 g reported².

Fresh grape leaves are reported to be good for many diseases. They cuts diarrhea, effective in healing wounds and sore, more efficient and that helps strengthen the memory and³, the raw cellulose in plant-derived foods, hardening of the arteries, spinal diseases and reduce the formation of large intestine cancer⁴.

Fatty acids are an important factor that contribute to the development of cardiovascular, cancer and degenerative diseases⁵⁻⁹. Therefore, increased consumption of monounsaturated

fatty acids and polyunsaturated fatty acids and decreased consumption of saturated fatty acids are linked to positive health outcomes¹⁰.

However, a comparative study on fatty acid profile of harvest time of grape leaf samples in Turkey has not been reported up to now. It can be change fatty acid composition in harvest period of grape leaves. Determining the fatty acid profile of them will improve the nutritional information available to consumer. Thus, the present study was carried out to determine and compare the fatty acid profile of them.

EXPERIMENTAL

Müsküle grape leaf samples *Vitis vinifera* L. were collected from six different time in Konya in Turkey in 2010. Six samples were collected from different times for each cultivar. The leaf was excised from product and air-dried at room temperature under shaded conditions. It was stored at room temperature until analysis.

Oil extraction: The oil extraction of the dried and powdered aerial plants (10 g) of each harvest time was carried out at 80 °C for 6 h by Soxhlet extractor, using petroleum ether as a solvent. The solvent was evaporated by a rotary evaporator.

Fatty acid methyl esters preparation: The fatty acids were converted to corresponding methyl esters according to the reported method¹¹. A solution of NaOH in methanol (2 N,

4 mL) was added to 200 mg of oil in a 25 mL round bottomed flask attached to a reflux condenser. The mixture was heated to reflux for 10 min and then allowed to cool to room temperature. 5 mL of BF₃-methanol complex (14 %, w/w) was added to the mixture, it was heated to reflux for 10 min again and then allowed to cool to room temperature. 2 mL of heptane and 4 mL of NaCl solution (0.6 %, w/v) is added. Organic phase is separated with a Pasteur pipette, dried over Na₂SO₄ and filtered.

GC conditions: At the beginning of each analysis, the samples were allowed to equilibrate to room temperature and analyzed by gas liquid chromatography (GLC) (Agilent-6890N), equipped with dual flame ionization detector and a 30 m in length, 0.32 mm ID, film thickness 0.25 µm DB-23 capillary column. Column temperature was 60 °C for 2 min. and then raised progressively 5 °C/min up to 250 °C where it was maintained for 20 min at 250 °C. The carrier gas was hydrogen (2 mL/min). The injector and detector temperatures were 270 and 280 °C, respectively. Conditions to separate fatty acids of carbon chain length from 8 to 24 were determined. The fatty acids in samples were identified by comparison of retention times with known external standard mixtures (Sigma), quantified by a HP Chem Station software and the results were expressed as percentage distribution of fatty acid

methyl esters. Each of the experiments was repeated three times.

Statistical analysis: GLC analyses were repeated by three times. In fatty acid analysis, seven data (n = 7) were obtained for each variety. The results are reported as means ± SD. Statistical comparisons were made using SPSS Software (version 15.0). Analysis of variance (ANOVA) was used to compare four different samples with each other. *p*-value < 0.05 was considered to be significant. The mean values were compared by Tukey's test.

RESULTS AND DISCUSSION

The fatty acid profiles are presented in Table-1. Twenty eight fatty acids were detected for six different crop times grown in Turkey grape leaves. The major fatty acid was found to be oleic acid (18:1, ω9) for all times.

Myristic acid is the major saturated fatty acid and the amount of fatty acids in Müsküle grape leaves was found to be between 34.19 and 28.01 %. In accordance with our results, in the fatty acid composition of *Vitis vinifera* L. leaves, myristic acid was shown to have the highest proportion in saturated fatty acids¹². However, it has been determined that the myristic acid in the seeds of eight *Cephalaria* species is lower than that

TABLE-1
FATTY ACID PROFILES OF MÜŞKÜLE GRAPE LEAVES (%)

Fatty acids	16.05.2010 (n=7)	31.05.2010 (n=7)	16.06.2010 (n=7)	03.07.2010 (n=7)	20.07.2010 (n=7)	07.08.2010 (n=7)
C 8:0	0.01 ± 0.01b	0.00 ± 0.00b	2.00 ± 1.73ab	2.67 ± 2.31ab	0.00 ± 0.00b	4.00 ± 3.46a
C 10:0	0.01 ± 0.00a	0.01 ± 0.00a	0.01 ± 0.00a	0.00 ± 0.00b	0.00 ± 0.00b	0.00 ± 0.00b
C 12:0	0.01 ± 0.01a	0.02 ± 0.01a	0.02 ± 0.01a	0.02 ± 0.01a	0.02 ± 0.01a	0.02 ± 0.01a
C 14:0*	34.19 ± 0.08a	32.25 ± 0.09b	31.14 ± 0.10c	30.86 ± 0.22d	30.20 ± 0.08e	28.01 ± 0.09f
C 15:0	0.00 ± 0.00d	0.08 ± 0.01b	0.06 ± 0.01c	0.00 ± 0.00d	0.00 ± 0.00d	0.12 ± 0.02a
C 16:0	3.72 ± 0.06a	3.47 ± 0.08b	3.20 ± 0.01c	2.05 ± 0.04d	1.96 ± 0.05de	1.87 ± 0.04e
C 18:0*	16.45 ± 0.10a	11.97 ± 0.09b	10.72 ± 0.00c	8.42 ± 0.07d	7.65 ± 0.12e	6.40 ± 0.06f
C 20:0	0.00 ± 0.00b	0.79 ± 0.04a	0.00 ± 0.00b	0.00 ± 0.00b	0.03 ± 0.01b	0.01 ± 0.35b
C 21:0	0.12 ± 0.01b	0.15 ± 0.01a	0.12 ± 0.01b	0.01 ± 0.00d	0.06 ± 0.01c	0.01 ± 0.01d
C 22:0	0.07 ± 0.01bc	0.08 ± 0.01ab	0.06 ± 0.01c	0.09 ± 0.01a	0.09 ± 0.01a	0.02 ± 0.01d
C 24:0	0.05 ± 0.01b	0.05 ± 0.01b	0.07 ± 0.01a	0.03 ± 0.01c	0.02 ± 0.01d	0.01 ± 0.00d
ΣSFA	54.63	48.87	45.38	41.45	40.01	36.47
C 14:1 ω5	0.25 ± 0.02a	0.09 ± 0.01c	0.06 ± 0.01d	0.03 ± 0.01e	0.09 ± 0.01c	0.17 ± 0.02b
C 16:1 ω7	0.80 ± 0.03a	0.66 ± 0.04b	0.75 ± 0.10ab	0.48 ± 0.05cd	0.39 ± 0.02d	0.55 ± 0.05c
C 18:1 ω9*	36.30 ± 0.10f	40.14 ± 0.10e	43.42 ± 0.07d	49.40 ± 0.23c	51.10 ± 0.11b	54.19 ± 0.04a
C 20:1 ω9	0.03 ± 0.01d	0.47 ± 0.03b	0.79 ± 0.04a	0.02 ± 0.01d	0.10 ± 0.01c	0.04 ± 0.01d
C 22:1 ω9	0.05 ± 0.01b	0.06 ± 0.01b	0.16 ± 0.03a	0.14 ± 0.02a	0.06 ± 0.01b	0.02 ± 0.01c
ΣMUFA	37.43	41.43	45.18	50.05	51.75	54.96
C 18:2 ω6*	2.89 ± 0.06f	3.36 ± 0.06e	4.63 ± 0.01d	7.67 ± 0.16c	7.76 ± 0.03b	8.34 ± 0.04a
C 18:2-T	0.00 ± 0.00b	1.39 ± 0.07a	0.00 ± 0.00b	0.00 ± 0.00b	0.01 ± 0.01b	0.02 ± 0.01b
C 18:3 ω3	4.27 ± 0.07a	4.22 ± 0.01a	4.01 ± 0.03b	0.18 ± 0.02c	0.14 ± 0.01c	0.04 ± 0.01d
C 20:2 ω6	0.06 ± 0.00c	0.19 ± 0.01a	0.08 ± 0.00b	0.01 ± 0.00e	0.05 ± 0.01c	0.02 ± 0.01d
C 20:3 ω3	0.14 ± 0.01a	0.09 ± 0.01b	0.04 ± 0.00c	0.02 ± 0.01d	0.10 ± 0.01b	0.03 ± 0.01c
C 20:4 ω6	0.12 ± 0.01a	0.06 ± 0.02b	0.07 ± 0.01b	0.04 ± 0.01c	0.08 ± 0.01b	0.01 ± 0.00d
C 20:5 ω3	0.02 ± 0.01d	0.08 ± 0.01c	0.17 ± 0.01a	0.13 ± 0.01b	0.03 ± 0.01d	0.02 ± 0.01d
C 22:2 ω6	0.05 ± 0.01a	0.06 ± 0.01a	0.02 ± 0.01a	0.33 ± 0.41a	0.02 ± 0.01a	0.03 ± 0.01a
C 22:3 ω3	0.05 ± 0.01c	0.10 ± 0.01a	0.11 ± 0.01a	0.08 ± 0.01b	0.00 ± 0.00e	0.02 ± 0.01d
C 22:4 ω6	0.07 ± 0.01b	0.06 ± 0.01c	0.15 ± 0.01a	0.03 ± 0.01d	0.02 ± 0.01e	0.02 ± 0.01de
C 22:5 ω3	0.05 ± 0.01b	0.06 ± 0.01b	0.10 ± 0.01a	0.00 ± 0.00d	0.02 ± 0.01c	0.01 ± 0.01cd
C 22:6 ω3	0.20 ± 0.01a	0.04 ± 0.01c	0.07 ± 0.01b	0.03 ± 0.01cd	0.02 ± 0.01de	0.01 ± 0.00e
ΣPUFA	7.93	8.30	9.44	8.49	8.23	8.55

*a-f Means that are in the same row as each other and that do share a superscript letter in common are not significantly different from each other at *p* < 0.05.

found in our study, but myristic acid is the major fatty acid identified in saturated fatty acids¹³.

Linoleic acid content ranged from 2.89-8.34 % in leaves of six different crop times grown in Turkey. Also, linoleic acid was the major fatty acid in polyunsaturated fatty acids. The fatty acid composition of *Vitis vinifera* L. leaves grown in Turkey has been investigated previously and the linoleic acid content was determined amount of around in our study¹². The fatty acid composition of mazmura oils has shown that oleic acid was the major fatty acid¹⁴. Linoleic acid was identified as the major fatty acid in Razaki and Narince, which contained 66.40 and 66.11 %, respectively¹⁵. Linoleic acid, one of the essential fatty acids, is very important for the nutritional value of oils¹⁶. The high linoleic acid content makes the oil of *Vitis vinifera* L. *Centaurea* species nutritionally valuable.

An inverse correlation exists between C18:2 and C18:3 in grape leaves. The presence of linoleic acid, one of essential fatty acids, is very important factor for nutritional quality of oils. The higher linoleic acid content makes grape leaves oil nutritionally more valuable. The results of the present study are agreement with work of Demir and Namli¹⁷ and Pardo *et al.*¹⁸.

Oleic acid is the highest composition of fatty acids found in monounsaturated fatty acids, ranging from 36.30-54.19 % in the *Vitis vinifera* L. In a recent study. It has been reported that oleic acid levels varied from 20.26-32.01 % in different localities and varieties of *Amaranthus cruentus*¹⁹.

It has also been reported that monounsaturated fatty acids and saturated fatty acids were higher than polyunsaturated fatty acids, as investigated in leaves of six different crop times Müsküle grape leaves. Polyunsaturated fatty acids were determined to be between 7.93 and 9.44 % in Müsküle grape leaves. Monounsaturated fatty acids content on 07.08.2010 (54.96 %) was higher than in the other times. The level of saturated fatty acids and monounsaturated fatty acids were determined to be in the range of 36.47-54.63 and 37.43-54.96 %, respectively. Similarly, Goren *et al.*²⁰, have found that polyunsaturated fatty acids are higher than saturated fatty acids and monounsaturated fatty acids in *Salvia* species. According to these results, the oil of Müsküle grape leaf may be a good source of saturated fatty acids.

Conclusion

The fatty acid compositions of six different harvest times grown in Turkey were determined and compared. The results

clearly indicate that there are differences in fatty acid compositions between them. Müsküle grape leaves are more and more thicker after the date of 16.06.2010. That's why, the most suitable time to collect Müsküle grape leaves in terms of fatty acids can be recommended the dates between 16 May and 16 June 2010. Consequently, Müsküle grape leaves may be a good fatty acid and dietary fiber source.

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REFERENCES

1. A. Baysal, General Nourishment, Ankara Hatiboglu Publications, pp. 1-214 (1993).
2. A. Unver, M. Ozcan, D. Arslan and A. Akin, *J. Food Process. Preservation*, **31**, 73 (2007).
3. M. Akin, Asma Yapraginin Faydalari, From <http://bitkiselumit.blogcu.com/asma-yapraginin-faydalari/7796152> (2010).
4. O.L. Gurses, *Gida*, **3**, 43 (1980).
5. U.N. Das, *Prostagl. Leukot. Essent. Fatty Acids*, **63**, 351 (2000).
6. S.M. Grundy, *Am. J. Clin. Nutr.*, **66**, 988 (1997).
7. S.C. Larsson, M. Kumlin, M. Ingelman-Sundberg and A. Wolk, *Am. J. Clin. Nutri.*, **79**, 935 (2004).
8. M.L. Slattery, J.D. Potter, D.M. Duncan and T.D. Berry, *Int. J. Cancer*, **73**, 670 (1997).
9. C. Von Schacky, P. Angerer, W. Kothny, K. Theisen and H. Mudra, *Annals Internal Med.*, **130**, 554 (1999).
10. R.N. Lemaitre, I.B. King, D. Mozaffarian, N. Sotoodehnia, T.D. Rea and L.H. Kuller, *Circulation*, **114**, 209 (2006).
11. H.T. Slover and E. Lanza, *J. Am. Oil Chem. Soc.*, **56**, 933 (1979).
12. R. Demir and B. Otludil, *Biochem. Arch.*, **13**, 223 (1997).
13. S. Kirmizigül, N. Böke., H. Sümbül, R.S. Göktürk and N. Arda, *Pure Appl. Chem.*, **79**, 2297 (2007).
14. D.I. Batovsk, I. T. Todorov, V.S. Bankov, S.P. Parushev, A.I. Atanassov, T.D. Hvarleva, G.J. Djakova and S.S. Popov, *Nat. Prod. Res.*, **22**, 1231 (2008).
15. S.G. Tangolar, Y. Ozogul, S. Tangolar and A. Torun, *Int. J. Food Sci. Nutri.*, **60**, 32 (2009).
16. C.O. Eromosele and I.C. Eromosele, *Bioresour. Technol.*, **82**, 303 (2002).
17. R. Demir and S. Namli, *Int. J. Agric. Biol.*, **5**, 615 (2006).
18. J. E. Pardo, E. Fernández, M. Rubio, A. Alvarruiz and G.L. Alonso, *Eur. J. Lipid Sci. Technol.*, **111**, 188 (2009).
19. B.E. Berganza, A.W. Moran, M.G. Rodriguez, N.M. Coto, M. Santamaria and R. Bressani, *Plant Foods Human Nutri.*, **58**, 1 (2003).
20. A.C. Goren, T. Kilic, T. Dirmenci and G. Bilsel, *Biochem. System. Ecol.*, **34**, 160 (2006).