Variation in Fatty Acid Composition of 'Tulameen' Red Raspberry Seed Oil by the Application of Nitrogen Fertilizers and Organic Manure

R. GERCEKCİOGLU[1] N. YILMAZ[2] O.Faruk BAYRAK[3] Fikrettin SAHIN[3]

[1] Department of Horticulture, Faculty of Agriculture, Gaziosmanpasa University, 60240 Tokat,TURKEY

[2] Department of Biology, Faculty of Science and Art,Gaziosmanpasa University, 60240 Tokat,TURKEY

[3] Department of Genetics and Bioengineering, Faculty of Engineering and Architecture, Yeditepe University, Istanbul, TURKEY

Corresponding Author
E-mail: resulg@gop.edu.tr

Received : 15 December 2006
Accepted : 05 February 2007

Abstract

In this study, the application of nitrogen fertilizer and organic manure in various rates on the percentage and the composition of the fatty acids in'Tulameen' Red Raspberry (Rubus idaeus L.) seed oil were investigated. A randomized complete block design with there different rates 0, 56, 112; and 112 split kg N ha-1and two different rates (30 ton ha-1 and 40 ton ha-1) ofsheep manure was utulized. Linoleic acid (49.25%), alpha linolenic acid (38.33%) and palmitic acid (4.97%) were typically the most abundant fatty acids; and composition of these fatty acids in seed oil were different in all treatments. However, the changes in percentage of the fatty acids in Tulamen seeds with these treatments were not significant. The effects of either application time or rate of the fertilizers tested on fatty acid percentage in Tulameen seeds was not significant.

Key words: Red raspberry, Fatty acid, Turkey

INTRODUCTION

Recent findings indicates that the small and insignificant berry seeds may very well prove to be a treasure chests of valuable, health supporting nutrients, as well as cosmetically beneficial phytochemicals. These studies could elevate revalueate the importance of the berry seeds as a valuable nutrient sources for human [1].

Scientists now suspect that berry seeds contain high levels of life prolonging essential fatty acids (EFA), known to be an effective against heart diseases, diabetes and cancers. These essential fatty acids such as omega 3 fatty acids found in walnuts, fish, flaxseeds, borage and evening primrose are all familiar to health conscious consumers [2].

Recent studies of red raspberry seeds by Oomah et al. [3] show that the most abundant fatty acids found in raspberry oil are linoleic, alpha linolenic and oleic acids. The omega-3 and omega 6 fatty acids has been shown to reduce negative effects of oxidative stresses in the body. Thus raspberry seed oil shows favorable nutritional implications and beneficial physiological effects for the prevention of coronery heart diseases and many cancers. Johansson et al. [4] found that linoleic, alpha linoleic, oleic, and palmitic acids were typically the most abundant fatty acids in seed oil of 22 common edible wild northern berries, including raspberry. Johansson et al. [4] assuming that 10% seed in fresh berries and 23% oil content of seeds [4]. Winton & Winton [5] found that the percentage of oil extracted from raspberry seeds was ranging between 14.6-18% of the whole seed and contained 0.73-1.10% phytosterol, and had a saponification value of187-192 [5].

Nitrogen fertilizer is an important input in raspberry and blackberry production. Much research has focused on its effect on growth and yield. Although the plant yields were increased by N and K, postharvest quality response varied. Martin &Nelson's [6], reported that under the application rates of N and K of 0, 67, and 135 kg ha^{-1}, respectively, soluble solid concentration (SSC) was lowest and individual berry mass was the highest with N at 67 kg ha^{-1}. However, fruit*p*H and titratable acidity (TA) were not affected by N and K rates[6].

In general, it is assumed that only 50% of the nitrogen in most manures or composts will be available during the year of application. The availability of nitrogen in fresh poultry manure may be closer to 90% in the first year. However, the remaining nitrogen will become available in subsequent years and should be credited accordingly [7]. The need of large amounts of organic matter added to the soil for maximum bramble production, however, as pointed out under site preparation, large amounts of organic matter will require additional nitrogen added to the plant. This organic matter can be added to the crop as strawy manure. Nine to ten tons of manure can be added per acre each year. Even though the manure will provide nitrogen to the crop, 30 to 40 pounds of nitrogen should still be broadcast in late March [8].

The objective of this work was to test the effects ofapplication times and doses of Nand organic manure [sheep manure (SM)] on the percentage and composition of the fatty acids in 'Tulameen' Red Raspberry (*Rubus idaeus* L.) seed oil.

MATERIAL AND METHODS

These study was conducted at the experimental area of Horticultural Department, Gaziosmanpasa University, Faculty of Agriculture, Tokat, Turkey (40°13′- 40°22′N, 36°1′- 36°40′ E, altitude 525 m) in the 2004. The some climatic data for the research area are given Table 1.

Table 1. Climatic data for the experimental area in the year 2004* at Tokat-Turkey

	Temperature (°C)			
Month	Min.	Max.	Mean	Rainfall (mm)
April	-8.3	30.5	11.3	32.0
May	0.7	30.1	14.9	48.0
June	6.5	32.4	18.7	27.2
July	7.0	36.2	20.6	0.4
August	9.9	36.4	21.9	4.8
September	1.1	33.3	16.8	0.0
Mean Total	*16.9*	*198.9*	*104.2*	*112.4*

*:The meteorological station of general directorate of rural services (the altitute is 585 m)

Organic matter was determined by Walkley-Black method as suggested by Black[9]. The experimental soil was slightly alkaline in reaction (*p*H 7.79), medium in calcium carbonate content (11.9%), poor in P content (2.06 mg P_2O_5 100 g^{-1} soil), rich K content (28.7 mgK_2O 100 g^{-1} soil) and poor in organic matter (1.47 %). The characteristic ofexperimental SM is alkaline (8.21), rich N content (9.34 %), poor in Pcontent (%1.79), rich K content (84.47 mgK_2O 100 g^{-1} soil) and rich in organic matter (55 %).

Nitrogen fertilization and organic manure application

A randomized complete block design with there different doses of 0, 56, 112 and 112kg N $ha^{-1}$and two different doses (30 ton. ha^{-1} and 40 ton. ha^{-1}) of Sheep manure were used. Doses of the first treatment (nitrogen)were 0 kg. ha^{-1}(no nitrogen) as control group, 56 kg N ha^{-1} as a single spring application in late March and 112 kg N ha^{-1} as a single spring application in late March and112 kg N ha^{-1}applied as split with equal portions (56 kg ha^{-1} in late March and 56 kg ha^{-1}late June). Ammonium nitrat (26%) as N source was applied near the base of the canes in 2004. In the second treatment, 30 ton ha^{-1} or 40 ton ha^{-1} of dry sheep manure [before planting in late winter (every year in three consequtive years)] in single applications. The seedlings of red raspberry cv. Tulameen (a year-old) were planting on March, 2004.Each plot had six canes.

Determination of fatty acid composition

The fatty acid analysis performed according to proceduredescribed by Stead et al [10]. A fused silica capillary column SP™- 2380 (30m x 0.25 mm with a 0.25-µm film thickness) from Supelco (Bellefonte, PA, U.S.A.) was used with helium as the carrier gas at 1 mL/min. The temperature was programmed at 165 ^{0}C for 20 min followed by a 5 ^{0}C/min increase to 185 ^{0}C, which was then held for 10 min.

Statistical analysis

Data were analysed with analysis of varience (ANOVA) procedures using the Statistical Soft-ware Package [11]. The mean variation among treatment means for application times and doses of the fertilizers were obtained using the LSD test. The most abundant fatty acid percentage calculations were considered to be significant for P-values < 0.05.

RESULTS AND DISCUSSION

Relative abundance of fatty acids were was similar in all five variable fatty acids (unknown fatty acids were the exception) in Tulameen red raspberry cultivar (Table 2). There was no significant effect of application times and doses of the fertilizers tested on fatty acid percentages in Tulameen seeds. The results showed that linoleic acid (mean 49.25%), alpha linolenic acid (mean 38.33%) and palmitic acid (mean 4.97%) were typically the most abundant fatty acids from seed oil. These findings results were similar to those by Johanson et al [4].Oomah et al. [3] found 47.28% C18:2 (linoleic acid)and 14.35% C18:3 (alpha linolenic acid). Linoleic acid findings values are also show similarity, however, alpha linolenic acid findings results are greater than the values reported in literature [3].

Seeds from red and black raspberries had 1000 seed-weights of approximately 1.5 g, the other caneberries were 2.4 (Marion blackberry) and 3.6 g boysenberry and evergreen blackberry [12]. Red raspberry seed was reported to contein 12.2% protein and 11-23% oil. The composition of red raspberry seed oil was 54.5% of linoleic acid (18:2), 29.1% of alpha linolenic acid (18:3), 12% of oleic acid (18:1) and 4% of satured fatty acids [3,4].

It is common to use N rich fertilizer in raspberry growth. Organic fertilizers are is also beneficial for raspberry. Use of commercial fertilizers is being abondoned due to negative the effect on the human health and environment. On the other hand, it is undisputable the effects of raspberry seeds on the human health [7,8].It is important to know the effects of the kinds, amounts and time of the fertilizersbeing used on the fatty acid contents of seeds.There are no reports on the relationship between fertilizer and fatty acid contents of raspberry seeds. However, much works have been published on the crops such ascorn, sunflower, and linen which are processed directly for oil [13].

In Knapp's [14] progress report, fatty acids were given as follows:palmitic acid (2.43 g kg^{-1}), stearic acid (1.00 g kg^{-1}), oleic acid (11.51 g kg^{-1}), linoleic acid (53.73) g kg^{-1} and Linolenic acid (15.22 g kg^{-1}) in the seeds ofred raspberry[14]..In this study,22 fatty acids were found in all applications as shown in Table 2.Three of them were not identified. Six of them were encountered in all applications and theiramountcorresponds to an average 96.39 % of total fatty acids. These fatty acids and their contents were as follows: unknown (2.13%),21:0 ISO (0.93%), palmitic acid (4.97%),γ-Linolenic acid (0.79%),α-Linolenic acid (38.33%) ve linoleic acid (49.25%), as shown in Table 3. These results showed that oleic acid was low, palmitic acid was high and the others were similarto those reported in Knapp's [14] report [14]. The reason for differences can be attributed to differences in kind, soil and climatic factors.

Table 2. Effect of rate and timing ofN fertilizer and organic manure application on seed oil fatty acid composition of 'Tulameen' red raspberry (*Rubus idaeus* L.) cultivar

Variable	Fatty acids	%
	Unknown	0.37
	Unknown	0.07
	Unknown	**3.87**
	11:0 ante ISO	0.70
	12:0	0.36
	14:0	**0.43**
	16:0	**5.63**
	16:1 ω7 C	0.82
	18:0	1.29
	18:0 2OH	0.65
N-application[x]	18:0 3 OH ISO	0.57
None (Control)	18.1ω 7 C DMA	1.10
	18:1 ω 8 t	0.93
	18:2 ω 6 C	**50.43**
	18:3 ω 3 C	**35.88**
	18:3 ω 6 C	**0.64**
	19:1ω 6 C	0.45
	19:1ω 8 t	0.93
	20:0	0.34
	21:0ISO	**0.71**
	21:1ω 6 C	0.92
	C 22Primer alcohol	0.40
	Unknown	**2.29**
	12:0	0.24
	14:0	0.36
	16:0	4.88
	16:1 ω 7 C	**0.49**
	17:0 3 OHISO	0.49
	18:0	0.72
N-application[x]	18:1 ω 7 C DMA	0.57
Half /Spring	18:1 ω 8 t	1.31
	18:2 ω 6 C	**47.75**
	18:3 ω 3 C	**40.11**
	18:3 ω 6 C	**1.12**
	19:1ω 6 C	0.50
	21:0 ISO	1.31
	C 20 N alcohol	0.66
	C 22Primer alcohol	**0.70**
	Unknown	**1.78**
	12:0	0.59
	12:1ω 9 C	0.83
	14:0	0.41
	16:0	**5.09**
	16:1 ω 7 C	0.46
	18:0	0.72
N-application[x]	18:1 ω 7 C DMA	0.55
Full /Spring	**18:2 ω 6 C**	**48.84**
	18:3 ω 3 C	**38.61**
	18:3 ω 6 C	**0.67**
	19:1ω 6 C	1.15
	19:1 ω 8 t	0.43
	21:0 ISO	**1.06**
	C 20 N alcohol	0.66
	C 22Primer alcohol	1.01

Continuing of Table 2

N-application[x] Full /Split	Unknown	1.66
	Unknown	**0.42**
	12:0	0.22
	14:0	**0.39**
	16:0	**4.46**
	16:1ω 7 C	0.49
	18:0	**0.90**
	18:2 ω 6 C	**49.88**
	18:3 ω 3 C	**39.91**
	18:3 ω 6 C	**0.77**
	19:1ω 6 C	**0.60**
	21:0 ISO	**0.98**
	C 20 N alcohol	0.59
Organic manure[y] SM_1(Full/Winter)	**Unknown**	**2.50**
	12:0	0.21
	14:0	0.39
	16:0	**4.65**
	16:1ω 7 C	0.32
	16:1ω 7 C DMA	0.62
	17:0 3 OHISO	0.61
	18:0	0.91
	18:1 ω 8 t	1.09
	18:2 ω 6 C	**47.99**
	18:3 ω 3 C	**39.08**
	18:3 ω 6 C	**1.00**
	19:0 ω 6 C	0.61
	19:1ω 6 C	0.35
	21:0 ISO	**0.86**
	21:0ω 6 C	0.94
	C 20 N alcohol	0.42
	C 22Primer alcohol	0.96
Organic manure[y] SM_2 (Full/Winter)	Unknown	2.50
	Unknown	**0.39**
	12:0	**0.20**
	14:0	**0.34**
	16:0	**5.09**
	16:1ω 7 C	0.37
	18:0	1.04
	18:1ω 7 C DMA	0.56
	18:2 ω 6 C	**50.61**
	18:3 ω 3 C	**36.40**
	18:3 ω 6 C	**0.51**
	19:1ω 6 C	**0.56**
	20:0	**0.34**
	20:0 3 0H	0.21
	21:0 ISO	**0.66**
	22:0	0.36
	C 20 N alcohol	**0.34**
	C 22Primer alcohol	**0.49**
	C :25 N alcohol	0.15

[x] Half application: 56 $kg.ha^{-1}$, full application : 112 $kg.ha^{-1}$

[y] SM_1 : 30 $ton.ha^{-1}$, SM_2 : 40 $ton.ha^{-1}$

Table 3. Effect of rate and timing ofN fertilizer and organic manure application on seed oil fatty acid composition of 'Tulameen' red raspberry (*Rubus idaeus* L.) cultivar

	Variable+						
	N-applicationx				Organic manurey		
Fatty acids (%)	Control (none)	Half/ spring	Full/ spring	Full/ split	SM_1	SM_2	*Mean*
Unknown*	3.87 a	2.29 ab	1.78 ab	0.42 c	2.50ab	1.89bc	*2.13*
16:0 (ns)	5.63	4.88	5.09	4.46	4.65	5.09	*4.97*
18:2 ω 6 C (ns)	50.43	47.75	48.84	49.88	47.99	50.61	*49.25*
18:3 ω 3C(ns)	35.88	40.11	38.61	39.91	39.08	36.40	*38.33*
18:3 ω 6C (ns)	0.64	1.12	0.67	0.77	1.00	0.51	*0.79*
21:0ISO(ns)	0.71	1.31	1.06	0.98	0.86	0.66	*0.93*
Total (%)	**97.16**	**97.46**	**96.05**	**96.42**	**96.08**	**95.16**	*96.39*

+ Means with same letter are not different at the p=0.05(*) and p=0.01(**); ns: no significant
x Half application: 56 kg.ha^{-1}, full application : 112 kg.ha^{-1} ,y SM_1 : 30 ton.ha^{-1}, SM_2 : 40 ton.ha^{-1}

Different results were obtained depending on differences in kind and application of the N fertilizers. Jackson [13] studied the effects of N and sulphide on seed oil content in spring canola. He found that seed oil content varied from 370 to 510 g kg^{-1} and that increasing N decreased the oil content due to increasing yield and fatty contents. Optimum seed and oil yield occured at about 200 kg N ha^{-1}.About 20 kg S ha^{-1} was adequate for optimum seed and oil yields[13].

Strasil & Vorlicek [15] investigated three doses of amonium nitrate were used 0,40, and 80 kg.ha^{-1} in safflower (*C. tinctorius* L.). The average values of oil content were highest in seeds of CW-74 variety (27.2%). Gila variety had the lowest oil content (24.5%). Linoleic acid was dominant in all three safflower varities. Variation in linoleic acid content of in different years was not so high in oil content. The highest average content of linoleic acid was found in Gila variety (81.2%), the lowest in Sironaria variety (77.4%). Neither were thousand seeds weight nor number of flower heads were influenced significantly by different N rates or different in plant density[15]. The effect of N fertilization on oil content in seeds was not significant. Zaman[16] and Ekshinge *et a* . [17] obtained the highest oil content with N rate of 60 kg ha^{-1} in safflower (*C. tinctorius* L.) in conditions of West Bengal or Parbhani [16,17].

Ozguven et al. [18] investigated the effects of tobacco churn obtained from tobacco factory on rape seed and sesame, used as fertilizer. Tobacco powder (milled tobacco leaves) contains 1-3 % N. They further have used 80 kg.ha^{-1} N and 120 kg.ha^{-1} N in fertilizing programme for sesame and rape seed, respectively. In trials progressing for two years, it was found that the most fat ratio in both rape seeds and sesame seeds obtained in the lowest tobacco dose (7500 kg ha^{-1}) [18].

As seen in the investigations on the fatty acid-fertilization relationship for various plants,it was found that increased N dosage, application time, different organic fertilization and yearsdo not affect fatty acid contents significantly. The effects of variety, sort, kind and ecology on fatty acid percentage are more important than the other parameters (such as application times and dosage of the fertilizers).

As conclusion, there was no effect of application times and dosage of the fertilizers tested on fatty acid percentage in Tulameen seeds.

ACKNOWLEDGEMENT

The authors greatly appreciates the contributions of Assoc. Prof.Dr. Sabit Ersahin and Assist. Prof. Saban Tekin.

REFERENCES

[1]. Pourrat, H. & Pourrat, A. 1973. Compositions cosmétiques et pharmaceutiques. French patent 7345501.

[2]. Pourrat, M. & Carnat, A.P. 1981. Chemical composition of raspberry seed oil (Rubus idaeus L.). Rev. Fr. Corps Gras 28, pp. 447-479.

[3]. Oomah, B., Ladet, S., David, V., Godfrey, D., Liang, J., &, Girard, B. 2000. Characteristic of raspberry (Rubus idaeus L.) seed oil. Food Chem. 69, 187-193.

[4]. Johansson, A., Laakso, P. & Kallio, H. 1997. Characterization of seed oils wild, edible Finnish berries. Z. Lebensm. Unters. Forsch. A. 204, 300-307.

[5]. Winton, A.L. & Winton, K.B. 1935. The structure and composition of foods. Vol. II. Vegetables, legumes, fruits. New York, NY: John Wiley and Sons.

[6]. Martin, L.W. & Nelson, E. 1986. Effects of Between-Row Spacing and Training Method on Yield and Plant Characteristics of Mechanically Harvested 'Meeker' Red Raspberry. Fourth International Rubus and Ribes Symposium.. Number 183, pp.167-172.

[7]. Kuepper, G.L., Born, H. & Bachman, J. 2003. Organic Culture of Bramble Fruits Horticulture Production Guide. ATTRA-National Sustainable Agriculture Information. http://attra.ncat.org/attra-pub/pdf/bramble,

[8]. Anonymous. 1999. Brambles-Production Management and Marketing. Bulletin 782-99. Chapter 3 Selection and Care of Plants. http://Ohioline.osu.edu/b782/b782-9htm/.

[9]. Black, C.A. 1965. Methods of Soil Analysis, Part 2. American Society of Agronomics, Madison, WI, pp.1372-1376.

[10]. Stead, D.E., Sellwood, J.E., Wilson, J. & Viney, I. 1992. Evaluation of a commercial microbial identification system based on fatty acid profiles for rapid accurate identification of plant pathogenic bacteria. J.App. Bacterial 72,315-321.

[11]. SAS Institue. 1985. SAS User's Guide. Statistics. SAS Institute, Cary, NC.

[12]. Bushman, B.S., Philips, Bliss., Isbell, T., Ou, B., Crane, J.M. & Knapp, S.J. 2004. Chemical composition of caneberry (Rubus spp.) seeds and 0ils and their antioxidant potential. Journal of Agricultural and Food Chemistry. Oregan State University, Corvallis, Oregon 97331(in pres). E-mail. sbushman@cc.usu.edu

[13]. Jackson, G.D. 2000. Effects of Nitrogen and Sulfur on Canola Yield and Nutrient Uptake. Agronomy Journal 92:644-649.

[14]. Knapp, S.J. 2002 .Progress report on the chemical and nutritional characteristics of berry seed oils. .Department of Crop and Soil Science, Oregon State University, Corvallis, OR 97331-3002, 9 .September .

[15]. Strasil, Z. & Vorlicek, Z. 2002. The effect of nitrogen fertilization sowing rates and site on yields and yields components of selected varities varities of safflowers (C. tinctrius L.). Rostlinna Vyroba,, 48, 307-311.

[16]. Zaman, A. 1988. Effect of N,P,K fertilizers on seed yield and oil content of safflower (Carthamus tinctorius L.) in drought prone lateritic tract of West Bengal. J. Oil Seeds Res., 5: 179-182.

[17]. Ekshinge, B.S., Sondge, V.D. & Raikhellar, S.W. 1993. Effect of advance application of fertilizer on oil content and oil productivity in safflower (Carthamus tinctorius L.) at the 003 Tula irrigation district.Hidalgo.Revta-Chapingo,. 12: 60-61, 45-48.

[18]. Özgüven, M., Kaya, Z., Yılmaz, M.A., Kırıcı, S. & Tansı, S. 1999. Sigara fabrikası tütün atıklarının gübre olarak değerlendirilmesi. T.J. of Agricultural Forestry, 23, Ek Sayı 1: 43-51.