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Fatty Acid, Sugar and Vitamin Contents in Rose Hip Species

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The content of oleic and linoleic acids, glucose, fructose, sucrose, maltose, vitamin C and E (α and δ-tocopherol) was determined in seeds and fruits of *Rosa iberice, Rosa canina, Rosa villosa, Rosa dumalis* and *Rosa pisiformis*. The highest level of the compounds in fruits was determined as follows: linoleic acid in *R. dumalis* (3.150 µg/g), oleic acid in *R. canina* (0.57 µg/g), δ-tocopherol in *R. dumalis* (10.12 µg/g), α-tocopherol in *R. pisiformis* (17.60 µg/g), vitamin C in *R. canina* (2855.33 µg/g), fructose in *R. dumalis* (18.44 mg/g), glucose in *R. dumalis* (10.04 mg/g), sucrose in *R. canina* (5.61 mg/g) and maltose in *R. dumalis* (1.92 mg/g). The highest level of the compound in seeds was also determined and sequenced as follows: linoleic acid in *R. canina* (3.97 µg/g), oleic acid in *R. dumalis* (10.50 µg/g), δ-tocopherol in *R. canina* (7.15 µg/g), α-tocopherol in *R. iberice* (11.01 µg/g), vitamin C in *R. iberice* (952.10 µg/g), fructose in *R. pisiformis* (17.20 mg/g), glucose in *R. iberice* (9.83 mg/g), sucrose in *R. dumalis* (14.96 mg/g) and maltose in *R. canina* (2.46 mg/g).

Key Words: Fatty acids, Rose hip, Sugars, Vitamins.

INTRODUCTION

Roses belong to one of the most popular groups of ornamental plants and mainly have been grown for the beauty of flowers, production of rose oil and water, cosmetic and medicinal purpose[1]. The fruits of rose species are used in food products such as tea, jam, marmalade, fruit juice, food colorants and soup. In addition of high content of vitamin C (130-6694 mg/100 g)[2-4] rose hips contain high amount of carotenoids[5], phenolic compounds[6] and folates[7]. Antiinflammatory properties[8], high antioxidant capacity[9], antimutagenic activities[10] and inhibition of cancer cell proliferation[11] of rose hips have been demonstrated. The rose species contains

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considerable amount of vitamin A, E, B_1, B_2, K and carbohydrates and rich in calcium, phosphorus and potassium minerals[12,13]. In addition, rose hips contain other vitamins and minerals, tocopherol, bioflavonoids, fruit acids, tannins, pectin, amino acids and essential oils[4,14-19]. Equal amount of rose hip contains 30-40 times more vitamin C than orange[20]. Rose hips have also been reported to be a good source of easily digestible carbohydrates[18]. Seed oil extracts of roses contain unsaturated fatty acids, linoleic, archodonic and oleic acids[18,19].

Because of the nutritional, medicinal, industrial and pharmaceutical properties, rose hips have gained more interest in recent years. Health-conscious consumers are also interested in natural, tasty and healthy products. Rose hip also requires less maintenance and has an extended ecologically-tolerant range. It is generally able to be grown in suboptimal climatic and soil condition. It is therefore a potentially profitable crop for both food and nutraceutical industries. The analytical studies about rose hip have been mainly focused on vitamin C determination. Investigation on the content of the other vitamins, carbohydrates and fatty acid are limited in the literature. The present study was therefore focused on the determination of fatty acid, carbohydrate and vitamin E and C analysis in fruit and seed of five widespread species in the region and the chemical composition of the species were compared in both fruit and seed to take attention on the economical and medicinal importance of rose hips species.

EXPERIMENTAL

Fruits of *Rosa iberice, Rosa canina, Rosa villosa, Rosa dumalis* and *Rosa pisiformis* were collected from Lake Van basin, Turkey and kept in deep freeze at -80 °C. The compounds were extracted from seeds and fruits and their levels were determined as µg/g-fresh weight except for sugars (mg/g-FW).

Determination of vitamin C: The extraction method of ascorbic acid was modified from Cerhata *et al.*[21]. 2 g of samples either fruits or seeds were ground to powder in liquid nitrogen and mixed with 0.5 M of 2 mL perchloric acid. Then the sample volume was made up to 10 mL with double distilled water and shaken well for a few minutes. The sample was centrifuged for 10 min at 8000 rpm. The supernatant was transferred to clean vials and 20 µL of sample was introduced to HPLC equipped with C_{18} column (µBondapak); Waters 6000 A pump (Waters, Hicrom Ltd. Uk); Ultraviolet detector (Unicam Analitical Systems, Cambridge, UK). Mobile phase was adjusted with 0.05 M phosphate buffer (pH: 4), 1.0 mL/min flow rate. The absorption wavelength was chosen 246 nm[22]. Under this condition the retention time of vitamin C standard was determined to be 3.5 min.

Determination of vitamin E: Sample were ground in liquid nitrogen and extracted in 20 mL of 2:1 chloroform: methanol. In this procedure the tissue homogenate was washed twice with water in a separatory funnel. Combined organic layers were dried. Then it was redissolved in 1 mL methanol and 20 µL samples injected to HPLC equipped with C_{18} column (250 × 4.6 mm, ACE 5 C18, Scotland); Waters 6000 A pump (Waters, Hicrom Ltd. Uk); Ultraviolet detector (Unicam Analitical Systems, Cambridge, UK). As the mobile phase, methanol-water (98:2) was used in 1.5 mL/min flow rate. For the quantitative determination of the vitamin E, the absorption wavelength was chosen 290 nm wavelengths[23]. Under this condition the retention time of δ and α-tochopherol standard were determined to be 11.27 and 16.1 min, respectively.

Determination of fatty acids: Quantitative determination of oleic and linoleic acid was performed by methods of Selcuk[24], Christie[25] and Hamrouni *et al.*[26]. 2 g of sample were ground to powder in liquid nitrogen and homogenized in 20 mL chloroform and methanol (2:1) mixture. It was diluted 1/5 by adding 4 mL of double distilled water. The mixture was separated in two phases. The upper layer was removed and the lower layer where fatty acid remained was evaporated by vacuum. The residue was dissolved by methanol and injected into HPLC equipped with µ Bondapak C_{18} column (Waters, Hicrom Ltd. UK); Waters 6000 A pump (Waters, Hicrom Ltd. Uk); Refractive Index detector (Waters 2414). As the mobile phase, acetonitrile (78 %) was used in 1.5 mL/min flow rate. The absorption wavelength was chosen 412 nm wavelengths[23]. Under this condition the retention time of linoleic and oleic acid standard were determined to be 6.2 and 9.04 min, respectively.

Determination of free sugars: The analysis of free sugars was done by the modified methods[27,28]. 5 g of sample was ground into powder in liquid nitrogen and 40 mL of methanol was added. The mixture was incubated on a magnetic stirrer at 65 °C for 0.5 h. It was centrifuged at 4 °C, 1300 rpm for 40 min. The supernatant was transferred in clean tube and made up to 50 mL with methanol. The methanol was then removed by rotary evaporator and the residue was dissolved in 25 mL double distilled water. Extract was passed through Sep-Pak C_{18} cartridge and 2.5 mL of filtrate was mixed with 7.5 mL acetonitrile. Then it was filtrated by 0.45 µm membrane filter and injected into HPLC equipped with Spherisorb 5 µm NH_2 column (250 × 4.6 mm; waters, Ireland); waters 6000 A pump (waters, Hicrom Ltd. Uk); Refractive Index detector (waters 2414). As the mobile phase, acetonitrile (80 %) was used in 1.5 mL/min flow rate. The absorption wavelength was chosen 412 nm wavelengths. Under this condition the retention time of fructose, 5.12; glucose, 5.65; sucrose, 7.87 and maltose, 9.2 min were determined, respectively.

All analysis was carried out at least in triplicate. Results were expressed as means of different experiments ± standard deviation.

RESULTS AND DISCUSSION

The chemical composition of rose hips has been studied and has been found to differ depending on species, climate and maturation degree. In various studies, mainly the vitamin content of rose hips and partly carbohydrate and fatty acids were investigated. However, the comprehensive studies on the carbohydrate, vitamin E and C and fatty acids in fruits as well as in seeds of different rose hips species are limited. Therefore the present study was performed to determine and compare medicinally and economically important compounds in five different rose hips species in the same time.

The oleic and linoleic acid levels were determined to be different in each species and also in fruits and seeds. The highest linoleic acid level in fruits was found in *Rosa dumalis* (3,150 µg/g) while the highest oleic acid level was detected in *Rosa canina* (0.57 µg/g). In seeds, the highest linoleic and oleic acid levels were determined in *R. canina* (3.95µg/g) and *R. dumalis* (10.50 µg/g), respectively (Table-1).

TABLE-1
FATTY ACIDS LEVELS IN FRUIT AND SEED OF ROSA SPECIES
(µg g^{-1} FW and ± SD, n = 3)

Rosa species	Fruit		Seed	
	Linoleic acid	Oleic acid	Linoleic acid	Oleic acid
R. iberice	2.030 ± 0.250	0.52 ± 0.05	0.85 ± 0.07	1.03 ± 0.12
R. cannina L.	1.930 ± 0.180	0.57 ± 0.04	3.95 ± 0.40	3.70 ± 0.27
R. pisiformis	0.730 ± 0.081	0.36 ± 0.02	1.21 ± 0.10	2.45 ± 0.29
R. villosa L.	0.850 ± 0.065	0.22 ± 0.02	0.65 ± 0.05	0.87 ± 0.09
R. dumalis	3.150 ± 0.290	0.45 ± 0.03	0.73 ± 0.06	10.50 ± 1.21

Although the fatty acid levels were reported to differ depending on period of development and environmental factors[29,30]. The results indicate significant differences between species as well as organs. Seed oil extracted from roses contain unsaturated fatty acids and can be used efficiently against contact dermatitis and in food products[19,31]. Seed oils extracted from roses contain 50.08 % linoleic and 19.31% oleic acid. Thus has a higher oxidative stability than other unsaturated oils. This oil could conceivable be used as a gourmet oil, adding a special flavour to food products and providing consumer with a new health-promotional oil product[18]. The fatty acids in rose hips extracts were reported to lower the colonic pH and control proliferation of pathogens and colonic carcinogenesis[8]. The present study showed

that the fatty acid contents of *R. canina* and *R. psiformis* seeds are higher than fruits. Naturally, seeds contain more fatty acids than fruits. But the findings attract the attentions on the utilization of seeds instead of discard.

The significant changes in the vitamin E levels in seeds and fruits were also observed among Rosa species. The highest δ and α-tocopherol levels in fruits were found in *R. dumalis* (10.12 μg/g) and *R. pisiformis* (17.60 μg/g) whereas they were in highest level in seeds of *R. canina* (7.15 μg/g) and *R. iberica* (11.01 μg/g), respectively (Table-2). The δ and α-tocopherol levels were determined higher in the seeds of *R. iberica* than those of fruits. While only δ-tocopherol level was higher in seeds of *R. canina*, whereas the fruit of the remaining species was found to contain higher vitamin E than those of seeds. It can be thought that Rosa species are good source in terms of not only vitamin C but also vitamin E. The result was supported by the report of Szentmihalyi *et al.*[19]. The present results also showed that seeds are also as important as fruits for vitamin E.

TABLE-2
VITAMIN E LEVELS IN FRUITS AND SEEDS OF ROSA SPECIES
(μg g^{-1} FW and ± SD, n = 3)

Rosa species	Fruit		Seed	
	δ-tokoferol	α-tokoferol	δ-tokoferol	α-tokoferol
R. iberice	1.44 ± 0.13	5.75 ± 0.54	2.55 ± 0.21	11.01 ± 1.20
R. canina L.	2.50 ± 0.21	6.40 ± 0.59	7.15 ± 0.69	4.03 ± 0.89
R. pisiformis	9.07 ± 0.87	17.60 ± 1.50	1.92 ± 0.17	10.05 ± 1.12
R. villosa L.	3.95 ± 0.32	3.57 ± 0.31	0.74 ± 0.06	3.85 ± 0.35
R. dumalis	10.12 ± 1.10	10.25 ± 1.21	2.60 ± 0.23	7.50 ± 1.67

The highest vitamin C level in fruit and seed was determined in *R. canina* (2855.33 μg/g) and *R. iberica* (952.10 μg/g), respectively (Table-3). Among vegetable and fruits, Rosa genus is one of the highest vitamin C containing plants. The vitamin C level of rose hips was well documented in the literature. Although the results in the present work on vitamin C levels are in good agreement with the literature, the vitamin C levels were determined lower than those of previous studies. The vitamin C content in seeds was reported to be in trace level[32]. However, the results in this work demonstrated that the vitamin C levels in seeds are considerably important. The difference might attribute to harvesting time and environmental factors[33].

Vitamin C is important in collagen synthesis, boosts the immune system against common cold and flu, strengthens arteries along with polyphenols, potentially lowers the risk of cancer due to antioxidant properties, helps dietary iron to function in the body, works in adrenaline synthesis and lowers the blood cholesterol[18, 34-36].

TABLE-3
VITAMIN C LEVELS IN FRUITS AND SEEDS OF ROSA SPECIES
(μg g^{-1} FW and ± SD, n = 3)

Rosa species	Fruit	Seed
R. iberice	1747.66 ± 165.01	952.10 ± 89.13
R. cannina L.	2855.33 ± 275.11	243.50 ± 21.32
R. pisiformis	1533.10 ± 149.03	395.20 ± 35.43
R. villosa L.	870.12 ± 79.21	790.20 ± 70.01
R. dumalis	1715.20 ± 165.42	225.33 ± 19.22

The sugar contents varied from species to species in fruits and seeds. The highest fructose, glucose, sucrose and maltose levels in fruits were determined in *R. dumalis* (18.44 mg/g), *R. dumalis* (10.04 mg/g), *R. canina* (5.61 mg/g) and *R. dumalis* (1.92 mg/g) (Table-4a) while the highest levels in seeds were found in *R. pisiformis* (17. 20 mg/g), *R. iberice* (9.83 mg/g), *R. dumalis* (14.96 mg/g) and *R. canina* (2.46 mg/g) (Table-4b), respectively. Sucrose and maltose levels were determined to be higher in seeds of rose species than those of fruits. Fructose levels of *R. canina* and glucose level of *R. iberice* and *R. psiformis* were also found to be higher in seeds than those of fruits. The findings are not surprising due to the important role of carbohydrate in seed germination and also seed is an important

TABLE-4a
SUGAR LEVELS IN FRUITS OF ROSA SPECIES
(mg g^{-1} FW and ± SD, n = 3)

Rosa species	Fructose	Glucose	Sucrose	Maltose
R. iberice	15.06 ± 1.40	8.65 ± 0.75	4.11 ± 0.39	0.87 ± 0.09
R. canina L.	15.92 ± 2.35	7.45 ± 0.69	5.61 ± 0.50	1.74 ± 0.19
R. villosa L.	13.58 ± 1.87	8.33 ± 0.75	2.58 ± 0.22	0.21 ± 0.03
R. dumalis	18.44 ± 2.95	10.04 ± 1.10	0.57 ± 0.49	1.92 ± 0.21
R. pisiformis	15.24 ± 2.85	6.89 ± 0.59	3.21 ± 0.27	0.62 ± 0.07

TABLE-4b
SUGAR LEVELS IN SEEDS OF ROSA SPECIES
(mg g^{-1} FW and ± SD, n = 3)

Rosa species	Fructose	Glucose	Sucrose	Maltose
R. iberice	14.41 ± 1.53	9.83 ± 0.99	10.79 ± 1.20	1.92 ± 0.20
R. canina L.	16.66 ± 1.18	7.56 ± 0.82	9.22 ± 1.00	2.46 ± 0.30
R. villosa L.	10.58 ± 1.97	4.85 ± 0.53	10.27 ± 1.19	0.81 ± 0.09
R. dumalis	13.48 ± 1.45	6.26 ± 0.69	14.96 ± 1.61	1.98 ± 0.16
R. pisiformis	17.20 ± 1.67	9.15 ± 0.79	8.67 ± 0.93	1.47 ± 0.19

storage organ. The results indicated considerable sugar levels differing between species and organs are in a good agreement with the literature[29,37-39]. The variations between results might attribute to environmental differences and the genetics of plants.

When plants are exposed to drought stress, solid compound particularly sugar synthesis increases to regulate osmotic potential. Therefore, the sugar content of plants varies depending on climate and soil structure[40]. The high sugar levels in rose hips may provide an advantageous opportunity to employ them for industrial purpose. The rich sugar assortment and concentration give a pleasent taste to rose hips[30]. The high fructose levels determined in rose hips grown in Lake Van basin, Turkey provide them different quality. The rose hip seeds might be used as food additives in powdered form.

Kadakal *et al.*[18] reported easily digestible carbohydrates as 7.55-21.29 % reducing sugars, 1.08-2.01 % sucrose and 8.68-22.44 % total sugars in rose hips.

The results obtained from the present study are in accordance with literature.

The level of biochemical compounds in plants varies depending on species, period of development, harvesting time, altitude of region, soil structure and drying, processing, storage and maintenance methods. Moreover, considering the rich bioactive compound content of rose hips, they are important source for food and food additives, used for boosting body immune system and medicinal treatment because of vitamins which have antioxidant and antiinflammatory effects[41-43]. The seed powder of *R. canina* was reported to use against osteoarthritis[8]. Because of the rich sugars and fatty acid content of seed, it may be suggested that the seeds can be ground to make beneficial instead of wasting. A comprehensive study is needed to investigate the inhibiting and activating factors of the compounds to control the synthesis of chemicals.

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