

# CHARACTERIZATION OF BIOACTIVE COMPOUNDS IN ROSEHIP SPECIES FROM EAST ANATOLIA REGION OF TURKEY

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## ABSTRACT

The objective of this work was to determine some bioactive compounds for four different rosehip species (*Rosa* L.), growing in the East Anatolia region of Turkey. It was determined that the average fruit weights of the species varied between 9.8 g (*R. dumalis*) and 34.5 g (*R. canina*). The total soluble solids showed statistically significant variations among the rosehip species (14-22 °Brix). The acidity was inversely proportional to total soluble solids and ranged between 1.00% (*R. canina*) and 2.67% (*R. villosa*). The highest total phenolic, L-ascorbic acid contents and the highest total antioxidant capacity were found in *R. canina*. The total phenolic, total anthocyanin, total dry matter, and L-ascorbic acid contents and the total antioxidant capacity of the rosehip species ranged as follows 1081-6298 mg gallic acid equivalent/100 g, 2.43-3.72 mg/100 g, 40.1-56.7%, 24.93-754.48 mg/100 g, and 10.04-97.95 mmol trolox equivalent/g, respectively. Glucose was the most common sugar in *Rosa* species (5.99-12.48 g/100 g), the major organic acid in the rosehip species was citric acid (0.48-1.05 g/100 g). A dendrogram based on some pomological and biochemical characteristics of the rosehip species were grouped into 2 main clusters. Findings on the biochemical characteristics of the species will provide insights to plant breeders / growers and for further research.

*Keywords:* antioxidant, organic acids, phenolic, rosehip, sugars

## 1. INTRODUCTION

Rosehip plants are not selective in terms of climate and soil requirements and grow in several areas, including Europe, Africa, Middle and West Asia and Russia (NILSON, 1997; ILISULU, 1992). Rosehips grow in almost all regions of Turkey and are well-known and consumed fruits in Anatolia. They are perennial plants belonging to the genus *Rosa* in the Rosaceae family. The genus *Rosa* includes numerous species and varieties, and each country has its own endemic rosehip species. *Rosa pisiformis* and *Rosa dumalis* subsp. *antalyensis* are endemic species for Turkey (ERCISLI, 2005). Out of about 100 rosehip species occurring all around the world, 27 species grow in Turkey (TURKBEN, 2003; ERCISLI and GULERYUZ, 2005).

Red fruits are rich in phytochemicals such as phenolic substances, flavonoids, anthocyanin and carotenoids (QIAN *et al.*, 2004; TRAPPEY *et al.*, 2005; CIESLIK *et al.*, 2004). Rosehips contain more and a greater variety of phytochemicals compared to other fruit species (HALVORSEN *et al.*, 2002; OLSSON *et al.*, 2004). Also, they contain minerals, high-capacity antioxidants, carotenoids, phenolic compounds, tocopherol, bioflavonoids, tannins, pectins, organic acids, amino acids, ascorbic acid, and fatty acids (GAO *et al.*, 2000; DEMIR and OZCAN, 2001; LARSEN *et al.*, 2003; CHRUBASIK *et al.*, 2008; JABLONSKA *et al.*, 2009; BARROS *et al.*, 2010). The fact that *Rosaceae* fruits have important physiological functions may be due to abundant phenolic substances, because it is known that the spectra of biochemical activity of phenolic substances, including their antioxidant activity, antimutagenic and anti-carcinogenic effects, are wide (TAPIERO *et al.*, 2002; NAKAMURA *et al.*, 2003). These compounds also contribute to the quality and nutritional value of the plant (ERCISLI, 2007). Moreover, it has been reported that rosehip fruits are used to cure illnesses such as influenza, other infections, inflammatory diseases, chronic pain and ulcer and that they have a protective effect on health (GUIMARAES *et al.*, 2010).

Despite species variation, rosehips contain about 20- to 30-fold more vitamin C compared to oranges. Besides, rosehips, which are a valuable source of minerals, are quite rich in phosphorus and potassium (NOJAVAN *et al.*, 2008; SZENTMIHALYI *et al.*, 2002; KOVACS *et al.*, 2004). Therefore, rosehip fruits are widely used in food and pharmaceutical industries. In Turkey, numerous foods such as marmalade, jam, churchkhela, nectar and tea are made from rosehip fruits (ERCISLI and GULERYUZ, 2005; YILDIZ and ALPASLAN, 2012). Besides, rosehip fruits are added to probiotic beverages, fruit yogurts and soup (DEMIR *et al.*, 2014).

Recently, naturalness and bioavailability have been considered among the most important characteristics of food products (ERCISLI, 2007). In Turkey, rosehip fruits grow naturally, without requirement of chemical compounds and fertilizers. In this study, we aimed to determine and compare some important bioactive compounds and biochemical features of four different rosehip species growing naturally in high altitudes of Ardahan city located in Eastern Anatolia in Turkey. So far, there is little information about sugar and acidity in rosehips, and no previous scientific studies have been carried out on rosehip species in the region.

## 2. MATERIALS AND METHODS

### 2.1. Plant material

Mature fruits of *R. pimpinellifolia*, *R. villosa*, *R. canina*, and *R. dumalis* were collected at the same ripening stage in two locations in Ardahan Province in September 2014 (Table 1). The fruits were immediately transferred to the laboratory in polyethylene bags and stored

at  $-20^{\circ}\text{C}$  until analysis. Rosehip species have been identified based on fruit, flower and leaf of the collected genotypes as described by DAVIS (1972). All analyses except sugar analyses were carried out in triplicate. In total, 75 fruits were used for each species, and each replicate consisted of 25 fruits.

**Table 1:** The collection areas of species.

Species	Collection Areas
<i>R. pimpinellifolia</i> L.	Ardahan, Çıldır, Gölebakan village, Fields, 2010m, September 2014
<i>R. canina</i> L.	Ardahan, Posof Baykent village, Alaybeyi located, 1950m, September 2014
<i>R. villosa</i> L.	Ardahan, Posof Baykent village, Alaybeyi located, 1950m, September 2014
<i>R. dumalis</i> L.	Ardahan, Posof Gönülçalan village, Gönülçalan forest, 2000m, September, 2014

## 2.2. Fruit weight, total soluble solids, total dry matter, pH and titratable acidity

Ten hips of every species were weighted on a digital scale with a sensitivity of 0.01 g (TX-4202L, Shimadzu, Japan). The seeds of hips of every species were counted ( $n=10$ ). Total soluble solids in ten hips of every species were determined using a digital refractometer (Mettler Toledo 30P, USA) and expressed in °Brix at  $22^{\circ}\text{C}$ . The total dry matter in ten hips of every species was measured according to the AOAC (1984) reference method. Acidity in ten hips of every species was determined titrimetrically according to CEMEROGLU (1992) and expressed as a percentage of citric acid.

## 2.3. Total anthocyanin, total phenolic content and total antioxidant capacity

Determination of the total anthocyanin content was done according to GIUSTI and WROLSTAD (2001) with slight modifications. Fresh fruits (5 g) were homogenized in 10 mL of methanol containing 1% HCl for 2 min, then kept overnight, and filtered through Whatman No. 2 filter paper. Two extracts were prepared, one with potassium chloride buffer, pH 1.0 (1.86 g of KCl in 1 L of distilled water), and the other with sodium acetate buffer, pH 4.5 (54.43 g of  $\text{CH}_3\text{CO}_2\text{Na}\cdot 3\text{H}_2\text{O}$  in 1 L of distilled water). Absorbance of the extracts was measured at 510 and 700 nm (SQ2800, Unico UV visible Spectrophotometer, USA) after 15 min of incubation at room temperature. The content of total anthocyanin was calculated from the molar absorption of cyanide 3-glucoside.

The total phenolic content was determined by the Folin-Ciocalteu method (SPANOS and WROLSTAD, 1992). A fruit sample (5 g) was homogenized (T18, IKA Homogeniser, Germany) in 25 mL of ethanol and centrifuged (NF 400, Nüve, Turkey) at  $3.500\text{ g}$  for 3 min. The supernatant was collected, purified by filtration through filter paper, and 2 mL of 10% Folin-Ciocalteu reagent was added to 0.4 mL of the extract, followed by incubation for 2-3 min. Then, 1.6 mL of 7.5%  $\text{Na}_2\text{CO}_3$  solution was added to the mix and incubated for 1 hour in the dark. Absorbance was measured at 765 nm on a spectrophotometer (SQ2800, Unico UV visible Spectrophotometer, USA) against the blank solution (0.4 mL of water, 2 mL of Folin-Ciocalteu reagent, and 1.6 mL of  $\text{Na}_2\text{CO}_3$ ). The total amount of phenolic

compounds was calculated as a mg gallic acid equivalent (GAE)/100 g by using the gallic acid standard.

The ferric reducing antioxidant power (FRAP) assay was performed according to BENZIE and STRAIN (1996). Samples (1 g) were homogenized in 50 mL of 80% methanol solution in a flask wrapped in aluminum foil. The flasks were incubated in an incubator shaker (IKA, Germany) at 30°C and 150 g for 24 hours. The samples were centrifuged at 3.200 g for 20 min. The supernatant was collected, and 200  $\mu$ L of supernatant was mixed by vortexing (IKA, Germany) with 3 mL of the FRAP reagent (300  $\mu$ M acetate buffer, pH 3.6, 10  $\mu$ M 2,4,6-tripyridyl-s-triazine (TPTZ) in 40  $\mu$ M HCl, and 20  $\mu$ M FeCl<sub>3</sub>, 10:1:1 (v/v/v)). The samples were incubated in a water bath (ST30, Nüve, Turkey) at 37°C for 30 min, and the absorbance was determined at 593 nm. Standard curve was prepared using different concentrations of trolox and expressed in mmol trolox equivalent (TE)/g frozen sample.

## 2.4. Sugar and organic acid contents

Determination of sugar contents in rosehips was done according to MIRON and SCHAFFER (1991) by HPLC (HP Agilent 1100 series, USA) using a Shim-Pack HRC NH<sub>2</sub> column (300  $\times$  7.8 mm, 5  $\mu$ m) with a refractive index detector (RID). Frozen samples (1 g) were powdered in liquid nitrogen with a mortar and pestle, transferred to an Eppendorf tube, and 20  $\mu$ L of aqueous ethanol (80%, v/v) was added. The mixture was placed in an ultrasonic bath (Sonorex Digital 10P, Switzerland) sonicated for 15 min at 80°C, then filtered, and the procedure was repeated three times. All filtered extracts were combined and evaporated to dryness in a boiling water bath. The residue was dissolved with 2 mL of distilled water and filtered before HPLC analysis. The sugar contents in the samples were calculated using calibration curves plotted by using external standards.

Identification of organic acids and determination of their contents were done by HPLC using an HPX 87H (300  $\times$  7.8 mm, 5  $\mu$ m) column and a UV detector. For carboxylic acid and L-ascorbic acid detection, 1 g of a frozen sample was powdered in liquid nitrogen with a mortar and pestle and mixed with 20 mL of aqueous meta-phosphoric acid (3%) at room temperature for 30 min on a shaker. The acidic extract was filtered, made up to 25 mL with the same solvent, and then used for HPLC analysis. External standards were used to identify and calculate organic acid contents from the retention times and calibration curves (BOZAN *et al.*, 1997).

## 2.5. Statistical analysis

All results were analyzed using the SPSS (version 15) statistical analysis package and the mean  $\pm$  standard error values obtained from triplicate measurements. Data were subjected to analysis of variance (ANOVA) and significant differences between the groups were determined by the multiple comparison procedure according to DUNCAN (1955). Differences at  $p < 0.05$  were considered significant. The Cluster analysis applied to evaluate relationships among species was performed by Ward's method using Euclidean distances.

# 3. RESULTS AND DISCUSSIONS

## 3.1. Pomological and biochemical characterization

The fruit weights of the samples, total soluble solids, total dry matter, pH and acidity values are given in Table 2. Statistically significant differences ( $p < 0.05$ ) in these parameters between the species were determined (Table 2). It was also determined that the average

fruit weights of the species varied between 9.8 g (*R. dumalis*) and 34.5 g (*R. canina*), and that the average seed numbers of 10 hips varied between 10 (*R. villosa*) and 23 (*R. canina*). The total soluble solids showed statistically significant variations among the rosehip species (Table 2). The lowest total soluble solids were found in *R. villosa* (14 °Brix), while the highest value was found in *R. canina* (22 °Brix), followed by *R. pimpinellifolia* and *R. dumalis* (20 °Brix). The total dry matter content of the fruits was between 40.1% (*R. villosa*) and 56.7% (*R. canina*) (Table 2). Demir and Ozcan (2001) found that total dry matter amounts in *R. canina* were in the range between 20.5 and 23.5%. Ercisli (2007) reported that the total soluble solids of different rosehip species growing in the Erzurum region ranged between 29.4 and 37.3 °Brix, and that the highest total soluble solids were determined in *R. dumalis* (37.3 °Brix), while the lowest content was found in *R. villosa*. The author also found that the highest total dry matter content was shown by *R. dumalis* (40.4%) and the lowest total dry matter content was shown by *R. villosa* (29.4%). The total soluble solids of rosehip species were reported to range between 14 and 40 °Brix in several studies carried out in different regions of Turkey (Sen and GuneS, 1996; Misirli *et al.*, 1999; Demir and Ozcan, 2001). In our study, the acidity was inversely proportional to total soluble solids and ranged between 1.00% (*R. canina*) and 2.67% (*R. villosa*). The lowest pH value was observed in *R. villosa* (2.86), while the highest pH value was observed in *R. canina* (3.50). Demir and Ozcan (2001) demonstrated that the acidity of *R. canina* hips collected from two different regions was 1.17% in Hadim and 1.44% in Kastamonu, while the pH values were 5.12 in Hadim and 4.34 in Kastamonu. Different rosehip species, cultivars, climate and geographical conditions are known to affect total soluble solids, acidity and pH values (ERCISLI, 2007). Also, high altitude causes acidity levels to increase in fruits.

**Table 2:** Some pomological and biochemical properties of rosehip species.

Species	Localities	Fruit Shape	Flesh Colour	Peel Colour	Fruit weight (g)
<i>R. pimpinellifolia</i>	Çıldır	Round	Purple	Black	17.3±0.6 <sup>b</sup>
<i>R. villosa</i>	Posof	Round	Orange	Red	10±0.1 <sup>c</sup>
<i>R. canina</i>	Posof	Elliptic	Orange	Red	34.5±0.9 <sup>a</sup>
<i>R. dumalis</i>	Posof	Elliptic	Orange	Red	9.8±0.2 <sup>c</sup>
Species	Average seeds/1 hip	Total soluble solids (°Brix)	Total Dry matter (%)	Acidity (%)	pH
<i>R. pimpinellifolia</i>	11±0.9 <sup>ab</sup>	20±0.9 <sup>ab</sup>	55.3±2.5 <sup>a</sup>	1.30±0.10 <sup>b</sup>	3.00±0.05 <sup>b</sup>
<i>R. villosa</i>	10±1.1 <sup>ab</sup>	14±0.8 <sup>b</sup>	40.1±6.7 <sup>b</sup>	2.67±0.09 <sup>a</sup>	2.86±0.04 <sup>c</sup>
<i>R. canina</i>	23±0.7 <sup>a</sup>	22±1.4 <sup>a</sup>	56.7±5.5 <sup>a</sup>	1.00±0.02 <sup>b</sup>	3.50±0.05 <sup>a</sup>
<i>R. dumalis</i>	18±0.8 <sup>a</sup>	20±1.6 <sup>ab</sup>	55.9±8.2 <sup>a</sup>	1.45±0.02 <sup>ab</sup>	3.06±0.01 <sup>b</sup>

Different letters (a-d) for same line are statistically significantly differences among sampling dates by Duncan's multiple range test at  $p < 0.05$ .

### 3.2. Determination of total anthocyanin, total phenolic content and total antioxidant capacity

The total anthocyanin, total phenolic content and total antioxidant capacity of the rosehip species are given in Table 3. *R. pimpinellifolia*, known as a 'black rosehip' in the region, had

the highest anthocyanin content (3.72 mg/100 g), whereas *R. dumalis* and *R. villosa* had the lowest values (2.43 and 2.45 mg/100 g, respectively). It was previously reported that the major anthocyanin in *R. canina* fruits was cyanidin-3-O-glucoside (Guimaraes *et al.*, 2013). Guerrero *et al.* (2010) found that the total anthocyanin content in rosehip fruits was 0.38 mg/100 g, and the total phenolic content was 145.7 mg/100 g. Anthocyanins give color to fruits and they have therapeutic and antioxidant activity. Cyanidin-3-O-glucoside was reported to have the highest oxygen radical scavenging effect (WANG *et al.*, 1997).

In our study, the lowest total phenolic content was found in *R. pimpinellifolia* (1081 mg GAE/100 g), and the highest content was found in *R. canina* (6298 mg GAE/100 g). Various researchers determined that the amounts of total phenolic compounds were between 176–9600 mg GAE/100 g in ripe rosehips (ERCISLI, 2007; Su *et al.*, 2007; EGEA *et al.*, 2010; FATTAHI *et al.*, 2012; ROMAN *et al.*, 2013). Similar to our data, YOO *et al.* (2008) found the total phenolic content in rosehips to be 815.5 mg GAE/100 g, and FATTAHI *et al.* (2012) reported it to be 176.48–225.65 mg GAE/100 g. DEMIR *et al.* (2014) detected the highest total phenolic content among rosehip samples collected in Gumushane, Turkey in *R. dumalis* subsp. *boissieri* (5200 mg GAE/100 g) and the lowest total phenolic content in *R. canina* (3100 mg GAE/100 g). The total phenolic content results obtained in our study were found to be higher than those reported in the literature. The differences may be due to different extraction methods, the ripening stage of the hips, environmental conditions, the harvest season, altitude or plant genotype.

The FRAP (Ferric reducing antioxidant power) method was developed by BenziE and Strain (1996) and is based on the reduction by antioxidants of Fe<sup>3+</sup> complexed by TPTZ (tripyridyl triazine) to Fe<sup>2+</sup> in a low-pH environment. The results showed that there were statistically significant differences ( $p < 0.05$ ) in the total antioxidant capacities between the rosehip species. *R. pimpinellifolia* was found to have the lowest antioxidant capacity (10.04 mmol TE/g), and *R. canina* was found to have the highest antioxidant capacity (97.95 mmol TE/g). The values found in our study were lower than those found by DEMIR *et al.* (2014). The authors reported that the total antioxidant capacity of *R. dumalis* subsp. *boissieri* was 194.36 mmol TE/g and that of *R. canina* was 103.56 mmol TE/g. These differences may be due to factors such as the geographical area, the degree of ripening, climate conditions and experimental conditions. CUNJA *et al.* (2015) reported that the highest antioxidant capacity was observed in *R. canina* fruits harvested in September and that frost damage occurring in the following months decreased antioxidant capacity. In addition, it was shown that antioxidant capacities of *R. canina* fruits ranged from 63.35 to 127.8  $\mu$ M TE/100 g as determined by the DPPH method.

**Table 3:** Total anthocyanin, total phenolic content, total antioxidant capacity (FRAP), organic acid and sugar contents of rosehip species.

	<i>R. pimpinellifolia</i>	<i>R. villosa</i>	<i>R. canina</i>	<i>R. dumalis</i>
<i>Total anthocyanin (mg/100g)</i>	3.72±0.06 <sup>a</sup>	2.45±0.03 <sup>c</sup>	2.75±0.07 <sup>b</sup>	2.43±0.09 <sup>c</sup>
<i>Total phenolic (mg GAE/100g)</i>	1081±12.8 <sup>d</sup>	2944±70.8 <sup>c</sup>	6298±116.7 <sup>a</sup>	4411±16.9 <sup>b</sup>
<i>FRAP (mmol TE/g)</i>	10.04±0.47 <sup>d</sup>	37.84±1.55 <sup>b</sup>	97.95±2.12 <sup>a</sup>	26.45±6.98 <sup>c</sup>
<i>L ascorbic acid (mg/100g)</i>	24.93±4.0 <sup>d</sup>	119.83±3.3 <sup>c</sup>	754.48±100.2 <sup>a</sup>	254.81±12.5 <sup>b</sup>
<i>Sucrose (g/100g)</i>	0.38 <sup>c</sup>	0.42 <sup>b</sup>	0.55 <sup>a</sup>	0.41 <sup>b</sup>
<i>Glucose (g/100g)</i>	5.99 <sup>d</sup>	12.48 <sup>a</sup>	8.05 <sup>b</sup>	6.79 <sup>c</sup>
<i>Fructose (g/100g)</i>	4.38 <sup>c</sup>	4.90 <sup>b</sup>	5.03 <sup>a</sup>	4.15 <sup>d</sup>
<i>Sorbitol (g/100g)</i>	4.17 <sup>c</sup>	6.25 <sup>a</sup>	5.15 <sup>b</sup>	3.94 <sup>d</sup>
<i>Total sugar(g/100g)</i>	14.92 <sup>d</sup>	24.05 <sup>a</sup>	18.78 <sup>b</sup>	15.29 <sup>c</sup>
<i>Oxalic acid (g/100g)</i>	0.14±0.01 <sup>c</sup>	0.25±0.02 <sup>b</sup>	0.38±0.04 <sup>a</sup>	0.29±0.01 <sup>b</sup>
<i>Tartaric acid (g/100g)</i>	0.21±0.08 <sup>c</sup>	0.26±0.03 <sup>c</sup>	0.65±0.13 <sup>a</sup>	0.30±0.01 <sup>b</sup>
<i>Malic acid (g/100g)</i>	0.65±0.04 <sup>b</sup>	0.45±0 <sup>c</sup>	0.48±0.06 <sup>c</sup>	0.73±0.16 <sup>a</sup>
<i>Citric acid (g/100g)</i>	0.48±0.05 <sup>c</sup>	1.05±0.05 <sup>a</sup>	0.94±0.14 <sup>b</sup>	0.95±0.3 <sup>b</sup>
<i>Succinic acid (g/100g)</i>	0.092±0.01 <sup>a</sup>	0.007±0 <sup>c</sup>	0.010±0 <sup>b</sup>	0.006±0 <sup>c</sup>
<i>Fumaric acid (g/100g)</i>	0.015±0 <sup>b</sup>	0.011±0 <sup>c</sup>	0.033±0.01 <sup>a</sup>	0.014±0 <sup>b</sup>

Different letters (a-d) for same line are statistically significantly differences among sampling dates by Duncan's multiple range test at  $p < 0.05$ .

### 3.3. L-Ascorbic acid, sugar and organic acid contents

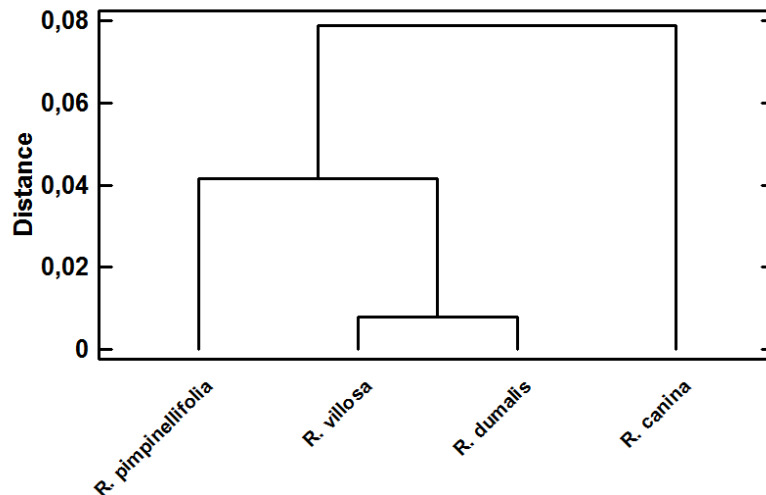
Sugar and organic acid contents are the most important factors determining fruit quality and taste. Organic acids increase bioavailability of ascorbic acid by inhibiting ascorbic acid oxidation (PADAYATTY and LEVINE, 2001; KOBUS *et al.*, 2005). In this study, there were significant differences in L-ascorbic acid, sugar and organic acid contents between the rosehip species, as presented in Table 3. The L-ascorbic acid contents of the species were found to range between 24.93 mg/100 g (*R. pimpinellifolia*) and 754.48 mg/100 g (*R. canina*). The L-ascorbic acid values obtained in our study were higher than those reported in the literature. ROMAN *et al.* (2013) revealed that the ascorbic acid contents in ripe rosehips ranged between 112.2 and 360.2 mg/100 g. Barros *et al.* (2010) found the ascorbic acid content in *R. canina* to be 68.04 mg/100 g. NOJAVAN *et al.* (2008) determined that the ascorbic acid content increased upon ripening to 417.5 mg/100 g in rosehip species and that the value was 6-fold of that found in oranges. DEMIR *et al.* (2014) determined that the ascorbic acid content was lowest in *R. dumalis* (65.75 mg/100 g) and highest in *R. gallica* (160.30 mg/100 g). In addition, CELIK *et al.* (2009) found the ascorbic acid contents in rosehip species in Van, Turkey to be 604-1.032 mg/100 g. It was also shown that there were significant differences between rosehip species in ascorbic acid content, which could be affected by ecologic factors, the degree of ripening and soil conditions (MABELLINI *et al.*, 2011; ADAMCZAK *et al.*, 2012). Rosehip species growing in high altitude regions are rich in ascorbic acid due to higher light exposure and lower oxygen amounts. Light exposure increases the amount of carotene and thus protects ascorbic acid in the fruit, while the lack of oxygen reduces oxidative stress and lessens ascorbic acid breakdown (YAMANKARADENIZ, 1983). Ascorbic acid contents of rosehips vary depending on climate conditions, fruit types and years (DEMIR and OZCAN, 2001).

Glucose was found to be the most common sugar in *Rosa* species, and the lowest glucose content was found in *R. pimpinellifolia* (5.99 g/100 g) while the highest content was found in *R. villosa* (12.48 g/100 g). The amounts of sucrose ranged in the species between 0.38–0.55 g/100 g (*R. pimpinellifolia* and *R. canina*, respectively), the fructose contents ranged between 4.15-5.03 g/100 g (*R. dumalis* and *R. canina*, respectively), and the sorbitol contents ranged between 3.94-6.25 g/100 g (*R. dumalis* and *R. villosa*, respectively). Also, the lowest total sugar amount was found in *R. pimpinellifolia* (14.92 g/100 g) while the highest amount was found in *R. villosa* (24.05 g/100 g). Other studies reported glucose contents in rosehip fruits to range between 7.45-12.94 g/100 g and fructose contents to range between 7.96-18.44 g/100 g. Similar to our results, sucrose contents were reported to range between 0.88-5.61 g/100 g and total sugar contents were reported to range between 12.05-20.46 g/100 g (YORUK *et al.*, 2008; BARROS *et al.*, 2011; ROSU *et al.*, 2011; OZRENK *et al.*, 2012). Likewise, DEMIR *et al.* (2014) revealed glucose amounts in *Rosa* species to range between 9.54 g/100g (*R. dumalis*) and 17.25 g/100g (*R. gallica*) and fructose amounts to range between 10.78 g/100g (*R. dumalis*) and 18.84 g/100g (*R. canina*). The fructose and sucrose values obtained in our study were found to be lower than those reported in the literature, whereas the total sugar amounts were found to be higher. The differences in organic acid and sugar values between our and other studies might be due to different soil and climate conditions of the region and the differences in experimental analysis. Also, differences in the harvest season are thought to affect the results.

The major organic acid in the *Rosa* species was citric acid (0.48 to 1.05 g/100 g). In this study, we found that oxalic acid was most abundant in *R. canina* (0.38 g/100 g) and least abundant in *R. pimpinellifolia* (0.14 g/100 g). Fumaric acid was also most abundant in *R. canina* (0.033 g/100 g) and least abundant in *R. villosa* (0.011 g/100g). The tartaric acid values among the species were 0.21-0.65 g/100 g, the malic acid values were between 0.45 and 0.73 g/100 g, and the succinic acid values were between 0.006-0.092 g/100 g. In a



previous study, the citric and malic acid amounts in *Rosa* species were 4.76-9.12 g/100 g and 0.45-1.10 g/100 g, respectively (DEMIR *et al.*, 2014). ADAMCZAK *et al.* (2012) found that the citric acid content in *R. tomentosa* was 4.34 g/100 g. Thus, the organic acid values obtained in our study were lower compared to those found in previous studies. A dendrogram based on the pomological and biochemical characteristics studied of the rosehip species can be seen in Fig. 1.



**Figure 1:** Cluster analyse of rosehip species according to their pomological and biochemical properties.

The species were grouped into 2 main clusters. In the first cluster, *R. villosa* and *R. dumalis* were found to be the closest species based on the characteristics analyzed. Fruit weights, total anthocyanin contents and succinic acid contents of both species were low. *R. pimpinellifolia* was found in the same cluster, while *R. canina* fell in a separate cluster, for its characteristics were different from those of the other species.

This study aimed to determine and compare some important bioactive compounds and biochemical features of 4 different rosehip species growing naturally in Ardahan (Eastern Anatolia, Turkey) and mostly consumed by the locals. The differences in acidity and sugar contents of the species, compared with previous studies, are thought to be due to different altitudes. Moreover, it was found that the L-ascorbic acid, total anthocyanin and total phenolic content values, known to increase with altitude, were high in this study. The total antioxidant capacities of these species were also high. This study is important as a foundation for further research. Besides, knowing biochemical characteristics of the species will facilitate the work of plant breeders and growers. It is known that bioactive components of fruits positively affect health. It is suggested that rosehip fruits are good sources of bioactive compounds and phytonutrients. Their consumption may prevent some illnesses and protect health.

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