



Morphological and Biochemical Diversity in Fruits of Rowanberry (*Sorbus aucuparia* L.) Genotypes

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Abstract

The world is presently over-dependent on a few fruit species. Diversification of production and consumption habits to include a broader range of fruit species, particularly those currently identified as underutilized, could significantly contribute to improve human health and nutrition, livelihoods and ecological sustainability. Rowanberry or Mountain ash (*Sorbus aucuparia*) is one of the neglected fruit species and needs to be studied on it. In present study wild grown rowanberry genotypes in Coruh valley located Northeastern part of Turkey were evaluated first time for some important human health and nutrient content. Results indicated that fruit color indices, pH, organic acids, specific sugars, total phenolic content, vitamin C and antioxidant activity of rowanberry fruits significantly differed among genotypes. *L*, chroma and hue values of genotypes were found between 30.12–41.04; 27.07–32.81 and 30.44–44.06, respectively. Glucose was the dominant sugar in rowan fruits and among organic acids, Malic acid was found to be the highest. Total phenolic content, vitamin C and antioxidant activity varied from 161–204 mg GAE per 100 g, 28.4–38.2 mg per 100 g and 2.93–5.68 mM trolox equivalent per 100 g fresh weight basis. Results indicated that developing the market potential for rowan fruits depend on better marketing and reliable supply of the end product. Because rowanberry fruits rich in nutrients and vitamins, more detailed research and scientific investigation needed to explore those properties.

Keywords *Sorbus aucuparia* L. · Rowanberry · Diversity · Biochemical content

Morphologische und biochemische Vielfalt in Früchten von Genotypen der Vogelbeere (*Sorbus aucuparia* L.)

Schlüsselwörter *Sorbus aucuparia* L. · Vogelbeere · Vielfalt · Biochemischer Gehalt

Introduction

Wild edible fruits including rowanberry, rose hip, cornelian cherry, barberry, myrtle, elderberry etc. have played an important role in supplementing staple foods by supplying vitamins, minerals, and trace elements in order to obtain a balanced diet for healthier life (Vijayan et al. 2008; Ercisli et al. 2008; Serce et al. 2010; Fazenda et al. 2019). Their interest as a source of nutraceuticals has been highlighted in recent studies (Leonti 2012; Guney et al. 2019).

The wild edible fruit species have been part of the human diet and they can be an interesting genetic resource for the development of new food products (Tardio et al. 2006; Butiuc et al. 2019). Wild edible fruits may have great potential as a source of unusual colours and flavours, bioactive compounds and as sources of dietary supplements or functional foods (Salvatore et al. 2005). In addition due to continuous seed propagation of wild edible fruit species, there is a great variability for most of the morphological and biochemical characteristics of wild non-cultivated fruits that could be important for breeding activities to obtain healthier fruits.

Sorbus is a large genus including nearly 250 species and among *Sorbus* species, rowanberry (*Sorbus aucuparia*) and service tree (*Sorbus domestica*) are more common for fruit production. *Sorbus aucuparia* found between 800–2500 m and *Sorbus domestica* found between 100–1300 m in Turkey

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and have been using as ornamental plants in botanical gardens and park areas. In rural areas and forests, fruits of particularly rowanberry (*Sorbus aucuparia*) and service tree (*Sorbus domestica*) are used as food or food ingredients (Gil-Izquierdo and Mellenthin 2001). Also, alcoholic drinks, for example, wine, beer, and spirits, such as brandy or vodka, were made from or flavored with rowanberry juice or their fruits (Mikulic-Petkovsek et al. 2017) mostly because of the biochemical compounds that help to clear and preserve alcoholic drinks, adding flavor, astringency, bitterness, and extra sugars. In Turkey, rowanberry fruits have been consumed due to their nutritive and medicinal properties. It is surmised that it reduces diabetic symptoms, the reason of which these fruits have been traditionally used as an antidiabetic agent (Termentzi et al. 2008).

In literature, there was limited information about morphological and biochemical diversity among rowanberry fruit. Thus, in this study, we aimed to determine and compare some important morphological and biochemical features of *Sorbus aucuparia* genotypes, naturally growing in Coruh valley in Turkey due to their potential application in functional foods.

Materials and Methods

Plant Material

Commercially ripe fruit of *Sorbus aucuparia* L. is naturally grown in Coruh valley located in the Northeastern part of Turkey were sampled. During commercial harvest period, red color *Sorbus aucuparia* L. fruit was collected in 2017 from 12 pre-selected genotypes that show higher yield, pest and diseases free and more attractive bigger fruit.

Morphological Characteristics

The external fruit color of 40 fruit from each genotype was determined with a Minolta Chroma Meter CE-400, recording L^* , a^* , and b^* color coordinates on the berry surface. Color parameters a^* and b^* extend from -60 to 60 ; a^* negative is for green and a^* positive is for red and b^* negative is for blue and b^* positive is for yellow. The data were expressed as h° (hue angle) calculated as $\tan^{-1}(b^*/a^*)$ in 0 to 360° (0° =red, 90° =yellow, 180° =green, and 270° =blue). Minolta a^* and b^* values were used to compute values for chroma as well.

Biochemical and Bioactive Composition

Sample Preparation and Extraction

For the analyses of the organic acids, specific sugars, total phenolic contents, and total antioxidant capacity analyses, the harvested fruit was immediately frozen and stored at -20°C until further analysis. During the analysis, the fruits were taken from refrigerator and thawed to 24 – 25°C . A laboratory blender was used to homogenise the fruit samples (100 g lots of fruits per genotypes) and a single extraction procedure taking 3 g aliquots transferred inside tubes and extracted for 1 h with 20 mL buffer including acetone, water (deionized), and acetic acid (70:29.5:0.5 v/v) was carried out (Singleton and Rossi 1965).

Extraction of Sugars and Organic Acids

Five grams of samples slurries were mixed with deionized water or metaphosphoric acid (2.5%) for the analysis of individual sugar and organic acid, respectively. The obtained homogenates were centrifuged at 10,000 rpm for 10 min. The samples were filtered into HPLC vials using $0.45\ \mu\text{m}$ PTFE membrane filter for analysis. All HPLC solvents were sonicated. All samples and corresponding standard injection were repeated three times and the mean values were calculated.

Chromatographic Conditions

The Perkin Elmer HPLC system controlled by Totalchrom navigator software (version 6.2.1), consists of a pump and UV detector was used for analysis of the samples. Organic acids separation and determination were performed as on the method reported by Shui and Leong (2002). The sugars were determined using the method of Bartolome et al. (1995) with help of HPLC with refractive index (RI) detector. The separation was carried out on SGE SS Exsil amino column ($250 \times 4.6\ \text{mm}$ ID). The isocratic elution was performed using acetonitrile (80%) and deionized water (20%) with a flow rate of $0.9\ \text{mL}/\text{min}$. The column was operated at 30°C and the sample injection volume was at $20\ \mu\text{L}$. Quantification of organic acids and sugars were performed against the reference standards.

pH and Vitamin C

Flesh parts of fruit samples were used to assess pH and vitamin C. Vitamin C was determined with the reflectometer set of Merck Co (Merck RQflex). pH of fruit was determined by a pH-meter.

Total Phenolic Content

The total phenolic content (TPC) of the samples was evaluated using the method of Singleton and Rossi (1965). In this procedure, each extract (1 mL) was mixed with Folin-Ciocalteu's reagent and water 1:1:20 (v/v). The samples were incubated for 8 min. Then sodium carbonate (10 mL) having a concentration of 7% (w/v) was added. After incubation for 2 h, the absorbance at 750 nm was measured. The total phenolic content was calculated against the reference standard calibration curve of Gallic acid. The TPC was expressed as mg of gallic acid equivalents (GAE) per 100 g of sample (fresh weight (FW)).

Ferric Reducing Antioxidant Power Assay

FRAP (Ferric reducing antioxidant power) assay was used for antioxidant capacity analysis. For this purpose, acetonitrile fruit extract (50 µL), FRAP reagent (2.95 mL), acetate buffer (0.1 mol/L), TPTZ (10 mmol/L), and ferric chloride of 20 mmol/L (10:1:1 v/v/v) were used. The values of samples absorbance were compared with those of the reference standard calibration curves in the range of 10–100 µmol/L of Trolox was used to determine FRAP values of samples. The FRAP was expressed as mM per 100 g of Trolox equivalent on the basis of the fresh weight of the fruit (Benzie and Strain 1996).

Statistical Analysis

The experiments were performed in quintuplicate. For analysis of variance, the obtained data were used for means calculation. Duncan multiple range tests were performed at the significant level of $p < 0.05$.

Results and Discussion

Tree growth habit and fruit external color indices (L , chroma and hue) were shown in Table 1. As indicated Table 1, there were statistical significant differences among rowanberry genotypes. Most of the genotypes (eight genotypes) had vase growth habit and rest of the 4 genotypes had upright growth habit (Table 1). Fruit external color indices as L , chroma and hue values were found between 30.12–41.04; 27.07–32.81 and 30.44–44.06, respectively (Table 1). Mikulic-Petkovsek et al. (2017) reported L , chroma and hue values in 2 rowanberry cultivars between 33–45; 28–30 and 33–43 indicating some similarities with our results. The term sensory quality of fruits is mainly related to their visual appearance (e.g. colour, size), and can be affected by many factors including genotype. Thus the most important factor affecting red color develop-

Table 1 Growth habit, and color indices (L , Chroma and Hue) of rowanberry genotypes

Genotypes	Growth habit	L	Chroma	Hue
RW1	Upright	33.26c	29.11bc	38.47bc
RW2	Vase	41.04a	27.69cd	30.44d
RW3	Upright	37.05b	30.42bc	35.83bc
RW4	Upright	40.27ab	27.07d	43.32ab
RW5	Vase	34.45bc	28.68c	39.56bc
RW6	Vase	30.12d	30.89b	40.02b
RW7	Vase	35.22bc	31.15ab	41.12ab
RW8	Vase	36.55bc	27.44cd	44.06a
RW9	Vase	38.36ab	32.81a	35.17c
RW10	Upright	37.22ab	28.18cd	42.44ab
RW11	Vase	34.88bc	28.73c	34.14cd
RW12	Vase	39.31ab	30.02bc	40.11ab

Different letters indicate the statistical difference within the same column among genotypes at 5% level

ment in fruits was genetic background. Some cultivars or genotypes lack the ability to synthesize large quantities of anthocyanin. The other factors are light and temperature, tree nutrition, crop load and stress. Among consumers highly colored fruits are desired. Thus it is important for fruit cultivars that are genetically programmed to produce highly colored fruits (Ozturk et al. 2009).

Sugar contents of rowanberry genotypes are given in Table 2. Glucose and fructose are the predominant sugars for all rowanberry genotypes. Sorbitol was also found high level in rowanberry fruits after glucose and fructose. In the experimental material, the respective sugars were estimated overall as follows: Glucose (30.15–43.28 g/kg FW), fructose (22.40–34.12 g/kg FW), sorbitol (16.24–21.13 g/kg FW) and sucrose (0.62–2.04 g/kg FW) (Table 2). Similar results have been reported by Mikulic-Petkovsek et al. (2017) for rowanberry cultivars grown in Czech Republic, with glucose, fructose, sorbitol and sucrose content were between 33.29 and 47.68%; 20.62 and 38.50%; 26.83 and 27.80% and 0.45 and 1.67%, respectively. Sugars are important compounds for quality evaluation of rowanberry fruits, and in terms of sugar composition, fructose and glucose are the predominant sugars for most rowanberry cultivars (Mikulic-Petkovsek et al. 2017). Results suggesting that the *Sorbus* fruits has high sugar content and could be advantage for processing and also genotype/cultivar is one of the most important factors that have a direct impact on sugar content of rowanberry. The result also suggesting that content of sorbitol in *Sorbus* genotypes was very high in comparison with other fruit species including different berry species (Mikulic-Petkovsek et al. 2012).

Malic acid was the dominant organic acid in fruits of rowanberry genotypes with high percentage (19.41–25.38 g/kg FW) and followed by citric acid (1.94–

Table 2 Specific sugars of rowanberry genotypes (g/kg FW)

Genotypes	Sucrose	Glucose	Fructose	Sorbitol
RW1	1.12 ^{NS}	38.62cd	27.12bc	17.14ab
RW2	0.87	43.28a	25.46cd	16.24b
RW3	1.44	34.20ef	22.40d	20.40ab
RW4	1.56	35.78e	26.89c	20.78ab
RW5	0.62	41.22b	24.60cd	19.36ab
RW6	0.95	30.15g	30.56b	17.88ab
RW7	0.78	32.98f	29.50bc	21.13a
RW8	1.67	40.76bc	34.12a	19.96ab
RW9	2.04	39.62c	32.81ab	16.96b
RW10	1.52	38.66cd	30.10bc	19.20ab
RW11	1.20	37.50d	33.88ab	17.50ab
RW12	1.02	40.10bc	31.20ab	18.65ab

Different letters indicate the statistical difference within the same column among genotypes at 5% level
NS Non significant

Table 3 Organic acids in rowanberry fruits (g/kg FW)

Genotypes	Citric	Tartaric	Malic
RW1	5.31b	1.72 ^{NS}	23.32b
RW2	1.94ef	1.02	22.89bc
RW3	4.67bc	1.11	24.33ab
RW4	3.45d	1.30	20.80cd
RW5	5.44ab	1.26	19.41d
RW6	4.89bc	1.81	20.20cd
RW7	5.18bc	1.44	21.23c
RW8	6.23a	1.34	24.88ab
RW9	6.04ab	1.15	22.06bc
RW10	2.22e	1.50	25.38a
RW11	5.69ab	1.64	20.44cd
RW12	4.37c	1.70	19.88cd

Different letters indicate the statistical difference within the same column among genotypes at 5% level
NS Non significant

6.23 g/kg FW) and tartaric acid (0.78–1.34 g/kg FW), respectively (Table 3). There were statistically significant differences on citric and malic acid considering all genotypes. However tartaric acid not significantly differed among rowanberry genotypes (Table 3). Mikulic-Petkovsek et al. (2017) found that almost 60–88% of total organic acids in *Sorbus* fruits were Malic acid. They also point out that content of citric and tartaric acid together represented 7–39% of total analyzed acids. The result suggesting that rowanberry fruits could be used in purees and jams if it is necessary to slightly increase the acidity of the products. A large number of different organic acids are present in the fleshy parts of all fruits, but the contents of these can vary greatly both between fruits of different species and their cultivars. The presence of organic acids in the fleshy parts of fruits affects both their palatability and their utilization in fruit products. The organic acids, Malic, Citric, Oxalic,

Table 4 PH, acidity and vitamin C values of rowanberry genotypes

Genotypes	pH	Total phenolic content (mg GAE/100g FW)	Antioxidant capacity (mM trolox/100g)	Vitamin C (mg/100g)
RW1	3.84 ^{NS}	193b	4.52bc	28.9bc
RW2	3.71	161d	2.93d	28.4c
RW3	3.80	190b	4.24bc	37.0ab
RW4	3.41	165cd	3.07de	33.0bc
RW5	3.55	175c	3.38cd	36.6ab
RW6	3.60	198ab	4.69b	33.2b
RW7	3.50	180bc	3.44cd	29.8bc
RW8	3.72	185bc	3.83c	30.4bc
RW9	3.56	204a	5.68a	31.2bc
RW10	3.61	170cd	3.24de	37.3ab
RW11	3.47	200ab	5.22ab	38.2a
RW12	3.80	185bc	3.78c	36.9ab

Different letters indicate the statistical difference within the same column among genotypes at 5% level
NS Non significant

and Tartaric are very abundant in some fruits (Walker and Famiani 2018). pH, vitamin C (ascorbic acid), total phenolic and total antioxidant capacity of the rowanberry genotypes are given in Table 4. pH and ascorbic acid content of genotypes were between 3.41–3.84 and 28.4–38.2 mg/100 g FW (Table 4). Previously pH and vitamin C content of rowanberry were determined as 3.68 and 35 mg/100 g (Baltacioglu 2006). The results suggesting that rowanberry berries had average vitamin C content. Piir and Niiberger (2003), who indicated the range of vitamin C content in rowanberry fruits of 12–86 mg/100 g FW and Kampuss et al. (2009) reported 10–51 mg/100 g in rowanberry fruits.

The rowanberry genotypes show wide variation of total phenolic content. Total phenolic content varied from 161 mg GAE per 100 g to 204 mg GAE per 100 g. Previously, total phenolic content was found between 134–220 mg GAE per 100 g fresh samples between Rowanberries (Baltacioglu 2006; Mikulic-Petkovsek et al. 2017). The result indicates total polyphenol richness of rowanberries and high polyphenol content of rowanberries comparable with high polyphenol included fruits such as elderberry, blackberry, and raspberry.

Total antioxidant capacity (FRAP values) of genotypes are shown in Table 4 and results indicated statistically significant differences ($p < 0.05$) among genotypes in the total antioxidant capacities. Total antioxidant capacity was the lowest as 2.93 mM TE per 100 g FW and was the highest as 5.68 mM TE per 100 g. Mikulic-Petkovsek et al. (2017) reported total antioxidant capacity of rowan cultivars between 3.4–4.9 mM TE/100 g fresh weight bases. These differences may be caused by factors such as sample type used, the

species differences, geographical area, and the degree of ripening, climate conditions and experimental conditions. It has been reported that *Sorbus* fruits could contribute to the health. When compared with other fruit species, *Sorbus* fruits have higher or similar antioxidative activity like plums, which have 2.91–5.87 mM trolox/100 g (Kaulmann et al. 2014).

Conclusion

In conclusion, it seems that there are many benefits to consuming rowanberry fruits daily. We know from the healthy plate example, that rowanberry will not supply us with everything our body needs but it does make up a good portion of it. Hopefully this provided us with plenty of information that will encourage not just eating more rowanberry fruits but living a healthy lifestyle and eating a healthy diet that includes rowanberries.

Conflict of interest M.R. Bozhuyuk declares that he has no conflict of interest.

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