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Evaluation of Wild Raspberry (*Rubus idaeus* L.) Ecotypes from Northeastern Türkiye

Gulce Ilhan *

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Evaluation of Wild Grown Raspberry (*Rubus idaeus* L.) Ecotypes from Northeastern Türkiye

Gulce Ilhan

Department of Horticulture, Faculty of Agriculture, Ataturk University, 25240 Erzurum, Türkiye
* Correspondence: gulceilhan07@gmail.com

Abstract: Located between Asia and Europe, Türkiye is very rich in terms of wild edible fruit diversity. One of the most important wild edible fruits in Türkiye is red raspberries and they widely found in cold to warm temperate regions in the country. The wild raspberries in general have red fruit color but yellow fruited genotypes are evident. A total of 11 wild grown red raspberry genotypes sampled in Northern Anatolia region was used in this study and their sensory, biochemical and antioxidant characteristics were determined. For sensory analysis, aroma, taste and juiciness were used as the main criteria to compare ecotypes each other. Biochemical and antioxidant characteristics of ecotypes included SSC (Soluble Solid Content), vitamin C, organic acids, total anthocyanins (TA), total phenolic content, total flavonoid content and total antioxidant capacity. FRAP (Ferric Reducing Antioxidant Power) assay was used to determine antioxidant capacity. There were significant differences among ecotypes and years for most of the searched parameters even they found similar growing condition. The ecotypes had fruit weight between 1.04 g and 1.33 g in first year and 0.97 g and 1.27 g in second year. Fruit Chroma value were between 26.11 and 33.70 in 2021 and 23.17 and 30.19 in 2022 year. Vitamin C content of ecotypes were quite variable and ranged from 29.3 mg/100 g to 44.4 mg/100 g among ecotypes in both years. The total phenolic content, total anthocyanin content and total flavonoid content was between 164-390 mg gallic acid equivalent/100 g; 17.3-33.2 mg cyanidin-3-glucoside equivalent/100 g and 10.3-17.6 mg quercetin equivalent/100 g, respectively in both years. Citric acid was the dominant organic acid for all genotypes. According to the results V-4, V-8, V-3 and V-10 had attractive bigger fruits indicate their suitability for fresh consumption. V-3 and V-5 had sweeter fruits indicated their suitability for processing and V-6, V-7 and V-11 had richer for human health promoting compounds (higher total phenolic content and antioxidant capacity) making them suitable for future use as functional foods and as promising sources of natural antioxidants.

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1. Introduction

Fruits that human beings have used for centuries, especially as food and medicine, are grown in different continents of the world including temperate, subtropical and tropical climates with a great variety of species. Fruit are sources of vitamins and minerals, have an important place in the healthy nutrition chain. Thanks to their high antioxidant content, fruits help protect us from metabolic diseases such as heart, cancer and diabetes and helps keep digestive system healthy [1-6].

Wild edible fruits (WEFs) show greater variability compared to cultivated relatives. Most of the WEFs found in rural areas and in forests and they importantly contribute biodiversity. They also an important source of food and income for rural communities. WEFs very rich for non-nutritive, nutritive, and bioactive substances and have perfect flavor and excellent medicinal value [7-12].

Recently, several governmental agencies have been conducting several trainings and demonstrations to impart knowledge and skills to the local people on wild edible fruits particularly to obtain and improve knowledge on value added products such as such as jam, vinegar, jam, pickles, etc. [13].

Berries are the easiest to grow among the fruit species. It is especially preferred in fresh and processed products with its attractive and aromatic fruits. The most produced and consumed berries

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around the world are strawberries, blueberries, blackberries, raspberries, aronia and elderberries. There is a wide range of colors in berries (usually from red to purple or black), depending on the species. In addition to fresh consumption, its fruits are also used as different food products such as jam, marmalade, jelly, beverages etc. [14-16]. *Rubus* L. is indigenous to six continents and is one of the most diverse genera in the plant kingdom with its main constituents being raspberries and blackberries. The raspberry, which belongs to the Rosaceae family, Rubus genus, and Idaeobatus focke subgenus, is divided into three groups: *Rubus idaeus* (Red raspberries), *Rubus occidentalis* (Black raspberries) and *Rubus neglectus* (Purple raspberries) [17].

Red raspberry, *Rubus idaeus* L, originated from western Türkiye including Kaz (Ida) mountains and called as red forest fruits. In many regions in Türkiye it is also known locally as more, mudimak, kavuklu etc. Since, Türkiye is the homeland of red raspberries, there is many suitable areas for raspberry cultivation. Today, its cultivation has gained importance in large enterprises, family businesses and as an intermediate production option [13-16].

In addition to their attractive and colorful fruits, red raspberry fruits are preferred by consumers and consumed with pleasure due to their distinct tastes and flavors. With the increasing greenhouse production in recent years, its presence in the market throughout the year increases the consumption rate. In addition to its fresh consumption, raspberry is a fruit species that is a candidate to become an important branch of cultivation in the country, with its industrial consumption such as fruit juice, ice cream, pastry and deep freezing. In particular, in Black sea, Marmara and Northern Anatolia forests covered by wild red raspberry plants and they include many valuable ecotypes. The most suitable places for raspberry cultivation in the country are cool in summer and rainy do not exist during harvest time. Plant grow slows down in places with hot, dry and windy summers. The best cultivation occurs at an altitude of 800-1500 m with an average annual rainfall of 750-800 mm [14,17-18]. In different parts of the world wild red raspberries are found as populations and are an important genes source for breeding activities for new raspberries [19]. Approximately 60 major gene traits are reported, important for the selection process for red raspberries those are resistance to raspberry root rot (*Phytophthora rubi*), raspberry spur blight (*Didymella applanata*) and large raspberry aphid (*Amphorophora ideai*). For red raspberry breeding the most important berry characteristics are berry size and shape, firmness, colour, taste, aroma, suitability to different processing methods, postharvest response and ease of harvest [14,20].

Characterization is the identification of plant germplasm. Identification by morphological, biochemical or molecular markers indicates highly heritable characters. Characterization of germplasm is necessary to provide end users with information about the properties of genotypes. Recording and compiling data on important characteristics that distinguish genotypes within a species allows easy and rapid discrimination between phenotypes. It provides a better understanding of the composition and genetic diversity of the collection, allowing simple grouping of genotypes, development of core collections and generation of valuable germplasm for breeding programs [21-22].

In recent years, biochemical analyzes on fruits have become more important due to their properties for human health [22-24]. Although there are many biochemical studies on cultivated red raspberry fruits [14,17,18,22], biochemical analysis and study results on wild red raspberry fruits are limited in literature.

Thus, the study investigates the content of compounds which might have health-promoting potential of a large number of wild grown red raspberry fruits from Vercenik plateau of Türkiye and they can be used in specific breeding purposes (to obtain cultivars had high human health promoting substances). The obtained data will provide more detailed information to the researchers about to determine ecotypes for multipurpose use such as for food, cosmetics, pharmaceutical, and functional product sectors as well.

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2. Materials and Methods

2.1. Plant materials

Vercenik plateau is located between Erzurum and Rize provinces and very rich in terms of wild grown red raspberries (Figure 1). Vercenik mountain covered by wild grown red raspberry shrubs obtained by seeds. The fruits at full maturation stage of 11 red raspberry ecotypes were sampled according to pre-selection criteria (high yield, attractive fruits, free of pest and disease) in both 2021 and 2022 years. The samples taken each from one plant (ecotype). The climate of district is described as Humid Continental by the Köppen Climate System, abbreviated as Dfb. All genotypes are found similar climate and soil conditions in Vercenik plateau. For 11 wild grown plant materials, they were given a code number. These code numbers were preceded by the abbreviation of the Vercenik plateau name (V), and finally the shrub number. Thus, each ecotype was named from V-1 to V-11.

Figure 1. Wild raspberry plants in Vercenik mountain.

2.2. Sensory analysis of fruits

The harvested red raspberry fruits belong to 11 ecotypes were subjected to sensory analysis by 10 experts (5 males and 5 female students) based on taste, juiciness and aroma parameters. The classical and widely used 0 to 9 bipolar hedonic scale was used to rate overall pleasantness of aroma, taste and juiciness [25].

2.3. Morphological traits

Fruit weight, the number of drupelets per fruit and Chroma were morphological parameters determined on 30 fruits per genotype with three replicates. Fruit weight were determined with a digital balance (±0.01 g) (Scaltec SPB31, Scaltec Instruments GmbH, Goettingen, Germany). The number of drupelets in the raspberry fruits was determined by counting. The skin color of the wild red raspberries was measured using chromameter (Minolta CR-400, Konika Minolta Inc. Tokyo, Japan). The results were expressed in terms of L*, a*, and b* values and by using those values Chroma value were obtained [26]. Berry firmness was determined with a non-destructive Acoustic Firmness Sensor (Aweta B.V., Pijnacker, The Netherlands) expressed as N.

2.4. Biochemical parameters

Around 300 g wild red raspberry fruits were harvested from each ecotypes and the fruit juices obtained with a juice presser and stored at −80 °C immediately until further biochemical analyses (SSC, vitamin C, organic acids, total anthocyanins, total phenolics, total flavonoids and antioxidant capacity).

SSC in juice was determined by a digital refractometer (Kyoto Electronics Manufacturing Co. Ltd., Japan, Model RA-250HE) at 22 °C and expressed as % [26].

Vitamin C (ascorbic acid) was quantified using a reflectometer (RQFlex, Merck, Darmstadt, Germany) and expressed in mg/100 g fresh fruit base.

In the study, organic acid was determined with HPLC by Bozan et al. [27]. For the organic acid extractions, 1 g of the sample was mixed with 4 ml of 3% metaphosphorical acid. The mixture was placed in an ultrasound bath at 80 °C for 15 minutes and centrifuged at 5500 rpm for 15 minutes and obtained mixture was filtered. The organic acids extract was analyzed using HPLC (Shimadzu LC 20A vp, Kyoto, Japan) equipped with a UV detector and an 87 H column. The identification of organic acids and peak determination is based on peak retention times and the comparison of spectral data in accordance with standards. Results were expressed as g/100 g FW base.

Total phenolic content was determined using the Folin–Ciocalteu reagent in the modified method Singleton and Rossi [28]. The obtained results are expressed in mg gallic acid equivalents (GAE) per 100 g fresh fruit sample (FW).

For total flavonoid analysis, a total of 40 fruits were used. The fruit was crushed in a blender and homogenized. About 30 mL of the homogenate was obtained and placed in a 50-ml falcon tube. Pulp and juice were separated from each other by a centrifuge at 12.000 × g at 4 °C for 35 min. Total flavonoids were determined as described in the study by Marinova et al. [29] and was expressed as mg QE (quercetin equivalent)/100 g FW.

The total anthocyanin content of fruits was determined using Krawczyk and Petri [30]. Anthocyanins were extracted from 2 g of fruits with 0.1% HCl (2 mL) in 96% ethanol. The mixture was centrifuged at 5.500 rpm for 10 min. Results were expressed as mg of cyanidin-3-glucoside equivalents/100 g of FW.

FRAP (Ferric Reducing Antioxidant Power) [31] assay was used for antioxidant capacity analysis. Results are given as mmol Trolox equivalent/ 100 g FW.

2.5. Data analysis

SPSS Statistics, version 19.0 was used for analysis. Duncan multiple range tests were performed at the significant level of $p<0.05$.

3. Results and Discussion

3.1. Sensory analysis

Main sensory characteristics of 11 wild grown red raspberry ecotypes are shown in Table 1. Most of the genotypes had high aroma (V-1, V-4, V-5, V-6, V-8, V-9 and V-10), sweet fruit taste (V-2, V-3, V-4, V-5, V-9 and V-10) and high juiciness (V-1, V-4, V-5, V-7, V-8 and V-9) in 2021(Table 1). In 2022 year, 8 ecotypes had high aroma (V-1, V-2, V-4, V-6, V-7, V-8, V-9 and V-10), 6 ecotypes had sweet taste (V-2, V-3, V-4, V-5, V-9 and V-10) and 7 ecotypes had high juiciness (V-1, V-2, V-4, V-5, V-7, V-8 and V-9) (Table 1).

Fresh fruit quality is determined by nutritional and bioactive composition but aroma, taste, flavour and juiciness parameters in fruits are also very important and expressed as sensory properties [32,33]. Sugars, acids and volatile components have an important role, especially on taste and flavor parameters [34,35]. Alibabic et al. [25] examined the morphological, chemical, and sensory properties of four cultivars of raspberry (Meeker, Willamette, Fertödi and Polka) in Bosnia and Herzegovina and found great variability on sensory characteristics. Rambaran and Bowen-Fobes [36] indicated sensory differences among red raspberries and cv. Polka found the best properties for most of the sensory properties. Yu et al. [37] made sensory analysis (including appearance, color, flavor, taste and overall acceptability) on 22 red raspberry cultivars in China and found that sensory properties are cultivar dependent and the general sensory evaluation showed that the fruits of 'Rerille', 'DNS4', 'Ruby', 'Haritage' and 'Beijing 32' are excellent in quality and they could be recommended to consumers and traders. Kaplan et al. [32] evaluated sensory fruit quality characters (appearance, juiciness, aroma, flavor and overall taste) of 17 elderberry cultivars and and found significant differences among cultivars.

Table 1. The sensory characteristics in the fruits of 11 wild red raspberry ecotypes in 2021 and 2022 years.

	Aroma		Taste		Juiciness	
Ecotypes	**2021**	**2022**	**2021**	**2022**	**2021**	**2022**
V-1	High	High	Sweet-Sour	Sweet	High	High
V-2	Moderate	High	Sweet	Sweet	Moderate	High
V-3	Moderate	Moderate	Sweet	Sweet	Moderate	Moderate
V-4	High	High	Sweet	Sweet	High	High
V-5	High	Moderate	Sweet	Sweet	High	High
V-6	High	High	Sweet-Sour	Sweet	Moderate	Moderate
V-7	Moderate	High	Sweet-Sour	Sweet-Sour	High	High
V-8	High	High	Sweet-Sour	Sweet-Sour	High	High
V-9	High	High	Sweet	Sweet	High	High
V-10	High	High	Sweet	Sweet	Moderate	Moderate
V-11	Moderate	Moderate	Sweet-Sour	Sweet-Sour	Moderate	Moderate

High bioactive content as well high sensory profiles are the main aim of special breeding programmes. Aroma and taste combination is accepted formation of flavor. In addition, sugars, acids, phenolics, and hundreds of volatile compounds contribute to the fruit flavor [38, 39, 40, 41].

3.2. Morphological traits

Table 2 presents fruit weight, number of drupelets per fruit, fruit firmness and Chroma values in fruits of 11 wild grown red raspberry ecotypes. Significant differences(p<0.05) was evident among genotypes for all searched parameters.

In 2021-year, fruit weight was the highest in ecotype V-4 as 1.33 g and followed by V-3 (1.21 g), V-1 and V-6 (1.14 g) while it was the lowest in ecotype V-2 as 1.02 g (Table 2). In 2002 year in general same trends on fruit weight was observed. Fruit weight was a little bit lower in 2022. In 2022 the average temperature from fruit set to harvest was lower. Fruit weight was the highest in ecotype V-4 as 1.27 g and followed by V-3 (1.15 g) and V-6 (1.11 g) while it was the lowest in ecotype V-10 as 0.97 g (Table 2).

Number of drupelets were between 56 (V-2) and 76 (V-4) in 2021 year and 50 (V-2) and 69 (V-8). Previously, quite variable fruit weight (between 1.0 and 6.0 g) among wild and cultivated raspberries sampled different agro-climatic regions of Türkiye were reported [18,42,43]. In other studies, based on wild red raspberry samples, the fruit weight varied from 1.1 to 1.6 g [44], while the fruit weight in cultivated varieties is between 3.0 and 6.0 g [45]. In another study conducted on wild red raspberry fruits, fruit weight was found as 1.01 g [46]. Cekic and Ozden [43] reported berry weight between 0.7-1.2 g among wild grown raspberries in Turkiye. Karaklajic-Stajic et al. [47] showed a significant effect of genotype on the fruit weight, and the number of drupelets in red raspberries. They found the highest fruit weight in 'Tulameen' (5.17 g), while the lowest value of this parameter was obtained from 'Willamette' (3.57 g).

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Table 2. Fruit morphological characteristics of wild grown red raspberry ecotypes.

	Fruit weight (g)		**Number of drupelets**		**Firmness (N)**		**Chroma**	
Ecotypes	**2021**	**2022**	**2021**	**2022**	**2021**	**2022**	2021	**2022**
V-1	1.14±0.18bc	1.09±0.16bc	65±3.8cd	61±3.3ab	0.41±0.2bc	0.45±0.2ab	29.34±1.7abc	25.22±1.6ab
V-2	1.02±0.12d	0.99±0.10d	56±3.1ef	50±3.2c	0.44±0.4bc	0.51±0.3ab	32.11±1.9ab	30.19±1.8a
V-3	1.21±0.16b	1.15±0.17b	68±4.2c	63±4.0ab	0.52±0.5a	0.56±0.3a	30.03±1.6b	26.14±1.4ab
V-4	1.33±0.24a	1.27±0.20a	76±3.0a	68±3.1ab	0.40±0.2bc	0.43±0.4ab	28.30±1.4ab	26.56±1.3ab
V-5	1.11±0.10c	1.08±0.11bc	62±3.2d	60±3.4b	0.41±0.3bc	0.46±0.5ab	26.36±1.4c	27.19±1.6ab
V-6	1.14±0.12bc	1.11±0.10bc	66±3.0cd	59±3.0b	0.47±0.6abc	0.53±0.5ab	33.70±2.1a	29.06±2.4ab
V-7	1.04±0.10cd	1.02±0.11c	58±2.7e	54±2.8bc	0.40±0.2bc	0.45±0.3ab	27.30±1.5bc	28.25±1.4ab
V-8	1.07±0.10cd	1.03±0.10c	72±3.8b	69±3.5a	0.41±0.2bc	0.47±0.2ab	29.11±1.8bc	25.44±1.6ab
V-9	1.04±0.10cd	1.00±0.09cd	68±3.4c	62±3.0ab	0.38±0.9c	0.40±0.5b	31.18±1.9ab	30.10±2.2a
V-10	1.06±0.10cd	0.97±0.08d	70±4.4bc	67±4.0ab	0.46±0.7b	0.47±0.4ab	30.33±1.9b	27.41±1.8ab
V-11	1.10±0.09c	1.06±0.11bc	60±3.7de	55±3.8bc	0.43±0.8bc	0.49±0.6ab	26.11±2.1c	23.17±2.0b

There were significant (p<0.05) differences among the different letters in the same columns.

They reported the number of drupelets of cultivars were between 80-96 which higher value than our samples. Titirica et al. [22] reported the number of drupelets between 55-105 on cultivated and selections of red raspberries in Romania. Maro et al. [48] reported the number of drupelets in raspberry cultivars between 51-61, respectively that indicate similarities with our results. The influence of the cultivation place upon the number of drupelets may be associated to pollination-inherent factors. The low mass of the wild red raspberries allows to infer about the reduction in the sizes of these drupelets [48]. In present study, berry firmness and chroma were between 0.38 N (V-9)-0.52 N (V-3) and 26.11 (V-11)-33.70 (V-6) in 2021 and 0.43 N (V-4)-0.56 N (V-3) and 23.17 (V-11)-30.19 (V-2) in 2022, respectively (Table 2). Berries, like other fruit groups (pome fruits, stone fruits, nuts etc.) have an attractive appearance. Fruit size and color (external appearance) have a significant impact on quality evaluation by consumers. Cekic and Ozden [43] indicated that berry color is depends of wild and cultivated red raspberries and wild ecotypes showed lower Chroma values.

Firmness is an important factor in particular for fruit resistance to the transportation and handling and marketing [49]. As a berry crop, raspberries are very delicate [50]. Bañados et al. [49] reported firmness differences among the raspberry cultivars. They indicated that firmness was 0.73 in cv. Heritage and 0.24 in cv. Autumn Bliss. In contrast Maro et al. [48] reported lower berry firmness as average 0.12 N in raspberries in Brazil. These results explain how growing conditions and genotypes affects berry firmness on raspberries.

3.3. Biochemical content

Table 3 presents organic acid content of wild grown red raspberry ecotypes. There were statistically differences among ecotypes at p<0.05 level for citric and malic acid content (Table 3). The citric acid was the predominant organic acid in all wild grown red raspberry ecotypes, with the contents from 1.93 (V-9) to 2.46 g/100 g (V-1) in 2021 and 1.99 (V-9) and 2.73 (V-1) in 2022, followed by malic acid with the contents from 0.50 (V-4) to 0.66 (V-1) g/100 g and 0.55 (V-3)-0.75 (V-7) g/100 g, respectively. Tartaric acid detected in some ecotypes with minor contents.

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Table 3. Organic acid content in fruits of 11 wild grown red raspberries.

Ecotypes	Citric acid (g/100 g)		Malic acid (g/100 g)		Tartaric acid (g/100 g)	
	2021	2022	2021	2022	2021	2022
V-1	2.46±0.09a	2.73±0.12a	0.66±0.2a	0.71±0.1ab	0.14±0.01^{NS}	0.17±0.01^{NS}
V-2	2.11±0.09bc	2.32±0.10bc	0.59±0.1ab	0.63±0.2ab	0.10±0.03	0.12±0.03
V-3	2.07±0.08bc	2.15±0.09bc	0.60±0.1ab	0.55±0.1b	0.15±0.03	0.15±0.01
V-4	2.04±0.11c	2.07±0.10bc	0.50±0.1ab	0.57±0.2ab	0.21±0.02	0.23±0.02
V-5	2.10±0.07bc	2.07±0.11bc	0.60±0.2ab	0.64±0.1ab	0.09±0.01	0.14±0.01
V-6	2.32±0.09ab	2.26±0.13bc	0.65±0.2a	0.68±0.2ab	0.12±0.02	0.10±0.02
V-7	2.41±0.09a	2.47±0.12ab	0.63±0.1ab	0.75±0.1a	0.16±0.00	0.22±0.00
V-8	2.22±0.06b	2.27±0.14bc	0.63±0.1ab	0.70±0.1ab	0.10±0.00	0.19±0.00
V-9	1.93±0.07bc	1.99±0.09c	0.57±0.2b	0.63±0.1ab	0.20±0.02	0.26±0.00
V-10	2.16±0.08bc	2.22±0.09bc	0.58±0.2ab	0.65±0.2ab	0.23±0.00	0.27±0.01
V-11	2.43±0.12a	2.37±0.14b	0.64±0.4ab	0.69±0.3ab	0.11±0.00	0.16±0.00

There were significant ($p<0.05$) differences among the different letters in the same columns; NS: Non significant.

In China Yu et al. [37] reported that 22 red raspberry cultivars dominantly (nearly 90% portion) include citric acid and followed by malic acid. In Turkiye, citric acid (1.14-1.82 g/100 g FW) was also identified as the major organic acids in wild red raspberry accessions and small amounts of malic acids (0.05-0.12 g/100 g FW) were also detected [43]. Our findings in agreement with this study results. Organic acids are abundant constituents of ripe fruits depending on species and cultivars and are responsible for their sourness. In addition, they contribute to their flavour. In many fruits, the most abundant organic acids are malic and citric acids. Oxalic, and tartaric acid are also found in fruits. In particular, the flavor of most fruits is formed by the acid-sugar balance. Organic acids are mostly free in the cell sap of fruits, but some are salts, esters, glycosides, etc. They are found in various compounds, but always dissolved in water [51].

Table 4 and 5 shows SSC, vitamin C, total phenolic, total flavonoid, total anthocyanin and total antioxidant capacity in fruits of 11 wild grown red raspberry fruits. There were statistically significant differences (p<0.05) among used ecotypes in terms of SSC, vitamin C, total phenolic, total flavonoid, total anthocyanin and total antioxidant capacity.

SSC content was in range of 9.8% (V-1) and 12.8% (V-5) in 2021 and 10.2% (V-8) and 12.9% (V-5). The all ecotypes were found similar soil and climatic conditions in sampling area thus the differences could be attributed to the genetic structure of the ecotypes. Tosun et al. [42] reported SSC content between 10.87-13.60% and Cekic and Ozden [43] reported SSC content from 10.87% to 13.60% among a number of wild grown red raspberry ecotypes naturally grown in different regions of Turkiye. Veljkovic et al. [52] found SSC as 7% in wild grown red raspberry fruits. Titirica et al. [22] reported the SSC between 8.72-9.15% on cultivated and selections of red raspberries in Romania. The present SSC results are comparable with the data from above studies. SSC content of fruits is affected by a lot of factors including species, cultivars, ecotypes, maturation time, altitude etc. [53]. For determining ripening, SSC and titratable acidity in particular SSC/titratable acidity used. Soluble solid content (SSC) is also influencing the taste and flavor of raspberry fruits [42, 43].

The highest vitamin C content was observed in ecotype V-11 as 41.4 mg/100 g FW, and followed by V-8 (40.1 mg/100 g FW) and V-4 (39.7 mg/100 g FW), respectively. The lowest vitamin C was obtained in ecotype V-2 (29.3 mg/100 g FW) in 2021. In 2022 year, the highest vitamin C content was observed in ecotype V-4 as 43.4 mg/100 g FW and the lowest vitamin C was obtained in ecotype V-2 (33.4 mg/100 g FW) (Table 4). The Vitamin C content was a little bit higher in 2022 year.

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Table 4. Soluble Solid (SSC) and vitamin C in the fruits of 11 wild red raspberry ecotypes.

Ecotypes	SSC (%)		Vitamin C (mg/100 g FW)	
	2021	2022	2021	2022
V-1	9.8±0.4cd	10.3±0.6de	36.3±1.9ab	41.3±2.1ab
V-2	11.2±0.6bc	11.5±0.7c	29.3±1.7c	33.4±1.8c
V-3	12.4±1.0ab	12.2±1.1b	33.6±2.1bc	36.0±2.0bc
V-4	11.7±0.8b	11.9±0.7bc	39.7±2.4ab	43.4±2.0ab
V-5	12.8±0.7a	12.9±1.3a	37.2±2.2ab	39.1±2.0abc
V-6	10.3±0.5cd	11.1±0.6cd	38.8±2.0ab	38.6±2.1b
V-7	10.5±0.4cd	10.8±0.5d	37.2±1.8ab	40.3±1.8ab
V-8	9.9±0.4d	10.2±0.3e	40.1±1.9ab	44.4±2.1a
V-9	11.9±0.6abc	11.8±0.7bc	35.5±2.5b	38.3±2.3bc
V-10	11.6±0.6abc	12.0±0.7bc	35.0±2.3bc	39.0±2.2abc
V-11	10.6±0.7c	10.9±0.5cde	41.4±3.1a	42.7±3.0ab

There were significant ($p<0.05$) differences among the different letters in the same columns.

Veljkovic et al. [52] reported relatively higher vitamin C content (49 mg/100 g FW) in wild grown red raspberry fruits. In Turkiye vitamin C content were reported between 21-36 mg/100 g FW in a number of raspberry fruits [42]. Vitamin C content in fruits of red raspberries previously had been reported e between 17 and 37 mg/100 g FW [54, 55]. In Croatia, Purgar et al. [56], studied on wild grown red raspberries and found vitamin C between 22.34 to 45.00 mg/100 g FW.

Total phenolic content (TPC) was the highest in ecotype V-6 (362 mg GAE/100 g FW) and followed by V-7 ecotype (355 mg GAE/100 g FW) and V-11 (340 mg GAE/100 g FW) while the lowest value was obtained from V-5 ecotype as 164 mg GAE/100 g FW in 2021 year. In 2022 year, it was the highest in V7 ecotype as 355 mg GAE/100 g FW while the lowest in V5 ecotype as 164 mg GAE/100 g FW (Table 5). The genotypes had higher total phenolic content in 2022 year than 2021 year indicating more stress conditions for plants in 2022 year. Results present diversity and richness for total phenolic content of wild grown red raspberries. Cekic and Ozden [43] reported variable TPC between 148-347 mg GAE/100 g among a number of wild grown raspberries in Turkiye. Milivojevic et al. [57] determined the total phenolics in raspberry to be 102–222 mg GAE/100 g in the cultivars and 110 mg/100 g in the wild sample. Pantelidis et al. [55] studied on raspberries in Greece and determined the phenolics of raspberry cultivars between 65.7-249 mg GAE/ 100 g FW.

The total flavonoid content of 11 ecotypes varied greatly and given in Table 5. The ecotype V-2 had the highest total flavonoid content with a value of 17.2 mg QE/100 g fresh weight base whereas the ecotype V-10 had the lowest value with 10.3 mg QE/100 g in 2021. In 2022 year, the ecotype V-6 had the highest total flavonoid content with a value of 17.6 mg QE/100 g fresh weight base whereas the ecotype V-10 again had the lowest value with 11.8 mg QE/100 g (Table 5). In Croatia, Purgar et al. [56], studied on wild grown red raspberries and reported total flavonoid content between 20.4-24.6 mg QE/100 g FW. Aglar et al. (2023) found total flavonoid content as 15.1 mg QE/100 g in one wild grown red raspberry ecotype in Turkiye. Sariburun et al. [14] used red raspberry cultivars (Aksu Kirmizisi, Rubin, Newburgh, Hollanda Boduru and Heritage) in biochemical analysis and reported total flavonoid content between 15.4-41.1 mg catechin equivalent/100 g FW. The total flavonoid content in our samples were comparable with above reports and differences could be due to extraction methods, cultivars, or growth and cultivated conditions. Environmental factors including light, temperature, soil nutrients, altitude also influence the flavonoid content in fruits [14].

Table 5. Total phenolic, total flavonoid, total anthocyanin and total antioxidant capacity in the fruits of 11 wild red raspberry ecotypes.

Ecotypes	Total phenolic (mg GAE/100 g FW)	Total phenolic (mg GAE/100 g FW)	Total flavonoid (mg QE/100 g FW)	Total flavonoid (mg QE/100 g FW)	Total anthocyanin (mg cy-3-g eq./ 100 g)	Total anthocyanin (mg cy-3-g eq./ 100 g)	FRAP (mmol TE/100 g	FRAP (mmol TE/100 g
V-1	255±13.2d	303±15.1c	14.8±0.9cd	15.4±0.8bc	27.0±0.8e	26.3±0.7bc	17.0±1.0e	19.3±1.1
V-2	210±10.9ef	232±14.2	17.2±1.1a	16.9±1.0ab	32.1±1.1b	28.3±1.0ab	14.8±0.9f	16.2±0.8
V-3	270±12.6cd	279±11.3d	15.0±0.7cd	15.5±0.8b	28.4±0.9d	26.2±0.7bc	18.2±1.3d	19.5±1.0
V-4	233±11.2e	256±12.0e	14.0±0.8de	15.2±0.9bc	23.2±0.6f	23.0±0.5c	16.2±0.8ef	17.9±0.7
V-5	164±9.8g	188±9.2g	12.9±0.6ef	13.3±0.5c	19.4±0.6g	20.3±0.4cd	13.4±0.4g	14.6±0.5
V-6	362±18.4a	390±17.1a	16.3±0.9b	17.6±1.1a	33.2±1.3a	31.0±1.1a	22.3±1.5a	23.3±1.4
V-7	355±17.6ab	367±18.0b	13.6±0.8e	14.2±0.9bc	22.0±0.8fg	21.4±0.7cd	21.0±1.3b	22.7±1.2
V-8	241±12.0de	250±11.1e	14.4±1.1d	14.8±1.0bc	17.3±0.5i	17.6±0.4de	12.0±0.6h	15.5±0.7
V-9	296±14.9c	308±15.2bc	15.2±1.2c	15.9±1.1ab	30.9±1.1c	27.9±1.0b	19.0±1.1cd	21.4±1.4
V-10	188±10.5f	204±11.0f	10.3±0.5f	11.8±0.5d	30.0±1.2cd	29.6±1.1ab	10.4±0.5i	12.3±0.8
V-11	340±16.7b	361±18.0b	15.9±1.1bc	14.6±1.0bc	18.0±0.5h	19.2±0.7d	19.6±0.9c	20.7±1.1

There were significant (p<0.05) differences among the different letters in the same columns.

The total anthocyanin content (TAC) of 11 wild grown red raspberry ecotypes were in range of 17.3 (V-8)-33.2 (V-6) mg cy-3-g eq./100 g in 2021 and 17.6 (V-8)-31.0 (V-6) mg cy-3-g eq./100 g indicating 2 times differences among the highest and the lowest anthocyanin included ecotypes (Table 5). Previous studies conducted on both wild and cultivated red raspberries indicated genotypic differences among samples. For example, Sariburun et al. [14] used 5 red raspberry cultivars and found quite variable total anthocyanin content between 12.4-69.5 mg cy-3-g eq./100 g according to cultivars and extraction solvents (water and methanol). Cekic and Ozden [43] reported significant differences (13.7-29.6 mg cy-3-g eq./100 g) among wild grown red raspberry samples. Purgar et al. [56], studied on wild grown red raspberries and reported total anthocyanin between 27.9-47.0 mg cy-3-g eq./100 g. Kostecka-Gugala et al. [58], reported TAC among red raspberries 29.69-81.13 mg cyanidin 3-glucoside equivalents per 100 g FW. It was found between 35.1 and 49.1 mg in Greece [55].

Total antioxidant capacity of 11 wild grown red raspberry samples is given in Table 5. In 2021 year, the ecotype V-6 gave the highest FRAP value (22.3 mmol/100 g FW) while V-10 genotype gave the lowest one (10.4 mmol/100 g). In 2022 year, the ecotypes in general displayed same trend and similar to first year V-6 gave the highest FRAP value (23.3 mmol/100 g FW) while V-5 genotype gave the lowest one (14.6 mmol/100 g). Previously FRAP value of wild grown red raspberry fruits are reported between 8.1-12.4 mmol/100 g FW [42], 11.2-19.7 mmol/100 g FW [43] which corresponds to the results we obtained. Studies on red raspberry fruits in different agri-ecological conditions in Turkiye and abroad showed that the red raspberry fruits had high antioxidant capacity determined by different assays such as FRAP, TEAC, CUPRAC etc. [14,37,43].

Fruit antioxidant capacity are affected by some factors including genetic background, maturity stage etc. However, genotype proved to be the most important factor influencing the fruits' bioactive capacity [59-66].

3.4. Principal Compenent Analysis (PCA)

The PCA to identify groups among the eleven wild grown red raspberries are shown in Figure 2. 68.85% of the variability observed in the characteristics was explained by the first three components

(33.32%, 20.11% and 15.42% for PC1, PC2 and PC3, respectively). The ecotypes divided into 4 group. The first principal component (PC1) explains 33.32% of the overall variance, is clearly identified with the fruit weight, SSC and vitamin C, while the second principal component (PC2) is related to the total phenolics, total anthocyanins and citric acid. The factors that most contributed to PC1 (positive side) were: fruit weight and the number of drupelets per berry. The PCA analysis revealed and confirmed significant differences in the morphological, biochemical and bioactive content of wild red raspberry ecotypes (Figure 2).

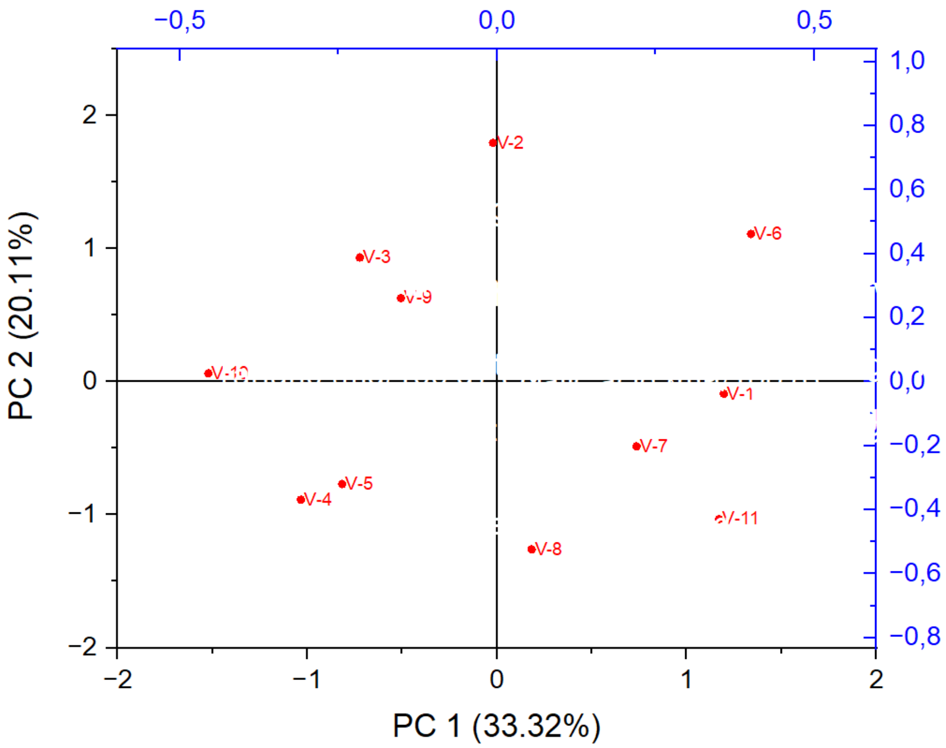

Figure 2. Distribution of wild red raspberries according to morphological, biochemical and bioactive characteristics determined by principal component analysis.

Conclusions

A detailed morphological, sensory and biochemical analysis was reported here for the first time in a large number of wild grown red raspberry in North eastern Turkey. The results indicated that even in same growing location, wild grown red raspberry ecotypes showed rich diversity on most of the morphological traits and nutritional and nutraceutical compositions. According to results V-4, V-8, V-V-3 and V-10 had attractive bigger fruits indicate their suitability for fresh consumption. V-3 and V-5 had sweeter fruits indicated their suitability for processing and V-6, V-7 and V-11 had richer for human health promoting compounds (higher total phenolic content and antioxidant capacity), making them suitable for future use as functional foods and as promising sources of natural antioxidants.

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