



Comparison of Some Parts of *Cotoneaster coriaceous* Franch. Plant in Terms of Phytochemicals and Antioxidant Capacity

Hatice Feyza Akbulut^{1,a,*}

¹Selçuk University, Çumra Vocational High School, Department of Medicinal and Aromatic Plant, Konya, Türkiye

*Corresponding author

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ABSTRACT

Cotoneaster spp. is a plant belonging to the Rosaceae family, which includes different genera and taxa. It is a woody plant that grows from shrubs to trees depending on its height (between 0.2-20 m) and grows in the temperate areas of Europe, North Africa and Asia. Many *Cotoneaster* species have become highly popular ornamental plants due to their striking leaves, dense flowers, and bright red-black fruits. These species have been used traditionally for the treatment of numerous diseases due to their rich bioactive components present in both their above-ground and below-ground parts. This study investigates the phytochemical and antioxidant properties of the above-ground parts of *Cotoneaster coriaceous* Franch., including its fruits, stems, and leaves. For this purpose, total phenolic content (TPC) and DPPH radical scavenging activity, organic acid and sugar profile, and mineral distributions were determined. According to the results, the highest amounts of macro-minerals identified were potassium (K) and calcium (Ca), while iron (Fe) and boron (B) were the predominant micro-minerals. The dominant organic acid in the fruit was malic acid, while succinic acid was prevalent in the stems and leaves. Sucrose and fructose, the sugars detected in the fruit, were found in equal levels in the stems and leaves. Fructose was identified as the dominant sugar in the leaves. It was determined that the fruit, stem, and leaf parts of the *Cotoneaster coriaceous* Franch. plant species were rich in TPC, with the stems exhibiting higher antioxidant capacity.

^a haticefeyza@selcuk.edu.tr

<https://orcid.org/0000-0001-6798-0953>



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Introduction

The Rosaceae family, encompassing herbaceous and climbing plants, woody trees, and shrubs, is a large plant family distributed from the temperate regions of the Northern Hemisphere to the subtropical zone. Globally, it comprises 104 genera, with approximately 297 taxa within 37 genera found in Türkiye (Hürkul & Köroğlu, 2021).

Species of the genus *Cotoneaster* belonging to the family Rosaceae range from shrubs extending up to 0.2 m to trees reaching 15-20 m and are found in temperate regions of Europe, North Africa, and Asia. The *Cotoneaster* genus comprises 84 species, predominantly found in Türkiye, Syria, Iran, Pakistan, and Europe (Ali et al. 2021). Many *Cotoneaster* species have become popular ornamental plants due to their attractive leaves, abundant flowers, and particularly bright red (or occasionally black) fruits. Numerous varieties have been developed and are frequently planted in temperate and warm temperate regions (Bartish et al., 2001).

Since the beginning of human existence, people have benefited from the healing properties of plants (Mann, 2000). Plants, with their diverse phytochemical structures, function as complex chemical industries, offering a wide

range of natural products (Choi et al., 2009). Herbal medicines are favored due to their minimal side effects, low cost, and ease of availability. There are approximately 450,000 plant species on Earth, with about 308,312 of these being vascular plants (Gupta et al., 2020; Ali et al., 2021; Pimm & Joppa, 2015). According to a World Health Organization report, approximately 80% of the population in the developing countries relies on herbal medicines to treat diseases (Ali et al., 2021).

Species of *Cotoneaster* have been used for long time in traditional medicine in East Asia, Iran, Pakistan, Türkiye, Mongolia, and Tibet. In Iran, the gum fraction obtained from the branches of *C. tricolor* and *C. discolor* is orally administered to mothers or newborns for the treatment of jaundice (Kicel et al., 2016; Ali et al., 2021). Some species of the *Cotoneaster* are used as laxatives, relaxants, hemostatic, and expectorants. This plant is also employed in the treatment of eye and bronchial infections, choking, thirst, itching, fever, lesions, hemorrhoids, and urinary stones (Swati et al. 2018; Khayam et al. 2019; Popoviciu et al. 2020). Methanolic extract of *C. orbicularis* has demonstrated both lipoxigenase inhibition and antioxidant

properties. Notable biologically active flavonoids and their glycosides have been extracted from this plant (Khan et al., 2007). Additionally, various phenolic compounds such as anisic acid, protocatechuic acid, *p*-coumaric acid, catechin, epicatechin, vitexin, 2-O- α -rhamnopyranosyl vitexin, rutin, isoquercetin, hyperin, and naringenin have been identified in *C. simmonsii* (Palme et al., 1996; El-Mousallamy et al., 2000; Khan et al., 2007). Furthermore, a lignan with significant antioxidant activity was isolated from the ethylacetate-soluble fraction of *C. racemiflora* (Boland & Donnelly, 1998).

There are many studies in the literature on different species of the genus *Cotoneaster* (Zengin et al., 2014; Ali et al., 2021; Holzer et al., 2013). To the best of our knowledge, this is the first study on the species *Cotoneaster coriaceus* Franch. Therefore, this study aimed to determine the phenolic contents, organic acid and sugar distributions, DPPH radical scavenging activity, and macro- and micro-mineral contents of the fruits, stems, and leaves of *Cotoneaster coriaceus* Franch..

Materials and Methods

Plant Materials

The fruits, stems, and leaves of *Cotoneaster coriaceus* Franch. plant species were used. Samples were obtained from the Selçuk University Alaaddin Keykubat Campus and immediately brought to the laboratory. The fruits were cleaned, and stems were separated from fruits. Additionally, the leaves of the plant were also used. The fruits, stems, and leaves were firstly kept at -80°C for 24 hours and dried in a freeze-dryer (Labogene ScanVac Coolsafe110-4, Lynge, Denmark). The lyophilized samples were stored in a closed package at -30°C until used. These samples were used to prepare methanol extracts. The methanol extracts were used for determining in vitro antioxidant activity and total phenolic content, while the ground lyophilized forms were used for determining macro and micro minerals. Aqueous extracts of the samples were used in sugar and organic acid analyses.

Extraction Process of Fruits, Stems and Leaves of *Cotoneaster coriaceus* Franch.

Twenty grams each of lyophilized and ground *Cotoneaster coriaceus* Franch. fruit, stem, and leaves were separately mixed with 150 mL of methanol. The samples were shaken at 200 rpm in a flask for 24 hours. The solution was filtered through coarse filter paper, and the solvent was removed from the filtrate using a rotary evaporator at 40°C . Fifty milliliters of pure water was added to the extracts, and they were held at -80°C for 24 hours, then dried by removing the water in a freeze-dryer. The methanol extracts were used to analyze the total phenolic content and assess the antioxidant capacity.

Total Phenolic Content Analysis

The total phenolic content was measured using the Folin-Ciocalteu colorimetric method. 0.5 mL of methanol extract solution (prepared by diluting 1 g of lyophilized extract powder with 50 g methanol) was mixed with Folin solution (2.5 mL; 0.2N) followed by the addition of 2 mL of a saturated Na_2CO_3 solution. This mixture was allowed to react for 2 hours. Then, the absorbance of the samples was

measured with a spectrophotometer set at 765 nm. The results were calculated using the gallic acid curve and expressed as mg gallic acid equivalent (GAE) per gram dry weight of extract (Binici et al., 2024; Singleton & Rossi, 1965).

DPPH Radical Scavenging Activity

The DPPH radical scavenging activity of the fruit, stems, and leaves extracts of *Cotoneaster coriaceus* Franch. was analyzed using the method described by Brand-Williams et al. (1995), with some modifications in sample preparation according to Akbulut & Akbulut (2023). In this method, 0.1 mL of the methanolic extract solution (by diluting 1 g extract powder with 50 mL of methanol) was added to 3.9 mL of DPPH solution (6×10^{-5} M). After 30 minutes of incubation in the dark at room temperature, the absorbance of the samples was read using a spectrophotometer set at 515 nm. The results were expressed as μmol Trolox equivalent (TE)/g dry weight extract.

Macro and Micro Mineral Analyses by ICP-AES

Approximately 0.2 g of dried and ground samples were placed into a combustion vessel, followed by the addition of 15 mL pure HNO_3 and 2 mL H_2O_2 . The samples were incinerated at 200°C in a MARS 5 Microwave Oven, and the dissolved ash was diluted to a specific volume with ultra-pure water and filtered. The concentrations of macro and micro mineral elements were determined using an ICP-AES (Skujins, 1998; Kahve et al., 2024).

Determination of organic acid and sugar profiles by HPLC

Four grams of dried and ground samples were extracted in 50 mL of ultra-pure water using a homogenizer (WiseMixTM HG-150; Daihan Scientific, Korea). The mixture was centrifuged at $4000 \times g$ for 15 minutes (NF 800R, Nuve, Türkiye) and the supernatant was filtered. The organic acid and sugar analyses were conducted using an Agilent 1260 Infinity Series HPLC system equipped with DAD and RID detectors, respectively. Separation was achieved using an Aminex HPX-87H column (Bio-Rad, 300 x 7.8 mm). The mobile phase was 0.005 N sulfuric acid at a flow rate of 0.6 mL/min. The DAD detector was set to 210 nm for organic acids, and the temperature was maintained at 50°C (Coklar et al., 2018; Akbulut et al., 2024). Identification of organic acids and sugars was based on retention times, and the data were analyzed using ChemStation software.

Statistical analysis

The total phenolic content, DPPH radical scavenging capacity, organic acid and sugar profile, and macro- and micro-mineral results of fruits, stems, and leaves of *Cotoneaster coriaceus* Franch. plant species were subjected to analysis of variance (ANOVA) using the MINITAB Software version 19 (Minitab Inc., PA, USA) for statistical evaluation. Tukey's test was used to determine whether the differences between group means were significant. A significance level of $P < 0.05$ was considered. All results are presented as "mean value \pm standard deviation." To differentiate between the experimental groups such as fruits, stems, and leaves, methods such as principal component analysis (PCA), and hierarchical cluster analysis (HCA) were employed.

Results and Discussion

Total Phenolic Content

The total phenolic content (TPC) of methanol extracts from the fruit, stems, and leaves of *Cotoneaster coriaceous* Franch. is shown in Table 1. According to the results, the highest total phenolic content was found in the stems extract at 38.35 ± 3.28 mg GAE/g, followed by the leaves extract at 24.21 ± 2.07 mg GAE/g, and the fruits extract at 9.055 ± 0.607 mg GAE/g.

Kicel et al. (2016) determined the TPC in the leaf samples of 12 *Cotoneaster* Medik. plant species to be between 5.2 and 15.4 g GAE/100 g. They identified *C. bullatus*, *C. zabelii*, *C. hjelmqvistii*, *C. divaricatus*, *C. lucidus*, and *C. splendens* as the species with the highest phenolic content and suggested that these species' leaves, showing the highest phenolic levels, could be the most promising natural antioxidant sources and have significant potential for pharmaceutical applications due to their high antioxidant capacities.

The TPC of *Cotoneaster nummularia* Fisch. extracts ranged from 81.11 to 266.39 mg GAE/g extract, with the water extract showing the highest phenolic content, followed by the methanol and ethyl acetate extracts (Zengin et al., 2014).

A study on various extracts of *Cotoneaster microphyllum* have found that the phenolic content in the aerial parts of the plant ranged from 33.00 to 83.96 mg GAE/g dry extract. The ethyl acetate fraction exhibited the highest phenolic content at 83.96 mg GAE/g, while the n-hexane fraction showed the lowest at 33.00 mg GAE/g (Ali et al., 2021).

Phenolic compounds are bioactive substances with high antioxidant potential. Therefore, a strong relationship between antioxidant capacity and TPC was emphasized in literature (Skotti et al., 2014; Alwazeer & Sally, 2019).

The results of our study also revealed a strong correlation between the TPC and antioxidant activity (Figure 1). The stem of *Cotoneaster coriaceous* Franch. fruit exhibited higher levels of both antioxidants and total phenolics compared to the fruit and leaf extracts.

DPPH Radical Scavenging Activity

The results of the DPPH radical scavenging activity of the fruit stems, and leaves extracts of *C. coriaceous* Franch. are shown in Table 1. It was determined that the DPPH radical scavenging activity of the *C. coriaceous* Franch. stems extract (1142.6 ± 43.8 μ mol TE/g) was higher than that of the fruit and leaf extracts (35.33 ± 0.20 , 348.3 ± 40.8 μ mol TE/g).

In the study of the antioxidant capacity of the water, methanol, ethyl acetate, dichloromethane, and hexane extracts of the young shoots, old stems, and leaves of *Cotoneaster melanocarpus* Lodd, it was determined that the methanol extract of the young shoots showed the highest DPPH radical scavenging activity (IC₅₀ 30.91 \pm 2.97 μ g/mL) (Holzer et al., 2013).

Uysal et al. (2016) reported in their study on the methanol and water extracts of the branches and fruits of *Cotoneaster integrimus* that antioxidant activities determined by the DPPH radical scavenging method was ranked as follows: branch-methanol (B-Me) > branch-water (B-W) > fruit-water (F-W) > fruit-methanol (F-Me).

They noted that of B-Me exhibited twice the reducing ability of F-Me.

For the polar and apolar solvent extracts of *Cotoneaster pannosus* Franch. fruits, the DPPH radical scavenging activity values (IC₅₀) were reported as 47.3 and 54.9 μ g/ml respectively, while ascorbic acid had a value of 2.7 μ g/ml, indicating that polar solvent extracts exhibited higher antioxidant capacity (Les et al., 2017). Sokkar et al. (2013) showed that the ethanol extract of the branches of *Cotoneaster horizontalis* Decne. demonstrated DPPH radical scavenging activity with an IC₅₀ value of 19.3 μ g/ml in a dose-response curve.

Macro- and Micro-Mineral Profile

The macro-mineral values of the fruit, stems, and leaves of *Cotoneaster coriaceous* Franch., including phosphorus (P), potassium (K), calcium (Ca), magnesium (Mg), sulfur (S), and sodium (Na), are shown in Table 2. In the fruits of *Cotoneaster coriaceous* Franch., the highest macro-mineral value was found for K (19910 ± 461 mg/kg), followed by Ca (6486 ± 165 mg/kg), P (2391.3 ± 83.1 mg/kg), Mg (1143.8 ± 55.7 mg/kg), S (1143.8 ± 55.7 mg/kg), and Na (53.01 ± 5.90 mg/kg). In the fruit stems of *Cotoneaster coriaceous* Franch., six macro-minerals were detected as in the fruits, and their amounts were in the following order: K > Ca > Mg > P > S > Na.

In the fruits, stems, and leaves of *Cotoneaster coriaceous* Franch., five micro-minerals were detected: iron (Fe), copper (Cu), manganese (Mn), zinc (Zn), and boron (B). The quantities of these micro-minerals are presented in Table 3. Among the micro-minerals identified in the fruits, B (65.87 ± 1.11 mg/kg) was the most abundant, while Mn (7.090 ± 0.006 mg/kg) was the least abundant.

When examining the stems and leaves of *Cotoneaster coriaceous* Franch., it was determined that Fe was the micro-mineral with the highest concentration in both samples. The Fe content was found at 336.5 ± 14.8 mg/kg for the fruit stem and 401.9 ± 32.5 mg/kg for the leaves. Other micro-minerals in the fruit stem were in the following order: B > Zn > Mn > Cu, while in the leaves, they were found to be in the following order: B > Mn > Zn > Cu.

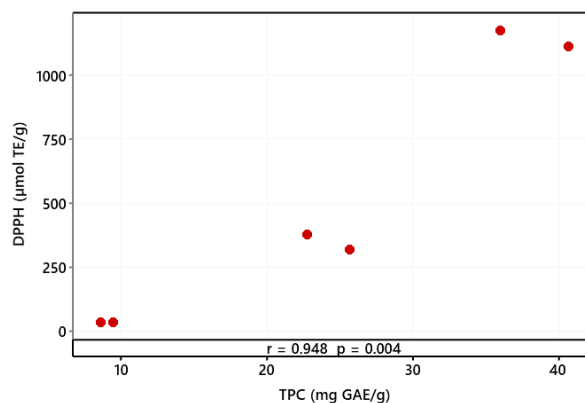


Figure 1. Pearson correlation between TPC and DPPH radical scavenging capacity in the fruits, stems and leaves extracts of *Cotoneaster coriaceous* Franch

Table 1. Total phenolic content (TPC) and DPPH radical scavenging activity of fruits, stems, and leaves extracts of *Cotoneaster coriaceous* Franch.

Plant parts	Total Phenolic Content (mg GAE/g)	DPPH radical scavenging activity ($\mu\text{mol TE/g}$)
Fruits	9.06 \pm 0.61 ^c	35.33 \pm 0.20 ^d
Stems	38.35 \pm 3.28 ^a	1142.6 \pm 43.8 ^b
Leaves	24.21 \pm 2.07 ^b	348.3 \pm 40.8 ^c
Ascorbic acid	-	4679.3 \pm 76.4 ^a

The values are expressed as "mean \pm standard deviation."; Values denoted by different letters in the same column indicate a statistically significant difference between them ($p < 0.05$).

Table 2. Macro-minerals of fruits, stems and leaves extracts of *Cotoneaster coriaceous* Franch.

Plant parts	Macro minerals (mg/kg DW)					
	P	K	Ca	Mg	S	Na
Fruits	2391.3 \pm 83.1 ^a	19910 \pm 461 ^a	6486 \pm 165 ^c	1143.8 \pm 55.7 ^b	656.9 \pm 24.3 ^b	53.01 \pm 5.90 ^c
Stems	1397.0 \pm 50.2 ^c	19539 \pm 405 ^a	11298 \pm 130 ^b	1481.0 \pm 20.9 ^b	647.1 \pm 5.7 ^b	182.84 \pm 4.32 ^a
Leaves	2172.9 \pm 60.8 ^b	15985 \pm 347 ^b	16360 \pm 538 ^a	1674.0 \pm 64.8 ^a	882.2 \pm 24.0 ^a	157.91 \pm 2.80 ^b

The values are expressed as "mean \pm standard deviation."; Values denoted by different letters in the same column indicate a statistically significant difference between them ($p < 0.05$).

Table 3. Micro-minerals of the fruits, stems and leaves extracts of *Cotoneaster coriaceous* Franch.

Plant parts	Micro minerals (mg/kg DW)				
	Fe	Cu	Mn	Zn	B
Fruits	64.82 \pm 5.09 ^c	7.863 \pm 0.129	7.090 \pm 0.006 ^c	7.424 \pm 0.317 ^c	65.87 \pm 1.11 ^a
Stems	336.5 \pm 14.8 ^b	7.563 \pm 0.358	14.53 \pm 0.67 ^b	35.84 \pm 0.20 ^a	37.13 \pm 0.50 ^b
Leaves	401.9 \pm 32.5 ^a	7.795 \pm 0.154	23.83 \pm 0.99 ^a	23.38 \pm 0.52 ^b	26.30 \pm 0.46 ^c

The values are expressed as "mean \pm standard deviation."; Values denoted by different letters in the same column indicate a statistically significant difference between them ($p < 0.05$).

It has been determined that the fresh fruits of *C. pannosus* were rich in macro elements; particularly high amounts of Ca (2950.9 mg/kg) and K (2607.9 mg/kg), while Mg (277 mg/kg) and Na (49.3 mg/kg) were detected in lower amounts (Les et al., 2017). In a study on the fruits of *Cotoneaster nummularia* Fisch., it has been stated that the levels of macro-minerals such as potassium, phosphorus, magnesium, calcium, sodium, and sulfur decrease with the age of the plant (Ali et al., 2018).

Among the microelements in the fresh fruits of *C. pannosus*, significant levels of Fe (13.0 mg/kg), Cu (1.5 mg/kg), and Zn (7.4 mg/kg) have been found (Les et al., 2017). In the investigation of micronutrients in the fruits of *Cotoneaster nummularia* Fisch., it has been observed that iron levels increased with maturity, from 188 $\mu\text{g/g}$ in the pre-reproductive stage to 281 $\mu\text{g/g}$ in the post-reproductive stage. Copper and zinc levels have decreased in the post-reproductive stage, while molybdenum and cobalt have not been detected in either phenological stage. Manganese levels have increased from 24 $\mu\text{g/g}$ in the pre-reproductive stage to 30 $\mu\text{g/g}$ in the post-reproductive stage (Ali et al., 2018).

Ca and K are the most abundant macro minerals in the fruits of *Cotoneaster* species, followed by Mg and P (Les et al., 2017). Potassium, making up about 70% of the positive ions in cells, is crucial for regulating the acid-base and water balance in cells (Senhaji et al., 2020). Ca, K, and Mg are essential for cell repair, strengthening bones and teeth, and red blood cell function. Fe is a vital micro mineral for oxygen and electron transfer in the human body (Özcan and Akbulut, 2008). Cu and Zn are important for human nutrition due to their roles in enzymatic and redox systems (McLaughlin et al., 1999). Additionally, B in the diet is a significant micronutrient as it influences metabolic enzymes, steroid hormones, and the metabolism of

nutrients like calcium, magnesium, and vitamin D, while also potentially enhancing brain functions (Devirian & Volpe, 2003).

Minerals play important roles in our body, from the formation of strong bones essential for healthy and long life to the transmission of nerve impulses. The presence of a variety of minerals not only helps in producing different hormones but also regulates basic functions like maintaining a regular heartbeat. Certain macro and microelements are crucial for the structure and function of teeth and bones. For instance, Ca, P, and F are essential for teeth, while bones also contain Mg, Mn, B, and F. On the other hand, microelements like Cu, Fe, Mn, Mg, Se, and Zn are vital for various enzyme functions. Macro-elements such as Ca, Mg, P, Na, and K are important for nerve function and signaling, whereas trace elements like I are crucial for specific functions like erythrocyte formation, glucose regulation, and antioxidant defense. Additionally, minerals like Ca and K can help manage blood pressure, while Ca, Mg, Cu, Se, and Zn support immune functions, and Cr and Mn contribute to brain health. The results obtained from our study indicate that the fruits, fruit stems, and leaves of *C. coriaceous* Franch. contain significant macro- and micro-minerals essential for human metabolism.

Distribution and Concentration of Organic Acids and Sugars

The distribution and concentrations of organic acids and sugars in fruits, stems, and leaves of *Cotoneaster coriaceous* Franch. are presented in Table 4. In the fruit of *Cotoneaster coriaceous* Franch., three organic acids were identified: citric, tartaric, and succinic acid. In the stems, four organic acids were found: citric, tartaric, malic, and succinic acid. Lastly, six organic acids were detected in the

leaves: citric, tartaric, malic, formic, succinic, and acetic acid. The most abundant organic acids in the plant's fruit, stems, and leaves were observed to be malic acid (544 mg/kg), succinic acid (3790 mg/kg in stems and 8840 mg/kg in leaves).

Table 5 displays the sugar distribution and concentrations in the fruit, stems, and leaves of *Cotoneaster coriaceus* Franch. Two sugars, sucrose and fructose, were identified in the fruit, stems, and leaves of the plant. The concentrations of sucrose and fructose were found to be similar in the fruit, whereas fructose was dominant in the fruit stems and leaves. The stems exhibited the highest sugar content, followed by leaves and then fruits.

The distribution and concentrations of organic acids and sugars in fruits are fundamental quality characteristics that significantly influence taste and aroma. This balance is significant for determining the ripeness level of fruits and also plays a crucial role in the production of products like jam, marmalade, and preserves (Toker et al., 2013). Factors such as variety, harvest time, ecological conditions, and cultivation methods affect the distribution and concentrations of organic acids and sugars in fruits (Chen et al., 2008).

Metabolic changes occurring in fruits affect color, texture, and flavor development, thereby enhancing nutritional and sensory quality. These changes not only increase consumer acceptability but also extend the fruit's shelf life. Changes in organic acid concentrations are examples of these metabolic changes during ripening (Toker et al., 2013). Organic acids are major determinants of the taste and flavor of fruits (Cao et al., 2009; Chen et al., 2009; Xu et al., 2010; Toker et al., 2013). During ripening, organic acids decrease in concentration as they are utilized as an energy source in respiratory metabolism and contribute to sugar production as a carbon source. The main sugars accumulated during ripening are glucose, fructose, and sucrose (Hasegawa et al., 2010; Toker et al., 2013).

While numerous studies have investigated the phenolic acid content of various *Cotoneaster* species' aboveground parts (Zengin et al., 2014; Les et al., 2017; Kicel et al., 2018; Kicel et al., 2019; Krzemińska et al., 2022), no studies regarding the organic acid and sugar profile such as malic, tartaric, citric, succinic acids have been found.

Krzemińska et al. (2022) identified numerous phenolic compounds, mannitol (6834 ± 249 and 3104 ± 123 $\mu\text{g/g}$ dry weight, respectively), and ascorbic acid (298 ± 10 and 1726 ± 66 $\mu\text{g/g}$ dry weight, respectively) in the leaves of two different *Cotoneaster* species, *C. hissaricus* and *C. hsingshangensis*.

Dmitruk et al. (2022) reported that different *Cotoneaster* species' flower nectars contain an average of 16.90-36.60% (w/w) sugars, predominantly glucose and fructose, with varying dominance depending on the species, along with a small amount of sucrose.

Our study findings indicate that succinic acid is the dominant organic acid in the stems and leaves of *Cotoneaster coriaceus* Franch., while malic acid dominates in its fruits. Succinic acid is naturally found in nearly all plant and animal tissues, as well as in fermentation products like beer, and foods such as meat, eggs, fruits, honey, and molasses (Vaghela et al. 2002). It plays a significant role in intermediary metabolism and the Krebs cycle (Saxena et al. 2017). Citric, malic, and tartaric acids are natural acids mostly found in fruits. Citric acid is the primary acid in citrus fruits like lemons, oranges, mandarins, and grapefruits. Malic acid predominates in fruits such as cherries, apples, and juniper, while tartaric acid is abundant in grapes. Sugars and organic acids indirectly affect the production of phenolic compounds by altering pH and serving as building blocks (Perkins-Veazie & Collins, 2001).

PCA and HCA analyses regarding phytochemical and antioxidant capacity values

In this study, statistical methods such as Principal Component Analysis (PCA) and Hierarchical Clustering Analysis (HCA) were employed to visually and comprehensively evaluate the analyses from different perspectives. PCA mitigates correlations among numerous variables under investigation by reducing them to linear combinations of fewer components. In this study, PCA was utilized to visualize differences in phytochemical components and antioxidant capacity. Figure 2 illustrates a scatter plot (labeled as Figure 2A) where points represent fruits, stems, and leaves of the studied *Cotoneaster coriaceus* Franch. plant species, while vectors represent phytochemical and antioxidant parameters. PC1 explains 68.7% of the variance, and PC2 explains 31.3%, as shown in Figure 2.

Table 4. Organic acids in the fruits, stems and leaves of *Cotoneaster coriaceus* Franch. plant

Samples	Organic acids (mg/kg)					
	Citric	Tartaric	Malic	Formic	Succinic	Acetic
Fruits	195.0 \pm 9.9 ^a	-	544 \pm 59 ^a	-	nd	-
Stems	17.70 \pm 3.25 ^b	nd	325 \pm 35 ^b	-	3790 \pm 212 ^b	-
Leaves	nd	17.60 \pm 0.59	303 \pm 24 ^b	31.81 \pm 1.13	8840 \pm 305 ^a	508 \pm 37

The values are expressed as "mean \pm standard deviation."; Values denoted by different letters in the same column indicate a statistically significant difference between them ($p < 0.05$); nd: not detected; -: absent

Table 5. Sugars in the fruits, stems and leaves of *Cotoneaster coriaceus* Franch. plant

Samples	Sugars(g/kg)			
	Sucrose	Fructose	Total Sugar	F/S
Fruits	0.766 \pm 0.055	0.778 \pm 0.045 ^b	1.544 \pm 0.061 ^b	1.016 \pm 0.139 ^b
Stems	0.609 \pm 0.016	2.800 \pm 0.212 ^a	3.409 \pm 0.174 ^a	4.598 \pm 0.548 ^a
Leaves	0.682 \pm 0.035	2.340 \pm 0.057 ^a	3.022 \pm 0.260 ^a	3.431 \pm 0.099 ^a

The values are expressed as "mean \pm standard deviation."; Values denoted by different letters in the same column indicate a statistically significant difference between them ($p < 0.05$); F/S: Fructose/Sucrose

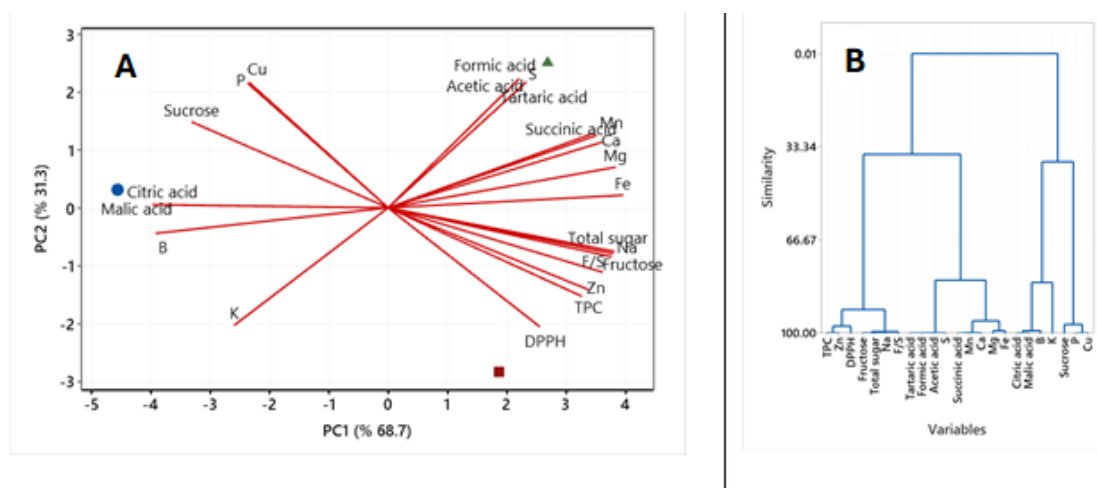


Figure 2. Graphs obtained through the evaluation of phytochemicals and antioxidant activity of the fruits, stems, and leaves of the plant *Cotoneaster coriaceus* Franch.: Biplot (A) and Dendrogram obtained via hierarchical cluster analysis (HCA) (B).

Table 6. PCA results regarding phytochemicals and antioxidant activity of the fruits, stems and leaves of the plant *Cotoneaster coriaceus* Franch.

Items	PC1	PC2
Eigenvalue	18.81	7.19
Variance percentage (%)	68.7	31.3
Cumulative variance (%)	68.7	100
Eigenvectors		
Total Phenolic Content (TPC)	0.207	-0.212
Antioxidant capacity (DPPH)	0.162	-0.285
Citric acid	-0.251	0.008
Tartaric acid	0.147	0.302
Malic acid	-0.251	0.007
Formic acid	0.147	0.302
Sucrose	0.220	0.180
Fructose	0.147	0.302
Total sugar	-0.210	0.206
Fructose/Sucrose	0.229	-0.155
Phosphor (P)	-0.148	0.301
Potassium (K)	-0.164	-0.282
Calcium (Ca)	0.228	0.157
Magnesium (Mg)	0.243	0.097
Sulphur (S)	0.140	0.310
Sodium (Na)	0.241	-0.105
Iron (Fe)	0.251	0.030
Copper (Cu)	-0.150	0.299
Manganese (Mn)	0.223	0.174
Zink (Zn)	0.214	-0.197
Boron (B)	-0.248	-0.061

The leaves of *Cotoneaster coriaceus* Franch. showed close relationships with formic acid, acetic acid, tartaric acid, succinic acid, S, Mn, Ca, Mg, and Fe, clustering on the positive right side of PC1. In contrast, the fruit stems, which exhibit strong associations with fructose, total sugar (TS), fructose/sucrose (F/S) ratio, antioxidant capacity (DPPH), total phenolic content (TPC), Na, and Zn, were clustered on the negative right side of PC1. The fruits of these species, unlike the leaves and fruit stems, clustered on the negative left side of PC1 with Cu, P, B, K, sucrose, citric acid, and malic acid (Figure 2-A). In Figure 2-B, it is observed that the fruits, stems, and leaves of the *Cotoneaster coriaceus* Franch. plant species are divided into 3 clusters based on phytochemical and antioxidant capacity parameters. The first cluster consists of stems

along with TPC, Zn, DPPH, fructose, total sugar (TS), Na, and fructose/sucrose (F/S). The second cluster consists of leaves along with formic acid, acetic acid, tartaric acid, succinic acid, S, Mn, Ca, Mg, and Fe. The third cluster consists of fruits along with Cu, P, sucrose, citric acid, malic acid, B, and K. This situation indicates that the fruit, fruit stem, and leaves of this species can be clearly differentiated from each other based on the determined phytochemical parameters and antioxidant capacity. It is understood that PC1 explains 68.7% of the variance and PC2 explains 31.3% of the variance, and that all variances can be explained 100% by PC1 and PC2 (Table 6).

Many researchers have conducted close analyses of various plants and have generally found that plants are either positively correlated with each other based on their

specific characteristics and grouped together or show negative correlations and are separated from each other (Prasad et al. 2010; Ramdath et al. 2020; Khan et al. 2024). The results of our study are supported by many similar reports in literature.

Conclusion

The results of this study have shown that the fruits, stems, and leaves of the *Cotoneaster coriaceous* Franch. plant have high total phenolic content. Additionally, the results indicate that the antioxidant capacities of the above-ground parts of *Cotoneaster coriaceous* Franch. are quite high compared to many plant species. Among the different parts of *Cotoneaster coriaceous* Franch., the stems have the highest values in terms of total phenolic content and antioxidant capacity, followed by the leaves and fruits. In the tested parts of the *Cotoneaster coriaceous* Franch. plant, a total of 11 elements were detected, including 6 major elements (P, K, Ca, Mg, S, and Na) and 5 minor elements (Fe, Cu, Mn, Zn, and B). The most abundant major element was found to be K (in fruits and stems) and Ca (in leaves), while the most abundant minor element was Fe (in stems and leaves) and B (in fruits).

Three organic acids (citric, malic, and succinic) were identified in the fruits of this species; while 4 organic acids (citric, malic, tartaric, and succinic acids) in the stems, and 6 organic acids (citric, malic, tartaric, formic, succinic, and acetic acids) in the leaves were shown. The most dominant organic acids were malic acid in the fruits and succinic acid in both the stems and leaves. In the fruits of *Cotoneaster coriaceous* Franch., sucrose and fructose were found in equal amounts, while fructose was more dominant in the stems and leaves.

As a result, the above-ground parts of *Cotoneaster coriaceous* Franch., such as the fruit, stems, and leaves, are rich in organic acids, sugars, total phenolic content, and minerals compared to many plant species and are thought to be valuable in terms of human health and nutrition.

Declarations

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Author Contributions

Hatice Feyza AKBULUT: Data collection, investigation, formal analysis, and writing the original draft, project administration, supervision, conceptualization, methodology, review and editing.

The Declaration of Conflicts of Interest

There are no conflicts to declare.

The Declaration of Ethics Committee Approval

Ethics committee permission is not required for this study.

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