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Evaluating the Nutritional and Safety Aspects of *Pyracantha coccinea*: Antioxidant Activity, Mineral, and Heavy Metal Content

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ABSTRACT

In this study, the fruits of *Pyracantha coccinea*, known for their ornamental and medicinal properties, were analysed to evaluate their antioxidant capacity, mineral content, and heavy metal concentrations. The antioxidant potential of *Pyracantha coccinea* was determined using DPPH, CUPRAC, and ABTS tests. Inductively Coupled Plasma Optical Emission Spectrometry (ICP-OES), Atomic Absorption Spectroscopy (AAS), and Gerthard Dumatherm techniques were used to determine the mineral and nutrient composition of the plant. As a result of the evaluation, calcium (0.25±0.02%), protein (4.29±0.47%), potassium (0.39±0.01%), magnesium (0.197±0.01%), sodium (0.08±0.01%), iron (0.012µg/g DW), aluminium (138±9.6 µg/g DW), cobalt (0.541±0.11 µg/g DW), chromium (0.422±0.05 µg/g DW), manganese (20±1.7 µg/g DW), zinc (43.9±4.6 µg/g DW), % DPPH (76.92±0.48) % ABTS value (77.52±0.39) and CUPRAC values (0.771±0.045 for 100ppm) were determined. In particular, the high levels of chromium (Cr) and zinc (Zn) in the fruits exceed the thresholds considered safe for medicinal applications and suggest that the heavy metal content in plants for medicinal use should be critically evaluated within acceptable limits. This study aims to explore the nutritional value and safety of *Pyracantha coccinea* by examining its antioxidant properties, mineral content, and potential heavy metal contamination. The findings will help shed light on its potential benefits and risks, offering valuable insight for its use in health, nutrition, and environmental applications.

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Introduction

Pyracantha coccinea, a perennial shrub belonging to the Rosaceae family, is characterized by its evergreen leaves and ability to grow up to three meters tall. This shrub, known as firethorn, thrives in many altitudes in regions including the Balkans, Europe, the Caucasus, and Turkey. The fruits of this plant are known for their bright red color, small size, and sweet taste. They are not only used to make jam. Still, they are also highly valued in traditional medicine for their beneficial effects on the body's overall health, heart function, and ability to increase urine production (Kambur & Tilki,2010; Keser, 2014). Furthermore, it has been determined that *Pyracantha coccinea* fruits have cytotoxic and antioxidant effects (Sharifi-Rad et al., 2020). The infiltration of ambient pollutants, such as toxic metals, into the world of plants poses a significant obstacle to the safe utilization of medicinal herbs. The buildup of metals such as chromium (Cr) and zinc (Zn) within cells can lead to many health issues, including the onset of cancer (Rodriguez-Fragosa et al., 2008). Therefore, medicinal plants must maintain heavy metal levels below predetermined safety thresholds. Heavy metal pollution is caused by industrial activities as well as the use of chemical pesticides and fertilizers in crops. These activities contribute to the movement of these metals into the surrounding ecosystem (Stanojkovic et al., 2015). In developing countries, medicinal plants have become an essential resource for over 80 percent of the population seeking relief from health complications concomitant with the rise in diseases that include cancer, diabetes, and cardiovascular disease (Seagel et al., 2019; Fridlender et al., 2015).

Macro and micronutrients are crucial for plant life as they are integral to the composition of enzymes that participate in different metabolic processes. Plants exhibit numerous morphological and physiological alterations in their deficits, particularly chlorosis, color changes, leaf curving, leaf spotting, and leaf abscission. The concentrations of major and trace elements in plants are determined by the geochemical characteristics of the soil and the plant's capacity for selective element

accumulation. Furthermore, contaminating plants with environmental constituents from air or water enables certain plants to be bioindicators of environmental pollution (Queralt et al., 2005). The permissible concentrations of elements in plants are crucial for human consumption, as these elements can be transferred to people via the food chain (Peralta-Videa et al., 2009). Examining medicinal plants has revealed that several sources can lead to their pollution with heavy metals. Consequently, it is imperative to recognize these plants' mineral and heavy metal composition to effectively utilize their healing properties while avoiding the dangers of toxicity. The antioxidant capacity of plants can be accurately measured using CUPRAC, ABTS, and DPPH methods, which are well-established methods known for their reliability and cost-effectiveness (Sharifi-Rad et al., 2020)

This research evaluates the elemental and heavy metal composition and investigates the antioxidant activity of Pyracantha coccinea fruits obtained from Van, Turkey. It aims to explore the nutritional value and safety of *Pyracantha coccinea* by examining its antioxidant properties, mineral content, and potential heavy metal contamination. The findings will help shed light on its potential benefits and risks, offering valuable insight for its use in health, nutrition, and environmental applications.

Materials and Method

Firethorn fruits were collected in July 2023 from the garden of a site 20km from the motorway at an altitude of 1720 m (latitude: 38°34'47.65 'N and longitude: 43°16'18.24 'E) in Van, Turkey. The plant was identified by specialist Dr. Murat Ünal and preserved at Van Yuzuncu Yil University Van F Herbarium with the code VANF 20555. For 25 days, the fruits of *Pyracantha coccinea* were dried at room temperature in a location shielded from direct sunlight. To inhibit moisture ingress into the fruits, a freeze-drying procedure was employed using a lyophilization device, operating at a pressure setting of 50 millitorr and a temperature of -80°C, extended over 72 hours.

Treatments Applied to the Plant Material

Two hundred milligrams of dried plant material were dissolved in Teflon tubes at a temperature of 180°C and a pressure of 32 bar for 40 minutes using the Milestone Ethos Microwave digestion system. Following microwave treatment, the samples were placed into 50 ml tubes and adjusted with Ultrapure Milli-Q water until they reached a final volume of 50 ml. The plant samples were evaluated using atomic absorption spectrometry (ICP-OES icap 6000 series, Thermo Scientific).

Procedure for ICP-OES and AAS Analyses

Concentrations of Al, Ag, Cd, As, Cr, Co, Cu, Mo, Pb, Ni, Zn, Fe, Mn, and Se were determined by using ICP-OES. Na, Mg, Ca, and K levels were measured using the AAS instrument. Analyses were conducted in triplicate. All elemental concentrations are expressed in mg kg^{-1} dry weight.

Protein Analyses

The fruits were desiccated in darkness at ambient temperature (22±2°C) and ground using a grinding mill (IKA, A11 basic Analytical mill). Following the grinding of the desiccated plant materials, the quantities of nitrogen and protein were assessed using the Dumatherm Nitrogen-Protein apparatus (Gerhardt Analytical System, Germany). Approximately 50 mg of pulverized plant sample was weighed and composted in aluminum tin cups at 1000°C.

Extract Preparation

The fruits of *Pyracantha coccinea* were dried at room temperature and extracted using ethanol. The 1/10 w/v weighed lyophilized plant sample was kept in a magnetic stirrer in 80% acidified ethanol at 25°C for 24 hours. After centrifugation at 8000 rpm for 15 min at 20°C, the supernatant was evaporated at 110 rpm at 48°C to remove ethanol. The antioxidant capacity was determined by the CUPRAC, DPPH (2,2-diphenly-1-picrylhydrazyl), and ABTS (2,2'-azinobis-(3-ethylbenzothiazoline-6-sulfonic acid)) methods. The extracts obtained were used in different volumes (25µg/ml, 50 µg/ml, 100 µg/ml, 125 µg/ml, 150 µg/ml) for DPPH, CUPRAC and ABTS assays. All extraction procedures were conducted three times. The standard antioxidant (BHT (butylated hydroxytoluene)) and extract were dissolved at a concentration of µg/mL.

Determination of antioxidant capacity by CUPRAC method

The antioxidant properties of the ready extracts were assessed at four distinct concentrations using the CUPRAC method. Cu (II), Neocuprin, and NH_4Ac (Ammonium acetate) buffer were added to the prepared samples and standards at final concentrations of 10, 25, 50, and 100 µg/mL, and absorbance was measured at 450 nm after one hour (Apak et al., 2008). The absorbance levels of the samples were assessed in comparison to the standards. BHT (Butylated Hydroxy Toluene) and α-tocopherol served as standards, with total antioxidant capacity and TEAC (Trolox equivalent antioxidant capacity) values measured in µg/ml for all extracts.

DPPH and ABTS analyses

The DPPH (2,2-diphenly-1-picrylhydrazyl) method was performed according to the procedures described by Brand-Williams et al. (1995). For the analysis, DPPH radical solution was produced in 0.1% (m/v) ethanol and stored in a light-free condition. The samples tested were produced at varying concentrations, and 2 mL of DPPH radical solution was combined with 2 mL of sample solution. These combinations were incubated at room temperature (24°C) in the dark for 30 min. The scavenging capability was evaluated in triplicate. The sample vials were kept in darkening and exposed to continuing rotation for thirty minutes. The absorption spectra of the mixtures at 517 nm were determined using a Multiskan SkyHigh microplate spectrophotometer (Thermo Fisher Scientific, USA). Ethanol was utilized for the blank. The DPPH capacity was measured according to the equivalent:

$$RSC = [(A0 - A1) \div A0] \times 100$$

RSC : DPPH Radical Scavenging Capacity (%)
A0 : Absorbance of DPPH solution
A1 : Absorbance of sample and DPPH mixture

The ABTS method described by Re et al. (1999) is used to evaluate a substance's antioxidant capacity. This method measures the capacity of $ABTS^{\bullet+}$ (2,2'-azinobis-(3-ethylbenzothiazoline-6-sulfonic acid)) radical cation to be neutralized by an antioxidant. The $ABTS^{\bullet+}$ solution (2 mM) was mixed with potassium persulfate $K_2S_2O_8$ (2.5 mM) and incubated at room temperature in the dark for 4 hours. The prepared solution was diluted with ethanol before analysis; the absorbance was adjusted to 0.72± 0.02 at a wavelength of 734 nm. Pyracantha coccinea fruit extracts were prepared for analysis at 10, 25, 50, and 100 mg/mL concentrations. 20 µL of each solution was mixed with 200 µL of $ABTS^{\bullet+}$ solution, and the mixtures were incubated in the dark for 25 min. After infusion, the absorbance of the mixtures was measured using a Multiskan SkyHigh microplate spectrophotometer (Thermo Fisher Scientific, USA). The % ABTS capacity was measured in the same way as DPPH. The extracts' free radical scavenging capacity was expressed in both tests as percentage inhibition. BHT served as the reference antioxidant for antiradical properties.

Statistical Analysis

The statistical analyses were conducted using IBM SPSS 25 software. Data obtained from three independent replicates were summarized as mean ± standard deviation (SD). Differences among groups were evaluated using One-Way ANOVA. When significant differences were detected, Duncan's multiple range test was applied for pairwise comparisons at a significance level of $p < 0.001$. For comparisons between two groups, independent t-tests were applied following Levene's test for homogeneity of variances.

Results and Discussion

Mineral, Heavy Metal and Protein Amounts

The acceptable limits of plant elements are essential for plant growth and human health. Excessive or deficient concentrations of elements in plants can affect agricultural productivity and health risks in the food chain. Table 1 presents the acceptable concentration ranges for various essential elements and minerals in plants, as reported in the literature (Corlett et al., 2002; Kabata-Pendias, 2010; Taiz & Zeiger, 2010). These limits indicate the typical ranges found in healthy plants and are critical for evaluating nutrient status and potential toxicity. In this study, the amounts of Ca, K, Mg, and Na are within acceptable limits, as shown in Table 2. However, the amounts of Zn and Cr are above acceptable values (Table 3). This result was attributed to firethorns growing near motorways and being contaminated with vehicle exhaust gases. Likewise, it may be due to wastewater pollution and chemical fertilizers accumulated in the soil (Liu et al., 2023). Further studies are planned to clarify this result. Toxic levels of heavy metals in plants intended to be used as medicinal plants prevent the plant's therapeutic properties.

The elements identified in the medicinal plants examined in this study are essential and recognized for their significant physiological and biochemical functions in humans. Mn, Zn, and Cu are known for being very important for keeping the body's redox balance because they can eliminate harmful reactive oxygen species (Silva et al., 2019). Mn was detected below the values reported for realizing metabolic activities in plants. Iron is a crucial element of hemoglobin (Vogt et al., 2021). The low levels of Fe and Mn, which play an essential role in synthesizing primary and secondary metabolites in plants and participate in the structure of enzymes, may be associated with geographical, physiological, and genetic factors (Grace et al., 2003; Özay & Pehlivan, 2024). The lower than expected amount of Fe and Mn may also reduce the antioxidant capacity of the plant during its use as a medicinal plant. Cobalt is a component of vitamin B12 essential for red blood cell formation (Osman et al., 2021). Cadmium (Cd), copper (Cu), chromium (Cr), and zinc (Zn) can accumulate in the cells and cause health problems when they are at toxic levels (Rodrigez-Fragoso et al., 2008; Liu et al., 2023). Magnesium (Mg) and Calcium (Ca) are the predominant metallic constituents in numerous plants, as they are integral to chlorophylls, metalloenzymes, and secondary metabolites (Olukayode et al., 2003). Potassium (K) is crucial as a cofactor for various enzymes, particularly in maintaining cell turgor pressure and electroneutrality (Taiz & Zeiger, 2008).

The composition of the soils in which the plants are grown determines the heavy metal and mineral contents. Plants such as *Pyracantha coccinea* can accumulate heavy metals and minerals depending on environmental conditions and soil quality. The amount of accumulation may also differ based on the pollution level of the plant's growth location (Zhang et al., 2022). The protein amount (% 4.29) is determined with previous studies (Song et al., 2023).

Table1. The acceptable limits for some essential elements and minerals in plants

Elements in plants	Acceptable limits
Aluminum (Al)	1-200 (µg/g)
Zinc (Zn)	20-150 (µg/g)
Nickel (Ni)	0.1-5 (µg/g)
Chromium (Cr)	0.006-18 (µg/g)
Cadmium (Cd)	0.05-0.2 (µg/g)
Cobalt (Co)	0.02-0.5 (µg/g)
Copper (Cu)	5-30 (µg/g)
Iron (Fe)	50-250 (µg/g)
Manganese (Mn)	30-300 (µg/g)
Calcium (Ca)	0.2%-1.0%
Potassium (K)	1.0%-3.0 %
Sodium (Na)	0.01%-0.2%
Magnesium (Mg)	0.1%-0.5%

Table 2. Percentage (%) Values of Elements in *Pyracantha coccinea* Fruits

% Amounts of Elements and protein	Mean ±SD (%)
Protein	4.29±0.47
Ca	0.25±0.02
K	0.39±0.01
Mg	0.197±0.02
Na	0.08±0.04

The data were analyzed over three replicates. Values are presented as mean (Mean) ± standard deviation (SD), representing the percentage composition of elements and protein in *Pyrachantha coccineae* fruits. The values presented in this table are based on descriptive statistics."

Table 3. Mineral and Heavy Metal Concentrations in *Pyracantha coccinea* Fruits (µg/g)

Minerals and heavy metals	Mean ±SD (µg/g)
Fe	0.012±0.03
Al	138±9.6
Co	0.44±0.11
Cr	20±0.05
Cu	6.5±1.02
Mn	20±1.7
Zn	200±4.6

The data were analysed over three replicates. Values are presented as mean (Mean) ± standard deviation (SD), representing the percentage composition of elements and protein in *Pyrachantha coccineae* fruits. The values presented in this table are based on descriptive statistics."

Table 4. Percentage (%) antioxidant (DPPH and ABTS) values of *Pyracantha coccinea* Fruits

Method	*P.coccinea* (Mean ± SD)	BHT (Mean ± SD)
DPPH	76.92 ± 0.48^{b}	94.27 ± 0.21^{a}
ABTS	77.52 ± 0.39^{b}	93.36 ± 0.41^{a}

Values are represented as mean ± standard deviation (SD) for three replicates. Letters (a,b) denote significant differences between groups ($p < 0.001$) according to the independent t-test.

Table 5. Trolox Equivalent Antioxidant Capacity TEAC (µmol TE g−1 DW) of *P.coccinea*, BHT, and α-Toc determined by the CUPRAC method

Concentration (ppm)	*P.coccinea (Mean±SD)*	BHT (Mean±SD)	α-Toc
10 ppm	0.246 ± 0.005^{c}	0.95 ± 0.007^{a}	0.35 ± 0.005^{b}
25 ppm	0.495 ± 0.005^{c}	1.48 ± 0.003^{a}	0.69 ± 0.004^{b}
50 ppm	0.989 ± 0.03^{c}	$2.35 \pm 0{,}006^{a}$	1.17 ± 0.004^{b}
100 ppm	1.791 ± 0.04^{c}	$3.28 \pm 0{,}007^{a}$	2.18 ± 0.004^{b}

Values are represented as mean ± standard deviation (SD) for three replicates. Letters (a, b, c) denote significant differences among groups ($p<0.001$) according to One-way ANOVA and Duncan's multiple-range test.

Conducting is crucial for evaluating the capacity of plants to remediate polluted soil through bioremediation. Investigating plants' mineral and heavy metal composition is essential in environmental science, botany, and ecology. These studies help us better understand how plants react to environmental stress, how they play a part in the natural detoxification process of pollutants, and how they ultimately affect human health. The formulation of medicinal herbal concoctions necessitates employing botanicals harboring heavy metal concentrations below specified safety thresholds. Consequently, the surveillance and control of hazardous metal quantities in medicinal vegetation and their derivatives are imperative for preserving both the effectiveness and security of phytotherapeutic agents.

Antioxidant Capacity

A critical factor in assessing the health benefits attributed to dietary components is their ability to yield antioxidative agents. Multiple assays, such as CUPRAC, ABTS, and DPPH, are conducted to determine this potential. All three assays are recommended as uncomplicated, rapid, reliable, and affordable techniques for evaluating the antioxidant activity of plant-based herbal medicines. The capacity of antioxidants depends on numerous external elements, similar to the chemical profile of plants. These factors include inherited potential, preservation, soil composition, agronomic techniques, weather conditions, stressors during the growth season, and industrial procedures.

A common technique for evaluating the antioxidant capacity of natural extracts is the DPPH method, which is based on the reduction of DPPH by an antioxidant solution. According to the DPPH method obtained, the antioxidant activity value of *Pyracantha coccinea* in our study is 76.92 ± 0.48 % DPPH (100 µg/mL), the % ABTS (100 µg/mL) value is 77.52 ± 0.39 (Table 4), and CUPRAC is 0.77 TEAC (µmol TE g^{-1} DW) (for 100ppm) (Table 5). In acccordance with our study, the DPPH antioxidant activity of *Pyracantha coccinea* collected from different locations and extracted with different solvents was reported as 78.73 µg/mL (Keser, 2014), 500 µg/mL (Kerasioti et al., 2019) and 36.53 µg/mL (Semerci et al., 2020) in previous studies. In another study (Tüysüz et al., 2020), the CUPRAC value of *Pyracantha coccinea* was 2.871 µmol TE g^{-1} Dry Weight, DPPH antioxidant capacity as % 83.13 and ABTS as %79.34 TEAC (µmol TE g^{-1}DW) are reported. These differences in results could be explained by differences in

the geographical origin of plants, the time it takes to collect them, and the condition of extraction. Such factors, like temperature, time, and extraction methodology, are known to significantly affect the levels of antioxidant activity measured, which could explain discrepancies in these studies. The safe use of *Pyracantha coccinea* fruits depends on reducing heavy metal absorption. With proper precautions, they may serve as a significant resource for both dietary and medicinal purposes. Given the corroborative findings consistent with existing literature, our research recommends further research to thoroughly evaluate the medicinal effects of Pyracantha coccinea fruits for dietary applications.

Conclusion

Increased concentrations of certain heavy metals, especially chromium (Cr) and zinc (Zn) that are higher than recommended safety thresholds may make these plants less viable for use as medicinal plants. However, because of its high phenolic content, *Pyracantha coccinea* is well known for having vigorous antioxidant activity. This emphasizes its usefulness as a healthy dietary component. In conclusion, *Pyracantha coccinea* is promising in cytotoxic and antioxidant applications and has proven uses as a heavy metal biomonitor. However, there are still a lot of uncharted territories for future research. Notably, molecular identification and enhancement of bioactive compounds and broader pharmacological assessments are warranted.

Declarations

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References

Ajasa, A., Bello, M., Ibrahim, A., Ogunwande, I., & Olawore, N. (2004). heavy trace metals and macronutrients status in herbal plants of Nigeria. food chemistry, 85(1), 67-71. https://doi.org/10.1016/j.foodchem.2003.06.004

Akguc, N., Ozyigit, I., Yasar, U., Leblebici, Z., & Yarci, C. (2010). Use of *Pyracantha coccinea* Roem. as a possible biomonitor for the selected heavy metals. International Journal of Environmental Science & Technology, 7(3), 427-434. https://doi.org/10.1007/BF03326152

Apak, R., Güclü, K., Özyürek, M., & Celik, S. E. (2008). Mechanism of antioxidant capacity assays and the CUPRAC (cupric ion reducing antioxidant capacity) assay. Microchimica Acta, 160(4), 413-419. https://doi.org/10.1007/s00604-007-0777-0

Brand-Williams B, Cuvelier M, Berset C 1995. Methods used to evaluate the free radical scavenging activity in foods and biological systems. Food Sci Technol Int 28: 25-30. https://doi.org/10.1016/S0023-6438(95)80008-5

Corlett JL, Clegg MS, Keen CL, et al. (2002) Mineral content of culinary and medicinal plants cultivated by Hmong refugees living in Sacramento, California. Int J Food Sci Nutr 53:117–128. https://doi.org/10.1080/09637480220132139

Davis, P. Flora of Turkey, Volume 4, Edinburgh: Edinburgh University Press, (1984). https://doi.org/10.1515/9781474466097

Everette, J.D., Bryant, Q.M., Green, A.M., Abbey, Y.A., Wangila, G.W. & Walker, R.B. (2010). A thorough study of reactivity of various compound classes towards the Folin-Ciocalteu reagent. Journal of Agricultural and Food Chemistry, pp. 58, 8139–8144. https://doi.org/10.1021/jf1005935

Fico, G., Bilia, A.R., Morelli, I., Tome F. (2000). Flavonoid distribution in *Pyracantha coccinea* plants at different growth phases. Biochemical Systematics and Ecology, 28, 673–678. https://doi.org/10.1016/S0305-1978(99)00109-X

Fridlender, M., Kapulnik, Y., & Koltai, H. (2015). Plant-derived substances with anti-cancer activity: from folklore to practice. Frontiers in plant science, 6, 799.https://doi.org/10.3389/fpls.2015.00799

Ghandilyan, A., Barboza,L., Tisne, S., Granier, C., Reymond, M., Koornneef, M., Schat, H., Aarts, M. G. M. (2009). Genetic analysis identifies quantitative trait loci controlling rosette mineral concentrations in Arabidopsis thaliana under drought. *New Phytologist*, 184(1),180-192. https://doi.org/10.1111/j.1469-8137.2009.02953.x

Görmez, G., Battal, A., Dalar, A., & Türker, M. (2022). The Investıgatıon of The Medıcınal Potentıal of Alcea Kurdıca Alef. In Nature and Tıssue Culture. Farmacia, 70(6). https://doi.org/10.31925/farmacia.2022.6.23

Kabata-Pendias A, Pendias H (2010). Trace elements in soils. 4th Ed. Boca Raton, London, New York, CRC Press pp. 1–548. eBook ISBN9780429192036. https://doi.org/10.1201/b10158

Kambur, S., Tilki, F. (2010). *Pyracantha coccinea* Roem. tohumunun çimlenme özelliklerinin belirlenmesi. III. Ulusal Karadeniz Ormancılık Kongresi 20-22 Mayıs 2010 Cilt: II Sayfa: 785-791. https://doi.org/10.1080/14786419.2014.942304

Kerasioti, E., Apostolou, A., Kafantaris, I., Chronis, K., Kokka, E., Dimitriadou, C.& Stagos, D. (2019). Polyphenolic composition of *Rosa canina*, *Rosa sempervivens,* and *Pyracantha coccinea* extracts and assessment of their antioxidant activity in human endothelial cells. Antioxidants, 8(4), 92. https://doi.org/10.3390/antiox8040092

Keser, S. (2014). Antiradical activities and phytochemical compounds of firethorn (*Pyracantha coccinea)* fruit extracts. Natural Product Research, 28(20), 1789-1794. https://doi.org/10.1080/14786419.2014.942304

Liu, H., Tang, J., Chen, T., Zhu, P., Sun, D., Wang, W. (2023). Assessment of heavy metals contamination and human health risk assessment of the commonly consumed medicinal herbs in China. Environmental Science and Pollution Research, 30(5), 7345-7357. https://doi.org/10.1007/s11356-022-22647-z

Okatch, H., Ngwenya, B., Raletamo, K. M., & Andrae-Marobela, K. (2012). Determination of potentially toxic heavy metals in traditionally used medicinal plants for HIV/AIDS opportunistic infections in Ngamiland District in Northern Botswana. Analytica chimica acta, 730, 42-48. https://doi.org/10.1016/j.aca.2011.11.067

Osman, D., Cooke, A., Young, T., Deery, E., Robinson, N., & Warren, M. (2021). the requirement for cobalt in vitamin b12: a paradigm for protein metalation. Biochimica et biophysica acta (BBA) - molecular cell research, 1868(1), 118896. https://doi.org/10.1016/j.bbamcr.2020.118896

Özay, C., & Pehlivan, E. (2024). Bitki Sekonder Metabolitlerinin Biyosentezini Ve Akümülasyonunu Etkileyen Faktörler. Journal of Faculty of Pharmacy of Ankara University, 48(3), 1248-1263. https://doi.org/10.33483/jfpau.1488042

Peralta-Videa, J. R., Lopez, M. L., Narayan, M., Saupe, G., & Gardea-Torresdey, J. (2009). The biochemistry of environmental heavy metal uptake by plants: implications for the food chain. The international journal of biochemistry & cell biology, 41(8-9), 1665-1677. https://doi.org/10.1016/j.biocel.2009.03.005

Queralt, I., Ovejero, M., Carvalho, M. L., Marques, A. F., & Llabrés, J. M. (2005). Quantitative determination of essential and trace element content of medicinal plants and their infusions by XRF and ICP techniques. X-Ray Spectrometry: An International Journal, 34(3), 213-217. https://doi.org/10.1002/xrs.795

Re R, Pellegrini N, Proteggente A, Pannala A, Yang M, Rice-Evans C. 1999. Antioxidant activity applying an improved ABTS radical cation decolorization assay. Free Radic Biol Med. 26(9- 10): 1231-7. https://doi.org/10.1016/S0891-5849(98)00315-3

Rodriguez-Fragoso, L., Reyes-Esparza, J., Burchiel, S. W., Herrera-Ruiz, D., & Torres, E. (2008). Risks and benefits of commonly used herbal medicines in Mexico. Toxicology and applied pharmacology, 227(1), 125-135. https://doi.org/10.1016/j.taap.2007.10.005

Semerci, A., Tunç, K., & Okur, İ. (2020). Antioxidant activity of the fruits of *Pyracantha coccinea* using ethanolic extract method. , 6, 35-40. https://doi.org/10.3153/fh20005.

Sharifi-Rad, J., Taheri, Y., Ayatollahi, S. A., Naderi, N., Kumar, N. V. A., Koirala, N., ... & Martins, N. (2020). Biological activities and health-promoting effects of Pyracantha genus: a key approach to the phytochemical's potential. Cellular and Molecular Biology, 66(4), 20-27. https://doi.org/10.14715/cmb/2020.66.4.4

Silva C.S, Moutinho C., Ferreira da Vinha A., Matos C. (2019). Trace minerals in human health: iron, zinc, copper, manganese, and fluorine. International Journal of Scientific Research Methodology, 13:57–80. http://hdl.handle.net/10284/8105

Song, S., Li, J., Liu, H., Qi, Y., Subbiah, V., Sharifi-Rad, J., ... & Suleria, H. A. (2023). Pyracantha as a promising functional food: A comprehensive review on bioactive characteristics, pharmacological activity, and industrial applications. *Food Frontiers*, *4*(4), 1720-1736. https://doi.org/10.1002/fft2.300

Stanojkovic-Sebic, A., Pivic, R., Josic, D., Dinic, Z., & Stanojkovic, A. (2015). Heavy metals content in selected medicinal plants commonly used as. Journal of Agricultural Sciences, 21(3), 317-325. https://doi.org/10.1501/Tarimbil_0000001334

Teiz, L., & Zeiger, E.(2010). Plant Physiology (5th ed., Chapter 5: Mineral Nutrition). Sunderland, Massachusetts, USA: Sinauer Associates Inc., Publishers.285-297.

Tüysüz, B., Çakır, Ö., & Dertli, E. (2020). Bazı yabani meyve türlerinin antioksidan kapasitesi, toplam fenolik madde içeriği ve fenolik asit profilinin belirlenmesi. *Avrupa Bilim ve Teknoloji Dergisi*, (21), 191-197. https://doi.org/10.31590/ejosat.818925

Ulmer, R., Couty, A., Eslin, P., Gabola, F., & Chabrerie, O. (2020). The firethorn (*Pyracantha coccinea*) is a promising dead-end trap plant for the biological control of the spotted-wing Drosophila (Drosophila suzukii). Biological Control, 150, 104345. https://doi.org/10.1016/j.biocontrol.2020.104345

Vahabi, L., Monajemi, R., & Hosseini, S. A. (2015). The cytotoxic effect of methanolic extract of *Pyracantha coccinea* M. Roemer fruit on Hela cell line, antioxidant capacities, and total phenol contents of methanolic and aquatic extract of this fruit. Biomedical and Pharmacology Journal, 8(March Spl Edition), pp. 99-103. http://dx.doi.org/10.13005/bpj/564

Vogt, A.C., Arsiwala, T., Mohsen, M., V ogel, M., Manolova, V., Bachman, M.F. (2021). On iron metabolism and its regulation. International Journal of Molecular Sciences, 22 (9), 4591. https://doi.org/10.3390/ijms22094591

Zhang, J., Guan, Y., Lin, Q., Wang, Y., Wu, B., Liu, X., Wang, B., Xia, D. (2022). Spatiotemporal differences and ecological risk assessment of the heavy metal pollution of roadside plant leaves in Baoji City, China, *Sustainability*, 14(10),5809. https://doi.org/10.3390/su14105809