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Investigation of Heavy Metal Pollution in Some Non-Edible Wild Plant Species Around the Old Chromium Mine Site in Köyceğiz District of Muğla Province

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Abstract: In this study, the possible presence of heavy metals in the soil and some non-edible wild plant species around the old Cr deposits in the Koycegiz-Dalyan Special Environmental Protection Area (Sandras Mountain) was investigated. Possible anthropogenic risk factors to which wild plant biodiversity is exposed are also the subject of this study. The Iron (Fe), Cadmium (Cd), Cobalt (Co), Chromium (Cr), Nickel (Ni) and Lead (Pb) contents of soil, shoots and roots of five non-edible wild plant species [*Alyssum masmenaeum* (AM), *Cytisopsis pseudocytisus* (CP), *Centaurea ensiformis* (CE), *Fumana aciphylla* (FA) and *Phlomis angustissima* (PA)] growing naturally in the region were investigated and it was determined that heavy metals except Ni, Cr and Co were within normal limits. Especially the Ni content of the soil was extremely high (1715-2338 mg kg^{-1}). Heavy metals except Ni, Cr and Fe were within normal limits in shoot and root samples. For Ni, one species (AM) (max. Ni content: 18267 mg kg^{-1}) and for Fe, three species (FA, PA and CE) showed different levels of hyperaccumulators with max. Fe contents: 10096, 8564 and 7007 mg kg^{-1}, respectively. The Ni hyperaccumulation detected in *Alyssum masmenaeum* (AM) species is a particularly striking but well-known phenomenon. Since the research area has a special environmental protection status, the soil structure and endemism of the region must be protected.

Keywords: Heavy metal, hyperaccumulation, wild plant species.

Muğla İli Köyceğiz İlçesinde Eski Krom Madeni Sahası Çevresindeki Bazı Yenilemeyen Yabani Bitki Türlerinde Ağır Metal Kirliliğinin Araştırılması

Öz: Bu çalışmada Köyceğiz-Dalyan Özel Çevre Koruma Bölgesi (Sandras Dağı) eski Cr yatakları çevresindeki toprakta ve yenilemeyen bazı yabani bitki türlerinde ağır metallerin olası varlığı araştırılmıştır. Yabani bitki biyoçeşitliliğinin maruz kaldığı olası antropojenik risk faktörleri de bu araştırmanın konusunu oluşturmaktadır. Bölgede doğal olarak yetişen beş yenilemeyen yabani bitki türünün [*Alyssum masmenaeum* (AM), *Cytisopsis pseudocytisus* (CP), *Centaurea ensiformis* (CE), *Fumana aciphylla* (FA) ve *Phlomis angustissima* (PA)] toprak, sürgün ve köklerinde Demir (Fe), Kadmiyum (Cd), Kobalt (Co), Krom (Cr), Nikel (Ni) ve Kurşun (Pb) içerikleri araştırılmış ve Ni, Cr ve Co dışındaki ağır metallerin normal sınırlar içerisinde bulunduğu belirlenmiştir. Özellikle toprağın Ni içeriği son derece yüksektir (1715-2338 mg kg^{-1}). Sürgün ve kök örneklerinde Ni, Cr ve Fe dışındaki ağır metaller normal sınırlarda bulunmuştur. Ni için bir tür (AM) max. Ni kapsamı:18267 mg kg^{-1}ve Fe için üç tür (FA, PA ve CE) sırasıyla max. Fe kapsamı: 10096, 8564 ve 7007 mg kg^{-1} farklı seviyelerde hiperakümülatör özellik göstermiştir. *Alyssum masmenaeum* (AM) türlerinde tespit edilen Ni hiperakümülüyonu özellikle çarpıcı ama iyi bilinen bir olgudur. Araştırma alanı özel çevre koruma statüsüne sahip olduğundan bölgenin toprak yapısının ve endemizminin mutlaka korunması gerekmektedir.

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Anahtar kelimeler: Ağır metal, hiperakümülasyon, yabani bitki türleri.

INTRODUCTION

Turkey possesses a wide variety of vegetation types and a very rich floristic structure due to its geographical location, geomorphological structure, diverse soil types and the influence of different climatic characteristics. Muğla City plays an important role in this diversity. The Köyceğiz-Dalyan Special Environmental Protection Area is a protected natural reserve in the Turkish province of Muğla. In June 1988 it was determined and declared the first protected area of its kind of Turkey. Sandras Mountain is rich in habitat types and this is reflected in the number of plant species and the endemism rate. A large part of Sandras Mountain and its near surroundings consists of serpentine rocks (Minerals in this group, which are rich in magnesium and Silisium). Regions with serpentine bedrock are not very suitable for plant growth. However, certain wild plant species (mostly endemic taxa) demonstrate very good growth and adaptation in such areas. Ultramafic rocks are common rocks that generally contain intense Fe and Mn elements. Elements such as Ni, Co and Cr are detected quite frequently in soils consisting of these rocks. The mineral groups that form the rocks are composed of hydrous Mg silicate minerals known as serpentines, and these soils are called serpentine soils (Reeves and Adıgüzel, 2008). The fact that Sandras Mountain possesses different vegetation series, ranges from 80 m to 2294 m in height and consists of mostly serpentine bedrock leads to high plant diversity, which also affects the endemism rate. As a result of the literature review performed, it was determined that there is a close relationship between endemism rate and rock structure. Our study area is a tourism region and there is no significant industrial activity. However, there are old chrome deposits in the Sandras mountain region and there is also olivine mineral processing. Olivine mineral is 95–99 % pure and the remaining 1-5 % is composed of pyroxene, serpentine, chlorite and spinel. They are mainly composed of Mg^{+2} and Fe^{+2} silicates and are in the ortho-silicate group. It contains low amounts of Cr and Ni in its chemical composition.

Most of the plants growing wild in nature are edible and are consumed intensively by local people. The level of exposure of these plants to toxic heavy metals should be considered as an important factor in this respect. In a study conducted on this subject, Cu, Cr, Cd, Co and Pb contents of *Malva sylvestris* (mallow), *Falcaria vulgaris* (sickle grass) and *Chenopodium botrys* (goose foot) edible wild plants growing in Van province were determined. It was reported that Cu content (mg kg^{-1}) was determined in the range of 24.67-59.24, Cr (0.10-1.22), Cd (0.18-0.22), Co (0.77-1.41) and Pb (0.30-0.44 mg kg^{-1}) (Tunçtürk et al., 2018).

Many plant species are collected from nature throughout Muğla province and some of them are used as vegetables. These naturally grown plants are offered for sale as raw vegetables by the local people in the neighborhood markets all year round. In a study conducted in this context, the amounts of Cu, Mn, Zn, Fe, Ni, Pb and Cd of 8 different species consumed as raw vegetables were determined by ICP-OES and it was determined that the amounts of toxic metals (Pb and Cd) were low and below the limits determined by WHO/FAO (2007) (Sarıkürkçü et al., 2010).

The usability of the naturally occurring *Lepidium draba* L. (wildcress) species in phytoremediation method was investigated. Heavy metal amounts in the plant were determined as Fe>Mn>Ni>Co. Ni was found below the toxic limit in plant and soil, Fe was found above the toxic limit in plant and soil, Co was found above the toxic limit in plant and soil and Mn was found above the toxic limit in urban, roadside and suburban locations. The bioaccumulation capacity of *L. draba* species was determined and it was found to have hyperaccumulatory properties (Kılıç et al., 2019).

Another study was conducted in Kahramanmaras province to determine the ecology and heavy metal tolerance limit of *Alyssum pateri* plant, known as hyperaccumulator, in a different growing environment. According to the findings of the study, it was understood that ultramafic geo-ecology may have different characteristics in the same macroclimate but in different geographies. While the total amount of nickel (Ni) in the soil was 7.08 mg kg^{-1} on average, the highest amount of Ni in *Alyssum pateri* plant was found to be 4061 mg kg^{-1}. In the research area, the general hyperaccumulator feature of *Alyssum pateri* plant was determined (Dindaroğlu et al., 2019).

Plants have the ability to accumulate heavy metals in some cases. These plants, known as hyperaccumulators, are not exposed to any toxic effects due to the heavy metals they accumulate. This has given them a unique ability. Various metal ligands play key roles in this mechanism. In this mechanism, heavy metals in the rhizosphere are transported very quickly from the roots to the shoots and stored safely. This is a type of heavy metal compartmentalization. Such plants are used in the remediation of soils contaminated with heavy metals (Sytar et al., 2021). Inclusion of species in the database is based on accepted definitions of trace element hyperaccumulation. In their natural environment, they contain > 100 mg kg^{-1} cadmium, > 300 mg kg^{-1} cobalt or chromium, > 1000 mg kg^{-1} nickel or lead in leaf dry matter (Reeves et al., 2018). Metal accumulation has been widely detected in angiosperms, especially in Brassicaceae family (Kramer, 2010). The phytoremediation potential of some species has been well researched. For example: Alyssum for Ni, Helianthus for Pb, Thlaspi for Cd and Zn. During the phytoremediation process, metal tolerance has developed in many plants due to adaptation in plants grown in soils that have been contaminated for a long time. It is aimed to find various plant species with phytoremediation potential (Sainger et al., 2011).

Our study area, which is a tourism and special environmental protection zone, is a pollution-sensitive area that should be especially protected from anthropogenic and natural sources of pollution. Considering the existence of many wild species, both endemic and non-endemic to the region, the goal of sustainable environment should be prioritized by raising awareness for the protection of local biodiversity. This is the main purpose of this study. None of the 5 species investigated in our study and growing naturally in the region are edible/consumable. In addition; some of the plants growing naturally in the region are known to be medicinal-aromatic and edible. Another aim of this study is to raise awareness about the presence of an old chromium mine in the region and the fact that the soils of the region are ultramafic and rich in heavy metals. Since there are also wild plants of consumable species in the same region, the inclusion of toxic heavy metals, especially Ni and Cr, into the biological cycle through living organisms and the potential risks they pose should also be considered. Another one, is to determine the extent to which the wild plants, some of which are endemic to the region, which are distributed on ultramafic soils in the region of the old mine deposit, reflect the specific elemental character of the soils. However, in the natural field study process, as in this study, important random results can be reached that were not planned beforehand.

MATERIAL AND METHOD

Study area and species studied: Sandras Mountain, located in the southwest of Turkey within the borders of Muğla province, consists of ultramafic, slightly weathered serpentine rocks. Below, is a brief introduction of the studied plants, and Table 1 shows the wild plant species included in this study. *Cytisopsis pseudocytisus* subsp. *reeseana* (Guyot) Lassen is an endemic species from the Legume (Fabaceae) family. The name Pseudocytisus (Cytisus) is derived from the Greek word pseudes, meaning liar. They grow naturally in stony soils with calcareous or serpentine rocks and often in maquis. *Phlomis angustissima* Hub.-Mor. is an endemic species from the Lamiaceae family. It is mainly distributed in the Aegean and Antalya sub-regions. Phlomis is derived from the ancient Greek word phlox, meaning 'flame'; It refers to the ancient use of plant leaves as suppositories. *Alyssum masmenaeum* Boiss. is an endemic and perennial herbaceous plant from the Brassicaceae family and its name comes from the Greek words "a" (absence) and "lyssa" (anger) and is believed to have been used to treat dog bites in ancient times. *Centaurea ensiformis* P.H. Davis is a perennial herbaceous plant from the Astraceae family, endemic to the Eastern Mediterranean and Coastal Aegean regions. *Fumana aciphylla* Boiss. is a shrub-shaped species from the Cistaceae family. It usually grows on limestone soils and sunny areas. The flowers of the plant are yellow.

Table 1. The scientific and local names of the collected species.

Species	Family	Local Names	Code Names
Cytisopsis pseudocytisus	Fabaceae	Yalan kedi tırnağı	CP
Phlomis angustissima	Lamiaceae	İnce çalba	PA
Alyssum masmenaeum	Brassicaceae	Çam kuduz otu	AM
Centaurea ensiformis	Asteraceae	Ece sarı başı	CE
Fumana aciphylla	Cistaceae	Kır güneş otu	FA

Soil samples (from 0-30 cm depth) were collected for each plant species from the locations where the plants were collected in addition to shoot samples from the 5 specified plant taxa. The soil and plant samples collected were subjected to pre-treatment in the laboratory. The soil samples were collected from 5 different locations to represent the field before being mixed, brought to the laboratory, subjected to pre-treatment and made ready for analysis. Lime was determined calcimetrically, organic matter was determined using the method proposed by Kacar (2012) pH and Electrical Conductivity (EC) were determined by a combined pH-EC meter. Total heavy metal content in soil samples, although varying according to the elements, was determined by reading the amount of heavy metals in the solution from the soil sample burned with HF, HNO_3, H_2SO_4 and $HClO_4$ by Atomic Absorption Spectrometry (AAS) (Kacar, 2012).

The plant samples brought to the laboratory were washed with tap water and then rinsed with distilled water. Afterwards, the samples were dried in an oven at 70 °C for 72 hours. The dried plant parts were ground in a mortar and placed in paper bags to be analyzed for analysis. Subsequently, the dried plant samples were ground, and 1 g was taken from these samples before being decomposed in a muffle furnace at 550°C for 6 hours. The ash obtained after the decomposition process was extracted with 2 N of warm HCl and distilled water, and then heavy metal contents of the plants (Fe, Cd, Co, Cr, Ni and Pb) were made in the same samples according to Kacar and Inal, (2008) and determined by the ICP-OES. The results were given as mg kg^{-1} in dry matter.

***Risk factors for the heavy metals investigated*:** Naturally growing wild plants may contain toxic levels of heavy metals depending on the parent material characteristics of the soils in which they grow. As a result of the consumption of these plants by local people, a number of serious health problems develop, and the consumption of foods contaminated with heavy metals can seriously deplete essential nutrients in the body, causing a decrease in immunological defense, growth retardation, psychosocial disorder, malnutrition-related disabilities and high prevalence of gastrointestinal cancer. They have hematotoxic, immunotoxic, neurotoxic, genotoxic, nephrotoxic, hepatotoxic, reproductive and pulmonary toxic effects. Therefore, heavy metal contamination is a global problem that requires regular monitoring of these toxic chemicals in the food chain (Mansour, 2017). The amount of heavy metals accumulated in the food chain is assessed using various factors or indices such as trophic transfer factor, biota-sediment accumulation factor, transfer factor, metal transfer factor, accumulation factor, bioaccumulation factor and bioconcentration factor (Kumar et al., 2019).

Increasing the risk awareness of heavy metals ingested through the consumption of vegetables consumed as food, including naturally growing plants, is of great importance for food safety. In this context, for nickel, the European Food Safety Authority (EFSA) has updated its scientific advice and dataset on the risks of nickel in food to human health, raising the safe level, known as the tolerable daily intake (TDI), from 2.8 µg kg^{-1} body weight to 13 µg kg^{-1}. The TDI for chromium III, a naturally occurring, essential nutrient and the main form of chromium found in foods, was set at 0.3 µg kg^{-1}. Dietary exposure in all age groups is well below the TDI and therefore not of public health concern. The current average dietary cadmium exposure for adults is 2.5 µg kg^{-1}, which is the tolerable weekly intake (TWI) for cadmium. Lead concentrations in food are typically between 10-200 mg kg^{-1}. The average daily intake of lead in adults in Europe is 0.36-1.24 mg kg^{-1} body weight. Absorption of inorganic lead compounds after exposure depends on a number of factors such as age, nutritional status, diet composition, iron and calcium status (EFSA, 2021).

RESULTS AND DISCUSSION

Heavy metal content and some properties of the soils: The EC (dS m^{-1}), pH, $CaCO_3$ (%) and organic matter (%) contents of the soils and heavy metal content where the species grow are given in Table 2 and Table 3. Considering some general soil properties of the localities, EC (0.30-0.61), pH (5.46-6.08), $CaCO_3$ (0.08-0.23 %) and organic matter contents were determined in the range (1.55-5.12 %). It is seen that the pH values of the soils where wild plant species grow are in moderate acid reaction. In particular, the solubility or mobility of heavy metals increases in acidic soil environments. The EC (total soluble salt concentration) and $CaCO_3$ % contents of the soil samples were also found at very low levels. In addition, organic matter contents were found to be moderate except for one sample (Table 2).

Parolly, (2021) in his study investigating the Serpentine vegetation of Mount Sandras reported that in higher elevations of Sandras Mountain, peridotites prevail, which contain ortho-pyroxenes and magnesian olivines. They develop into acidic soils (pH values 4.5-5.5) with very low percentages of $CaCO_3$ (1-2 %). These results, as we have determined, are an indication that wild species thrive in acid conditions. In the study conducted by Allison and Moodie, (1965) on $CaCO_3$ % content in soils the proposed range of values was reported as 5-15 %. Based on this range of values it was determined that all of the soils examined had very low lime content. In addition, except for the soil sample where *Phlomis angustissima* and *Fumana aciphylla* species spread, organic matter % was detected at low-moderate level in other soil samples. Altınözlü et al., (2012) reported that the soils where the wild plant species spread on serpentine soils of Turkey, contain organic matter in between of 0.62-0.90 %. A few soils containing high levels of organic matter were of woodland origin. The same researchers determined the EC values of the soils as 0.07-0.42 dS m^{-1}. These results are in agreement with our data. Besides; In a study investigating in the areas where polluted due to anthropogenic activities in Southern Italy; soil electrical conductivity (EC) values were found between as 0.20-0.70 dS m^{-1} in the eight soil samples (Brunetti et al., 2009). In another; in a study investigating wild plants growing in mafic and ultramafic terrain in Northern Pakistan, the average EC values of soil from 3 different regions with ultramafic character were reported as 0.157-0.233 dS m^{-1} (Muhammad et al., 2013). Compared with these researches it was revealed through analyses that the localities investigated were within the 100 % salt-free class and that there was no problem in relation to salinity (Table 2).

Table 2. Some analysis parameters of the soil where the species were collected.

Parameters	CP	PA	AM	CE	FA
EC (dS m^{-1})	0.57	**0.61**	**0.30**	0.46	0.44
pH	5.90	**6.08**	5.80	**5.46**	5.70
$CaCO_3$ (%)	0.19	0.17	**0.23**	0.08	**0.08**
Organic matter (%)	**1.55**	**5.12**	2.93	2.63	4.43

In addition; Table 3 shows the total heavy metal content of the samples collected from the soils where the plant species naturally spread. Micro elements Fe investigated in this study are also plant nutrients and have important roles in plant metabolism. According to Güneş et al., (2000) the total Fe content of the soils was 0.02-10.0 % (200-100 000 mg kg^{-1}), while Yağmur and Okur, (2011) reported that the total Fe content of the soils varied between 0.5-5.0 %. In this study, according to these reference values, Fe contents of the soils are at low levels. However, no signs of Fe deficiency were observed in the collected wild plant species.

Cadmium (Cd) is nonessential to plant nutrition but it is of significant concern because may be taken up by plants in sufficient amounts to be potentially harmful to humans and livestock that eat them. As shown in Table 3, the Cd elements obtained in the soil samples collected around the Sandras chromium mineral deposit were found to be between 0.93-1.52 mg kg^{-1}. When the results are compared with these limit values, it is seen that the average Cd value is 1.27 mg kg^{-1} in a total of 5 soil samples. This shows that there is no pollution caused by cadmium. Compared to the target values set by FAO, there is no pollution problem in terms of Cd element in the soils of the study area (Table 4). Junior et al., (2009) indicated that, the impact of Candiota coal-fired thermal power plant was evaluated during two years by means of passive biomonitoring and collection of leaf samples of *B. dracunculifolia, E. mollis, E. horridum, P. notatum, P. montevidense* and *B. trimera*. The Cd content in the species used for passive biomonitoring were between < 0.010 to 1.74 mg kg^{-1}, with a mean value of 0.171 mg kg^{-1}. In the other study conducted by Çiçek and Koparal, (2004) around the Tunçbilek thermal power plant it was found that the Cd values in the soils varied between 1.4 and 21.7 mg kg^{-1}.

Table 3. Total heavy metal contents of soil samples (mg kg^{-1}).

Heavy Metals	CP	PA	AM	CE	FA
Iron (Fe)	42	**89**	**36**	65	38
Cadmium (Cd)	1.3	1.2	1.4	**1.52**	**0.93**
Cobalt (Co)	93	73	118	**125**	**69**
Chromium (Cr)	500	**373**	587	**722**	548
Nickel (Ni)	**2338**	1956	2318	2235	**1715**
Lead (Pb)	**16**	16	14	14	**12**

CP: *Cytisopsis pseudocytisus,* **PA:** *Phlomis angustissima,* **AM:** *Alyssum masmenaeum,* **CE:** *Centaurea ensiformis,* **FA:** *Fumana aciphylla The min. and max. values of each metal are shown in bold.*

As seen in Table 3, the Co contents of the soils where 5 species grow are min. and max. it was found in the range of 69-125 mg kg^{-1}. The Co content of all the soil samples in the present study are above the specified limit range of 50-60 mg kg^{-1} and this indicates that the soils are at a mild pollution threshold in terms of Co. Alloway, (1990) stated that the normal Co limit value in soils is between 0.5-65 mg kg^{-1}. In addition to these reference values, when compared with the criteria determined by FAO, it was determined that the Co contents of the soil samples were within normal limits (Table 4).

Table 4. Maximum limits of heavy metals in soil (mg kg^{-1}) (FAO, 2004).

Heavy Metals	Target value	Ref. range in residental zone	Ref. range in industrial zone
Cadmium (Cd)	1	5	20
Cobalt (Co)	20	50	300
Chromium (Cr)	100	300	800
Nickel (Ni)	50	150	500
Lead (Pb)	50	300	600

In literature reviews, around *Pinus brutia* forest soils of Muğla province, the content of Fe: 1.3-2.5 %, Mn: 338-659 mg kg^{-1}, Cd: 0.49-1.60 mg kg^{-1}, Co: 15.1-31.7 mg kg^{-1}, Cr: 33-92.7 mg kg^{-1}, Ni: 45.6-78.3 mg kg^{-1} and as Pb: 24.5-58.2 mg kg^{-1} were reported (Tuna et al., 2005). Furthermore, Kılıç and Ortakcı, (2021) reported that average Co accumulation in species were found as *E. uncinatifolium*: 279 mg kg^{-1}, *G. acutidentatum*: 148 mg kg^{-1}, *A. pateri*: 129 mg kg^{-1} and *A. leptophlla*: 94 mg kg^{-1}.

The lowest and highest values in terms of Cr were determined as 373-722 mg kg^{-1}. The average Cr content of total 5 soil samples was determined as 546 mg kg^{-1}. The high Cr content of the soil is thought to be the result of the pollution originating from the chromium mineral deposit as the main source. The surrounding of Sandras Mountain, where the research was carried out, contains old Cr mineral deposits. It is also known that there are Olivine mineral deposits in the region. The Cr content was found to be high because the soils of the study area are ultramafic-serpentinic and spread in an old chromium mining area. The average Cr content of 546 mg kg^{-1} reflects the area affected by high or industrial pollution according to the criteria determined by FAO (Table 4).

In the study conducted by Uminska, (1988) around a Cr smelting plant in Poland, it was reported that the Cr concentrations exceeded 10.000 mg kg^{-1}. Similarly, it was reported that the Cr concentrations in wastewater-contaminated soils ranged from 214 to 727 mg kg^{-1} (Beckett et al., 1979). The literature review supports the findings of the present study.

The total Ni content of the soils included in the present study was determined as 1715 mg kg^{-1} at the lowest (in the soil where *Fumana aciphylla* grows) and 2338 mg kg^{-1} at the highest (*Cytisopsis pseudocytisus* grows). Also, the average Ni content was determined as 2112 mg kg^{-1}. Nickel element in the soil may be of parent material or anthropogenic origin. It is usually found around 50 mg kg^{-1}. In ultramafic serpentine origin soils, it may exceed 10.000 mg kg^{-1}. Rocks containing olivine, pyroxene, biotite and chlorite minerals are rich in Ni. Kabata-Pendias and Pendias, (1992) reported 100 mg kg^{-1}, as the limit values for total Ni in soils to have an effect on toxicity.

The high Ni contents detected in our study are specific to this region due to the main material of soil structure of the region. According to FAO classification, 500 mg kg^{-1} is the upper limit for Ni pollution in industrial areas (Table 4). In our study, values approximately 5 times higher than this upper limit were detected (Table 3). Nickel content in soils varied widely and have been estimated to range from 3 to 1000 mg kg^{-1}; for the world soils, the brand range is between 0.2 and 450 mg kg^{-1}, while the grand mean is calculated to be 22 mg kg^{-1} (Kabata-Pendias and Pendias, 2000; Cempel and Nikel, 2006). Duke (1980) also reported an average concentration of 86 mg kg^{-1} for the natural nickel content in the earth‟s crust (Iyaka, 2011). As can be understood from these literature data, the soil nickel contents determined in our study (1715-2338 mg kg^{-1}) are quite high (Table 3).

In the studies carried out to determine the heavy metal pollution of agricultural soils in our country, 50 mg kg^{-1} was accepted as the permissible limit value of Ni pollution (Yağmur and Okur, 2011). In another study, in the Muğla Sandras Mountain region, where our study was also conducted soil samples were collected from the habitats of serpentine endemic *Rosularia serpentinica* and *Teucrium sandrasicum*, which spread on serpentine substrate in the Sandras Mountain region of Muğla. As a result of the analysis, the total Ni, Cr and Fe contents were determined as Ni: 3045-3872, Cr: 350-494 and Fe: 35.769-51.920 mg kg^{-1}, respectively (Altıoğlu, 2009). Bani et al., (2013) investigated the total trace elements in some Albanian serpentine soils where *Alyssum bertolonii, A. markgrafii, A. murale* and *B. baldaccii* wild species grow, determined the Ni content of the soils in the range of 1658-3077 mg kg^{-1}. Altınözlü et al., (2012) aboveground parts of 413 herbaceous plants and the surface soil (0-15 cm) of 192 serpentine soil samples were collected and reported that the total nickel content of the 3 selected serpentine soils as 2636 mg kg^{-1}.

In our study, the Pb contents found in 5 soil samples varied between 12-16 mg kg^{-1}. According to the reference values given in the literature, there is no Pb pollution in the soil of the study area. Bergmann, (1993) under normal conditions, the Pb content of un-contaminated soils was reported as 1-20 mg kg^{-1}, while Alloway, (1990) reported 2-300 mg kg^{-1}. In addition, when compared with the values determined by FAO, there is no problem in terms of Pb pollution in the study soils (Table 4).

In a study investigating the total heavy metals contents of six wild plant species (*Carduus pycnocephalus, Ferula communis, Sinapis arvensis, Silybum marianum, Dasypyrum villosum, Stipa austroitalica*) in the areas where polluted due to anthropogenic activities in Southern Italy; Cd: 1.41, Cr: 1154, Cu: 280, Ni: 61, Pb: 325 and Zn were determined as 638 mg kg^{-1} (Brunetti et al., 2009). According to the heavy metal analysis results of the forest soils where *Cratoxylum sumatranum*, *Pneumatopteris leavis* and *Cratoxylum sumatranum* species grow, Cd: 0.1-0.47, Co: 0.45-4.9, Cr: 2.04-16.87, Fe: 2248-9414, Ni: 0.37-10.77 and Pb: 0.38-4.3 mg kg^{-1} were determined (Castanares and Lojka, 2020).

Hyperaccumulation characteristics and heavy metal content of the wild plant species: The heavy metal contents in the shoot and root parts of 5 plant species that are naturally spread around the Sandras Mountain chromium mineral deposit in Muğla located around the borders of the Köyceğiz specially protected environment area were examined. Table 5 shows the changes in the heavy metal content of the shoot and roots of the plants included in the present study. Although this study only aims to determine the heavy metal contents of some naturally occurring wild plant species, it is also aimed to reveal the possible hyperaccumulator properties of some examined species, according to the data obtained as a result of the analysis. For this reason, the Fe and Ni hyperaccumulator feature of two species in particular (*Fumana aciphylla* and *Alyssum masmenaeum*) were emphasized, respectively.

Table 5. Heavy metal contents and hyperaccumulator characteristics of the species (mg kg^{-1}).

Heavy metals	Part	CP	PA	AM	CE	FA
Iron	Shoot	244	8564	925	7007	**10096**
	Root	1551	7503	2252	2837	**9105**
Cadmium	Shoot	0.27	nd.	0.02	0.04	nd.
	Root	0.48	nd.	nd.	0.24	0.01
Cobalt	Shoot	31	10	21	8	12
	Root	8	12	3	6	10
Chromium	Shoot	5	36	40	76	97
	Root	9	34	36	41	97
Nickel	Shoot	40	288	**18277**	300	442
	Root	161	407	**2869**	253	407
Lead	Shoot	0.16	3.23	2.78	2.13	2.68
	Root	1.35	1.6	0.16	0.7	1.5

CP: *Cytisopsis pseudocytisus*, **PA:** *Phlomis angustissima*, **AM:** *Alyssum masmenaeum*, **CE:** *Centaurea ensiformis*, **FA:** *Fumana aciphylla*, **nd:** not determined, Bold text indicates hyperaccumlation

All of the plants collected in the study are endemic to the region and are not edible plants consumed as food. However, tolerable levels of heavy metals for edible vegetables have been determined by international health and food organizations. Tolerable levels (mg kg^{-1}) of some heavy metals determined by WHO/FAO (2007) organizations for vegetables are 0.2 for Cd, 0.3 for Pb, 67 for Ni and 425 for Fe (Mensah et al., 2009).

The term "hyperaccumulator" describes a number of plants that belong to distantly related families, but share the ability to grow on metalliferous soils and to accumulate extraordinarily high amounts of heavy metals in the aerial organs, far in excess of the levels found in the majority of species, without suffering phytotoxic effects. Three basic hallmarks distinguish hyperaccumulators from related non-hyperaccumulating taxa: a strongly enhanced rate of heavy metal uptake, a faster root-to-shoot translocation and a greater ability to detoxify and sequester heavy metals in leaves (Rascio and Navari-Izzo, 2011).

Considering the shoot Fe content of the 5 collected wild species, the highest Fe content was found in *Fumana aciphylla* species (10.096 mg kg^{-1}) and followed by *Phlomis angustissima* (8564 mg kg^{-1}) and *Centaurea ensiformis* (7007 mg kg^{-1}). *Fumana aciphylla* species contains approximately 1 % Fe and with this feature, it can be classified as Fe hyperaccumulator. According to Reuter and Robinson, (1986) the Fe element should be in the range of 100 to 300 mg kg^{-1} in the dry matter under normal conditions. However, this limit value has been determined for cultivated plants. It may differ for wild species. Considering this reference values, it was observed that the Fe content in the shoot and root of all the plants examined was well above the recommended values (Table 5).

Schettini et al., (2018) reported that 90.6-983.4 mg kg^{-1} Fe were found in 27 wild species collected from the ferruginous rocky outcrops land containing Fe parent material. The highest Fe content was found in *A. albicans* from the Asteraceae family. Muhammad et al., (2013) investigated the 16 wild plant species for their phytoremediation potential of macro and trace metals. According to the data obtained, the lowest average Fe content was found in *Dodonaea viscosa* species (365 mg kg^{-1}), while the highest Fe content was found in *Selaginella jacquemontii* species (4118 mg kg^{-1}). Looking at the recent studies, literature reports indicate that some wild plant species can contain Fe at the hyperaccumulatory limit. These reports are also consistent with our findings.

The Cd contents of the samples belonging to the species collected in our study are not within remarkable limits. Kabata-Pendias and Pendias, (1992) reported that plants naturally contain between 0.1-1 mg kg^{-1} of Cd. In five species, the Cd coverage is below the detection limit of the ICP-AES device. The highest detectable Cd content is 0.27 mg kg^{-1} in shoot and 0.48 mg kg^{-1} in root in *Cytisopsis pseudocytisus* species (Table 5).

Co contents of 5 species collected in this study showed moderate elevation. As can be seen from Table 5, the Co content of 5 species were shoot: 8-31 mg kg^{-1} and root: 3-12 mg kg^{-1}. Sauerbeek, (1982) reported that more than 20 mg kg^{-1} of Co in plants can have a toxic effect. According to the findings obtained in this study, it can be said that the Co contents in different parts of the plants included in this study are quite high compared to the sample findings in the literature. A study was conducted on *Carduus pycnocephalus, Ferula communis, Sinapis arvensis, Silybum marianum* and *Dasypyrum villusum* species grown on contaminated soils in Southern Italy. Cd contents of the soils were determined as 0.95-1.57 mg kg^{-1}, Cr contents as 459-1770 mg kg^{-1}, Ni contents as 36-89 mg kg^{-1} and Pb contents as 137-891 mg kg^{-1}. The shoot and root Cd, Cr, Ni and Pb contents of the collected wild plant species were also determined. According to the results obtained; shoot and root Cd content 0.04-0.71 and 0.18-0.43 mg kg^{-1}, shoot and root Cr content 1.7-5.7 and 3.8-18.7 mg kg^{-1}, shoot and root Ni content 0.8-5.3 and 0.9-3.7 mg kg^{-1} and shoot and root Pb content 0.5-1.3 and 0.7-7.1 mg kg^{-1}.

Based on the results obtained in the present study, the highest Cr content was observed in both the shoot (97 mg kg^{-1}) and root part of *Fumana aciphylla* (97 mg kg^{-1}) while the lowest Cr content was found in both the shoot and root part of *Cytisopsis pseudocytisus* (Table 5). Naturally occurring in soil, Cr ranges from 10 to 50 mg kg^{-1} depending on the parental material. In a study conducted by Rashed, (2010) in wild plants collected around a gold mineral deposit, chromium content in the structure of the plants collected from locations near the mineral deposit was reported as range of 10.1-18.1 µg kg^{-1}. Cr content in plants can vary widely, depending on the nature of the parent material and industrial activities. According to Castanares and Lojka, (2020) heavy metal contents (as mg kg^{-1}) in leaves of *Cratoxylum sumatranum* grown in forest soils; Cd: 0.12, Co: 0.45, Cr: 3.58, Fe: 608, Ni: 19, Pb: 1.77, in leaves of *Pneumatopteris leavis*; Cd: 0.13, Co: 2.65, Cr: 31.6, Fe: 447, Ni: 8.22, Pb: 1.61 and in *Cratoxylum sumatranum* leaves; Cd: 0.52, Co: 0.45, Cr: 2.3, Fe: 282, Ni: 57 Pb: 0.92 mg kg^{-1} were determined. Moreover, Lorestani et al., (2011) reported average shoot and root Pb content of 7 wild species were 945 and 1233 mg kg^{-1}, respectively in industrial town of the Hamedan province. Shoot and root Pb contents of 5 wild plant species collected in our study, found 0.16-3.23 mg kg^{-1} and 0.16-1.6 mg kg^{-1}, respectively. Kabata Pendias and Pendias, (1992) reported that plants naturally contain between 0.1-10 mg kg^{-1} of Pb. Accordingly, it can be said that the Pb contents of the species collected in our study were within acceptable limits. Adıgüzel and Reeves, (2012) reported that Muğla-Köyceğiz Sandras Mountain, where this research was conducted, is a soil of serpentine origin at an altitude of 1100-1750 m. According to authors, the serpentine flora is rich in species and in local endemics. In temperate climates, including the Mediterranean Region and Turkey, the families Brassicaceae, Asteraceae and the Caryophyllaceae are well represented in the serpentine floras. Alyssum is the most characteristic genus of this family and often shows Ni-hyperaccumlator features.

According to Scheffer and Schachtschabel, (1989) 0.1-3 mg kg^{-1} Ni is considered normal in plants. Alloway, (1990) reported the normal Ni value in the range of 0.02-5.00 mg kg^{-1}.

The Ni content of the 5 species collected in our study was found to be between 40-18.277 mg kg^{-1} in the shoot parts and 161-2869 mg kg^{-1} in the roots. It has been determined that wild plant species contain Ni in very wide ranges. According to the findings, the highest Ni content was determined in the shoot and root parts of the *Alyssum masmenaeum* plant (18.277 and 2869 mg kg^{-1}) (Table 5). In the field study conducted by Reeves and Adıgüzel, (2008) on Turkish serpentines in the period 1996-2003, Nickel content in *Alyssum masmenaeum* Boiss., which is an endemic species of Turkey and serpentine, was reported as 1640-24.360 mg kg^{-1}. The researchers' data appear to be consistent with our findings. Altınözlü et al., (2012) determined Ni: 13.778 mg kg^{-1} in *Alyssum caricum* and 12.273 mg kg^{-1} in *Thlaspi oxyceras*, which they collected from the serpentine soils of Muğla Köyceğiz region. Researchers also collected; *A. sibiricum, A. pateri, A. tortuosum, A. murale, A. peltarioides. A. peltarioides* species belonging to the genus Alyssum throughout Turkey and reported 431-9505 mg kg^{-1} Ni content. In this comprehensive study conducted by the researchers, *Alyssum masmenaeum*, which we define as a Ni hyperaccumulator, was not included. Ni hyperaccumulation feature of Alyssum species is a known phenomenon. Similarly, in our study, *Alyssum masmanaeum* was the species with the highest Ni content among the 5 species. In another study, Ni, Cr, Mn and Fe contents were investigated in the aboveground parts of serpentine endemic *Rosularia serpentinica* and *Teucrium sandrasicum* spreading on serpentine substrate in Muğla Sandras Mountain region. According to the analysis data, Ni: 26-137, Cr: 6-55, Mn: 39-125 and Fe: 355-1856 mg kg^{-1} were found (Altıoğlu, 2009). The values found are consistent with our study in terms of Cr and Mn. However, Ni and Fe contents are quite high in our study.

CONCLUSION

In conclusion, although some heavy metals were found above the limit values and hyperaccumulator properties, no physical evidence was found that the plants in the study were adversely affected metabolically by these high values. This can be considered as a potential indicator that these plants, which are widely found in nature, have genetically high antioxidant capacity. In addition, old or new mining sites and the nature of the bedrock can also cause serious heavy metal pollution. It was determined that there was an increase in some metal concentrations in the soil and vegetation near the mine deposit, and that it was especially important to take the necessary protective measures, considering that the region is a Special Environmental Protection Area.

In addition, although wild plants that are not consumed by the local people were investigated in this study, it should not be ignored that the edible wild plants that grow naturally in the same region and consumed by the local people may be contaminated with extremely high Ni and Cr heavy metals from the soil parent material. In similar studies to be conducted in the future, possible risk factors can be revealed by investigating the heavy metal contents of edible medicinal aromatic plants, especially Ni and Cr, which grow naturally in this region.

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