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Teucrium as a Novel Discovered Hyperaccumulator for the Phytoextraction of Ni-contaminated Soils

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Abstract
The success of phytoextraction, a promising new method that uses green plants to detoxify metals, depends upon the identification of suitable plant species that hyperaccumulate heavy metals in their above ground parts. In this study, the roots and above ground parts of the *Teucrium polium* plants grown in serpentine and non-serpentine soils as well as soil samples were analyzed for their Ni and Co concentrations. The Ni concentrations between 9,678 and 14,110 mg kg^{-1} in above ground parts of *Teucrium polium* plants grown in serpentine soils were found together with the translocation factors between 2.23 and 3.23, and enrichment coefficients between 5.9 and 9.2. The cobalt concentrations in the same samples were found to be in the ranges of 3.1 and 6.4 mg kg^{-1} together with the translocation factors 2.8 and 15, and with enrichment coefficients of 0.01 and 0.03. The Ni/Co-ratios in the ammonium chloride and ammonium acetate extracts of the soils were found to be higher than 4 and 10 fold for the serpentine soils, respectively, while the values were below or about 1-fold for both the extracts of the non-serpentine soils. The ratios of Ni/Co concentrations in the roots and aboveground parts of *Teucrium polium* grown in the serpentine soil were significantly higher (up to 12,857-times) than the ratios for *Teucrium polium* grown in the non-serpentine soils (up to 8.1-times). These values are highly important and the original results. Consequently, *Teucrium polium*, a new hyperaccumulator plant for nickel, has been discovered and suggested for phytoextraction of Ni in contaminated soils.
Keywords: Cobalt, hyperaccumulator, nickel, phytoextraction, *Teucrium polium*.

Ni ile Kirlenmiş Toprakların Phytoekstraksiyonu İçin Yeni Keşfedilen Bir Hiperakümülatör Olarak Teucrium

Özet
Metallerin zehir etkisini gideren yeşil bitkilerin kullanıldığı ve umut verici yeni bir metod olan phytoextraction' nın başarısı üst kısımlarında ağır metalleri aşırı toplayan uygun bitki türlerinin belirlenmesine bağlıdır. Bu çalışmada, toprak örneklerinin yanısıra serpentinik ve serpentinik olmayan topraklarda yetişen *Teucrium polium* bitkilerinin kök ve üst kısımları, Nikel ve Kobalt konsantrasyonları için analiz edildi. Serpentinik topraklarda yetişen *Teucrium polium* bitkisinin üst kısımlarında 9,678 ve 14,110 mg kg^{-1} arasında Ni konsantrasyonları, 2.23 ile 3.23 arasında değişen iletme faktörü ve 5,9 ile 9,2 arasında değişen zenginleşme katsayısı ile birlikte bulundu. Aynı örneklerdeki Co konsantrasyonları 3,1 ve 6,4 mg kg^{-1} aralığında, 2,8 ile 15 lik iletme faktörü ve 0,01 ile 0,03 lük zenginleşme katsayısı ile birlikte bulundu. Serpentinik toprakların amonyum klorür ve amonyum asetat ekstraktlarındaki Ni/Co oranları sırasıyla 4 ve 10 kat tan daha büyük bulunurken, serpentinik olmayan toprakların Ni/Co oranları 1 veya 1 den daha küçük kat civarında bulundu. Serpentinik topraklarda yetişen *Teucrium polium* bitkisinin kök ve üst kısımlarındaki Ni/Co oranları (12,857 e kadar), serpentinik olmayan topraklarda yetişenlerin oranlarından (8,1 e kadar) önemli oranda daha büyüktür. Bu bulgular büyük oranda önemlidir ve orjinal sonuçlardır. Sonuç olarak, nikel için yeni bir hyperaccumulatör bitki olan *Teucrium polium* keşfedildi ve kirlenmiş topraklardaki Nikelin phytoekstraksiyonu için önerildi.
Anahtar Kelimeler: Hiperakümülatör, kobalt, nikel, phytoekstraksiyon, *Teucrium polium*.

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INTRODUCTION

Heavy metal contamination generally originates from anthropogenic activities such as mining, smelting, mineral processing, metalliferrous electroplating, waste disposal, combustion of fossil fuels, agricultural applications of pesticides and biosolids, energy, and fuel production (Oliver 1997, Fairbrother et al. 2007). Removal of excess metal ions from polluted sites can be performed by chemical and conventional remediation methods as well as biological means. Chemical remediation involves the use of chemicals to clean the environment, but this method is not universal, and these techniques are highly costly and may cause

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secondary pollution (Shah and Nongkynrih 2007). The use of hyperaccumulators opens a new branch of phytoremediation technology that is an ecofriendly and scientific approach to remove, extract, or inactivate metal ions in the soil using plants (Fairbrother et al. 2007). The success of hyperaccumulator plants to remediate the soil and water depends on rapid accumulation of the metal in the plants, transfer of the metal from the root to the above ground parts, the rapid growth of the plant, and a large biomass and the high uptake capacity of the plants (Anonymous 2000, Shah and Nongkynrih 2007). Phytoextraction, the most effective strategy of phytoremediation, is the use of pollutant-accumulating plants capable of extracting and translocating pollutants to the harvestable parts (Pulford and Watson 2003). Briefly, this method is not only about 1000-fold cheaper than the conventional remediation methods such as excavation and reburial but also an environmentally friendly technology for contaminated soils (Anonymous 2000, Pulford and Watson 2003).

Trace elements can be accumulated by some plants growing naturally on polluted sites with up to 10–500 times higher levels, compared to the same plants growing on non-polluted soils (Zu et al. 2004, Kaya and Yaman 2008a, Kaya and Yaman 2008b, Kaya et al. 2010a, Kaya et al. 2010b). Although the standard for a hyperaccumulator has still not been defined scientifically, the current criterion defining a hyperaccumulator is; “a plant that can accumulate metal to a concentration that is 100 times greater than “normal” plants growing in the same environment” (Jaffre et al. 1976, Baker et al. 2000, Pilon-Smits and Pilon 2002, Pulford and Watson 2003). The bioavailability of metals for plant uptake and biomass can be increased by modulation in both plant and soil associated factors. Related with plant associated factors, natural-metal accumulating plants release metal chelating compounds to the rhizosphere which increase the bioavailability of metals that are tightly bound to the soil and helps to carry them into plant tissues. Chelating compounds (phytochelators) are usually low molecular weight organic compounds such as malic, malonic, oxalic, acetic and succinic acid, sugars, amino acids, and phenolics that can change the metal speciation and thus metal bioavailability. Related with soil associated factors, soil pH, fertilizers, and chelats in soil are the most important parameters. The complex mechanism of metal phytoextraction can be explained by several steps. These steps involves solubilization of metal from the soil matrix, secretion of $H+$ by roots to acidify the rhizosphere and increase the metal dissolution, secretion of ligands such as organic acids and enzymes into the rhizosphere to enhance metal desorption from the soil, transportation of metal from the root to the shoot by entering the xylem, and detoxification, and sequestration of the metal ion by converting to a less toxic form through chemical conversion, and by bonding with metallothioneins and phytochelatins (Anderson et al. 1999, Boominathan et al. 2004, Sheoran et al. 2009). Some families and genera are particularly well represented for Ni: *Brassicaceae* including *Alyssum* and *Thlaspi*, *Euphorbiaceae*, *Leucocroton*, and *Asteraceae* including *Senecio*, *Pentacalia*. Thus, they offer a sustainable treatment option for metal-contaminated sites (phytoextraction) and an opportunity to mine metal-rich soils (phytomining). The authors reported the methods to use the plants to "phytomine" nickel, cobalt, and other metals by cultivating Ni hyperaccumulator plants on metal-enriched soils and ashing the harvestable biomass to produce Ni ore (bio-ore), an economically viable alternative for metal recovery (Anderson et al. 1999, Reeves and Baker 2000, Boominathan et al. 2004, Sheoran et al. 2009).

In conclusion, in order to identify a plant as a hyperaccumulator, the plant should accumulate 0.1% of the elements such as Ni, Co, or Pb and 1.0% of Zn, Mn, and 0.01% of Cd, based on the dry mass (d.m.). The identification of hyperaccumulators is an imperious and important task as being the key to a successful implementation of phytoremediation. Currently over 400 metal hyperaccumulator species, belonging to 45 different families, have been identified, with the highest occurrence in the *Brassicaceae* family (Reeves and Baker 2000). The majority of these metal hyperaccumulators, at least 317, are nickel hyperaccumulators. These species containing 0.1 to 1.0% nickel in their aerial tissues are mainly found on serpentine soils. Hence, it is necessary to obtain more information about metal accumulation trends of the plants growing on serpentine soils to evaluate their potential for the management of metal polluted soils (phytoremediation) including metal extraction (phytoextraction), especially for the discovery of hyperaccumulators (Freitas et al. 2004). Furthermore, metal accumulators and metal tolerant plants are of considerable significance for biogeochemical prospecting of minerals.

In this study, a hyperaccumulator plant was discovered for nickel, namely Teucrium (*Teucrium polium* L.) growing in the serpentine soils around the abandoned copper mining area in Maden, Elazığ, Turkey. To confirm these results, the Ni and Co concentrations in the HNO_3/H_2O_2 digests, in the extracts of ammonium chloride and ammonium acetate of the soils, in the roots, and in the above ground parts of plants was determined. Furthermore, the Ni/Co-ratios for roots grown in the serpentine soils and in non-serpentine soils was also established. Similarly, the Ni/Co-ratios in aboveground parts of *Teucrium polium* L. plants grown in both serpentine and non-serpentine soils was calculated. In addition, cobalt concentrations in both soil and plant parts were determined and compared with the nickel concentrations.

MATERIAL AND METHODS

Geochemistry of the Study Site

The study area, Maden a town between Elazığ and Diyarbakir is situated between longitudes 39-40◦ east and latitudes 38–39° north. The town is 1054 m above sea level, and located on the slope of Mount Mihrap (1773 m), which is one of the main highest mountains of the county. The dominant climate in this region is the terrestrial climate and the winter seasons are cold and precipitant (low snow and rain) while summer seasons are hot and dry. The investigated region, a massive sulfide deposit, is located in the Ergani-Maden District of southeastern Turkey and has been a major source of copper for more than 4,000 years. The mineralization is hosted by strongly chloritized serpentinite, gabbro, diabase, and mudstone. The ore body mainly consists of pyrite and chalcopyrite, and locally contains abundant magnetite, pyrrhotite, and chromite. The gangue contains predominantly chlorite and rare quartz (Koptagel et al. 1998). The Maden copper deposit which is located between basaltic volcanic rocks and serpentinites was a Cyprus type deposit formed by the sea-floor spreading and being pushed onto the mainland by obduction and contains Cu, Pb, Zn, S, Fe, Ni, Au, and Co at relatively high concentrations. Although the Maden deposit has been in existence since 2000 B.C, the modern era of copper production at Maden began in 1939. As a result, this area has been heavily charged with metals by ancient and modern mining activities. The dominant vegetation in the study area is rare trees.

Sampling and Analysis

Teucrium polium L. is as plant which belongs to the Lamiaceae family. *Teucrium polium* L. is one of the wild-growing flowering species from this genus found in different geographical locations around the world. Its dry matter (biomass) was found as 82%. The investigated samples were obtained from the serpentine and copper-mining area in Maden-Elazığ-Turkey. Eight sites in this area where *Teucrium polium* L. plants are growing were selected as sampling points for soil and plants. Sites 1-5 around the abandoned mining area are Ni polluted and sites 6-8 are free from Ni pollution. The soil samples were collected from the surface at a depth of 10-20 cm and the stones and plant fragments were removed with a 2-mm sieve. The sieved samples were dried at 60°C and then ground in a mortar. The plant samples were thoroughly washed with tap water and rinsed with de-ionized water to remove any soil particles attached to the plant surfaces. The roots and the above ground parts (including stems and leaves) were separated and oven-dried at 105°C for 30 min, and then, at 70°C until a constant weight was achieved (Kaya and Yaman 2008a, Kaya and Yaman 2008b, Kaya et al. 2010a, Kaya et al. 2010b). Plants were identified with the help of local floras (Cakilcioglu et al. 2008). At each sampling site, soil samples at a depth of 10-20 cm were collected, dried at 80°C and sieved through a 100-mesh sieve (Kaya and Yaman 2008b, Kaya et al. 2010a, Kaya et al. 2010b). The soil and plant samples were analyzed for Ni and Co using the ATI-UNICAM 929 brand of an atomic absorption spectrophotometer. Some of the samples were randomly selected and analyzed by ICP-MS to check the accuracy of the measurements.

Digestion and Extraction of Plants and Soil

The dried plant samples were digested by using the dry ashing method as described in literature (Kaya and Yaman 2008a, Kaya and Yaman 2008b, Kaya et al. 2010a, Kaya et al. 2010b). In this method, dried samples were heated gradually in a furnace at 200ºC for 15 min, at 300ºC for 10 min, at 400ºC for 10 min, and finally ashed at 480ºC for 4.0 h. A 3.0 mL concentrated nitric acid/hydrogen peroxide (1/1, v/v) was added to 1.0 g of ashed sample and then heated to near the dryness. This procedure was repeated using the same acid mixture. After cooling, 3.0 mL of 1.0 M nitric acid was added and centrifuged. The clear solution was measured by FAAS for the determination of analytes. Blank measurements were also performed using the same procedure.

For the soil samples, 0.2 g of dried soil as described above was transferred into a flask (Pyrex) and 2.0 mL of a HNO_3/H_2O_2 mixture (1/1, v/v) was added. The mixture was heated on a hot plate, with shaking, to near dryness. This procedure was repeated once with the same sample. After cooling, 3.0 mL of 1.0 M HNO_3 was added and the sample was centrifuged. The clear solutions were analyzed by FAAS. Blank measurements for soil analysis were also performed using the same procedure given above. Other soil samples were extracted using 1.0 mol L^{-1} ammonium acetate (CH_3COONH_4) and ammonium chloride for the exchangeable and weakly adsorbed fraction and these fractions are also used to determine serpentine soil (Goncalves et al. 2007).

Analytical Performance

Linear calibration plots with the equations given below were obtained for Ni and Co and these calibration plots were used in the quantitative measurements.

$Y= 85 X + 0.5$ $R^2=0.99$ for Ni (0.2-2.0 mg/L)

$Y= 80 X + 0.6$ $R^2=0.99$ for Co (0.2-2.0 mg/L)

The accuracy of this method was checked by determining the recoveries of Ni and Co from the soil and plant leaves fortified with these elements. It was found that, at least, 95% of Ni and 94% of Co added to the samples was recovered. Furthermore, the standard additions method was done to find whether there is interference coming from the matrix or not. It was found that the slopes of direct calibration and the standard additions methods are very close to each other for soil and plant. Hence, linear calibration plots were used to determine Ni and Co in further studies. The effect of contamination was eliminated by subtracting the blank results.

Furthermore, the results obtained by FAAS were compared with the results obtained by ICP-MS to determine the validation. It was found that there are no significant differences (the recoveries higher than 93% were obtained for the results from FAAS and ICP-MS) between the data obtained by the FAAS and ICP-MS methods using t test at a confidence level of 90%.

RESULTS

In this study, the concentrations obtained for Ni and Co in the soil samples are given in Table 1. The results were found to be between 362-1875 mg kg^{-1} for Ni and 111-245 mg kg^{-1} for Co. The pH of the soils studied was in the range of 6.1-7.5. The mobile (available) Ni concentrations in the ammonium chloride and acetic acid-extracts were in the ranges of 0.34-47 and 0.53-37 mg kg^{-1}, respectively. Related with the plant leaves, the Ni concentrations were found to be between 9678 and 14110 mg kg^{-1} (d.m.) in the above ground parts of the *Teucrium polium* L. plants grown in serpentine soils together with the translocation factors between 2.23 and 3.23, and the enrichment coefficients between 5.9 and 9.2, respectively. Because plant Ni uptake is more important than the plant Ni concentration when the phytoremediation technique was considered, the Ni concentrations in the shoots and roots of the plant and the translocation factors are evaluated in the discussion section in detail.

DISCUSSION

Soils

The concentrations obtained for Ni and Co in the soil samples are given in Table 1. The results were found to be between 362-1875 mg kg^{-1} for Ni and 111-245 mg kg^{-1} for Co. These values are too heterogeneous and higher than the maximum permissible values (75 mg kg^{-1}) for agricultural soils in Turkey (Anonymous 2005). It was described that the ratio of Ni concentration to Co concentration in the soil changed between 5-10 times for the serpentine soil (Tappero et al. 2007). According to this definition, the first five soils given in Table 1 are serpentine soil, and the others are non-serpentine soils. Goncalves et al. (2007) reported Ni concentrations in ammonium acetate extracts of serpentine soils between 4.8 and 14.6 mg kg^{-1}, whereas, , the corresponding concentrations of the non-serpentine soils were significantly lower and varied between 0.6 and 3.0 mg Ni kg^{-1}. In this study, the Ni concentrations of the ammonium acetate extracts were found to be between 9 and 37 mgkg^{-1} for the first five (serpentine) soils, whereas, the corresponding levels in the last three (non-serpentine) soils were in the range of 0.53-1.4 mg kg^{-1}. On the other hand, the available Ni concentrations using the ammonium chloride

Table 1. Ni and Co concentrations (mg kg^{-1}) in the soil and Teucrium (*Teucrium polium* L.) plant parts (The results are an x ± standard deviation, n=3).

Sample	Ni	Co	TF*-Ni	EC**-Ni	TF*-Co	EC**-Co
Soil 1 total	1875±150	190±10	2.23	6.4	15	0.03
Soil 1-mobile NH_4Cl/a. ac	14±1 20±2	0.84±0.08 b.d				
Root 1	5400±378	0.42±0.05				
Above ground 1	12020±602	6.3±0.7				
Soil 2 total	1557±125	245±12	3.23	9.1	2.8	0.01
Soil 2-mobile NH_4Cl/ a. ac	47±5 37±4	2.2±0.3 0.75±0.09				
Root 2	4375±306	1.2±0.1				
Above ground 2	14110±708	3.4±0.4				
Soil 3 total	1535±123	226±11	2.5	6.3	3.5	0.02
Soil 3-mobile NH_4Cl/ a. ac	6.2±0.7 9±1	1.5±0.2 0.66±0.08				
Root 3	3856±259	1.1±0.1				
Above ground 3	9678±580	3.8±0.4				
Soil 4 total	1724±138	249±13	2.44	5.9	3.1	0.02
Soil 4-mobile NH_4Cl/ a. Ac	9±1 10±1	2.1±0.3 0.82±0.09				
Root 4	4163±250	1.3±0.2				
Above ground 4	10150±610	4.0±0.4				
Soil 5 total	1210±97	205±11	2.3	9.2	3.4	0.02
Soil 5-mobile NH_4Cl/ a. ac	11±1 13±1	1.5±0.2 0.42±0.06				
Root 5	4850±340	0.9±0.1				
Above ground 5	11140±780	3.1±0.3				
Soil 6 total	813±65	136±8	0.65	0.02	0.5	0.01
Soil 6-mobil NH_4Cl/ a. ac	0.34±0.04 0.53±0.06	0.30±0.04 b.d				
Root 6	20±2	3.2±0.3				
Above ground 6	13±2	1.6±0.2				
Soil 7 total	362±29	111±8	0.88	0.02	0.2	0.01
Soil 7-mobile NH_4Cl/ a. ac	3.3±0.4 1.4±0.2	5.7±0.5 2.1±0.3				
Root 7	8±1	7.6±0.6				
Above ground 7	7±1	1.2±0.1				
Soil 8 total	40±5	124±9	0.8	0.02	0.25	0.01
Soil 8-mobile NH_4Cl/ a. ac	2.2±0.3 1.3±0.2	3.5±0.3 1.1±0.1				
Root 8	10±1	5.2±0.4				
Above ground 8	8±1	1.3±0.2				

a. ac: NH_4CH_3COO, b.d: below detection
TF*: Metal concentration in the aboveground part/metal concentration in root
EC**: Metal concentration in the aboveground part/metal concentration in soil

reagent were found to be in the range of 6-47 mg kg^{-1} for the serpentine (first five) soils, whereas, the corresponding Ni concentrations were in the range of 0.34 and 3.3 for the non-serpentine (last three) soils. As a result, according to this definition, the first five soils are serpentine soils and the last three soils are non-serpentine soils.

Plants

The leaves of *Teucrium polium* L. have a relatively large biomass (%82) and include the constituents in the ranges of 11-25% for protein, 20-36% for cellulose, 1.2-2.8% for Ca, 1.5-6.2% for K, 840-2847 mg kg^{-1} for P, and 1199-2524 mg kg^{-1} for Mg (Bakoglu et al. 1999a, Bakoglu et al. 1999b). It was reported that the leaves of plants grown in serpentine soil contain Ni in the concentration range of 20-100 mg kg^{-1} while the Ni concentrations of non-serpentine plant leaves were in range of 0.1-5 mg kg^{-1} (Anderson et al. 1999, Reeves and Baker 2000, Boominathan et al. 2004, Sheoran et al. 2009). It has reported that some serpentine endemics can accumulate more than 1000 $mgkg^{-1}$ of Ni in their dried leaves (Anderson et al. 1999, Boominathan et al. 2004, Freitas et al. 2004, Goncalves et al. 2007, Tappero et al. 2007). Concentrations of 38,105 (3.81%) and 1465 mg Ni kg^{-1} (d.m.) in the aboveground parts of *Alyssum serpyllifolium* and *Bromus hordeaceus* were reported by Freitas et al. (2004), but, their enrichment coefficient was found to be 0.41 because Ni concentrations in the soils taken from around those plants was 93,808 and 3608 mg kg^{-1}, respectively. Tappero et al., (2007) reported 1610 mg Ni kg^{-1} in the shoots of *Alyssum Murale* after irrigation with Ni-enriched water (3 mg Ni L^{-1}) to represent the serpentine soil. Osma et al. (2012) determined the concentration of six toxic metals in the soil and in unwashed and washed samples of different vegetable species (*Petroselinum crispum*, *Brassica oleraceae* var. *acephala*, *Beta vulgaris* var., and cicla collected from six different sites (stream side, inner city, industrial, suburban, roadside, and rural) in Istanbul in 2007. They concluded that the total metal concentrations in the vegetables were as follows Pb > Cr > Ni > Zn > Cu > Cd. Demirayak et al. (2011) examined some trace metal accumulation (Pb, Cd, Zn, and Cu) in the leaves, needles, and twigs of some natural and exotic tree and shrub species, growing in the city centre and suburbs Atakum, Samsun, Turkey, which were selected and tested as biomonitors of these heavy metals polluted by the burning of fossil fuels and heavy traffic conditions from December 2007 to August 2008.

In this study, the Ni concentrations were found to be between 9678 and 14,110 mg kg^{-1} (d.m.) in the above ground parts of the *Teucrium polium* L. plants grown in serpentine soils together with the translocation factors between 2.23 and 3.23, and the enrichment coefficients between 5.9 and 9.2, respectively. According to our knowledge, the translocation factors and enrichment coefficients as high as the obtained values in this study have not been reported up to now. The cobalt concentrations in the same samples were found to be between 3.1 and 6.4 mg kg^{-1} together with the translocation factors being between 2.8 and 15, and the enrichment coefficients between 0.01 and 0.03. Moreover, the available Ni concentrations in the ammonium chloride and ammonium acetate extracts of those serpentine soils were found to be higher up to 10 times more than those in the non-serpentine soils. Consequently, the *Teucrium polium* L. plant has been considered as hyperaccumulator

Table 2. The ratios of the Ni concentrations to the Co concentrations in the studied soils sampled. Soil samples with the numbers of 1-5 are serpentine and 6-8 non serpentine soils.

Sample	Ratio of nickel to cobalt, -fold				
	soil	a.chl extract	a. ac extract	root	Aboveground part
Soil 1	9.8	16.7	-	12857	1907
Soil 2	6.3	21.4	49.3	3646	4150
Soil 3	6.8	4.1	13.6	3505	2547
Soil 4	6.9	4.3	12.2	3202	2538
Soil 5	5.9	7.3	31	5389	3594
Soil 6	5.9	1.1	-	6.3	8.1
Soil 7	3.3	0.6	0.7	1.1	5.8
Soil 8	0.3	0.6	1.2	1.9	6.2

a.chl: ammonium chloride
a. ac: ammonium acetate

plant and it can be used for phytoextraction of Ni-contaminated soil.

Nickel and cobalt are expected to interact antagonistically as a result of competition for the semiselective transport proteins in the roots. High Ni concentrations will increase competition for root metal transporters and for intercellular ligands participating in metal absorption (Sheoran et al. 2009). The ratios of Ni/Co concentrations using HNO_3/H_2O_2 digestion, in the extracts of the ammonium chloride and ammonium acetate of the soils, in the roots and in the aboveground parts of the plants are given in Table 2. The obtained Ni/Co-ratios for the roots grown in serpentine soils were found to be between 3202- and 12,825-fold while the values between 1.1- and 6.3-fold were found in the non-serpentine soils. Similarly, the ratios in the aboveground parts of the *Teucrium polium* L. plants grown in the serpentine soils were between 1907- and 4150-fold while the values were between 5.8- and 8.1-fold for the non-serpentine soils. The Ni/Co-ratios in the HNO_3/H_2O_2 digestions of soils were found to be between 5.9- and 9.8-fold for serpentine and 0.3- and 5.9-fold for non-serpentine soils. The Ni/Co-ratios in the ammonium chloride and ammonium acetate extracts of the soils were found to be higher than 4- and 10-fold for serpentine soils, respectively, whiles the values were below or about 1-fold for both extracts of the non-serpentine soils. As a result, the ratios of Ni/Co concentrations in the roots and aboveground parts of *Teucrium polium* L. grown in serpentine soil were significantly higher (between 2547-12,857-times) than the ratios for *Teucrium polium* L. grown in non-serpentine soils (between 1.1-8.1-times). These values are highly important and are the original results. Ni concentrations in the studied soils and plants are given in Table 1.

Translocation Factor and Enrichment Coefficient (bioconcentration factor)

One of the characteristics of hyperaccumulator plants is the ability to accumulate high concentrations of metal or metalloid in the aboveground biomass. Although the standard for a hyperaccumulator is still under dispute, the following consideration can be concluded (Zu et al. 2004, Cui et al. 2007). A hyperaccumulator plant must have the ability to translocate an element from the roots to the shoots at high rates. As a result, the metal concentrations in the aboveground parts of hyperaccumulator plants are invariably greater than those in roots, this is termed as the translocation factor, whereas, the metal concentrations in the roots are 10- or more-times higher than in the aboveground parts in normal plants (Chaney et al. 1997). This indicates an efficient ability to transport metals from the roots to the shoots and, most likely, the existence of tolerance mechanisms to cope with the high concentrations of metals. Translocation factors (TF) for a hyperaccumulator plant should be typically >1. Furthermore, the enrichment coefficient (EC) or bioaccumulation factor should be higher than 1, defined as the ratio of the metal concentration in the above ground parts of the plant to in the soil, which emphasizes the degree of plant metal uptake.

Translocation factors for Ni in the studied *Teucrium polium* L. plants were found to be between 2.2-3.2 for serpentine soil and 0.65-0.88 for non-serpentine soil (Table 1). These results show that the *Teucrium polium* L. plant has the ability to transfer the nickel from the roots to the aboveground parts. Tappero et al. (2007) reported the TF as 2 for Ni and 3 for Co in Alyssum murale treated with 50 μM (3 mg L^{-1}) of Ni, Co, and Zn in a nutrition solution. The enrichment coefficients for Ni in the studied *Teucrium polium* L. plant samples were found to be between 5.9-9.2 for serpentine soil and about 0.01-0.03 for non-serpentine soils (Table 1). These results also prove that the *Teucrium polium* L. plant has a high degree of Ni uptake from the soil. Due to the high toxicity properties of nickel (Yaman 2006), the discovered *Teucrium polium* L. plant has a high importance for the phytoextraction of contaminated soils with Ni.

CONCLUSION

The present study demonstrated that a new hyperaccumulator plant, *Teucrium polium* L., grown in serpentine soil can be considered for both bioremediation and biomining purposes. Nickel concentration up to 14,110 mg kg^{-1} basis on a dry mass was observed in the above ground part of the *Teucrium polium* L. plant. The Ni/Co-ratios in ammonium chloride and ammonium acetate extracts of the soils were found to be higher than 4- and 10-fold for serpentine soils, respectively, while the values were below or about 1-fold for both extracts of non-serpentine soils. The translocation factors and the enrichment coefficients for Ni in the studied *Teucrium polium* L. plants were found in the ranges of 2.2-3.2 and 5.9-9.2 for plants grown in serpentine soil, and 0.65-0.88, and about 0.02 for plants grown in non-serpentine soil, respectively. It was found that the ratios of nickel to cobalt in the roots and above ground parts of *Teucrium polium* L. plants grown in serpentine soils were evidently higher than those in non-serpentine soils. Consequently, taking into consideration the criteria for phytoextraction (Bhargava et al. 2012), the novel discovered *Teucrium polium* L. plant can be successfully used for phytoextraction of Ni-contaminated soils.

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