IN-SITU PHYTOEXTRACTION OF NICKEL BY *Odontarrhena serpyllifolia* ON ULTRAMAFIC SOILS OF PORTUGAL

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ABSTRACT

Serpentine soils derived from ultramafic rocks have a worldwide distribution. These soils are rich in nickel (Ni), chromium (Cr) and sometimes cobalt (Co), but are poor in essential nutrients, such as nitrogen (N), phosphorus (P), potassium (K) and calcium (Ca). Plants that hyperaccumulate Ni colonize serpentine soils as endemics. The rhizosphere of these endemic plants provides a complex and dynamic microenvironment where the roots, along with associated microorganisms, form unique ecosystem with an extraordinary phytoextraction function. This study considers the use of serpentine soils in Northeast Portugal as potential producers of Ni using an endemic species (*Odontarrhena serpyllifolia*) for phytomining. In this sense, the present study had the following objectives: (1) to investigate the relationship between Ni concentration in the soil and the concentration in the plant (*O. serpyllifolia*) in ultramafic areas of Portugal; and (2) to assess the potential of this species for Ni phytomining as bio-ore. This species is endemic to serpentine soils of the Bragança and Morais massifs and is a Ni hyperaccumulator that can accumulate more than 12,000 mg/kg (dry weight). Under natural conditions, the results show that considering only the aerial parts of the plant, the biomass varies between 4.08–9.37 t/ha, containing a range of 0.25–1.23% Ni, allowing a withdrawal of between 12.2–98.1 kg Ni/ha per crop under natural conditions. The amount of Ni extracted by the plant is similar in both ultramafic areas, having a mean value of approximately $US 500 per ha considering the January 2019 Ni prices. The results of this study indicate that some of the land in these ultramafic areas are potential producers of Ni from this endemic species.

Keywords: *agromining, bio-ore, hyperaccumulation, phytomining, serpentine flora*

INTRODUCTION

Ecosystems of serpentine soils are known to contain distinct communities of endemic plants [1]. These soils contain high levels of nickel (Ni), chromium (Cr), and cobalt (Co), and have a low calcium/magnesium (Ca/Mg) ratio [1]. Ni-hyperaccumulator species have been found in these serpentine soils, containing very appreciable amounts of this metal in their tissues, at concentrations higher than 5,000 mg/kg dry weight [2]. Hyperaccumulator plants can store high concentrations of specific metals in their aerial parts (0.01% to 1% of the dry weight, depending on the metal). For Ni, the threshold for hyperaccumulation in plant dry matter is 1,000 mg/kg [2]. The first discovery of Ni hyperaccumulation was documented in 1948 [3]. In this study the authors found Ni concentrations of 7,900 mg/kg in the leaves of *Alyssum bertolonii* Desv. These hyperaccumulator plants can be used for extraction of metals by phytomining (Figure 1). In phytomining, the plant biomass is processed, after harvesting, for extraction and recovery of commercially valuable metals, such as nickel (Ni) [4]. In an integrated approach, phytomining could provide to the local communities an alternative type of "agriculture" on degraded lands or low agricultural productivity soils, such as serpentine soils.

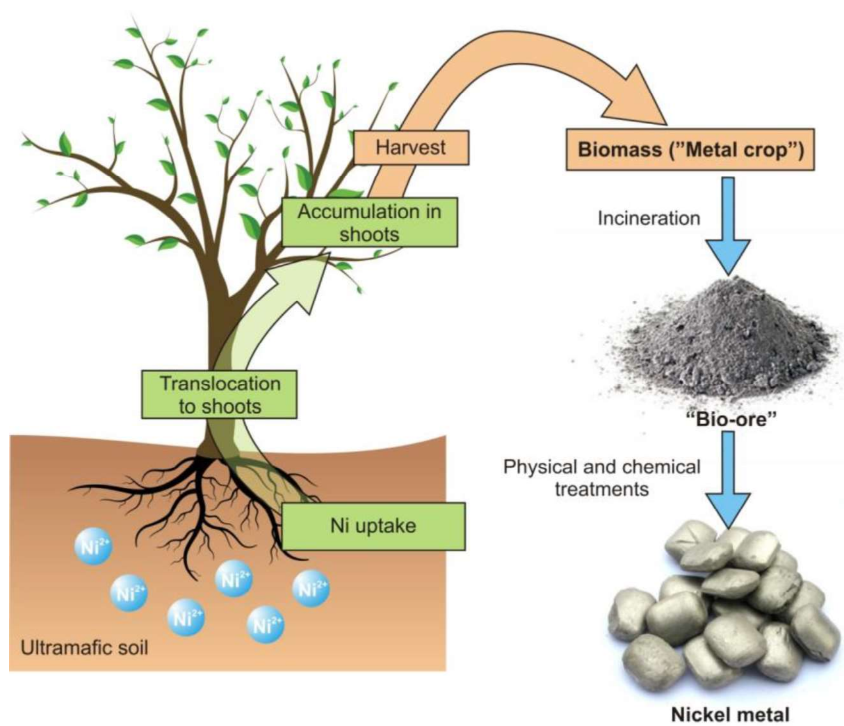

Figure 1. Phytomining operations with harvesting of biomass and processing of bio-ore to produce high-value Ni compounds or pure Ni metal.

In Portugal, the capacity of the *Odontarrhena serpyllifolia* (Desf.) Jord. & Fourr. species as Ni hyperaccumulator has been known by the scientific community, at least since the 1960s [5]. This species is also known by the synonyms *Alyssum pintodasilvae* T.R. Dudley and *Alyssum serpyllifolium* subsp. *lusitanicum* T.R. Dudley & P.C. Silva [6]. In this context, the present study had the following objectives: (1) to investigate the relationship between Ni concentration in the soil (total and bioavailable fraction) and the concentration in the plant (*Odontarrhena serpyllifolia*) at ultramafic areas of Portugal; and (2) to preliminarily assess the potential for this species in phytomining Ni as bio-ore.

MATERIALS AND METHODS

Study areas

The study areas are located in the Northeast of Portugal, in a serpentinized region with characteristic geology and flora. The ultramafic rocks of this region are represented in two massifs, namely, the Bragança massif and the Morais massif. Geologically, the Bragança and Morais massifs are composed of four main sequences [7], [8]: 1) a parautochthonous complex, with close palaeogeographic affinities with the underlying autochthon that are composed primarily of metasediments; 2) a lower allochthonous complex consisting of Ordovician metasediments, acid and basic metavolcanics and alkaline to peralkaline gneisses; 3) a middle allochthonous complex (or Ophiolitic Complex) formed by several thrust sheets made of ophiolitic rocks (amphibolites, serpentinized peridotites, gabbros, etc.) displaying variable outcropping extension in each of the massifs; and 4) an upper allochthonous complex made of partly oceanic high grade metamorphic rocks (paragneisses, eclogites, mafic granulites, pyroxenites and peridotites).

Sampling and sample preparation

Samples of *O. serpyllifolia* and soil were collected from 62 random sites in the study areas (Bragança and Morais massifs). On the selected sites, samples of soil and all plant specimens were collected in an area of approximately 1 m^2. At each location, four random partial soil samples weighing 0.5 kg each were collected from a 0 to 20 cm depth and mixed to obtain a single composite sample [9]. Plant sampling was focused on the whole plant, considering plants of similar maturity and the separation of the different tissue types (aerial parts and roots).

In the laboratory, the soil samples were oven-dried at a constant temperature of 40 ºC, manually homogenized and quartered. Samples for chemical analysis were sieved using a 2 mm mesh sieve to remove plant matter and were subsequently screened to pass through a 180 µm screen [9]. The vegetal material was washed thoroughly, first in running water followed by distilled water and then oven-dried at a temperature of 50 ºC. When dry, the material was weighed for an initial calculation of biomass. Then, the material was milled into a homogenous powder [9].

Analytical procedures

The determination of pseudo-total element content (Co, Cr, Cu, Ni, and Zn) in soil samples was performed using microwave-assisted aqua regia digestion in closed Teflon vessels (Multiwave 3000, Anton Paar) followed by atomic absorption spectrophotometry (AAS, Thermo Solar M2) [9]. We also performed a single (partial) chemical extraction to determine the bioavailable fraction, using ammonium acetate (1 M NH_4 acetate pH 4.5, 25 ºC, shake for 2 hours) [9]. The analytical processes for the plant samples involved microwave digestion with an HNO_3–H_2O_2 mixture in closed Teflon vessels. The analysis was performed in the same way as that for soil samples. Data were collected in triplicate. Certified reference materials were also used.

RESULTS AND DISCUSSION

As expected, the results showed that the soils of study areas have high levels of Co, Cr and Ni (Table 1). Regarding the potential bioavailable fraction, Ni was the most bioavailable metal. These results are in agreement with observations reported by other authors [9].

Table 1. Pseudo-total and bioavailable metal concentrations in the soils and metal concentrations in Odontarrhena serpyllifolia from the ultramafic sites of Bragança and Morais.

	Soil (n = 62)						*O. serpyllifolia* (n = 62)		
	Pseudo-total (mg/kg)			Bioavailable (mg/kg)			Aerial parts (mg/kg, dry weight)		
	Co	Cr	Ni	Co	Cr	Ni	Co	Cr	Ni
Minimum	34.2	107	706	0.01	0.55	5.01	0.14	0.06	2,497
Maximum	309	1,372	3,516	7.05	6.65	185	87.2	42.5	12,261
Mean	107	527	1,855	1.96	2.12	51.7	10.9	7.27	6,253
Median	101	462	1,766	1.39	1.73	39.8	9.68	5.69	5,600
S.D.	58.5	272	736	1.76	1.25	41.2	12.4	6.19	2,444

With regard to plant material, this study confirms the hyperaccumulation capacity of this species (Table 1). The concentrations of Co, Cr and Ni are of the same order of magnitude as obtained in other studies with the same species [9].

Over the past two decades some studies have been carried out to evaluate the phytotechnological potential of hyperaccumulator plant species, namely in order to assess their ability for phytoextraction of toxic metal(loid)s from soil and water [10], [11], [12], [13], [14], [15], as well as to evaluate its feasibility for the economic recovery of valuable metals, by phytomining [4], [9].

In this study, under natural conditions (i.e., without fertilizers or other agrochemicals addition), it is possible to obtain between 4.08 tonnes and 9.37 tonnes of biomass per hectare, with a Ni content that varies between 0.25 to 1.23% (Table 2). The results show that the production of Ni in kg per hectare (extraction rate) varies between 12.2 and 98.1 kg/ha (Table 2). Given the current Ni price (January 2019), it appears that the economic value obtained per hectare per crop varies from approximately $US 140 to $US 1,100, with a mean of $US 500 (Table 2).

Table 2. Values of the biomass, the Ni accumulation, the Ni extraction and their economic value (n = 62).

	Biomass (kg/ha)	Ni concentration in plants (mg/kg, DW)	Ni phytoextraction yield (kg/ha)	Economic value ($US/ha)
Minimum	4,080	2,497	12.2	138
Maximum	9,370	12,261	98.1	1,107
Mean	7,265	6,253	45.5	513
Median	7,920	5,600	43.0	486
S.D.	1,590	2,444	19.9	224

CONCLUSION

Under natural conditions, without fertilizers or other agrochemicals addition, the Ni concentrations found in the plants in the studied ultramafic massifs ranged from 0.25% to 1.23% Ni, with a mean of 0.63% Ni. In addition, this species shows a high biomass yield. The average biomass was calculated at 7.26 tonne/ha, with a minimum of 4.08 tonne/ha and a maximum of 9.37 tonne/ha, indicating a higher productivity. The amount of Ni extracted by the plant is approximately 45.5 kg/ha on average, having a mean value of approximately $US 500 per ha considering the January 2019 Ni prices. The results of this study suggest that some non-agricultural land, and land not suitable for agricultural uses in these ultramafic areas, are potential producers of Ni from *O. serpyllifolia* species. Although, it is necessary a better understanding of Ni accumulation mechanisms by this endemic species, to identify optimum cultivation practices, such as fertilization, addition of chelating agents, hybridization or cloning, to increase the Ni hyperaccumulation potential and biomass production. The Ni concentration in the aerial parts of *O. serpyllifolia* is higher than in the roots. So, realistically, any phytomining operations should use preferentially the aerial parts of the plants. Furthermore, this is a perennial species, which allows successive harvests without requiring seeding, greatly increasing the phytomining potential as a function of time.

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