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Assessing the agromining potential of Mediterranean nickel-hyperaccumulating plant species at field-scale in ultramafic soils under humid-temperate climate

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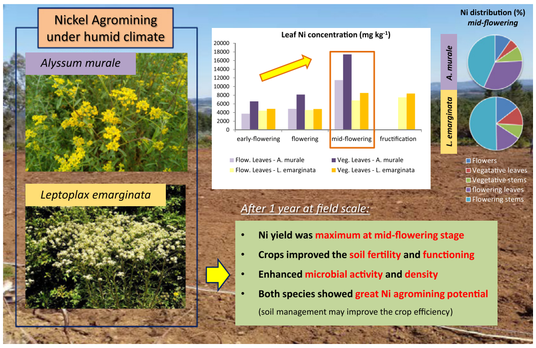
HIGHLIGHTS

- Agromining potential of *Alyssum murale* and *Leptoplax emarginata* was assessed.
- Ni bioaccumulation and yield was maximal at the mid-flowering stage.
- Crops increased soil nutrients and modified microbial community structure.
- Both species showed great potential for their use in Ni agromining systems.
- Optimising crop management practices could improve the Ni removal efficiency.

GRAPHICAL ABSTRACT



After 1 year at field scale:

- Ni yield was maximum at mid-flowering stage
- Crops improved the soil fertility and functioning
- Enhanced microbial activity and density
- Both species showed great Ni agromining potential (soil management may improve the crop efficiency)

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ABSTRACT

Nickel (Ni) agromining of ultramafic soils has been proposed as an eco-friendly option for metal recovery, which can also improve the fertility and quality of these low productive soils. The selection of adequate plant species and the analysis of their performance under the different climatic conditions are of interest for optimising the process and evaluating its full viability. A one-year field experiment was carried out to evaluate the viability of the two Ni-hyperaccumulating Mediterranean species, *Alyssum murale* and *Leptoplax emarginata*, for agromining purposes in ultramafic soils under a humid-temperate climate. Field plots of 50 m^2 were established and the soil was fertilised with gypsum and inorganic NPK fertilisers prior to cropping. *Alyssum murale* produced a slightly higher Ni yield than L. *emarginata*, but Ni bioaccumulation was dependent on the plant phenological stage for both species, being maximal at mid-flowering (4.2 and 3.0 kg Ni ha^{-1}, respectively). In both species, Ni was mainly stored in the leaves, especially in leaves of vegetative stems, but also in flowers and fruits in the case of L. *emarginata*. The main contributors to Ni yield of *A. murale* were flowering stems and their leaves, while for L. *emarginata* they were flowering stems and fruits. Implementing the agromining system increased soil nutrient availability, and modified microbial community structure and metabolic activity (due to fertilisation and plant root activity). The soil bacterial communities were dominated by *Proteobacteria*, *Actinobacteria*, *Acidobacteria* and *Chloroflexi*, and the agromining crops modified the relative abundance of some phyla (increasing *Proteobacteria*, *Bacteroidetes* and *Nitrospirae* and reducing *Acidobacteria* and *Planctomycetes*). Cultivating *A. murale* increased the densities of total culturable bacteria, while L. *emarginata* selected Ni-tolerant bacteria in its

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rhizosphere. In summary, both species showed great potential for their use in Ni agromining systems, although optimising soil and crop management practices could improve the phytoextraction efficiency.

1. Introduction

Ultramafic or serpentine soils develop from the weathering of ultramafic bedrocks, igneous and metamorphic rocks with a low silica content ($SiO_2 < 45\%$) but rich in mafic minerals (such as the olivine and pyroxene groups) and serpentine clays, with a high content in Mg, Fe and Ni (Bani et al., 2014). They are characterised by a high stoniness and poor water holding capacity, relatively high concentrations of Ni, Co and Cr, high Mg/Ca ratio and low to deficient levels of macronutrients (N, P and K) and micronutrients such as Mo and B (Nkrumah et al., 2016, 2018). As a result, ultramafic soils usually present a low fertility and productivity (Bani et al., 2007), making them unattractive for traditional agriculture.

Despite the elevated total Ni concentration in ultramafic soils, which reach values found in some mineral ores (between 0.1 and 1.5%; Nkrumah et al., 2016), conventional mining processes are not economically viable in these soils (Li et al., 2003a). On the other hand, Ni phytomining of ultramafic agricultural soils appears to be an ecological option with high economic potential to manage and re-valorise these low-productivity areas (Chaney et al., 2007; Bani et al., 2015a; Van der Ent et al., 2015a). It consists of cultivating hyperaccumulating plants that bioaccumulate Ni in their aboveground tissues, the subsequent harvesting of the aerial biomass, its drying, incineration and processing, generating bio-ores with up to 25% Ni (Chaney et al., 2007; Van Der Ent et al., 2015a). The concept of agromining was recently introduced to emphasise that phytomining forms part of an integrated agricultural chain (Van Der Ent et al., 2015a). Agromining allows for sustainable metal recovery without causing the environmental impacts associated with conventional mining activities, and at the same time, can improve soil fertility and quality and decrease Ni phytotoxicity over time, which would permit cultivation of conventional agricultural crops (Van der Ent et al., 2015a; Nkrumah et al., 2016). A recent study using life cycle assessment demonstrated that the whole agromining chain can offer the opportunity to promote new agricultural practices and preserve valuable resources (Rodrigues et al., 2016).

Numerous geobotanical studies have been carried out to identify suitable hyperaccumulator plant species (Reeves and Adigüzel, 2008; Bani et al., 2009; Van der Ent et al., 2015b). According to Nkrumah et al. (2018) only “hypernickelphores”, Ni-hyperaccumulators accumulating >1% Ni in their aerial biomass, are potential candidates for a successful agromining. On a global scale, Ni hyperaccumulation occurs in approximately 450 species, the majority endemic of ultramafic soils, and approximately 25% belong to *Brassicaceae* family (Pollard et al., 2014). Within the *Brassicaceae*, the Mediterranean species *Alyssum murale* Waldst. &Kit and *Leptoplax emarginata* (Bois) O.E.Schulz (syn. *Peltaria emarginata* (Boiss.) Hausskn), have been identified as having high Ni phytomining potential (Chardot et al., 2005; Lucisine et al., 2014; Zhang et al., 2014; Rue et al., 2015; Van der Ent et al., 2015a). Bani et al. (2009) recorded leaf Ni concentrations up to 2.0% for *A. murale* and 1.4% for L. *emarginata* in several sites of the Balkans. Zhang et al. (2014) evaluated the Ni content of 14 taxa of the genera *Alyssum*, *Leptoplax* and *Bornmuellera* collected from different localisations of Greece and Albania and found leaf Ni concentrations between 0.8 and 1.6% and 3.2–3.4% in *A. murale* and L. *emarginata*, respectively. However, in Europe, field-scale evaluations have only been carried out using *A. murale* (not L. *emarginata*) and in Mediterranean ultramafic regions (Bani et al., 2015a; Bani et al., 2015b). Field demonstrations of agromining potential, and assessing the performance of diverse agromining crops, in other geographical areas and under different climatic conditions are necessary if we are to optimise the process and determine its real viability and broader benefits to ecosystem services.

The aim of this work was to evaluate the performance of *A. murale* and L. *emarginata* within an agromining system during a one year field experiment in an ultramafic outcrop under a humid-temperate climate in NW Spain. For that, biomass productivity and Ni yields of both species were assessed, as well as the evolution in Ni concentration and distribution in the plant aerial organs over time. Additionally, the effect of implementing the agromining system on soil fertility, Ni availability, and microbial activity and diversity was evaluated.

2. Materials and methods

2.1. Experimental design

The experimental site is located in the Melide ultramafic complex close to the village of Eidián (NW Spain; 42°49′54.2”N 8°00′13.4”W). The soil had a sandy loam texture with 18.7% clay, 29.1% silt and 52.2% sand, 13.4% organic matter (OM) and was slightly acidic (pH 5.8; Table 1). The pseudo-total concentrations of Ni, Mn and Co were 967, 973 and 78 mg kg^{-1}, respectively (Table 1). The climate of this region is humid-temperate and during the experimental period (June 2015 to May 2016) the mean temperature was 12.2 °C, with a fluctuation between +37 °C and −3 °C and an accumulated precipitation of 1478 l m^{-2} (Melidemeteorological station (A Coruña); Agrometeorological report Meteogalicia).

Seeds of *A. murale* and L. *emarginata* were collected from Trigona (39°47′17.5”N, 21°25′19.1″E, Greece) and germinated in seed trays in ultramafic soil collected from the field site. Six plots of 50 m^2 (5 m × 10 m) were established in the field site and soil was amended with gypsum (1000 kg ha^{1}; supplied by Luis Perez Ulecia, s.l.) and NPK inorganic fertiliser (120:120:150 kg ha^{-1}). One month later, two month-old *A. murale* or L. *emarginata* were transplanted following a density of 4 plants per m^{-2} (according to Bani et al., 2015b) into 3 replicate plots per plant species (with a 1 m separation between plots) using a fully-randomised design.

Table 1
Physicochemical characterization of the soil at the field site. EC: electrical conductivity; CEC: cation exchange capacity; TOC: total organic C; TN: total N.

	Soil properties (mean ± standard error)
pH_{H2O}	5.8 ± 0.1
EC (mS cm^{-1})	78 ± 1
CEC ($cmol_c$ kg^{-1})	4.9 ± 1.0
TOC (g kg^{-1})	52.6 ± 8.0
TN (g kg^{-1})	3.0 ± 0.4
Pseudo-total element concentrations	
Al (g kg^{-1})	34.4 ± 1.3
Ca (g kg^{-1})	7.6 ± 0.6
Co (mg kg^{-1})	77.5 ± 3.3
Cr (mg kg^{-1})	1263.9 ± 64.6
Cu (mg kg^{-1})	14.8 ± 1.4
Fe (g kg^{-1})	71.0 ± 0.7
K (mg kg^{-1})	437.6 ± 25.1
Mg (g kg^{-1})	45.1 ± 2.3
Mn (mg kg^{-1})	972.9 ± 27.5
Ni (mg kg^{-1})	967.4 ± 13.0
P (mg kg^{-1})	261.1 ± 8.8
Pb (mg kg^{-1})	8.2 ± 0.3
Zn (mg kg^{-1})	42.7 ± 2.1