



Organic amendments for improving biomass production and metal yield of Ni-hyperaccumulating plants



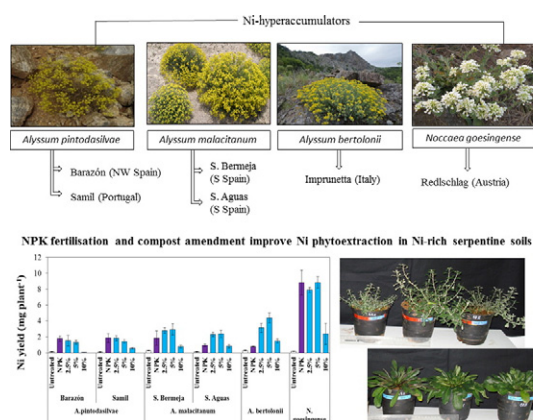
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HIGHLIGHTS

- The low biomass of most Ni-hyperaccumulator plants can limit the applicability of phytomining techniques at field scale
- The use of traditional agronomic practices such as fertilisation can improve plant establishment and biomass production of these plant species
- Changes in soil Ni availability, shoot biomass production and Ni accumulation by four Ni-hyperaccumulator species were evaluated after soil inorganic fertilisation (NPK) or compost amendment
- Intermediate doses of compost amendment had a stronger effect on Ni yield, mainly due to the enhanced shoot biomass
- The use of organic wastes as amendments in phytomining permits the recycling of these residues

GRAPHICAL ABSTRACT



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ABSTRACT

Ni phytomining is a promising technology for Ni recovery from low-grade ores such as ultramafic soils. Metal-hyperaccumulators are good candidates for phytomining due to their extraordinary capacity for Ni accumulation. However, many of these plants produce a low biomass, which makes the use of agronomic techniques for improving their growth necessary.

In this study, the Ni hyperaccumulators *Alyssum serpyllifolium* ssp. *lusitanicum*, *A. serpyllifolium* ssp. *malacitanum*, *Alyssum bertolonii* and *Noccaea goesingense* were evaluated for their Ni phytoextraction efficiency from a Ni-rich serpentine soil. Effects of soil inorganic fertilisation (100:100:125 kg NPK ha⁻¹) and soil organic amendment addition (2.5, 5 or 10% compost) on plant growth and Ni accumulation were determined. All soil treatments greatly improved plant growth, but the highest biomass production was generally found after addition of 2.5 or 5% compost (w/w). The most pronounced beneficial effects were observed for *N. goesingense*. Total Ni phytoextracted from soils was significantly improved using both soil treatments (inorganic and organic), despite the decrease observed in soil Ni availability and shoot Ni concentrations in compost-amended soils. The most promising results were found using intermediate amount of compost, indicating that these types of organic wastes can be incorporated into phytomining systems.

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1. Introduction

Metal hyperaccumulator plants present an extreme ability to accumulate trace elements in their harvestable tissues. To date, approximately 500 taxa of metal hyperaccumulators have been identified, of which over 90% accumulate nickel (Ni) (Pollard et al., 2014). The Brassicaceae family, and specifically the genus *Alyssum*, present the greatest number of Ni hyperaccumulators (Pollard et al., 2014). The vast majority of Ni-hyperaccumulating species are endemic to soils developed over ultramafic rocks (naturally rich in metals such as Co, Cr and Ni), and can present up to 1–3% dry weight Ni content in their shoots (Reeves 2006). This characteristic makes these plants particularly useful for application in phytoextraction or phytomining (Chaney et al., 2010). Phytoextraction techniques are focused on the remediation of degraded or contaminated soils using plants which are able to accumulate high amount of metals in their harvestable parts, while phytomining is the phytoextraction and recovery of metals from the harvested plant biomass for commercial gain (Chaney et al., 2007; Tang et al., 2012). Ni phytomining was proven to be economically feasible in the USA and Canada (Chaney et al., 2007; Li et al., 2003a,b), and more recently, in Europe (Bani et al., 2015; Bani et al., 2007). To date the Ni-hyperaccumulating *Alyssum* species, *Alyssum murale* and *Alyssum corsicum* (native to Mediterranean serpentine soils), have shown the highest potential application in phytomining processes (Chaney et al., 2007; Li et al., 2003a,b). Selected plants for Ni phytomining purposes should be able to hyperaccumulate this metal and also to tolerate and thrive under the adverse growth conditions which are frequently present in metal-rich environments (Harris et al., 2009). The use of native plant species has several advantages, both for practical reasons and also as a means of supporting the conservation of serpentine biodiversity. For example, *A. murale* occurs widely as a native weed of agricultural land on ultramafic Vertisols in Albania, and was successfully implemented in phytomining cropping systems in the Albanian regions of Pojské and Prenjas (Bani et al., 2015; Bani et al., 2007). However, a low biomass production can be an important bottleneck limiting the practical application of hyperaccumulators in phytomining (Kidd et al., 2015; McGrath and Zhao, 2003; Mench et al., 2010; Robinson et al., 1997).

Over the last decade several authors have highlighted the need to optimise the phytomining process using appropriate agronomic and crop management practices, such as pH adjustments, fertilisation regimes, design of cropping patterns (co-cropping or rotation), bioaugmentation using plant-associated microbial inoculants, or the use of plant growth regulators (PGRs) (Bani et al., 2015; Cabello-Conejo et al., 2014a; Cabello-Conejo et al., 2014b; Chaney et al., 2007; Kidd et al., 2015; Tang et al., 2012; Durand et al., 2015). In fact, the concept of agromining was introduced by van der Ent et al. (2015) to emphasise that phytomining forms part of an integrated agricultural chain. Several studies have shown the importance of soil fertilisation to increase plant yields and phytoextracted Ni. In field-scale experiments in serpentine soils, Bani et al. (2015) found a high positive response in biomass production of *A. murale* using NPK and Ca fertilisers. Robinson et al. (1997) enhanced the biomass of *A. bertolonii* with NPK fertilisers by 3-fold. Inorganic fertilisation also increased the biomass of *Alyssum serpyllifolium* ssp. *lusitanicum* growing on a mine-soil by a factor of 10; as a consequence the total Ni phytoextracted experimented a 6-fold increase (Kidd and Monterroso, 2005).

Municipal wastes such as sewage sludges (biosolids) have been used as an amendment to agricultural soils for many years now, and are an economically attractive alternative to disposal through landfill and incineration. However, few studies have considered the use of organic soil amendments to increase Ni phytomining efficiency. Ultramafic soils are characterised by a low fertility, low water holding capacity and poor structure. Compost addition could be a useful means of increasing soil fertility and improving soil structure (Bernal et al., 2007; Fagnano et al., 2011; Martínez-Fernández et al., 2014). Moreover, the use of composted solid organic residues, such as municipal solid wastes

(MSW) or sewage sludge, also constitutes a means of recycling waste products, thus contributing towards the objectives of the EU's Zero Waste Policy (2013).

The objective of this study was to evaluate the use of composted MSW as a means of increasing the phytoextraction efficiency of different Ni hyperaccumulating plants grown in ultramafic soils. The effects of different fertilisation regimes (compost addition or mineral fertilisers) on biomass production and Ni removal of the hyperaccumulators, *A. serpyllifolium* ssp. *lusitanicum* (two populations), *A. serpyllifolium* ssp. *malacitanum* (two populations), *Alyssum bertolonii* (one population) and *Noccaea goesingense* (one population), were compared.

2. Material and methods

2.1. Plant species, study soil and experimental set-up

Soil was collected from the ultramafic area of Barazón (Galicia) in NW Spain (where *A. serpyllifolium* ssp. *lusitanicum* is a native species). Soils were air-dried, sieved through an 8-mm stainless steel sieve and homogenised. Soil treatments included untreated control (no amendment), inorganic fertilisation (NPK), and different rates of an organic amendment addition (commercial compost based on municipal solid wastes (MSW)). Inorganic fertilisers were added as pure chemical reagents: N was added at 100 kg ha⁻¹ as NH₄NO₃, and P and K were added as KH₂PO₄ at 100 and 125 kg ha⁻¹, respectively (based on a soil depth of 20 cm and bulk density of 1.1 g cm⁻³). The compost amendment was applied at three different rates of addition: 2.5%, 5% and 10% w/w. The commercial compost used in this study was provided by a local SME company dedicated to the recycling of waste products (Tratamientos Ecológicos del Noroeste (TEN) s.l.) and is primarily based on municipal sewage sludges and wood chippings. The main properties of the compost are a slightly acidic pH (6.2), high amount of nutrients such as available P (P_{olsen}: 425 mg kg⁻¹), organic C (22.2%) or total N (1.6%), and high cation exchange capacity (CEC: 57.6 cmol_c kg⁻¹). The CEC was dominated by Ca (40.4 cmol_c kg⁻¹), and exchangeable K was 4.4 cmol_c kg⁻¹. After incorporation of amendments the soils were adjusted to 70% of their water holding capacity (which was measured according to the method of Guitián and Carballas (1976)) and left for 2 weeks to attain equilibrium, after which time aliquots were taken of each treatment for soil analyses.

Seeds of *A. serpyllifolium* ssp. *lusitanicum* Dudley and P. Silva (hereafter referred to as *A. pintodasilvae*) from Barazón (NW Spain) and Samil (Trás-Os-Montes, NE Portugal), *A. serpyllifolium* ssp. *malacitanum* Rivas Goday (hereafter referred to as *A. malacitanum*) from Sierra Bermeja and Sierra de Aguas (S Spain), *A. bertolonii* ssp. *bertolonii* Desv from Imprunetta (Italy), and *N. goesingense* Hálácsy from Redlschlag (E Austria) were collected and germinated on a 2:1 perlite:quartz mixture (2:1 v/v) in a growth chamber under controlled conditions (temperature 22–25 °C, PPF (Photosynthetic photon flux density) of 190 μmol m⁻² s⁻¹, under a 16/8 h light/dark cycle). Seeds were watered daily with deionised water until germination and thereafter with a Ni-rich serpentine-like macro-nutrient solution which consisted of 2 mM MgSO₄, 0.8 mM Ca(NO₃)₂, 2.5 mM KNO₃, 0.1 mM K₂HPO₄, 20 μM FeEDDHA, 10 μM H₃BO₃, 2 μM MnCl₂, 1 μM ZnSO₄, 0.5 μM CuSO₄, 0.2 μM Na₂MoO₄ and 300 μM NiSO₄ (based on Chaney et al. (2008)). Plastic pots were filled with 1 kg of soil and one four-week-old seedling of each of the six tested taxa was transplanted to each pot. Five replicate pots were established for each soil treatment (untreated, NPK, 2.5%, 5% and 10%) and plants were grown in an environmentally controlled growth chamber for five months (temperature 22–25 °C, PPF of 190 μmol m⁻² s⁻¹, under a 16/8 h light/dark cycle).

2.2. Plant and soil analysis

General physicochemical properties and the total and available metal concentrations of all soils were determined before planting. Soil

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