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Short communication

Evaluation of phytostabilizer ability of three ruderal plants in mining soils restored by application of organic amendments

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ABSTRACT

Abandoned mines involve a serious environmental problem because these soils contain high levels of trace elements. The aim of the study was selecting the most adequate autochthonous plant species to stabilize contamination and improve soil quality of a contaminated soil from an abandoned mine. For this purpose three plant species were studied: *Poa annua*, *Medicago polymorpha* and *Malva sylvestris*, in combination with two organic amendments (biosolid compost (BC) and "alperujo" compost (AC)) and a soil without amendments (CO). Soil pH increased due to the effect of amendments under all studied plants, which promotes the immobilization of trace elements in soil. Water soluble C (WSC) increased with the addition of both amendments and the highest concentration was found in soils under *M. polymorpha*. The evolution of trace element availability in soil depended on the amendment, plant species and characteristics of the element. The best treatment to stimulate biomass production was AC. The highest concentration of As and Pb was found in *P. annua* whereas the highest concentration of Mn and Zn was found in *M. sylvestris*. Considering chemical and biochemical properties, and concentrations in shoots *M. polymorpha* would be the most suitable plant to stabilize trace elements and improve soil quality. Nevertheless, the best results were obtained with the plant-amendment combination.

1. Introduction

Phytostabilization is a phytoremediation technique that tries to limit the mobility and bioavailability of pollutants in soil by plant roots, and involves the establishment of vegetation on the contaminated site that enhances the value of the land (Ali et al., 2013). This technique exploits plant transpiration and root growth to reduce leaching and control erosion. Selection of appropriate vegetal species is an important factor for the success of phytoremediation techniques. These plants should accumulate trace elements (TE) from substrate into their roots but restrict their transport and entry into their aerial parts (Sheoran et al., 2011). Moreover, the chemical and biological reactions occurring in the rhizosphere play an important role in the bioavailability of TE. Plant roots can change the physical, chemical, and biological conditions of the soil in the rhizosphere and enrich it with organic substances of plant and microbial origin (Bolan et al., 2011).

Currently there is an increasing tendency in using autochthonous or non-invasive plants to decrease any harmful effects on the ecosystems by introducing new plants to the environment (Suchkova et al., 2014). Therefore exploring the potential of different adapted native species to these environmental conditions for the phytostabilization of TE contaminated soils is compulsory.

Contaminated soils usually presented unfavorable conditions for plant growth, therefore, the addition of amendments into this type of soils has a durable and positive effect on plant growth (Pérez de Mora et al., 2011) and can also enhance key biological processes affecting the immobilization of heavy metals (Martínez-Fernández et al., 2014) resulting in an improved quality of soil properties (Pérez de Mora et al., 2006).

The aim of this study was to evaluate the ability of three different species to stabilize TE at rhizosphere level under different physical and chemical conditions created in the soil by the addition of organic amendments.

2. Materials and methods

2.1. Soil and compost characterization

The experiment was carried out using soil from an abandoned mining area located in Tharsis (Huelva), where the main activity was the extraction of pyrite (FeS_2) (Madejón et al., 2011). This is a loamy soil with an acid pH (pH 5.5) and a relatively high content

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in organic matter (9%). Soil was collected from the upper 0–25 cm of soil.

Biosolid compost (BC) and "alperujo" compost (AC) were used as amendments. The principal properties of both composts were reported in Madejón et al. (2014).

2.2. Plant species

Three plant species were selected for the experiment: *Poa annua* L. (PO), *Medicago polymorpha* L. (ME) and *Malva sylvestris* L. (MA). These species are common in the Mediterranean areas and are considered as ruderal plants. These plants have the ability to proliferate and persist in high intensity disturbance and stress conditions so they grow in human-modified environments. As a matter of fact, these species readily colonize contaminated soils (Madejón et al., 2006).

2.3. Experimental design

The experiment was carried out in containers situated outdoors arranged according to a complete randomized block design. Three treatments were established: biosolid compost (BC), alperujo compost (AC) and a control (CO) without amendment. Four replicates per treatment and plant species were performed. A single addition of amendments (75 g/pot) was made and afterwards seeds of the species were established. Soil samplings were performed at 3 (initial sampling) and 182 (final sampling) days after the amendment addition.

The experiment was carried out for 6 months (April–October) and the pots were regularly irrigated by dripping (3 days per week) to ensure the plants water demand. From mid-July to mid-September there was not irrigation to simulate natural conditions in Mediterranean area. Plants were harvested in June and October (final sampling). The second sampling was carried out after a reseeding in September. Biomass and TE concentration in vegetal tissues were measured in both samplings.

2.4. Chemical and biochemical analysis

Soil pH was measured according to Hesse (1971). The available Cd, Cu, Mn, Pb and Zn concentrations in soils were determined as described Houba et al. (2000). Soil available-As (0.5 M $NaHCO_3$ extracts, 1:10 w/v) was measured by hydride generation ICP-OES. Pseudo-total TE concentrations in soil samples were determined by digestion with aqua regia in a microwave oven and TE in the extracts were determined by ICP-OES (inductively coupled plasma-optical emission spectrometry). Total organic carbon (TOC) was determined according to Walkley and Black (1934) and total Kjeldahl-N (TKN) by the method described by Hesse (1971). Water-soluble carbon (WSC) content was determined on using a TOC-VE Shimadzu analyser after extraction with water using a sample-to-extractant ratio of 1:10. Available-P and available-K was determined according to Olsen et al. (1954) and Dewis and Freitas (1970), respectively.

Dehydrogenase activity was determined by the method of Trevors (1984), β-glucosidase activity as indicated by Tabatabai (1982) and Phosphatase activity by the method proposed by Tabatabai and Bremmer (1970).

Vegetal material was washed with a 0.1 N HCl solution and with distilled water, then they were oven dried at 70 °C and finally grounded and passed through a 500-μm stainless-steel sieve. Dried plant samples were digested by wet oxidation with concentrated HNO_3 under pressure in a microwave oven. Determination of TE in the extracts was performed by ICP-OES. The accuracy of the analytical methods was assessed through a plant reference sample (INCT-TL-1, Tea leaves).

Table 1
Evolution of TOC, WSC, TNK and enzymatic activities in soils (mean values ± standard error, $n = 4$; 1, initial sampling; 2, final sampling). For each column, treatment and sampling values followed by different letter differ significantly ($p < 0.05$).

Treatment	Plant	Sampling	Chemical parameters				Microbiological parameters		
			pH	TOC (%)	WSC (mg kg^{-1})	TNK (%)	Dehydrogenase (μg INTF g^{-1} dry soil h^{-1})	Phosphatase (μmol PNF g^{-1} dry soil h^{-1})	β-Glucosidase (μmol PNF g^{-1} dry soil h^{-1})
CO	PO	1	5.30	5.35	422	0.39	3.48	7.31	3.21
		2	6.14	5.30 ± 0.36	543 ± 63.5ab	0.43 ± 0.02b	1.56 ± 0.40	4.78 ± 0.64	3.19 ± 0.52
	ME	1	5.30	4.16	441	0.41	3.10	5.93	4.26
		2	5.90	5.20 ± 0.16	581 ± 36.5b	0.45 ± 0.03b	1.28 ± 0.32	8.79 ± 1.13	2.85 ± 0.38
	MA	1	5.30	5.73	500	0.37	2.90	5.87	3.89
		2	6.10	5.20 ± 0.07	373 ± 15.0a	0.31 ± 0.02a	0.32 ± 0.08	4.62 ± 0.38	2.11 ± 0.25
BC	PO	1	6.48	6.20	1300	0.54	7.25	3.56	1.80
		2	6.76	6.52 ± 0.27b	10,853 ± 93.1	0.53 ± 0.06	3.14 ± 1.14	3.06 ± 0.38	1.48 ± 0.41
	ME	1	6.48	5.07	1150	0.54	5.96	3.20	2.94
		2	6.52	6.06 ± 0.15a	1326 ± 140	0.45 ± 0.03	2.10 ± 0.57	2.56 ± 0.31	2.20 ± 0.56
	MA	1	6.48	5.12	1430	0.57	5.54	4.53	2.68
		2	6.57	5.53 ± 0.04a	1060 ± 48	0.50 ± 0.01	2.95 ± 0.33	1.35 ± 0.18	1.82 ± 0.08
AC	PO	1	6.58	5.97	715	0.49	7.24	3.42	2.70
		2	6.61	5.57 ± 0.19	978 ± 66.3b	0.57 ± 0.04c	4.92 ± 1.22	3.06 ± 0.35	2.20 ± 0.09
	ME	1	6.58	6.40	783	0.65	5.63	3.87	2.81
		2	6.47	5.20 ± 0.16	1127 ± 7.5b	0.45 ± 0.03b	4.88 ± 0.41	3.70 ± 0.36	2.17 ± 0.09
	MA	1	6.58	5.37	782	0.48	4.79	3.54	2.33
		2	6.38	6.04 ± 0.06	602 ± 34a	0.31 ± 0.02a	2.44 ± 0.44	3.35 ± 0.54	1.38 ± 0.16