# Adaptation, tolerance, and evolution of plant species in a pyrite mine in response to contamination level and properties of mine tailings: sustainable rehabilitation

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#### Abstract

*Purpose* The impacts of mining contaminations and physicochemical properties and geochemistry of mine tailings on the density, richness, biodiversity, evolution and succession of plant species and vegetation recovery in the mining area is very poorly reported in the literature. Therefore, the present study conducted an investigation on vegetation development and succession of plant communities at the abandoned São Domingos pyrite mining area.

Materials and methods We conducted the field survey to estimate the vegetation development and succession of plant communities, collect vegetation (plant species, lichen and moss) and tailing (and soil) samples, and finally analyzed the physico-chemical and geochemical properties and metal levels in mine tailings, soil and vegetation samples.

Results and discussion The results showed that the communities of low height and biomass like grass, legume, shrub, moss and lichen were dominating on the mine tailings and waste dumps at the inner sites and center of the mine, and the vegetation coverage was explicitly very poor. The reddish brown colluvia had poor soil quality, but high acidity and metal concentrations. However, at the outer edge of the

Vegetation cover

1 Introduction

For the past years, the inadequate environmental standards and lack of enforcement of environmental laws encouraged the dumping of toxic mine tailings and waste in the surrounding environment that ultimately contaminated the gardens, agricultural fields, human habitat, surface water, streams and groundwater due to acidity release, low pH and leaching of toxic metals as for example, Al, As, Cd, Cr, Cu, Fe, Sb, Zn, etc. The disposal of mining wastes has harmful effects on and a potential threat to soil and water quality, terrestrial and aquatic plants, organisms, ecology, and the functional diversity of microbial communities causing the decreasing trend in

mine the loamy soil and relatively lower acidity and metal

contamination favored the higher vegetation cover and a

gradual increase in the number of species and plant succes-

sion, where the taller, higher biomass and broad leaf trees

were abundantly grown forming a dense forest and canopy.

The succession of several plant communities dominating in

the mining area, vegetation coverage and species richness

were strongly related to the different levels of contamina-

Conclusions Although the high concentrations of toxic trace

elements and low pH soil are important factors for limiting

the plant growth, however, proper soil development with

enriched nutrients and properties on mining wastes, by

either natural or external soil aided process, can help to

promote the high vegetation growth, mine rehabilitation

and ecological restoration of the mining degraded lands.

**Keywords** Mine rehabilitation · Metal toxicity · Mining

activities · Species evolution · Tailing and soil property ·

tion, soil properties and adverse factors of mine tailings.

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plant, animal and ecological biodiversity (Wong and Wong 1990; Kandeler et al. 1996; Ives and Cardinale 2004; Hsu et al. 2006). High concentrations of metals in crops, cereals, vegetables, tropical plants and animals may cause food chain contamination and acute toxicity effects in humans and other vertebrates (Parmeggiani 1983). Soil contaminated by high concentrations of acidity, and low pH generated from oxidation of sulfidic minerals (e.g., pyrite) is unfavorable for plant growth, because plants cannot take up enough essential nutrients from soils at low pH. Some studies reported that high concentrations of metal deposition in soil have reduced the plant growth causing forest decline (Herrick and Friedland 1990). The physiological and chemical data of phytochelatins, indicators of metal stress, have also suggested that exposure to high concentrations of metals in soils has damaged the growth of tree resulting in decline of forests (Gawel et al. 1996).

Mining activities have already caused the loss of biodiversity in many parts of the world. As, for example, at Ishizu, Agatsuma (Gunma prefecture), and Matsuo sulfur mines of Japan (Iwate prefecture), which are abandoned since 1969 and 1971, respectively, the soil is still bare with sparse withered trees; and grass is the main vegetation over wide areas indicating the very slow vegetation recovery (Shimai 1984; Takeuchi and Shimano 2009). Therefore, before adopting a mine rehabilitation and ecological restoration program in a mining affected area, an environmental assessment is required in order to understand the inherent causes for the influences of mining activities on the vegetation evolution and coverage. Due to large-scale open pit mining activities, a number of environmental problems such as land subsistence, poor soil and water quality, high metal contamination, limited vegetation, groundwater lowering, geological hazards, tailings disposal and infrastructure damage are observed at many mining areas in arid and semi-arid regions (Vangronsveld et al. 1995, 1996; Bagatto and Shorthouse 1999; Vidic et al. 2006; Hernández and Pastor 2008; Takeuchi and Shimano 2009; Anawar et al. 2011, etc.). To combat the consequences of land degradation and pollution from mining activities, some remedial measures, as for example, vegetation development, eco-restoration and phytoremediation, are required to stabilize these mining waste dumps (Wong 2003). Phytostabilization process results in a stabilization of the metal contaminants in tailing substrates, and reduction in transfer of the metal from soil to plants (Blaylock and Huang 2000). In fact, plants have mechanisms for accumulation, and tolerance or alleviation of high levels of metals in contaminated soils (Khan et al. 2000). However, plants experience toxicity due to lack of soil moisture content, low nutrients and high concentration of toxic metals in mine tailings (Maiti and Nandhini 2006).

In general, flux of contaminants in the environment and habitats damages the land and resources therein, and makes some sensitive plant species extinct. However, it is related to the tolerance or sensitivity of the species (Salminen et al. 2001). Long-term continuation of this process will disturb the balance of plant species living in a community, and thus, eventually disrupt the frequency distribution of plant species in the ecological environments. Therefore, vegetation coverage may be a suitable indicator of the environmental effects of mining activities. Takeuchi and Shimano (2009) reported that the scarce and slow vegetation recovery in sulfur mine occurred due to past smoke, sulfur dioxide, and ferric sulfide from mining operations, and the landslide disturbance. However, no studies clearly reported yet the impacts of contamination levels, and bioavailability of metals, physicochemical properties (e.g., pH, organic carbon (OC), cation exchange capacity (CEC) and nutrients) and geochemistry of mine tailings on the density, richness, biodiversity, evolution and succession of plant species and vegetation recovery in the pyrite mining area. Therefore, it was necessary to conduct a study for understanding the successional stages in vegetation recovery at the São Domingos pyrite mine to reflect the phenomena occurring in the pyrite and sulfide-rich base metals, coal and other mines globally. The present investigation was carried out to screen the native plant species with a view to using them for the vegetation-based mine rehabilitation and ecological restoration of the area. The preliminary objective of the present study was to analyze the essential (Fe, Na, K, Se and Zn) and non-essential (environmentally toxic) metals (As, Cr, and Sb) in mine tailings and plant species. However, the main objectives were to (1) assess the adaptation, tolerance, evolution, vegetation development and comparative coverage of different plant species at the different study sites in response to mining activities, physico-chemical properties of mine tailings, soil development, degradation and landscape, (2) evaluate the effects of metal contents on biodiversity and species richness of the plant communities, and (3) predict the sustainable approach for vegetation-based rehabilitation or ecological restoration of mines over both local and regional scales.

### 2 Materials and methods

# 2.1 Study area

The São Domingos copper sulfide mine, with more than 25 Mt of ore extracted (Gaspar 1998) is a pyrite orebody located on south-eastern part of Portugal, in Baixo Alentejo Province. The mineral assemblage composed by pyrite, sphalerite, chalcopyrite, galena, arsenopyrite, and sulfosalts; and the products were pyrite, roasted pyrite, sulfur, and copper. The mining activities promoted the development of a railway and harbor for ore transportation, two pyrite burning factories, water reservoirs, cementation tanks, network channels for acid water evaporation, and the mining village. As a consequence of this historical mining activity since pre-Roman times to the



1930–1960, different type of materials, such as metallurgical slags, sub-grade ore, pyrite ash, weathered host rock, and materials from gossan were deposited in the surrounding area of the open pit and downstream to Telheiro area. Most of the area is covered by thin soils, sparse vegetation and natural rock outcrops.

### 2.2 Sampling

The mine tailings, plant species, lichen and moss samples were collected from the nine study sites at the inner sites of the mine and the remaining samples were collected from other eight sites at the outer edge of the mining operations center (Fig. 1). The surrounding vegetation at the outer edge of the mine was established on land where soil, imported from outside the mine, was mixed with mine tailings to improve the soil properties and help soil development on mine tailings. Mine tailings and soil samples, developed on waste materials and weathered rocks, were collected in such a way that are representative, in the area, of the different tailings and soil environments. Mine tailings and soil samples were collected (0–15 cm depth) in a restricted circle about 60 cm around the plant species and consisted of a homogeneous sample (around 4 kg) of four subsamples. After that, the samples were dried at 50 °C to a constant weight, mixed, homogenized, and sieved through a 2-mm screen.

One hundred six plant samples were collected from the 17 different study sites of the mining affected area in the summer of 2009. The root, stems, leaves, and flowers of

plant species were separated. Roots were washed with tap water and rinsed with 0.1 M HCl solution followed by several rinses with de-ionized water. The shoots (stems, leaves, and flowers) of plant species were washed with tap water followed by several rinses with de-ionized water to remove completely particulate material attached to stem, leaves, and flowers (shoots). The well-cleaned and washed plant materials were freeze-dried, ground in Teflon (balls and capsule) mills, thoroughly homogenized, and made into 250-mg pellets for neutron activation analysis, following the k0-standardized procedure.

### 2.3 Soil characteristics

The <2-mm fraction of tailing and soil samples was used to determine the main soil properties: pH was determined potentiometrically in a soil paste saturated with water; OC was determined by dichromate oxidation using the Tiurin method (Jackson 1960); CEC was determined according to the ammonium acetate method by extracting with a 1.0 M NH<sub>4</sub>OAc solution (pH 7.0) (Tan 1996); and particle size distribution (sand, silt, and clay) was analyzed by the pipette method (Gee and Bauder 1986).

### 2.4 Elemental analysis

All elemental determinations of bulk tailings (and soil) and plant samples (three replicates for every sample) were carried out at the Portuguese Research Reactor of the Technological

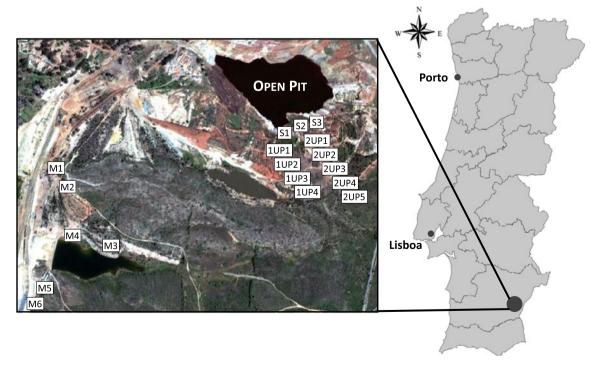


Fig. 1 A figure showing the position of the sampling sites in relation to the mine center



and Nuclear Institute (Sacavem; maximum nominal power: 1 MW), by K<sub>0</sub>-INAA. Typically, plant samples were irradiated for 5 h and soil samples for 1 h at thermal neutron fluxes of 2.25·10<sup>12</sup>n·cm<sup>-2</sup>·s<sup>-1</sup> for long irradiations, together with one disc (thickness: 125 μm; diameter: 5 mm) of an Al-0.1 % Au alloy as comparator. Gamma spectra were acquired on a liquid N<sub>2</sub>-cooled, ORTECÒ-calibrated, high-purity Ge detector (1.85 keV resolution at 1.33 MeV; 30 % relative efficiency). Samples were measured after 4 days and 4 weeks (long irradiations). The comparator was measured after 1 week (long irradiations). Elemental concentrations were assessed through the k0-IAEA program (version 3.21). Quality control was asserted by analyzing certified reference materials (IAEA-336 and soil-5) concurrently with the field samples. Deviations from certified values were generally within 1–15 %.

#### 3 Results

### 3.1 Physico-chemical properties

The results of OC, pH, CEC, and texture of mine tailings and waste from seventeen study sites are provided in Table 1. Mine tailings were extremely acidic due to pyrite oxidation, acid sulfate and poor soil development at the inner sites of the mine. The pH values varied from 2.8 to 4.8, a very lower range, at the sites adjacent to modern slags and waste dumps (e.g., sites 1up1, 1up2, 2up1, 2up2, 2up3, S1, S2, S3 and M1), while the values were near neutral and relatively higher (pH 4.8–7.0) at the outskirts/outer edge of

the mine. The texture of mine tailings at the inner sites of the mine was mostly thin layer of reddish brown coarse to medium grained sandy soil, clayey sand, nodular ferruginised colluvium and waste rocks, occasionally slopes, ditches and acid mine drainage (AMD), while those were sandy silt, loamy soil and clayey sand at the outer edge of the mine. The organic carbon contents and CEC in tailing samples at the inner sites of the mine were very low due to poor soil development over nodular ferruginised colluvium, while those were relatively higher at the outer edge of the mine (see Table 1).

### 3.2 Metal contents in mine tailings

The results of metal analysis revealed that mine tailings contained the high concentrations of potentially toxic trace elements like As, Sb and Cr (Fig. 2) as found in the previous study of Anawar et al. (2011). The concentrations of As, Sb and Fe varied 191–5984, 122–1933, and 66,335–298,300 mg/ kg, respectively, that were extremely high at sites 1up1, 1up2, 2up1, 2up2, 2up3, S1, S2, S3, M1, and M3 adjacent to modern slags. By contrast, the values ranged 5-648, 4-128, and 7,199-69,630 mg/kg at sites 1up4, 2up4, 2up5, M2, M4, M5 and M6, respectively, that were relatively lower but still 3–15 times higher than (1) guideline limit of As and Sb, (2) background concentration of 10 mgAs/kg in soils (Garcia-Sanchez and Alvarez-Ayuso 2003), (3) guideline limit of 50 mgAs/kg in agricultural soil (MAFF 1993), (4) average toxicity threshold of 40 mgAs/kg for crop plant (Sheppard 1992), (5) tolerable content of 5 mgSb/kg for agricultural soils

Table 1 Physico-chemical properties of tailings in São Domingos mine

Sample ID	Sites	pН	OC	CEC	Texture and description of tailings
1up1	Inner site	2.8	0.83	6.83	Thin layer of reddish brown coarse to medium grained sandy soil, nodular ferruginised colluvium
1up2	Inner site	3.4	1.60	7.46	Reddish brown coarse to medium grained clayey sand, nodular ferruginised colluvium
1up4	Outer edge	6.5	5.30	11.56	Reddish brown coarse to fine grained sandy silt
2up1	Inner site	4.8	1.20	5.26	Thin layer of reddish brown coarse to medium grained sandy soil, nodular ferruginised colluvium
2up2	Inner site	3.65	0.92	5.48	Thin layer of reddish brown coarse to medium grained sandy soil, nodular ferruginised colluvium
2up3	Inner site	3.49	3.10	7.54	Reddish brown medium to fine grained clayey sand
2up4	Outer edge	6.8	5.60	14.56	Reddish brown, coarse to fine grained sandy silt
2up5	Outer edge	7.0	4.80	11.42	Reddish brown, coarse to fine grained sandy silt
M1	Inner site	3.7	0.56	4.31	Thin layer of reddish brown coarse to medium grained sandy soil, nodular ferruginised colluvium
M2	Outer edge	5.8	4.80	11.85	Reddish brown, coarse to fine grained sandy silt
M3	Outer edge	4.8	3.10	10.52	Reddish brown, coarse to fine grained sandy clay
M4	Outer edge	6.5	2.10	15.24	Reddish brown, coarse to fine grained sandy silt
M5	Outer edge	6.8	7.50	13.14	Reddish brown, coarse to fine grained sandy silt
M6	Outer edge	6.9	7.60	17.65	Reddish brown, coarse to fine grained sandy silt
S1	Inner site	4.2	1.20	3.66	Thin layer of reddish brown coarse to medium grained sandy soil, nodular ferruginised colluvium
S2	Inner site	3.5	0.54	2.94	Thin layer of reddish brown coarse to medium grained sandy soil, nodular ferruginised colluvium
S3	Inner site	3.8	2.50	4.56	Thin layer of reddish brown coarse to medium grained sandy soil, nodular ferruginised colluvium

OC in wt% and CEC in meq 100 g<sup>-1</sup>



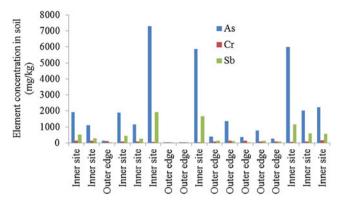


Fig. 2 Concentrations of As, Cr, and Sb in mine tailings

(Hammel et al. 2000) and (6) normal soil range of 10 mgSb/kg (Bowen 1979), at the outer edge of the mine. The amount of Fe was very high compared to other metals, and the concentrations of metals were in the order of Fe>As>Sb>Zn>Cr> Rb>Se>Sc>Co (Table 2). Zinc showed the opposite trend of distribution compared to other metals. Higher concentrations of Zn, near-neutral pH value and considerably lower iron (iron oxides) values at the outer edge of the mine can be related to rock weathering and soil formation processes with relatively less influences of AMD and mining activities. Other trace elements like Co, Cr, Rb, Sc, and Se were detected in relatively low quantities with maximum values of 26.8, 160, 207, 25, and 184 mg/kg, respectively; and the values were approximately same at all study sites indicating that these elements originated from the non-mineralization and rock weathering.

# 3.3 Landscape, evolution, and botanical composition of plants at different sites

All plant species at each study site were classified using Raunkiaer's life forms (Miyawaki et al. 1994) to understand the successional stages and relative cover of species of each life form of the communities (Shimano 2007). This method illustrates both quantitative and qualitative differences in plant communities according to the life cycle, life style, growth form, modes of dispersion, and light tolerance, described in Tables 3, 4 and 5.

# 3.3.1 Botanical composition of species on ferruginised colluvium, modern slags, and waste dumps at inner sites

The botanical composition of plant species at S1, S2, S3, M1, 1up1, 1up2, 2up1, and 2up2 sites is almost similar. The elevated metal levels (see Table 2) and lower pH, OC, and CEC values (see Table 1) of tailings as well as poor soil development at these sites had exerted a profound influence on the development of the plant species. These sites had sparse vegetation, predominantly composed of varying sized patches of the following species: *Agrostis castellana*,

ble 2 Average concentrations of trace elements and iron in tailings of São Domingos mine (mg/kg)

Table 7 Av	Table 2 Average concentrations of trace elements and non in tal	ations of t	iace elelli	ents and r	IOII III tai	ings on	Sao Dolli	ı) aııııı soğu	ng/kg)										
Sample ID	Sites	As	Std	Со	Std	Cr	Std	Fe	Std	Rb	Std	Sb	Std	Sc	Std	Se	Std	Zn	Std
1up1	Inner site	1,910	8.1	4.3	0.4	137	16.3	131,400	4,526	65	17.4	514	70.6	10.1	8.0	50.2	2	69	6.3
1up2	Inner site	1,095	35.2	2.6	0.2	151	3.6	212,250	7,425	89	15.7	280	9.5	9.8	0.2	31.4	4.1	98	1.9
1up4	Outer edge	132	3.2	4.6	0.1	110	0.5	43,445	191	207	2.2	27	17.7	24.2	0.4	5.9	0.2	172	1.8
2up1	Inner site	1,897	3.5	3.5	0.4	79	0.5	123,900	1,980	74	1	440	22.3	11.9	0.2	39.3	1.7	77	2.2
2up2	Inner site	1,169	111	2	0.1	85	1.3	116,950	5,162	92	4.7	265	11.7	14	0.3	17.3	3.6	71	6.6
2up3	Inner site	7,293	198	2.4	8.0	54	9.4	298,300	2,687	70	7.9	1,933	9.99	8.1	0.2	184	9.0	65	27.7
2up4	Outer edge	37	1.4	3.4	0.4	34	1.7	23,180	509	35	5.2	7	0.3	10.4	0.2	8.1	6.0	102	0.5
2up5	Outer edge	25	8.0	_	0.02	5	1.7	7,199	446	64	2.2	4	9.0	4.7	0.5	29	5.4	75	10.6
M	Inner site	5,874	93.5	5.5	9.0	43	9	196,950	3,041	92	0.4	1,668	4.9	6.4	0.5	125	3.7	229	11
M2	Outer edge	386	151	12.9	2.5	82	20.4	59,805	12,367	71	26.7	126	59	20.7	6.4	21.6	3.6	417	31
M3	Outer edge	1,372	105	2.4	0.1	137	2.1	66,335	389	59	2.3	122	12.6	25	1.5	14.9	1.9	171	34
M4	Outer edge	360	5.2	1.7	0.3	143	7.5	42,780	066	102	40.3	23	0.4	13.5	0.1	11.7	1.6	246	15
M5	Outer edge	771	308	11.4	1.5	83	38	61,305	20,145	73	51	128	2.79	18.5	4	22.9	7.7	433	9.8
M6	Outer edge	260	35.6	26.8	24.1	98	53.7	69,630	25,682	116	74	84	18.5	17.1	5.1	7.9	6.0	240	71
S1	Inner site	5,984	1,345	3.12	0.34	59	12.15	218,512	39,458	83	5.4	1,165	157	18	2.4	105.2	21.5	63	11.4
S2	Inner site	2,019	316	4.56	0.73	06	9.46	95,231	3,846	75	6.8	589	89	11.5	1.6	9.69	6.4	117	15.7
S3	Inner site	2,216	478	8.8	0.55	160	15.17	105,355	7,124	06	7.5	578	37	12.8	3.4	42.6	1.7	68	10



**Table 3** The life cycle, life style, growth form, mode of dispersion and light tolerance of plant communities at sites S1–S3

Site	Plant species	LC	LS	GF	MD	LT	Height
1	Agrostis castellana (Boiss. & Reut)	P	Н	AD	A	Н	30–50 cm
1	Andryala ragusina L.	P	Н	V	A	Н	30-50 cm
1	Carlina corymbosa L.	P	Н	V	A	Н	30-50 cm
1	Corrigiola telephiipholia Pourret	P	Ph	AD	Au	ST	1-30 cm
1	Cistus ladanifer L	P	Ph	V	Z	Н	>50 cm
1	Daphne gnidium L	P	Ph	V	Z	Н	>50 cm
1	Evernia prunastri (L.) Ach	A	Н	AD	Au	ST	1-30 cm
1	Erica australis L.	P	Ph	V	Z	Н	>50 cm
1	Erythraea pulchella Horn.	A	Ph	V	Z	Н	1-30 cm
1	Lavandula stoechas sub. Luisieri L.	P	Ph	AD	Z	Н	>50 cm
1	Moss	A	Н	AD	Au	ST	1-30 cm
1	Rumex induratus Boiss. & Reuter	P	Ph	AD	Z	ST	>50 cm
1	Rumex scutatusL.	P	Ph	AD	Z	ST	>50 cm
2	Agrostis castellana (Boiss. & Reut)	P	Н	AD	A	Н	30-50 cm
2	Cistus ladanifer L	P	Ph	V	Z	Н	>50 cm
2	Erica australis L.	P	Ph	V	Z	Н	>50 cm
2	Evernia prunastri (L.) Ach	A	Н	AD	Au	ST	1-30 cm
2	Lavandula stoechas sub. Luisieri L.	P	Ph	AD	Z	Н	>50 cm
2	Moss	A	Н	AD	Au	ST	1-30 cm
2	Rumex induratus Boiss. & Reuter	P	Ph	AD	Z	ST	>50 cm
2	Rumex scutatusL.	P	Ph	AD	Z	ST	>50 cm
3	Agrostis castellana (Boiss. & Reut)	P	Н	AD	A	Н	30-50 cm
3	Carlina corymbosa L.	P	Н	V	A	Н	30-50 cm
3	Cistus monspeliensis L.	P	Ph	V	Z	Н	>50 cm
3	Cistus ladanifer L	P	Ph	V	Z	Н	>50 cm
3	Erica australis L.	P	Ph	V	Z	Н	>50 cm
3	Erythraea pulchella Horn.	A	Ph	V	Z	Н	1-30 cm
3	Evernia prunastri (L.) Ach	A	Н	AD	Au	ST	1-30 cm
3	Helichrysum stoechas (L.) Moench.	P	Ph	AD	A	Н	>50 cm
3	Lavandula stoechas L.	P	Ph	AD	Z	Н	>50 cm
3	Rumex induratus Boiss. & Reuter	P	Ph	AD	Z	ST	>50 cm
3	Timus mastichina	P	Ph	AD	Z	Н	30-50 cm

LC life cycle, A species annuals, P Species perennials, LS life style, T therophytes, G geophytes, H hemicryptophytes, C Cametófito, Ph phanerophytes, GF growth form, V Vertical, R Rosetta, H Horizontal, AD all directions, MD modes of dispersion A Anemócora, Au Autocoro, B Barócora, Z Zoocóra, LT light tolerance, ST shade tolerant, H heliophilous

Andryala ragusina, Carlina corymbosa, Corrigiola telephiipholia, Cistus monspeliensis, Cistus ladanifer, Daphne gnidium, Daucus carota, Erica australis, Erythraea pulchella, Helichrysum stoechas, Lavandula stoechas, Rumex induratus, Scirpus holoschoenus, and Thymus mastichina. Out of all species, only Erica australis had established a greater cover than other species, but neither Eucalyptus globulus or Pinus pinaster or Pteridium aquilinum or Quercus ilex tree had grown at these sites. The more acidic nodular ferruginised colluvium and waste rocks at these sites favored the establishment of the weed grass, a few shrub, legume, semiaquatic plants, lichen and moss. The life cycles of the most species are perennial with an exception of three species as annual viz. E. pulchella, Evernia prunastri and moss (see Tables 3, 4, 5). The life styles of two third species are phanerophyte and one third is hemicryptophyte. The growth forms of about two third species are in all directions and one third is vertical. The modes of dispersion of about two third species are zoocóra and the rest of them are anemócora and autocoro. The light tolerance of about two third species is heliophilous and the rest are shade tolerant.

### 3.3.2 Botanical composition of species at sandy muddy sites

These sites (sites 1up3, 2up3, and M3) are low-lying lands and ditches with muddy tailings. The tailings are acidic (very low pH), but contain extremely high concentrations of As, Sb, and Fe (see Table 2). Only one plant species, *E. australis* and lichen, *E. prunastri* have predominantly developed; and the botanical composition of these species is given in Tables 4 and 5.



**Table 4** The life cycle, life style, growth form, mode of dispersion and light tolerance of plant communities at sites 1up1-1up4 and 2up1-2up5

Site	Plant species	LC	LS	GF	MD	LT	Height
1up-1	Agrostis castellana (Boiss. & Reut).	P	Н	AD	A	Н	30–50 cm
	Cistus ladanifer L	P	Ph	V	Z	H	>50 cm
	Cistus monspeliensis L.	P	Ph	V	Z	Н	>50 cm
	Erica australis L.	P	Ph	V	Z	Н	>50 cm
	Genista hirsuta Vahl	P	Ph	V	Z	ST	>50 cm
	Helychysum stoechas (L.) Moench.	P	Ph	AD	A	H	>50 cm
	Lavandula stoechas L.	P	Ph	AD	Z	H	>50 cm
1up-2	Agrostis castellana (Boiss. & Reut).	P	Н	AD	A	Н	30–50 cm
	Cistus salvifolius L.	P	Ph	V	Z	ST	>50 cm
	Cistus ladanifer L	P	Ph	V	Z	H	>50 cm
	Cistus monspeliensis L.	P	Ph	V	Z	H	>50 cm
	Daphne gnidium L	P	Ph	V	Z	H	>50 cm
	Daucus carota L.	A	Ph	V	Z	ST	>50 cm
	Lavandula stoechas L.	P	Ph	AD	Z	H	>50 cm
	Rumex induratus Boiss. & Reuter	P	Ph	AD	Z	ST	>50 cm
1up-3	Erica australis L.	P	Ph	V	Z	Н	>50 cm
	Evernia prunastri (L.) Ach	A	Н	AD	Au	ST	1-30 cm
1up-4	Carlina corymbosa L.	P	Н	V	A	H	30-50 cm
	Cistus ladanifer L	P	Ph	V	Z	H	>50 cm
	Daphne gnidium L	P	Ph	V	Z	H	>50 cm
	Erica australis L.	P	Ph	V	Z	H	>50 cm
	Eucalyptus globulus Labill.	P	Ph	V	Z	ST	>50 cm
	Evernia prunastri (L.) Ach	A	Н	AD	Au	ST	1-30 cm
	Lavandula stoechas L.	P	Ph	AD	Z	Н	>50 cm
	Pinus pinaster Ait	P	Ph	V	A	ST	>50 cm
	Quercus ilex L. subsp. Balota Samp.	P	Ph	V	Z	ST	>50 cm
2up-1	Cistus ladanifer L	P	Ph	V	Z	H	>50 cm
	Erica australis L.	P	Ph	V	Z	Н	>50 cm
	Eucalyptus globulus Labill.	P	Ph	V	Z	ST	>50 cm
	Evernia prunastri (L.) Ach	A	Н	AD	Au	ST	1-30 cm
2up-2	Erica australis L.	P	Ph	V	Z	Н	>50 cm
	Cistus ladanifer L	P	Ph	V	Z	Н	>50 cm
	Eucalyptus globulus Labill.	P	Ph	V	Z	ST	>50 cm
2up-3	Erica australis L.	P	Ph	V	Z	Н	>50 cm
	Evernia prunastri (L.) Ach	A	Н	AD	Au	ST	1-30 cm
2up-4	Carlina corymbosa L.	P	Н	V	A	Н	30-50 cm
	Cistus ladanifer L	P	Ph	V	Z	Н	>50 cm
	Daphne gnidium L	P	Ph	V	Z	Н	>50 cm
	Erica australis L.	P	Ph	V	Z	Н	>50 cm
	Eucalyptus globulus Labill.	P	Ph	V	Z	ST	>50 cm
	Evernia prunastri (L.) Ach	A	Н	AD	Au	ST	1-30 cm
	Lavandula stoechas L.	P	Ph	AD	Z	Н	>50 cm
	Pinus pinaster Ait	P	Ph	V	A	ST	>50 cm
	Quercus ilex L. subsp. Balota Samp.	P	Ph	V	Z	ST	>50 cm
2up-5	Cistus ladanifer L	P	Ph	V	Z	Н	>50 cm
-	Daphne gnidium L	P	Ph	V	Z	Н	>50 cm
	Eucalyptus globulus Labill.	P	Ph	V	Z	ST	>50 cm
	Lavandula stoechas L.	P	Ph	AD	Z	Н	>50 cm
	Pinus pinaster Ait	P	Ph	V	A	ST	>50 cm
	Quercus ilex L. subsp. Balota Samp.	P	Ph	V	Z	ST	>50 cm



**Table 5** The life cycle, life style, growth form, mode of dispersion and light tolerance of plant communities at sites M1–M6.

Site	Plant species	LC	LS	GF	MD	LT	Height
M1	Daucus carota L.	A	Ph	V	Z	ST	>50 cm
	Daphne gnidium L	P	Ph	V	Z	Н	>50 cm
	Lavandula stoechas L.	P	Ph	AD	Z	Н	>50 cm
	Rumex induratus Boiss. & Reuter	P	Ph	AD	Z	ST	>50 cm
M2	Daphne gnidium L	P	Ph	V	Z	Н	>50 cm
	Erica australis L.	P	Ph	V	Z	Н	>50 cm
	Eucalyptus globulus Labill.	P	Ph	V	Z	ST	>50 cm
	Helychysum stoechas (L.) Moench.	P	Ph	AD	A	Н	>50 cm
	Lavandula stoechas L.	P	Ph	AD	Z	Н	>50 cm
	Pistacia terebinthus L	P	Ph	V	Z	ST	>50 cm
	Pteridium aquilinum (L.) Kuhn	P	Н	V	Au	ST	>50 cm
M3	Erica australis L.	P	Ph	V	Z	Н	>50 cm
	Eucalyptus globulus Labill.	P	Ph	V	Z	ST	>50 cm
	Pinus pinaster Ait	P	Ph	V	A	ST	>50 cm
	Quercus ilex L. subsp. Balota Samp.	P	Ph	V	Z	ST	>50 cm
M4	Cistus ladanifer L	P	Ph	V	Z	Н	>50 cm
	Daphne gnidium L	P	Ph	V	Z	Н	>50 cm
	Erica australis L.	P	Ph	V	Z	Н	>50 cm
	Eucalyptus globulus Labill.	P	Ph	V	Z	ST	>50 cm
	Pinus pinaster Ait	P	Ph	V	A	ST	>50 cm
	Quercus ilex L. subsp. Balota Samp.	P	Ph	V	Z	ST	>50 cm
M5	Cistus ladanifer L	P	Ph	V	Z	Н	>50 cm
	Erica australis L.	P	Ph	V	Z	Н	>50 cm
	Eucalyptus globulus Labill.	P	Ph	V	Z	ST	>50 cm
	Pinus pinaster Ait	P	Ph	V	A	ST	>50 cm
	Quercus ilex L. subsp. Balota Samp.	P	Ph	V	Z	ST	>50 cm
M6	Cistus ladanifer L	P	Ph	V	Z	Н	>50 cm
	Erica australis L.	P	Ph	V	Z	Н	>50 cm
	Eucalyptus globulus Labill.	P	Ph	V	Z	ST	>50 cm
	Pinus pinaster Ait	P	Ph	V	A	ST	>50 cm
	Quercus ilex L. subsp. Balota Samp.	P	Ph	V	Z	ST	>50 cm

# 3.3.3 Botanical composition of species on sandy silt and loamy soil at outer sites

The botanical composition of plant species at 1up4, 2up4, 2up5, M2, M4, M5, and M6 sites is more diverse than the previous sites (see Tables 4 and 5). In addition to the weed grass, shrub, legume, semi-aquatic plants, lichen and moss, mentioned in section 3.3.1 and 3.3.2, the taller broad leaf trees including *E. globulus*, *P. pinaster*, *P. aquilinum*, *Q. ilex* etc. species had grown at these sites. The communities, developed here, represent the relatively lower influence of metal deposition and mining impacts.

### 3.4 Metal contents in plants

The plants demonstrated the differential accumulation pattern of metals and metalloids (Table 6). In general, with a

few exceptions, various plants and lichen showed the maximum accumulation of K in their shoot followed by Na, Fe, Zn, and Ba. Other elements such as As, Br, Co, Cr, Se, and Sb were very low in concentrations with the least concentration of Co. Among the various plants, C. telephiipholia was found to be the best accumulator of various elements, especially Fe, K, Zn, and As followed by T. mastichina, E. australis, and L. stoechas (Fig. 3). Other plant species such as, E. globulus, P. pinaster, P. aquilinum and Q. ilex showed comparatively very low potential of accumulating various metals. Although some plants accumulated relatively higher concentrations of elements, they are still not phytoaccumulator or hyperaccumulator, because they accumulate less than the threshold value for hyperaccumulation as mentioned below. A common threshold accumulation concentration for plants to be hyperaccumulator for most trace elements is 0.1 %. As



Table 6 Concentrations of different elements in the shoot of plant species grown at São Domingos mine (mg/kg)

Sampling site	Plant description		As	Ba	Br	Co	Cr	Fe	K	Na	Sb	Se	Zn
1up2, S1	Agrostis castellana	Average	3.4	19.2	6.8	0.4	7.9	247	6,339	354	0.6	0.8	35
		Std	0.2		4.0		2.9	11		12	0.0	0.14	2
S1	Andrela raqusina	Average	2.5	24.3	5.2	0.5	5.9	254	20,680	1,543	0.8	0.3	107
		Std	0.2	34.4		0.02	0.5	25	339	26	0.6	0.0	0.07
S1,3	Carlina corymbosa	Average	10.9	41	12.3	0.2	6.4	570	12,842	3,719	1.6	6.3	65.0
		Std	0.03	2.3	10.7	0.10	1.3	252	7,181	4,227	0.02	1.7	8.1
S3	Cistus hirsutus		5.5	62.2	4.9	0.3	3.0	237	7,797	236	0.8	2.5	116
2up1, S1-,3	Cistus ladanifer	Average	7.8	6.4	11.7	2.4	7.2	467	7,224	156	1.2	2.4	112
		Std	6.2	9.1	3.1	2.9	4.7	147	3,657	28	1.5	2.4	77
S1	Corrigiola telephiipholia	Average	16.1		42.7	1.9	5.8	986	24,520	5,491	3.6	1.8	246
		Std	0.3		1.8	0.02	0.6	27	1,146	165	0.3	0.1	4.5
S1,3	Daphne gnidium	Average	2.7	14.8	1.0	0.1	4.4	138	11,710	139	0.3	11	92
		Std	1.8		0.1	0.02	0.3	29	2,503	21.1	0.2	6.6	
S1,3	Erythraea pulchella	Average	14	29	2.0	0.2	4.9	611	13,435	349	2.7	5.0	37.0
		Std	2.0		1.0	0.1	0.5	18	177	1.1	0.9	0.8	2.3
1up1,3,4, M3,5	Erica australis	Average	6.4	18.3	1.3	0.2	6.3	360	4,926	398	0.70	0.9	25
2up2,3,4		Std	0.2	2.9	0.1	0.03	0.7	13	109	13	0.03	0.1	1.4
1up4, S3	Evernia prunastri	Average	12.4	15.1	8.2	0.3	10.6	701	1,640	318	1.5	0.4	41
		Std	5.1		2.1	0.06	2.2	97	263	3	0.46	0.027	0.72
2up1, 2up4	Eucalyptus globulus	Average	0.9	9.6	15.6	0.3	6.0	56	9,463	1,921	0.05	1.7	31
M2, M5		Std	0.5		0.17	0.07	0.4	7.41	82	21	0.02	0.14	1.6
1up1, S3	Helychysum stoechas	Average	6.2	26.7	9.3	0.3	8.1	331	17,123	1,360	1.7	4.4	91
		Std	0.6	6.1	0.1	0.1	1.4	11.1	74	24	1.7	0.3	0.8
S1	Lavandula stoechas		14.2	74.0	8.8	0.4	6.6	1,393	11,620	1,533	1.9	3.1	134
1up4, 2up4	Pinus pinaster	Average	3.0	7.5	2.9	0.4	4.6	106.9	5,066	361	0.10	0.4	48.3
M3,4,6		Std	0.1	0.7	0.1	0.04	0.7	71	314	46	0.1	0.3	3.8
M2	Pistacia terebinthus						2.6	90	11,220	138			17
M2	Pteridium aquilinum	Average	2.4	12.2	22.0	0.61	3.3	178	18,845	298	0.27	0.29	56
		Std	0.1		0.9	0.021	0.6	13	926	15	0.03	0.11	6
M3,4,5	Quercus ilex	Average	1.2	102	16.2	0.35	7.5	549	8,758	2,014	0.06	1.9	58
		Std	0.4		0.21	0.05	0.5	6.9	132	35	0.03	0.12	4.7
1up2, S1, S3	Rumex induratus	Average	1.3	15.9	28.8	0.2	7.1	75.2	14,055	413	0.1	3.1	23.7
		Std	1.9	14.3	1.40	0.05	0.4	21.2	3,677.8	207.5	0.2	1.8	5.1
S3	Timus mastichina		16.8	58.9	3.3	0.1	5.8	449	15,280	319	2.3	4.9	35

for example, the threshold concentration for arsenic hyperaccumulation is greater than 0.1 %, for zinc and manganese, the threshold concentration is 1 % and for cadmium, the threshold concentration is 0.01 % (Reeves et al. 1995; Ma et al. 2001). However, these plants are tolerant to extreme environmental conditions of mining area. Iron accumulation in all the plant species was found to be higher because of the high Fe content in mine tailings. Concentrations of As and Sb in plant species were generally low with some exceptions (see Fig. 3), that are higher than the toxicity threshold limit as suggested by Alloway (1990). The levels of As and Sb in the plants do not reflect their concentrations in mine tailings indicating the low bioavailability of these elements.

# 4 Discussion

4.1 Physico-chemical property and metal contents of tailings and vegetation development

The great variability of pH, OC, CEC, and texture in mine tailings at the inner sites and outer edge of the mine governed the variable cover of vegetation and diversity of plant species. Low pH soil prevented plant roots from absorbing minerals (Yuasa et al. 1995) resulting in low growth and cover of vegetation at the inner site of the mine. However, there is a greater extent and variety of vegetation cover under the nearneutral pH conditions at the outer edge of the mine, because that soil status improved seed germination and survival at even elevated soil metal concentrations. Higher organic



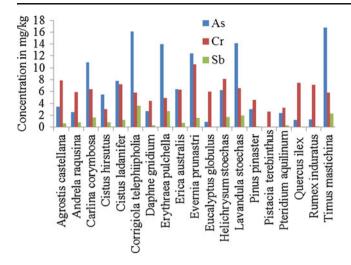


Fig. 3 Uptake of As, Cr, and Sb by different plant species

carbon contents and CEC in tailings at the outer edge of the mine provided a good nutrient source for plant growth (Chiu et al. 2006). At the inner sites of the mine, the patchy cover of vegetation, bare tailings, waste dumps and deforestation were widespread, that was vulnerable to degradation, aerial dispersion and rainfall transport; however, after the mine was abandoned for long time, the original broadleaf forest and tree-type plants with a canopy had recovered at the outer edge of the mine. Some previous studies observed the same phenomena in Agatsuma sulfur mine in Gunma prefecture of Japan (Oshima 1988; Oguro et al. 1990; Takeuchi and Shimano 2009). The adaptation, tolerance, evolution of different plant species, vegetation recovering, a gradual increase in the number of species and plant succession were positively correlated with soil development stage and properties at the São Domingos mine. The grass, legume, and shrub community that generally occurs in the early stages of succession, formed patchy communities on the bare mine tailings and wastes (Nakamura 1986).

At the center of the mine grass and shrub-type plants like E. australis, Erica andevalensis, Agrostis castellana, Agrostis delicatula, Rumex induratus, Genista hirsuta, and Daucus carota were the most abundant tolerant plant species. There were no tree-type taller plants, and the diversity of higher plant species was extremely low at the inner sites and center of the mine. By contrast, at the outer edge of the mine, the taller tree type plants, grass, legumes and shrubs formed the abundant and dense forest growth. The concentrations of metal contamination in mine tailings along with the pH, OC, CEC, and soil texture had significant effects on plant biodiversity and species richness of this study area. Hernández and Pastor (2008) also reported that soil heavy metal and Na concentrations, along with pH, had intense negative effects on plant biodiversity and species richness, followed by bioavailable and soluble metal contents in the topsoil layer of grasslands overlying an abandoned mine.

The present study also found that soil properties, metal and nutrient contents are the best soil-related predictors of species diversity, while soil contamination and nutritional disturbances contribute significantly to the vegetation damage in the ecosystems (Koptsik et al. 2003). The near-neutral pH values, high concentrations of OC, CEC, and Zn, enriched nutrient status as well as good soil quality (loamy soil, sandy silt, and silty clay), but relatively lower levels of toxic metals may result in significant establishment of taller and broadleaved tree-types plants, highly dense forest and the most diverse plant communities at the mining areas. The differences in post-mining soil development led to the several successional steps of plant communities. *E. australis* was found to grow abundantly throughout the mining area.

### 4.2 Sustainable vegetation development and phytostabilization

The proper selection of specific plant species out of all indigenous plants endemic to São Domingos metalliferous mine, which are often better in terms of survival, growth and reproduction, may be an adequate approach for sustainable vegetation development and phytostabilization of degraded mining areas (Pitchel et al. 2000; Yoon et al. 2006). Due to the high biomass, but relatively lower metal accumulation capacity in the aboveground tissues, the plant species such as E. globulus, P. pinaster, P. aquilinum and O. ilex etc. and some shrubs, legumes and semi-aquatic plants like E. australis, R. induratus, Cistus genus, and D. gnidium can be very effective for phytostabilization of the contaminated mining soils. The mine rehabilitation and ecological restoration could benefit from a broader perspective including different groups of plant species as they could perform distinct functional roles in the remediation process. For example, the use of legumes may enrich the soil nutrients content and the combined used of perennials and annuals grown in the present study areas of São Domingos mine can provide substantial inputs in terms of organic matter and nutrients recycling, thus contributing in distinct ways to the development of the soil (Hooper and Vitousek 1997, 1998). This study also clearly demonstrated that metal level is not the only adverse factor controlling the plant diversity and vegetation development on mine tailings, but also soil quality, pH level, OC, CEC, and nutrient status are the ancillary and main factors for the biodiversity, species richness and forest type vegetation development. Therefore, a proper management practice for soil development on mine tailings is recommended as follows: mixing loamy soil with nodular ferruginised colluvium, waste rocks and reddish brown sandy tailings at the center and inner sites of the mine. This management practice will dilute and reduce the metal levels and simultaneously, will augment the soil quality, pH level, OC, CEC, and nutrient status as well as promote the growth of tree type forest on tailings. This sustainable vegetation development will



present a biotechnology-based remediation option for the degraded mining sites; and simultaneously, this technique will be ancillary for the carbon-storage, and reduction of greenhouse gases and climatic change.

### **5 Conclusions**

The results of this study demonstrated that the tailings of poor soil quality, thin soil layer and high metal contents were the main restraints for vegetation growth at the center and inner sites of the mine, while the soil of good quality and low metal contents were highly favorable for plant growth, mine rehabilitation and ecological restoration of mining contaminated lands. However, some study sites indicated that although they had high metal contents, but their very good soil quality produced excellent vegetation growth, tree and forest development. The results of this study suggested that factors of soil quality predominated over high metal content for vegetation growth. Natural soil development on tailings, waste rock, spoils and dumps is a slow process. Therefore, mixing of soil (from outside of the mine) with these tailings is recommended, that will dilute the high metal content, accelerate the good soil development and ultimately result in excellent vegetation growth. These findings can be modeled for the ecological restoration and remediation of the mine tailings and contaminated areas.

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