Environ Geochem Health (2006) 28:487–498
DOI 10.1007/s10653-006-9058-9

ORIGINAL PAPER

Mercury accumulation in soils and plants in the Almadén mining district, Spain: one of the most contaminated sites on Earth

José Antonio Molina · Roberto Oyarzun · José María Esbrí · Pablo Higueras

Received: 27 December 2005 / Accepted: 19 July 2006 / Published online: 22 September 2006

Abstract Although mercury (Hg) mining in the Almadén district ceased in May 2002, the consequences of 2000 years of mining in the district has resulted in the dissemination of Hg into the surrounding environment where it poses an evident risk to biota and human health. This risk needs to be properly evaluated. The uptake of Hg has been found to be plant-specific. To establish the different manners in which plants absorb Hg, we carried out a survey of Hg levels in the soils and plants in the most representative habitats of this Mediterranean area and found that the Hg concentrations varied greatly and were dependent on the sample being tested (0.13–2,695 μg g^{-1} Hg). For example, the root samples had concentrations ranging from 0.06 (*Oenanthe crocata*, *Rumex induratus*) to 1095 (*Polypogon monspeliensis*) μg g^{-1} Hg, while in the leaf samples, the range was from 0.16 (*Cyperus longus*) to 1278 (*Polypogon monspeliensis*) μg g^{-1} Hg. There are four well-differentiated patterns of Hg uptake: (1) the rate of uptake is constant, independent of Hg concentration in the soil (e.g., *Pistacia lentiscus*, *Quercus rotundifolia*); (2) after an initial linear relationship between uptake and soil concentration, no further increase in Hg_{plant} is observed (e.g., *Asparagus acutifolius*, *Cistus ladanifer*); (3) no increase in uptake is recorded until a threshold is surpassed, and thereafter a linear relationship between Hg_{plant} and Hg_{soil} is established (e.g., *Rumex bucephalophorus*, *Cistus crispus*); (4) there is no relationship between Hg_{plant} and Hg_{soil} (e.g., *Oenanthe crocata* and *Cistus monspeliensis*). Overall, the Hg concentrations found in plants from the Almadén district clearly reflect the importance of contamination processes throughout the study region.

Keywords Biotypes · Mediterranean region · Mercury bioaccumulation · Plant uptake · Toxicity levels · ZAAS-HMF

J. A. Molina
Departamento de Biología Vegetal II,
Facultad de Farmacia, Universidad Complutense,
Madrid, Spain

R. Oyarzun (✉)
Departamento de Cristalografía y Mineralogía,
Facultad de Ciencias Geológicas, Universidad
Complutense, Avenida Complutense,
28040 Madrid, Spain
e-mail: oyarzun@geo.ucm.es

J. M. Esbrí · P. Higueras
Departamento de Ingeniería Geológica y Minera,
Escuela Universitaria Politécnica de Almadén,
Universidad de Castilla-La Mancha,
Plaza M. Meca 1, 13400 Almadén, Spain

Introduction

All plants accumulate metals at trace element levels (< 1000 μg g^{-1}), but some plant species are known for their ability to accumulate extremely high levels of metals, in some cases exceeding 10,000 μg g^{-1} [e.g., *Minuartia verna*: 11,400 μg g^{-1} lead (Pb); *Thlaspi caerulescens*: 43,710 μg g^{-1} zinc (Zn)] (Reeves, Baker, & Brooks, 1995). These plants can be used for the phytoextraction of metals from contaminated land on abandoned mining sites and districts (Chaney et al., 1995; Reeves et al., 1995; Cobbett, 2003).

The accumulation of mercury (Hg) in terrestrial plants has been reported to be related to both the concentration of the element in the soil and to the plant species (Crowder, 1991). Some studies have shown that plant uptake of Hg from the soil may be limited by the roots, which then function as a barrier between the plant and the element in the soil (Patra & Sharma, 2000; Boening, 2000), although Schwesig & Krebs (2003) suggested that this mechanism may only operate on highly polluted soils. However, the uptake of Hg by the plant cannot be regarded as being entirely related to the soil concentrations because there is also a continuous exchange of Hg between the atmosphere and the vegetation (Lodenius, Tulisalo, & Soltanpour-Gargari, 2003). There are relationships between the Hg absorbed by the canopy from the atmosphere (Hg_{atmos}), the Hg-enriched litterfall, and subsequent soil Hg enrichment. The deposition of Hg compounds onto the foliage occurs mainly by gaseous dry deposition and is closely linked to the transpiration of plants. The litterfall of mature trees has also been identified as a dominant pathway of total Hg deposition in forested catchments (Schwesig & Krebs, 2003). In addition to these pathways, we must also consider the reintroduction of Hg into the foliar system via the volatilization of Hg^0 from the soil (Patra & Sharma, 2000).

The toxic effects of Hg in plants differ according to the route of absorption (Patra & Sharma, 2000). Mercury affects both the light and dark reactions of photosynthesis and water transport through cell membranes. Another problem related to Hg in the plant-soil system is its incorporation into the human food chain, through the consumption of contaminated plants and animals (Loredo, Ordóñez, Gallego, Baldo, & García-Iglesias, 1999).

We here present the results of a preliminary phytogeochemical study of 53 plant taxa from the largest natural Hg accumulation site on Earth: the Almadén mining district, Spain (Fig. 1). The district is well known for widespread high levels of Hg in the air, water, soil, and stream sediments (Berzas Nevado, García Bermejo, & Rodríguez Martín-Doimeadios, 2003; Higueras, Oyarzun, Biester, Lillo, & Lorenzo 2003, Higueras et al., 2006; Gray, Hines, Higueras, Adatto, & Lasorsa, 2004). Our study is an update and a regional extension of previous studies (Hildebrand et al., 1980; Millán et al., 2004) to include the great variety of habitats present in the mining district.

Study area

The Almadén district in Central Spain (Higueras et al., 2000a, b) (Fig. 1) has produced one third of the total world production of Hg (Higueras et al., 2003, 2006). The district encompasses a series of Hg mineral deposits having in common a simple mineralogy dominated by cinnabar (HgS) and a minor component, pyrite (FeS_2). Almadén, the main mine of the district, has been active from Roman times to the present day, with almost no interruptions, except those caused by mining disasters (floods, fires) or external factors (wars). The Hg distribution in soils of the district reveals the existence of high to extremely high Hg concentrations (up to 8889 μg g^{-1}) (Higueras et al., 2003), whereas concentrations in stream sediments and waters reach up to 16,000 μg g^{-1} and 11,200 ng l^{-1}, respectively (Higueras et al., 2006). High concentrations of methylmercury, the most toxic form of mercury, have been detected in calcines (up to 3100 ng g^{-1}), sediments (0.32–82 ng g^{-1}), and waters (0.040–30 ng l^{-1}) (Gray et al., 2004).

The district is located within the so-called *Meseta Sur* (the Spanish southern mesa), which has a semi-continental Mediterranean climate with contrasting seasonal variations in mean temperatures: 6–8°C (winter) and 26–28°C (summer). The rainfall is concentrated in late autumn and early spring, with an annual total of 500–700 mm. The district is located within a region morphologically characterized by WNW trending valleys and sierras, within a landscape ranging in altitude between 200 and 1000 m a.s.l.

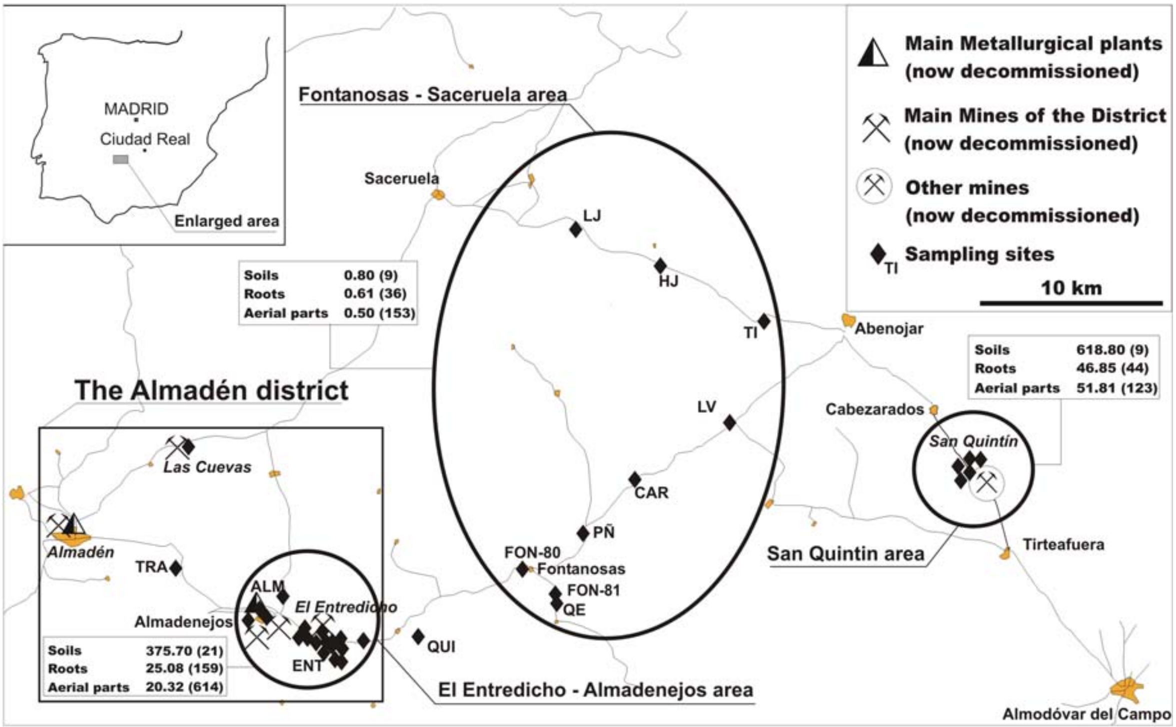

Fig. 1 Location map of the Almadén district, including mines, metallurgical sites, and sampling locations. Mean concentrations of Hg for all plant taxa (roots and aerial parts) and soils are shown in *boxes* (data in μg g^{-1} Hg). Abbreviations correspond to the project's internal data management

The soils of the district are mainly entisols, inceptisols, and alfisols, with a localized development of anthrosols (Soil Survey Staff, 1999). As shown by previous studies (Higueras et al., 2003) the Almadén soils are smectite-poor (main minerals: illite-pyrophyllite-chlorite-kaolinite), which severely reduces the possibility of cation exchange and increases the availability of metals from the inorganic matrix, leaving the organic matter as the sole agent for metal retention (Alloway, 2004).

The natural potential vegetation of the Almadén district corresponds to *Quercus rotundifolia* Lam. forest (Rivas-Martinez, 1987). The typical man-modified landscape from Central Spain constitutes the so-called *dehesa*, in which the initial evergreen oak forest, (*Q. rotundifolia* or *Q. suber* L.) is reduced to isolated groups of trees in a landscape dominated by perennial and annual grasses. Some of the wild plants of the Central Iberian Peninsula (Tardío, Pascual, & Morales, 2005) are edible (*Asparagus acutifolius* L., *Nasturtium officinale* R. Br., *Scolymus hispanicus* L., *Quercus rotundifolia*, etc.) and commonly consumed by the rural population or used for medical properties (*Marrubium vulgare* L., *Mentha pulegium* L., etc.)

Materials and methods

Experimental design and procedure

Samples of soils and plants were collected in three areas (Fig. 1): (1) the Almadén district, including highly polluted areas such as the Almadén, El Entredicho, Almadenejos, and Las Cuevas mine areas, and also sites with medium and low mercury contents east of El Entredicho; (2) the Fontanosas-Saceruela area, including sites with background levels of Hg in soil 20–50 km northeast of the Almadén district; (3) the San Quintín area, where Hg recovery tests were carried out at its flotation plant in 1987 and where, for this purpose, cinnabar ore was transported from Almadén and stockpiled for later treatment. Two field campaigns were carried out during the growing seasons (spring to autumn).

Soil and plant collection and management

Soils and plants were collected, ensuring that about one third of the total samples collected were in potentially contaminated areas. The soil samples (~1.5 kg) were collected from several sites in each locality, stored in plastic bags, and sieved in the laboratory. Given the type of soils (entisols, inceptisols, alfisols and anthrosols), we concentrated our efforts on the A horizon (20–30 cm depth).

Fifty-three plant taxa were studied. Plants were named using the standard regional flora designations (Castroviejo et al., 1986–2005; Valdés, Talavera, & Fernández-Galiano, 1987), plant life forms (biotypes) are according to the classification of Pignatti (1982), and habitats are defined as their characteristic plant-community following Rivas-Martínez, Fernandez-González, Loidi, Lousã, & Penas (2001). At least three replicates for each plant were collected at each sampling site. The plant organs were separated in the laboratory (roots, stems, leaves, flowers, and seeds), thoroughly cleaned with distilled water to avoid contamination, and kept in a dry and Hg-free environment until analysis.

Analytical methods

The total Hg content of the air-dried plant samples and soils was analyzed with a Lumex RA–915+ analyzer (Lumex, Arlington, Va.), a highly versatile instrument that is based on Zeeman atomic absorption spectrometry, with high-frequency modulation of light polarization (ZAAS–HFM) (Sholupov & Ganeyev, 1995). The application of Zeeman background correction and a multipath analytical cell provide a high selectivity and sensitivity to the measurements. The RP-91C (pyrolysis) attachment provides the capacity to measure Hg in solid samples. This attachment was used for the analyses of soils and dried plants at the laboratories of the Almadén School of Mines. The underlying principle is that total Hg in the samples is converted from a bound state to the atomic state by thermal decomposition in a two-section atomizer. As a first step, the sample is vaporized and the Hg compounds partly decomposed. This is followed by heating to 800°C, at which point the Hg compounds become fully decomposed, whereas organic compounds and carbon particles are catalytically transformed to carbon dioxide and water. The detection limits of total Hg are 0.5 μg kg^{-1} (soils) and 2 μg kg^{-1} (plants).

The detection of Hg by direct atomic absorption is complicated in samples with a complex matrix (plants, soils) because of the presence of organic compounds. However, the use of the background correction in the Zeeman atomic absorption mercury spectrometer RA-915+ overcomes the problem. In order to check the validity of the procedure, we ran analytical tests (Standard Addition Method, using the NIST 2710, NIST 2711, and BCR 146R standards; see below) on single (leaves) and composite (leaves plus a known amount of a standard) samples. The results showed that background correction effectively inhibited any major distortion (e.g., plant: *Eleocharis palustris* (L.) Roemer & Schultes: n=4 runs, variation on Hg concentration = 0.3%). Quality control at the laboratory was accomplished by analyzing replicate samples to check precision (see Table 1), whereas accuracy was obtained by using certified standards: (SRM) NIST 2710, (SRM) NIST 2711, and BCR 146R.

Vegetation

Table 2 shows the main plant communities identified in the area. The *dehesa*, an agro-silvo-pastoral system, is the most extensive landscape in the territory. It is an open evergreen oak woodland (*Quercus rotundifolia*) on a perennial grasslands of *Poa bulbosa* L. The shrublands are much extended in areas not used as pastures, and they commonly include *Pistacia lentiscus* L., *Cistus ladanifer* L., *Genista hirsuta* Vahl, *Cistus crispus* L., *Cistus salvifolius* L., *Cistus monspeliensis* L., and *Helichrysum stoechas* (L.) Moench. The streams of the area have riparian forests consisting mainly of *Fraxinus angustifolia* Vahl., *Salix salviifolia* Brot., and *Tamarix gallica* L. The prickly

Table 1 Biotypes, taxa, and concentration of Hg in soils, roots, stems, leaves, flowers, and seeds

Biotypes	Taxa	Soils (μg g^{-1} Hg)	Roots (g g^{-1} Hg)	Stems (g g^{-1} Hg)	Leafs (g g^{-1} Hg)	Flowers (g g^{-1} Hg)	Seeds (g g^{-1} Hg)
Suffruticose chamaeophyte	*Helichrysum stoechas*	0.14–49.50 (3)[a]	0.13–0.20 (2)	0.12–25.40 (9)	0.20–39.80 (9)	2.10–29.30 (6)	
Caespitose phanerophyte	*Crataegus monogyna*	0.13–49.50 (4)	[b]	0.06–1.54 (10)	0.43–55.70 (10)	0.08–8.16 (4)	
	Phillyrea angustifolia	4.55 (1)		0.27–0.35 (2)	1.40–1.80 (2)		
	Pistacia lentiscus	1.06–49.50 (5)	1.36–1.80 (2)	0.03–1.09 (10)	0.93–55.90 (12)		
Scapous phanerophyte	*Fraxinus angustifolia*	49.50 (1)	2.66–4.42 (2)	4.44–4.52 (2)	91.60–98.30 (2)		
	Olea europaea	49.50 (1)		7.69–9.54 (2)	37.80–40.30 (2)		
	Quercus rotundifolia	1.06–49.50 (5)	1.30–1.97 (2)	0.07–11.40 (13)	0.19–18.60 (15)		0.27–7.51 (4)
	Tamarix sp.	49.50 (1)		1.17–1.30 (2)	29.40–32.00 (2)		
Bulbous geophyte	*Allium* sp.	9.94–49.50 (2)	0.06–0.28 (5)	0.85–17.10 (6)	3.93–4.02 (2)	0.50–6.16 (6)	
	Ornithogalum narbonense	1,900.00 (1)		0.46–0.47 (2)	1.06–1.23 (2)		
Rhizomatous geophyte	*Asparagus acutifolius*	0.16–1,270.00 (7)	0.11–4.91 (8)	0.04–12.20 (14)	0.21–27.40 (11)		0.13–9.66 (4)
	Cyperus longus	0.13–37.10 (4)	0.66–6.34 (6)	0.09–1.61 (7)	0.16–4.07 (7)	2.37–4.55 (4)	
	Eleocharis palustris	1.98–37.10 (3)	2.64–24.40 (7)	1.30–1.74 (8)	3.84–28.20 (8)		3.00–6.39 (5)
	Juncus articulatus	0.13–15.90 (2)	8.13–19.50 (3)	0.07–6.01 (3)		8.94 (1)	
	Scirpus holoschoenus	1.06–1.98 (2)	0.49–6.70 (4)	0.34–4.22 (4)	0.70–3.56 (4)	10.90–11.70 (2)	
	Typha dominguensis	1.98 (1)	1.64 (1)	13.10 (1)	1.08 (1)		
Biennial hemicrytophyte	*Verbascum sinuatum*	20.70–2,695.00 (5)	0.13–96.80 (13)	0.22–11.10 (14)	8.34–125.00 (9)	0.36–6.30 (16)	
Scapous hemicrytophyte	*Marrubium vulgare*	1,270.00–2,695.00 (3)	4.62–85.50 (7)	0.74–4.49 (12)	5.14–30.60 (13)	1.81–2.61 (6)	
	Mentha pulegium	1.06–76.40 (3)	0.82–11.10 (8)	0.04–5.16 (8)	1.25–60.90 (10)	0.47–0.59 (2)	
	Mentha suaveolens	1.06–4.75 (2)	0.07–1.89 (4)	0.41–5.24 (6)	0.38–18.50 (6)	0.71–1.04 (4)	
	Nasturtium officinale	76.40 (1)	2.11–10.90 (4)	0.07–0.26 (5)	5.49–24.00 (4)	0.93–1.97 (3)	
	Oenanthe crocata	0.16–99.65 (4)	0.06–10.10 (8)	0.06–18.10 (8)	0.45–41.10 (7)	0.50–4.12 (4)	
	Rumex induratus	0.56–69.45 (6)	0.06–3.32 (14)	0.07–28.80 (13)	0.12–44.70 (9)	0.13–55.30 (7)	
	Scorzonera laciniata	23.30 (1)	9.97–33.10 (5)	0.93–5.19 (4)	47.80–63.10 (3)		1.40–2.47 (3)
	Scrophularia canina	1,730.00 (1)	13.90–25.40 (2)	37.40–52.90 (3)	67.20 – 81.90 (2)		
	Veronica anagallis-aquatica	37.10–76.40 (2)	2.25–6.20 (3)	1.55–1.55 (2)	1.78–11.50 (4)	0.84–1.08 (2)	
Rosulate hemicrytophyte	*Armeria alliacea*	4.78 (1)	0.50–0.55 (2)	0.18–0.26 (2)	0.58–0.78 (3)	2.10–5.25 (2)	
	Lemna sp.	0.13 (1)		0.43–0.50 (2)	0.30–0.37 (2)		
Hydrophyte radicant	*Callitriche stagnalis*	0.13–76.40 (2)	0.84–3.39 (4)	0.46–1.42 (2)	0.33–0.60 (2)		
	Ranunculus saniculifolius	0.13–76.40 (4)	0.62–15.57 (7)	0.21–4.09 (7)	0.39–21.85 (9)	0.57–8.47 (6)	
Nanophanerophyte	*Cistus crispus*	0.14–49.50 (5)	0.28–3.95 (7)	0.05–6.44 (6)	0.33–55.40 (7)		0.18–7.11 (7)
	Cistus ladanifer	1.06–49.50 (4)		0.07–3.09 (9)	0.44–9.73 (11)		0.40–0.49 (3)
	Cistus monspeliensis	4.55–49.50 (3)		0.04–11.90 (10)	7.53–106.00 (10)		0.27–14.50 (8)
	Daphne gnidium	9.94–49.50 (2)	4.49–6.37 (2)	3.58–46.30 (4)	5.40–52.10 (4)		
	Lavandula stoechas	0.71–69.45 (6)	0.06–2.44 (5)	0.09–8.47 (14)	0.38–33.60 (13)	0.25–86.60 (9)	
	Rubus ulmifolius	1,270.00 (1)	72.90–83.30 (2)	0.53–0.61 (2)	51.20 (1)		

Table 1 continued

Biotypes	Taxa	Soils (μg g^{-1} Hg)	Roots (g g^{-1} Hg)	Stems (g g^{-1} Hg)	Leafs (g g^{-1} Hg)	Flowers (g g^{-1} Hg)	Seeds (g g^{-1} Hg)
	Securinega tinctoria	0.13 –9.94 (3)		0.04–1.72 (7)	0.43–12.00 (7)	0.54 (1)	
	Thymus mastichina	0.16–49.50 (5)	0.10–5.35 (2)	0.07–8.72 (8)	0.40–82.20 (7)	0.38–6.22 (4)	
Caespitose therophyte	*Juncus hybridus*	297.50 (1)	57.70 (1)	3.30–16.30 (3)	82.20 (1)		
Scapous therophyte	*Carduus tenuiflorus*	21.20–2,695.00 (6)	1.65–175.00 (15)	0.31–3.81 (14)	1.17–24.50 (12)		0.71–1.84 (7)
	Daucus carota	4.55–49.50 (3)	1.39–4.68 (6)	1.44–15.40 (6)		2.09–2.97 (4)	
	Dittrichia graveolens	191.40 (1)	0.51–342.00 (3)	1.62–367.00 (4)	14.70–923.00 (3)	1.83–1,170.00 (4)	
	Gaudinia fragilis	4.56 (1)	9.15 (1)	0.09–0.10 (2)	2.63–2.79 (2)	0.82 (1)	
	Holcus setiglumis	33.85 (1)	15.70–19.50 (2)	0.47–0.56 (2)	10.50–14.70 (2)	5.92–7.10 (2)	
	Lolium rigidum	0.71–297.50 (4)	0.55–34.80 (7)	0.02–2.41 (7)	0.72–63.00 (4)	0.25–3.84 (8)	
	Petrorhagia nanteuilii	23.30 (1)	4.54–31.90 (4)	1.98–19.00 (7)	11.10–122.00 (9)	0.26–0.35 (4)	
	Polypogon maritimus	1.06–934.50 (5)	0.13–128.00 (8)	0.08–3.80 (7)	0.74–28.10 (8)	0.10–5.10 (8)	
	Polypogon monspeliensis	191.40 (1)	418.69–1,095.00 (2)	10.77–710.46 (4)	195.90–1,278.00 (2)	203.52–931.38 (4)	
	Reseda luteola	1,730.00 (1)	94.40 (1)	12.00–13.80 (2)	196.00–246.00 (2)	30.30 (1)	
	Rumex bucephalophorus	0.16–1,935.00 (9)	0.09–46.20 (18)	0.06–25.70 (19)	0.27–155.00 (15)	0.20–113.50 (12)	
	Silene inaperta	1,730.00 (1)	72.70–94.80 (2)	24.50–27.20 (2)	98.80–113.00 (2)	24.50–37.80 (2)	
	Spergularia purpurea	0.56–1,935.00 (9)	0.16–709.00 (21)	0.17–92.90 (24)	2.61–392.00 (14)	0.29–653.00 (21)	

[a] Number given in parenthesis in the body of the table denotes the number of analyzed samples

[b] An empty cell in the table indicates that data were not available

Table 2 Plant community types identified in the Almadén district

Community structure and environment	Representative species
Evergreen oak forest	*Quercus rotundifolia*, *Juniperus oxycedrus*
Silicicolous scrubland	*Lavandula stoechas*, *Genista hirsuta*, *Cistus monspeliensis*, *Cistus crispus, Cistus salviifolius*
Dwarf perennial grassland induced by adequate sheep pasture	*Poa bulbosa*, *Trifolium subterraneum*
Riparian scarcely flooded forest	*Fraxinus angustifolia*, *Populus nigra*
Riparian shrubland	*Salix salviifolia*, *Salix purpurea*, *Tamarix gallica*
Floodplain shrubland	*Securinega tinctorea*, *Rubus ulmifolius*
Prickly shrubland on wet soils	*Crategus monogyna*, *Rubus ulmifolius*
Meso-Eutrophic water-plant vegetation	*Ranunculus saniculifolius*, *Callitriche stagnalis*, *Callitriche brutia*
Water margin forb	*Oenanthe crocata*, *Cyperus longus*
Mediterranean rush	*Scirpus holoschoenus*, *Mentha suaveolens*
Annual grassland on seasonal pools	*Polypogon monspeliensis*, *Polypogon maritimus*, *Juncus hybridus*
Rupestrian scrubland	*Rumex induratus*, *Phagnalon saxatile*
Dwarf annual grassland	*Hordeum murinum* subsp. *Leporinum, Vulpia ciliata*, *Vulpia geniculata*, *Trisetaria panicea*
Subnitrophilous scrubland	*Marrubium vulgare*, *Urtica urens*
Perennial forbs and grasses on roadsides	*Dittrichia graveolens*, *Piptatherum miliaceum*
Sub-rupicolous and nitrophilous vegetation	*Sonchus tenerrimus*

shrubs of the wet areas are characterized by *Crataegus monogyna* Jacq. and *Rubus ulmifolius* Schott. The riverbeds host water-plant communities dominated by *Ranunculus saniculifolius* Viv. The water margins are colonized by forb communities of *Oenanthe crocata* L. and *Cyperus longus* L. Rush communities are dominated by the Mediterranean rush *Scirpus holoschoenus* L. Seasonal pools that dry up in late spring are floristically characterized by *Polypogon monspeliensis* L. or by *Juncus hybridus* Brot. Also, the territory presents screes where the vegetation is characterized by the shrub *Rumex induratus* Boiss & Reuter. Ephemeral plant communities of *Rumex bucephalophorus* L. develop on sandy-loam soils. Other noteworthy vegetation is that related with disturbed areas such as the subnitrophilous and therophytic grass *Hordeum murinum* L. subsp. *leporinum* (Link) Arcang. and the nitrophilous tall herb communities characterized by *Marrubium vulgare* L., *Carduus tenuiflorus* Curtis and *Dittrichia graveolens* L.

Results and discussion

The soil samples taken from the study area display a wide range of Hg concentrations, from 0.13 to 2695 μg g^{-1} Hg (Table 1). The background concentrations (0.13 μg g^{-1} Hg) are extremely anomalous when compared to baseline levels for soils, which are generally within the range 0.01–0.03 μg g^{-1} Hg (Senesi, Baldassare, Senesi, & Radina, 1999) (this range being 0.05–0.3 μg g^{-1} in agricultural soils; Alloway, 2004). They are, however, comparable to those found in soils from other mines or districts of Spain, such as Mieres, where Hg concentrations have been found to be in the range 1.7–29,304 μg g^{-1} Hg (Loredo, Ordóñez, Gallego, Baldo, & García-Iglesias 1999; Fernandez-Martínez, Loredo, Ordoñez, & Rucandio 2005), or the Azogue valley, with 6–1,400 μg g^{-1} Hg (Viladevall, Font, & Navarro, 1999). The areas subjected to intensive mining and/or metallurgical activities in the Almadén district (El Entredicho, Almadenejos) display the highest concentrations of Hg in both soils and plants. Surprisingly, the San Quintín decommissioned Pb-Zn mine, which only received a few truckloads of cinnabar from Almadén in 1987, shows very high Hg levels in plants and soils. The flotation of cinnabar at the San Quintín concentration plant proved to be ineffective and, therefore, most of the ore from Almadén remained on the mine site without later removal. On the contrary, the areas devoid of mining and/or metallurgical activities (away from the district and the San Quintín property) proved to be less contaminated. However, we use

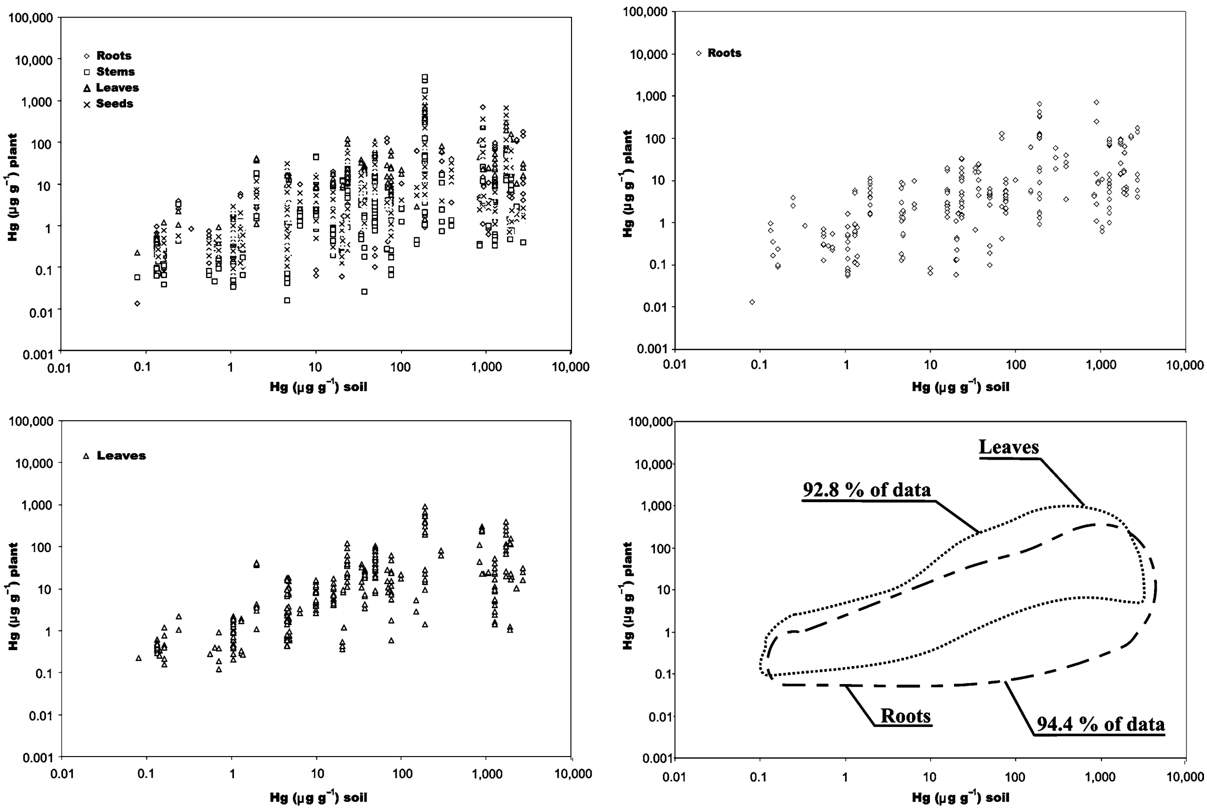

Fig. 2 Mercury in plant organs versus Hg in the soil samples taken from the Almadén district

'less-contaminated' here in the context of relative levels and not absolute values: the Hg levels measured in these localities are still well above baseline concentrations. For example, the soils from the Fontanosas-Saceruela area have a mean of 0.80 µg g^{-1} Hg, which is approximately 27- to 80-fold higher than the baseline levels for soils (Senesi et al., 1999).

Mercury concentrations in the studied plant taxa were extremely variable (Fig. 2). For example, roots were found to have concentrations of 0.06 (*Oenanthe crocata*, *Rumex induratus*) to 1095 (*Polypogon monspeliensis*) µg g^{-1} Hg, whereas in leaves, the concentrations ranged from 0.16 (*Cyperus longus*) to 1278 (*Polypogon monspeliensis*) µg g^{-1} Hg (Table 1). The response of plants to the high soil Hg concentrations varied, as expected, with taxon and organ. As a general rule, plant stems contained the lowest concentrations of Hg, whereas the highest levels were found in either roots or leaves (Table 1). To study the relationships between Hg concentrations in soils and plants we tested the working hypothesis that the Hg accumulated by a plant is largely related to amount of Hg present in the soil (Boening, 2000); that is, $Hg_{plant}=f(Hg_{soil})$. We found that the Almadén plant taxa can be grouped into four Hg-accumulating types (Fig. 3). In type 1, Hg_{plant} increases with increasing Hg_{soil}, a strict case of $Hg_{plant}=f(Hg_{soil})$. The best plant representatives of this type of relationship are *Pistacia lentiscus* and *Quercus rotundifolia*. Type 2 plants have a more complex behavior: after an initial linear relationship, no increase in Hg_{plant} is observed. Type 2 plants include *Asparagus acutifolius* and *Cistus ladanifer*. Type 3 plants initially show no increase in Hg_{plant} until a threshold is surpassed; above this threshold, a linear relationship between Hg_{plant} and Hg_{soil} exists. Plants that follow this behavior pattern include *Cistus crispus* and *Rumex bucephalophorus*. Type 4 plants are by far the most common, and they are characterized by

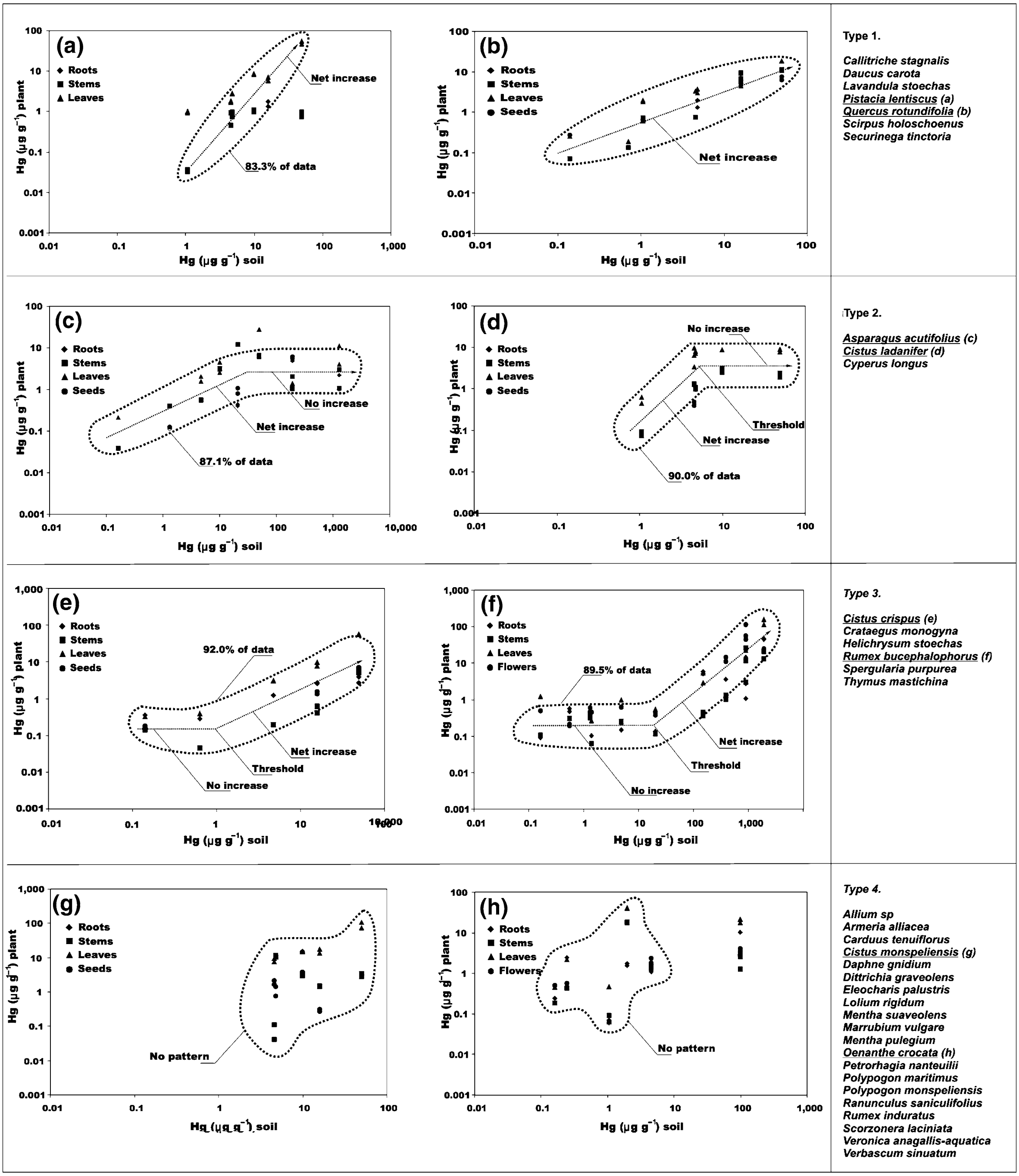

Fig. 3 Mercury accumulation types in the Almadén district and associated plant taxa

the absence of any relationship between Hg_{plant} and Hg_{soil}. Type 4 plants include *Cistus monspeliensis* and *Oenanthe crocata*.

Kovalevski (1987) established that about 95% of plant species and their organs have varying degrees of resistance to the uptake of elements present in large concentrations in the root zone. He subsequently classified plants in the following categories: (1) no barrier present, unlimited uptake; (2) practically no barrier present, allowing uptake to accumulate up to 100-fold the local background; (3) barrier present, limiting uptake to tenfold the local background; (4) background barrier present, preventing uptake beyond substrate background concentration. Our types 1 and 2 match Kovalevski's first two categories; however, types 3 and 4 depart from his classification. Type 3 plants (Fig. 3) are interesting because it appears that a certain Hg_{soil} concentration threshold must be surpassed before the plant starts accumulating Hg. Type 4 behavior is complex (Fig. 3) because uptake occurs, but in an unpredictable manner.

There is also a differential accumulation of Hg in the different parts of the plants (Fig. 2). For example, taxa such as *Carduus tenuiflorus*, *Polypogon maritimus*, or *Rubus ulmifolius* accumulate more Hg in the roots than in the leaves, whereas other species, such as *Helichrysum stoechas*, *Mentha pulegium*, or *Rumex induratus*, have the opposite behavior (Table 1). Nevertheless, in most cases, no clear tendencies of Hg accumulation are observed in either roots or leaves (Fig. 2). Since the concentration in the substrate is the dominant factor controlling the emission of Hg (Gustin et al., 2000), a naturally enriched realm, such as the Almadén district, is bound to be an important source of Hg_{atmos}. Large concentrations of Hg in ore deposits (>1,000 μg g^{-1} Hg) generate important anomalies of Hg_{atmos} (Gustin, 2003). Atmospheric Hg may utilize two main pathways in the Almadén district: (1) it may go directly to the plants via foliar uptake of Hg^0; (2) it may be reintroduced to the soils where Hg can be deposited as Hg^{2+}, either by the direct deposition of emitted Hg^{2+} or from conversion of emitted Hg^0 to Hg^{2+} through ozone-mediated processes (USEPA, 1997). The photolysis of inorganic Hg^{2+} to Hg^0 at the soil surface may in turn contribute significantly to the re-emission of Hg gas to the atmosphere (Scholtz, Van Heyst, & Schroeder, 2003) and, therefore, to the plants. We propose that factors other than that of soil contents may be playing an active role in the Hg cycle at Almadén, such as extremely high concentrations of Hg_{atmos}. It is known that plants do absorb Hg_{atmos} via leaves (Patra & Sharma, 2000; Lodenius, Tulisalo, & Soltanpour-Gargari, 2003); however, do all plants have the same capacity for Hg_{atmos} intake? Plant species do display contrasting behaviors: for example, Hg_{atmos} intake by leaves of gramineous species such as *Avena byzantina* C Koch, *Hordeum vulgare* L. and *Triticum aestivum* L. is fivefold larger than that of corn (*Zea mays* L.) or sorghum (*Sorghum* sp.). This variation could be attributed to a differential internal resistance to Hg_{atmos} binding (Patra & Sharma, 2000). Thus, it would appear that variations in Hg uptake do depend on a large number of factors, including Hg contents in the soil and atmosphere, the plant species and organs, and season. Additionally, uptake must be strongly influenced by temperature and light, which are key factors governing the rates of Hg emissions from the soil to the atmosphere (Gustin et al., 2002; Scholtz et al., 2003). This variety of factors and plant responses to Hg uptake may help to explain why the behavior observed in the Almadén plants is so different (types 1–4) (Fig. 3). In addition, we must also bear in mind that the larger the number of studied taxa, the more complex the answers will be. Fifty-three different plant taxa from a variety of plant habitats and communities are bound to generate contrasting behaviors.

Conclusions

Plants sampled from the Almadén district were found to have moderate to very high concentrations of Hg (0.03–1,278 μg g^{-1} Hg) in both the roots and aerial parts, thus reflecting the high concentration of this element in the soils of this area. Higher Hg concentrations in the roots, stems, and leaves were found in *Polypogon monspeliensis* (1095, 710, and 1278 μg g^{-1} Hg, respectively) sampled from seasonally flooded aquatic environments. Conversely, lower concentrations were found in the roots of *Allium* sp. (0.06 μg g^{-1} Hg), stems of *Pistacia lentiscus* (0.03 μg g^{-1} Hg), and the leaves of *Rumex induratus* (0.12 μg g^{-1} Hg). Four main patterns of mercury uptake by plants were detected: (1)

constant uptake, independent of Hg concentration in the soils; (2) no further increase in Hg_{plant} (flat behavior) after an initial linear relationship between uptake and soil concentration; (3) no increase in the uptake of Hg is recorded until a threshold is surpassed, and thereafter a linear relationship between Hg_{plant} and Hg_{soil} is established; (4) no relationship between Hg_{plant} and Hg_{soil}.

Acknowledgements The authors wish to express their gratitude to Prof. Michael Barbour (University of California at Davis) for reviewing the manuscript and providing constructive suggestions. Dr. José Manuel Murillo and an anonymous referee are thanked for helpful comments and suggestions. Valuable aid with sample collection was received from J.M. Ruiz, and from M.A. Verdonze with the chemical analyses. The research was funded by the European Union FEDER funds (Project 1FD97-0814), Ministerio de Ciencia y Tecnología (grants REN2002-02231 and PPQ2003-01902) and the University of Castilla–La Mancha.

References

Alloway, B. J. (2004). Bioavailability of elements in soil. In: O. Selinus, B. Alloway, J. A. Centeno, R. B. Finkelman, R. Fuge, U. Lindh, & P. Smedley (Eds.), *Essentials of Medical Geology* (pp. 347–372). Amsterdam: Elsevier.

Berzas Nevado, J. J., García Bermejo, L. F., & Rodríguez Martín-Doimeadios, R. C. (2003). Distribution of mercury in the aquatic environment at Almadén, Spain. *Environmental Pollution*, *122*, 261–271.

Boening D. W. (2000). Ecological effects, transport, and fate of mercury: a general review. *Chemosphere*, 40, 1335–10351.

Castroviejo, S, et al. (1986–2005) (eds) Flora Ibérica. Plantas vasculares de la peninsula Iberica e Islas Baleares, vol 1–8, 14, 21. Serv. Public. C.S.I.C., Madrid.

Chaney, R., Brown, S., Li, Y. M., Scott Angle, J., Holmer, F., & Green C. (1995). Potential use of metal hyperaccumulators. *Mining Environmental Management*, *3*, 9–11.

Cobbet, Ch. (2003). Heavy metals and plants—model system and hyperaccumulators. *New Phytologist*, *159*, 289–293.

Crowder, A. (1991). Acidification, metals and macrophytes. *Environmental Pollution*, *71*, 171–203.

Fernandez-Martínez, R., Loredo, J., Ordoñez, A., & Rucandio, M. A. (2005). Physicochemical characterization and mercury speciation of particle-size soil fractions from an abandoned mining area in Mieres, Asturias (Spain). *Environmental Pollution*, *142*, 217–226.

Gray, J. E., Hines, M. E., Higueras, P., Adatto, I., & Lasorsa, B. K. (2004) Mercury speciation and microbial transformations in mine wastes, stream sediments, and surface waters at the Almadén mining district, Spain. *Environmental Science and Technology*, *38*, 4285–4292.

Gustin, M.S. (2003). Are mercury emissions from geologic sources significant? *Science of the Total Environment*, *304*, 153–167.

Gustin, M. S., Lindberg, S. E., Austin, K., Coolbaugh, M., Vette, A., & Zhang, H. (2000) Assessing the contribution of natural sources to regional atmospheric mercury budgets. *Science of the Total Environment*, *259*, 61–71.

Gustin, M. S., Biester, H., & Kim, C. S. (2002) Investigation of the light-enhanced emission of mercury from naturally enriched substrates. *Atmospheric Environment*, *36*, 3241–3254.

Higueras, P., Oyarzun, R., Munhá, J., & Morata, D. (2000a) The Almadén mercury metallogenic cluster (Ciudad Real, Spain): alkaline magmatism leading to mineralization processes at an intraplate tectonic setting. *Revista de la Sociedad Geológica de España*, *13*, 105–119.

Higueras, P., Oyarzun, R., Munhá, J., & Morata, D. (2000b). Palaeozoic magmatic-related hydrothermal activity in the Almadén syncline (Spain): a long-lasting Silurian to Devonian process? *Transactions of the Institution of Mining and Metallurgy*, *109*, B199–B202.

Higueras, P., Oyarzun, R., Biester, H., Lillo, J., & Lorenzo, S. (2003) A first insight into mercury distribution and speciation in soils from the Almadén mining district. *Journal of Geochemical Exploration*, *80*, 95–104.

Higueras, P., Oyarzun, R., Lillo, J., Sánchez-Hernández, J. C., Molina, J. A., Esbrí, J. M., & Lorenzo S. (2006). The Almadén district (Spain): anatomy of one of the world's largest Hg-contaminated sites. *Science of the Total Environment*, *356*, 112–124.

Hildebrand, SG, Huckabee, JW, Sanz Díaz, F, Janzen, SA, Solomon, JA, Kumar, KD. 1980. Distribution of mercury in the environment at Almadén, Spain. Oak Ridge, Tenn., Oak Ridge National Laboratory, ORNL/TM–7446.

Kovalevski, A. L. (1987). *Biogeochemical exploration for mineral deposits* Utrecht: VNU Science Press.

Lodenius, M., Tulisalo, E., & Soltanpour-Gargari, A. (2003). Exchange of mercury between atmosphere and vegetation under contaminated conditions. *Science of the Total Environment*, *304*, 169–174.

Loredo, J., Ordóñez, A., Gallego, J. R., Baldo, C., & García-Iglesias, J. (1999). Geochemical characterization of mercury mining spoil heaps in the area of Mieres (Asturias, northern Spain). *Journal of Geochemical Exploration*, *67*, 377–390.

Millán, R., Gamarra, R., Schmid, Th., Vera, R., Sierra, M. J., Quejido, A. J., Sánchez, D. M., & Fernández, M. (2004). Mercury content in natural vegetation of three plots in the mining area of Almadén (Spain). *RMZ – Materials and Geoenvironment*, *51*, 155–158.

Patra, M., & Sharma, A. (2000). Mercury toxicity in plants. *Botanical Review*, *66*, 379–422.

Pignatti, S. (1982). *Flora d'Italia*. Bologna: Edagricole (pp. 1–3).

Reeves, R. D., Baker, A. J. M., & Brooks, R. R. (1995). Abnormal accumulation of trace metals by plants. *Mining Environmental Management*, *3*, 4–8.

Rivas-Martínez, S. (1987). *Memoria del mapa de series de Vegetación de España. 1: 400.000*. Madrid: Ministerio de Agricultura, Pesca y Alimentación, ICONA (p. 268).

Rivas-Martínez, S., Fernandez-González, F., Loidi, J., Lousã, M., & Penas, A. (2001) Syntaxonomical checklist of vascular plant communities of Spain and Portugal to Association level. *Itinera Geobotanica*, *14*, 5–341.

Scholtz, M. T., Van Heyst, B. J., & Schroeder, W. H. (2003) Modelling of mercury emissions from background soils. *Science of the Total Environment*, *304*, 185–207.

Schwesig, D., & Krebs, O. (2003) The role of ground vegetation in the uptake of mercury and methylmercury in a forest ecosystem. *Plant Soil*, *253*, 445–455.

Senesi, G. S., Baldassare, G., Senesi, N., & Radina, B. (1999). Trace element inputs into soils by anthropogenic activities and implications for human health. *Chemosphere*, *39*, 343–377.

Soil Survey Staff 1999 Soil Taxonomy: a basic system of soil classification for making and interpreting soil surveys; U. S. Dep. A. Agric. Handb. N.436.

Sholupov, S. E., & Ganeyev, A. A. (1995). Zeeman absorption spectrometry using high frequency modulated light polarization. *Spectrochim Acta*, *50B*, 1227–1238.

Tardío, J., Pascual, H., & Morales, R. (2005). Wild food plants traditionally used in the province of Madrid, Central Spain. *Economic Botany*, *59*(2), 122–136.

USEPA. (1997) *Mercury Study Report to Congress: Volume III Fate and Transport of Mercury in the Environment*. Washington: Office of Air Quality Planning & Standards and Office of Research and Development (p 376).

Valdés B., Talavera S., & Fernández-Galiano E. (Eds). (1987) *Flora vascular deAndalucía Occidental*. Barcelona: Ketres Edit.

Viladevall, M., Font, X., & Navarro, A. (1999) Geochemical mercury survey in the Azogue Valley (Betic Area, SE Spain). *Journal of Geochemical Exploration*, *66*, 27–35.