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The Phytomanagement of Trace Elements in Soil

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Trace elements (TEs) occur at low concentrations (<1000 mg kg^{-1}) in organisms, yet they have a large biological effect, both as essential nutrients and environmental contaminants. Phytomanagement describes the manipulation of soil-plant systems to affect the fluxes of TEs in the environment with the goal of remediating contaminated soils, recovering valuable metals, or increasing micronutrient concentrations in crops. Phytomanagement includes all biological, chemical, and physical technologies employed on a vegetated site. Successful phytomanagement should either cost less than other remediation or fortification technologies, or be a profitable operation, by producing valuable plant biomass products. This may include bioenergy or timber production on contaminated land, a practice that does not reduce food production. We review the components of phytomanagement and the underlying biogeochemical processes, with a view to elucidating situations where this technology may be successfully applied and identifying future research needs. Many full-scale operations have proved the efficacy of plants to reduce contaminant mobility in soils (phytostabilization), particularly when used in combination with other technologies. As a stand-alone technology, the oft-touted use of plants to extract TEs from contaminated soils (phytoextraction) or low-grade ore bodies (phytomining) is unsuitable for most, if not all, sites due to low-extraction rates and problems caused by site heterogeneity, the limited rooting depth of plants and the presence of contaminant mixtures. Unsubstantiated claims about phytoextraction have tarnished the reputation of all "phyto" technologies. Nevertheless, phytoextraction, as part of a larger environmental toolkit, has a role in phytomanagement. The growth, or lack thereof, of profitable companies that provide phytomanagement will indicate its value. A critical knowledge gap in phytomanagement is the integration of the processes that affect plant–TE interactions and the biophysical processes affecting TE fluxes in the root zone, especially the effect of roots on contaminant fluxes.

Keywords bioenergy, biofortification, phytoextraction, phytomining, phytoremediation

I. PHYTOMANAGEMENT TO CONTROL TRACE ELEMENT FLUXES

1.1 Trace Elements in the Soil-Plant System and their Importance to Humans

Biological trace elements (TEs) occur at minor concentrations (<1000 mg kg^{-1}) in organisms, yet they may have a major effect on life, both as essential nutrients and environmental contaminants. Figure 1 shows the periodic table of TEs that may occur in organisms, highlighting those that are essential for animals and higher plants, and those that are common environmental contaminants.

Trace element deficiencies affect most of the world's 6.5 billion human inhabitants. The World Health Organization (WHO) estimates that 3.7 billion people are Fe deficient, with 2 billion suffering from anaemia. Worldwide, some 35% of children between 0 and 5 years old suffer from Zn or Fe-deficiencies and 260 million suffer from iodine (I) or selenium (Se) deficiencies (2002). Most TEs that humans ingest come directly from plants or via animals, with a smaller fraction coming from geophagy (Oliver, 1997), the direct consumption of soil. Therefore, increasing the plant uptake of essential TEs, and rendering them in a form that can be absorbed by the human gut, would have a large beneficial effect on humanity in deficient areas (Yang *et al.*, 2007).

However, in many cases, it is not a deficiency, but excess consumption of and exposure to TEs that has a detrimental effect on humans. Human activities such as mining, industrial production, transport and agriculture release ever-increasing amounts of bioavailable TEs into the environment. TEs are immutable and mostly have a low mobility in soil. Therefore, they accumulate over time under specific environmental conditions. At high concentrations, all TEs are toxic to organisms, even those essential for life. Plants grown on contaminated soils may have reduced productivity and plant TE uptake may facilitate the entry of toxic elements into the food chain, possibly affecting human nutrition. Worldwide, some 22 million hectares of land are contaminated with trace elements (GACGC, 1994).

The increasing land area that is contaminated or otherwise degraded to the point where it can no longer be used for food production and the increase in the world's population mean that more food must be produced on a decreasing area of suitable land. Contaminated agricultural land has three negative consequences. Firstly, its usage results in food products that pose a human health risk. Secondly, nonagricultural land, such as that under forests, may be cleared for agricultural production, decreasing biodiversity and removing a sink for greenhouse gasses. Thirdly, agriculture will be intensified on noncontaminated agricultural land. This requires increased fertilizer and pesticide application, which may lead to further soil contamination, since TEs are unwanted constituents in many fertilisers (McLaughlin *et al.*, 1996) and key components in many pesticides (Mills *et al.*, 2005). Intensification may also lead to other forms of land degradation such as compaction, erosion, and salinization.

1.2 Phytomanagement of Trace Elements

Phytomanagement describes the engineering or manipulation of soil-plant systems to control the fluxes of TEs in the environment. Thus, the goal of phytomanagement may be to alleviate deficiencies of essential TEs or to reduce the environmental risk posed by contaminating TEs. A key component of

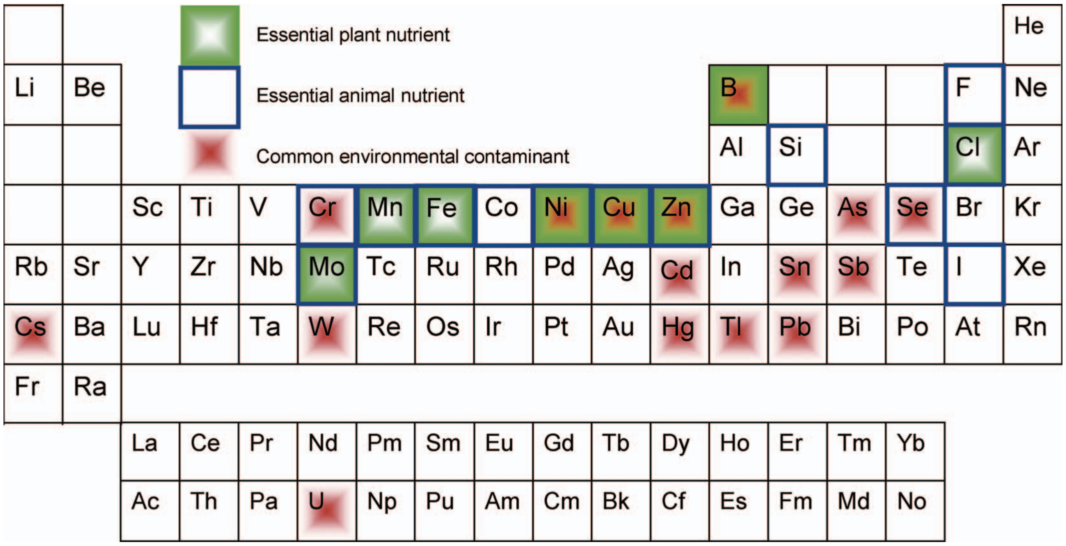

FIG. 1. Periodic table of the trace elements that may occur in living organisms. The empty fields are macronutrients.

phytomanagement is that it should either cost less than other remediation or fortification technologies, or be a profitable operation, by producing valuable plant biomass products. The term phytomanagement relates to the best site-specific management option, rather than a single technology. Trace element phytomanagement technologies are connected in that they all effect a change in TE fluxes in the soil-plant system.

Although the goals of individual phytomanagement scenarios may differ, the underlying processes in the soil-plant system are identical. Phytomanagement exploits plants as bio-pumps that use the sun's energy to remove water and perhaps TEs from the soil to the aboveground portions, while returning some of the products of photosynthesis back into the root-zone (Robinson *et al.*, 2003b). Transpiration is the cornerstone of phytomanagement because water is the main vehicle for the transport of many TEs into the soil–plant system. Clothier and Green (1997) described roots as "the big movers of water and chemicals in soil." Of the global average rainfall of 720 mm that falls on land each year (Sellers, 1965), some 410 mm is returned back to the atmosphere, to some degree directly by evaporation from the soil, but mostly by evapotranspiration from vegetation growing therein (Clothier and Green, 1997). Evapotranspiration reduces the average volume of soil solution than exits the root-zone and enters receiving waters by some 57%. In some cases, where evapotranspiration is greater than rainfall, leaching can be eliminated. TEs in soil may enhance or reduce plant growth and affect the quality of plant material as foodstuff both positively and negatively. Conversely, plants affect the movement and speciation of TEs in the environment. Figure 2 gives an overview of TE interactions with plants that are relevant to phytomanagement.

1.2.1 Terminology

Phytomanagement describes an assemblage of related technologies that include *phytomining* (Nicks and Chambers, 1994), *phytoremediation*, which is divided into *phytoextraction* and *phytostabilization* (Salt *et al.*, 1995), as well as *biofortification* (King, 2002). Phytomanagement of contaminated soils describes use of vegetation, possibly in combination with other technologies, to deliver the most cost-effective means of mitigating any environmental risks associated with the site. The role of vegetation in contaminated site phytomanagement is to add value to the land and possibly mitigate the negative effects associated with soil contamination. Phytoremediation may aim to immobilise the TEs in the soil, so that they neither leach nor enter the plant shoots (phytostabilization). Alternatively, one may engineer or manipulate the soil-plant system so that plants take up the contaminating TEs into the shoots, where they may be volatilised (phytovolatilization), or removed from the site when the plants are harvested (phytoextraction). Phytomining describes the phytoextraction of valuable TEs from ore bodies or metal-contaminated environments where the target metal concentration is too low for conventional mining. In soils that are deficient in essential TEs, the goal of phytomanagement is to increase the TE concentration in the edible plant organs (biofortification). The boundaries between these technologies are not distinct. Phytoextraction of TEs from a contaminated soil may produce biomass enhanced in essential TEs (biofortification) or saleable quantities of valuable TEs (phytomining), while reducing erosion and TE leaching from the site (phytostabilization). Figure 3 provides a schematic diagram connecting the phytomanagement technologies.

An important distinction in the TE distribution in soils is whether they are inert, able to interact with plants (*phytoavailable*) or able to interact with any biological organism, including humans (*bioavailable*). We use these terms to indicate a TE's physicochemical potential but not its biological action. For example, phytoavailable TEs may be toxic to plants, or accumulated in the plant's shoots, but they are not necessarily

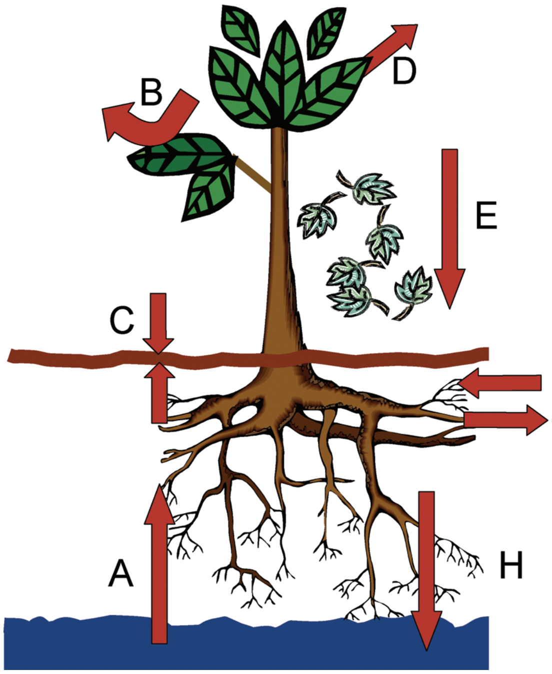

FIG. 2. Key plant—trace element interactions in phytomanagement. (A) Root water uptake and transpiration reduce the water flux through the soil profile. Since water is the vehicle for TE movement in soil, transpiration also reduces TE leaching. (B) Rainfall evaporates from the plant canopy, reducing water flux through the soil and consequently reduces TE leaching. (C) Roots stabilize the soil, reducing the transport of TEs on soil particles. (D) TEs accumulated by the plant may enter the food chain via herbivores. Alternatively, harvesting the plants may remove TEs from the site. (E) Leaf abscission returns TEs to the soil surface. Combined with root exudation and decay, this increases soil organic matter. Some of this organic matter is soluble and may increase TE leaching or uptake by the plant. (F) Toxic TEs in the soil may inhibit growth. (G) Roots change, soil pH, redox potential, aeration, promote the growth of soilborne organisms, and change the speciation of TEs. (H) Roots may create macropores that increase TE leaching by preferential flow processes. Root exudates may also solubilise TEs increasing their leaching or plant uptake.

so. Toxicity and accumulation are dependent upon plant species.

1.3 Phytoextraction for Soil Cleansing

Chaney (1983) suggested that hyperaccumulator plants, namely those that accumulate TEs to concentrations 10 to 100 times those found in 'normal' plants (Brooks *et al.*, 1977), could cleanse polluted soil by extracting the contaminating metals. McGrath *et al.* (1993) and Baker *et al.* (1994) demonstrated that the phytoextraction of metals was possible with the Zn hyperaccumulator *Thlaspi caerulescens* (J. & C. Presl.).

Repeated cropping of plants that accumulate contaminating TEs in soil should, in theory, lower the soil's TE concentrations to acceptable levels, provided the harvested amounts of TEs exceed further inputs. Phytoextraction employs plants that accumulate large amounts of one or more target TEs into the above-ground biomass. Each cropping would remove TE from the area. The TE-rich biomass would be burned, fermented or used in gasification to reduce its volume. Residual material that is rich in the contaminating TE could be reprocessed to recover the TE or stored in an appropriate area, such as a contained landfill that does not pose a risk to the environment.

Some plants transform soil contaminants into volatile compounds that disperse in the atmosphere (phytovolatilization). Plant-microbial systems have been discovered that volatilise Hg, As and Se (Brooks, 1998a). Drawbacks of phytovolatilization include the limited number of plant-microbial systems that volatilise a limited number of TEs and, more importantly, that there is no control on the destination of the volatilised elements. For essential TEs such as Se, however, phytovolatilization offers the possibility of redistributing this element from areas where Se toxicity exists to downwind areas where there is Se deficiency (Zayed *et al.*, 2000). Phytoextraction via volatilisation has the advantage over other forms of phytoextraction in that the vegetation does not require regular harvesting; the crop can be left onsite until the soil's TE concentrations comply with environmental regulations.

Phytoextraction is appealing because there are few technologies for economically removing contaminating TEs from soil, and many other strategies require harsh chemical or physical processes. Incineration, thermal desorption, and soil washing are expensive and leave the soil infertile. In contrast, phytoextraction would use the sun's energy to cleanse the soil using normal agronomic processes. Because of this, it is often incorrectly touted as a low-cost means to cleanse contaminated soils.

Phytoextraction for soil remediation requires that the concentrations of the contaminating TEs be reduced to levels that complies with environmental regulations. There is a conspicuous absence of successful phytoextraction field trials or commercial operations. Selenium volatilisation using genetically engineered *Brassica juncea* (L.) is one of the few examples of the successful field application of phytoextraction (Bañuelos *et al.*, 2005). The factor determining the duration of phytoextraction is the mass of TEs removed by the crop per unit of time (years) compared to the mass of TE in the soil.

Phytoextraction requires ongoing site management, harvesting and processing of the TE-rich biomass. Burning may reduce the volume of the biomass. Although specialized incineration facilities may be required to prevent TE-loss in the smoke (Keller *et al.*, 2005), Margni *et al.* (1997) reported most existing waste incinerators can safely process metal-enriched biomass. Alternatively, relatively volatile metals, such as Cd, could be removed from the biomass via pyrolysis, leaving uncontaminated bottom ash (Keller *et al.*, 2005).

Phytoextraction has fixed costs in the form of site assessment and geotechnical preparation for planting (Robinson *et al.*, 2006). In its pure form, the cost of phytoextraction increases with the time needed to reduce the soil's contaminant burden to below regulatory limits. This time is not simply the quotient of the soil's TE burden and the crop's TE content, since contaminant heterogeneity and the decline in plant uptake as soil TE

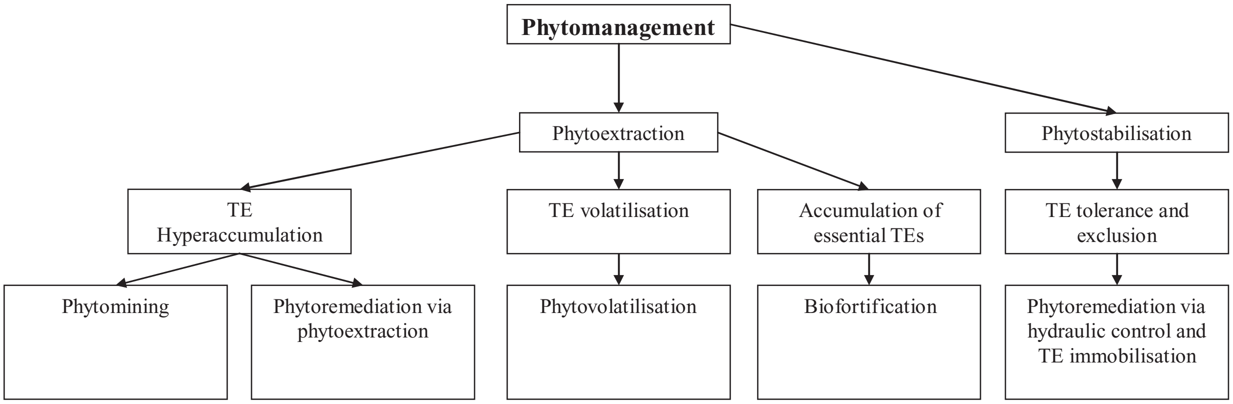

FIG. 3. The relationship of various “phyto” technologies to phytomanagement.

concentrations decline invariably increase the time needed. Most studies on plant-TE interactions used homogeneous growth media in greenhouse environments. In the TE-contaminated sites where phytoextraction would be applied, the distribution of TEs is typically highly heterogeneous, both spatially and temporally, as TEs leach or are phytoextracted from the soil. Unlike pot trials, roots in the field may not be in intimate contact with the TE-rich material, thus resulting in lower-than-expected TE uptake (Section 3.3). A clear example of this “greenhouse vs field” difference was observed with Se uptake (Bañuelos *et al.*, 1998). More importantly, the likely different uptakes between pot and field trials underscores the importance of minimizing expectations of phytoextraction under field conditions based upon observations in a controlled contaminated environment. Successive crops reduce the pool of TE that is available for plant uptake (Section 2.5), resulting in small amounts being extracted. “Hot spots” of high TE concentrations may reduce or inhibit plant growth, rendering phytoextraction ineffective in these zones. The time needed for phytoextraction may be calculated using the following formulae (Robinson *et al.*, 2006).

$$t = \frac{M_i(x)_{\max} - M_f}{P(E)B(E)} \qquad [1]$$

where t is the time in years, x is the spatial position (latitude, longitude) $M_i(x)_{\max}$ is the maximum initial TE burden (g ha^{-1}) in the affected area, M_f is the target TE soil burden (g ha^{-1}), P is the average crop TE concentration (g t^{-1}), and B is the biomass production (t ha^{-1} yr^{-1}), both of which are a function of the root exposure to bioavailable TE, E (g t^{-1}). E can be calculated thus:

$$E = \int_0^z \int_0^t R(t', z)\, C(M(t', z)) dt' dz \qquad [2]$$

where z is depth (m), R is the root fraction (dimensionless) that is in contact with the phytoavailable TE, C (g t^{-1}), which is a function of M.

For practical purposes, Eq. 2 requires a numerical solution. The function $R(t',z)$ needs to be determined using existing data that describes the root development of the species. Similarly, $C(M(t'z)$ is calculated using the TE’s absorption isotherm and an measurements of the TE in the soil profile. Calculations using Eq. 2 usually indicate that high spatial and temporal heterogeneity result in longer extraction times. This is because the maximum contaminant burden, $\mathrm{M}_i(\mathrm{z})_{\max}$, is always greater in heterogeneous environments than in homogeneous ones. The response of plant roots to heterogeneous patches of TEs (Section 3.3) will greatly affect the value of t. Plants with roots that forage contaminant hotspots, such as *Thlaspi caerulescens* (Whiting *et al.*, 2000), would remove TEs at a greater rate than those whose root systems are indifferent or avoid hotspots. Research into root behaviour may be as important for phytoextraction as the investigation of TE transporter systems and soil conditioners.

Equation 2 is applicable over a wide range of scales. The degree of heterogeneity, and therefore cleanup time, will increase as the scale decreases. Setting the scale for assessing soil concentrations, and therefore cleanup thresholds, is the domain of regulators.

Table 1 shows the time required to halve the soil concentration of a contaminating TE, at initial extraction rates of 5 and 50 kg TE ha^{-1} yr^{-1}. Assuming a constant extraction rate of 50 kg ha^{-1} yr^{-1}, soil concentrations of up to 200 mg kg^{-1} could be halved within a decade. However, incorporating temporal heterogeneity and a realistic root distribution (Eqs. 1 & 2), the calculated time quadruples.

Equation 1 calculates the time to cleanse a soil of only one TE. Many polluted soils contain more than one contaminant. Few plant species can extract high concentrations of more than one element. Consequently, remediation may require sequential crops of different species that target a range of contaminants. Additional TEs or other xenobiotic compounds in the soil may further reduce plant growth. Therefore, the time to cleanse soils with a suite of contaminants will be longer than that required for the removal of a single TE, and the time required for

TABLE 1
Time Required for Phytoextraction to Reduce the Contaminant Burden in the Soil by Half, Calculated Using Both a Constant Extraction Rate and Incorporating Temporal Heterogeneity, and given an Initial Extraction Rate of 5 or 50 kg ha^{-1} yr^{-1} in the above-ground Biomass. The Soil Depth is 20 cm and the Bulk Density is 1.2. The Temporally Heterogeneous Calculations, with a Realistic Root Distribution were Made Using the Phyto-DSS (Robinson *et al.*, 2003). $\phi = 1$, $K = 0.15$

	Constant extraction rate		Temporally heterogeneous	
Initial extraction rate (kg ha^{-1} yr^{-1})	**5**	**50**	**5**	**50**
Initial soil concentration (mg kg^{-1})				
50	12	2	50	5
100	24	3	99	10
200	48	5	199	20
400	96	10	398	40
800	192	20	797	80

phytoextraction to cleanse even a moderately contaminated soil is, in the best case, in the order of decades.

The application of phytoextraction in the field presents additional challenges. Commercial providers of seeds of hyperaccumulator species may find seed production unattractive because most hyperaccumulator species have a limited ecological range. Seed merchants would thus be required to produce small volumes of seeds of many hyperaccumulator species, each requiring specialised growing conditions. There are also ecological concerns that plant-TE uptake may provide an additional exposure pathway into food chains if local herbivores consume these plants (Wolfe and Bjornstad, 2002).

Were phytoextraction combined with a profit-making operation that is unaffected by any elevated plant-TE loadings, then the time to decontaminate the soil becomes less important. Revenue-generating operations may include forestry (Pulford *et al.*, 1995) and bioenergy production (Licht and Isebrands, 2005). Recent concern over global warming due to CO_2 emissions may provide economic incentives to produce plant-based fuels because such systems do not result in net CO_2 production. As neither humans nor animals consume biofuels, elevated TE concentrations in such fuel crops are of lower concern than they would be in food crops. Biofuel production may be an effective way for the land to be cleansed while providing a positive economic return (Bañuelos, 2009). The use of contaminated or marginal soils for biofuel production is economically more prudent than using good quality soils. Food production must not suffer for biofuel production.

1.4 Phytomining

Nicks and Chambers (1994, 1995) reported that it might be possible to use plants to extract Ni from low-grade ores that would otherwise not be economic to mine, and were unsuitable for agriculture due to their high Ni concentration. Low-grade Ni ores cover large areas of the Earth's surface, for example in Western Australia, Italy, Brazil, Canada, Russia, and many other countries and territories. Nicks and Chambers found that *Streptanthus polygaloides* (Gray) grown on ultramafic (serpentine) soils near Chinese Camp in California, could extract up to 100 kg ha^{-1} of Ni (worth \$550 ha^{-1} at the prices at that time). A large-scale industry with continuous incineration of the crop would add an additional \$219 ha^{-1} from the energy of combustion. They concluded that the return to a farmer growing a 'crop of nickel' (i.e., half the gross yield) would be roughly comparable, or superior to, that obtained for a crop of wheat. An obvious problem with the use of an incinerator to produce steam for power generation is that the crop harvesting would occur over a short period and therefore the power plant should be near an urban area where domestic waste might be used as a feedstock to keep the plant going the rest of the year. There is also the possibility of two crops a year that would not only increase Ni yield but would give more work to a nearby incineration plant.

Subsequent studies (Anderson *et al.*, 1999; Leblanc *et al.*, 1999; Robinson *et al.*, 1997a, 1997b) have shown other species could produce even greater profits from the land, and that phytomining could extract other metals, especially Tl and Au. An American company, Viridian Environmental, subsequently patented the phytomining process (U.S. patent Nos. 5711784 & 5944872).

Unlike phytoextraction for soil cleansing, the feasibility of phytoextraction for phytomining is based solely on the value of the TE that is extracted. Brooks *et al.* (1998) showed that, in principle, Ni should be feasible since there are many hyperaccumulator plants, such as *Alyssum* (Robinson *et al.* 1997b) spp. and *Berkheya coddii* (Robinson *et al.*, 1997a), that fulfil the criterion of a high biomass production ($>$10 t ha^{-1}year^{-1}) with a high shoot Ni concentration ($>$10,000 mg kg^{-1}). Chaney *et al.* (2007) reported that Ni phytomining using *Alyssum* could give a return of US\$ 16,000 ha^{-1}.

Yet, as with phytoextraction for soil cleansing, there are no reports of successful commercial phytomining operations. So, given the potentially high economic returns, what is preventing the commercialisation of phytomining? Since removal of Ni from soil using phytomining has been demonstrated to be viable in principle, and in field trials (Brooks *et al.*, 1998; Chaney *et al.*, 2007; Chaney *et al.*, 2005; Robinson *et al.*, 1997a, 1997b), either unaccounted costs are preventing the commercialization of phytomining, or it is only a matter of time before phytomining becomes widespread. Here, we investigate potential limiting factors.

Li *et al.* (2003) demonstrated that feasibility of Ni recovery from the ash (bio-ore) of *Alyssum* spp. Bio-ore contains higher

Ni concentrations (6–16%) (Koppolu *et al.*, 2004) than normal Ni-ores (ca. 3%). Bio-ores are free of oxides of Mn, Fe and Si that normally hinder the recovery of Ni from conventional ores (Li et al. 2003). Boominathan *et al.* (2004) showed that bio-ore generated from *Berkheya coddii* contained 34% Ca, some fifteen times more than bio-ore generated from *Alyssum bertolonii*. The Ca, present as hydroxyapatite, may reduce the efficiency of Ni phytomining using *Berkheya coddii*. Nevertheless, it is unlikely that metal recovery from bio-ore is preventing the commercialization of phytomining.

Chaney *et al.* (2007) discuss the agronomic aspects of phytomining and report that although phytomining crops require special fertilisation regimes, these are not excessively expensive compared to normal agronomic practices. Furthermore, as discussed in Section 1.3, there are currently no commercial providers of seed from the specialized hyperaccumulator species. However, such providers would no doubt rapidly appear as soon as money were made from large-scale phytomining operations.

More serious problems of phytomining are its inherently low efficiency with respect to land use and time, and, perhaps counter intuitively, its negative environmental effects. Compared to conventional mining, phytomining is inefficient because for every ton of metal produced, a much larger area of land is required. For example, 1 t of Ni, phytomined at an extraction rate of 0.4 t ha^{-1} $year^{-1}$ (a theoretical maximum (Chaney *et al.*, 2007)) requires the use 2.5 hectares for one year, followed by the processing of 37 t of dry biomass, assuming a Ni concentration of 2.7%. In contrast, conventionally mining 1 t of Ni from an ore containing 3% Ni requires the removal of 33 t of ore, which occupies just 22 m^3 of space, assuming a density of 1.5 t m^{-3}. Instead of taking a year, this process requires, at most, a few hours. Moreover, Ni-rich ultramafic soils can sustain between 3 and 18 phytomining crops before the Ni levels in the hyperaccumulator plants drop to sub-economic levels (Robinson *et al.*, 1999b). Then, phytomining can only continue once the topsoil has been removed or its geochemical properties have been modified so that more Ni becomes phytoavailable. Both these treatments are expensive. Given its low rate of metal extraction, phytomining has poorer economics of scale than conventional mining, which would result in increased transport and production costs.

The low efficiency of phytomining indicates that it would result in much greater environmental disturbance compared to conventional mining. While there is no debate that conventional open cast mining destroys habitats, contaminates the surroundings and leaves an ugly scar on the landscape, the area of disturbed land is relatively low compared to the disturbance that phytomining would cause for the same rate of extraction. In the example above, phytomining requires that 2.5 ha of land be cleared of its native vegetation and put into a monoculture of hyperaccumulator plants. In contrast, conventionally mining the same amount of metal would disturb just 0.0022 ha of land, assuming the first metre of soil is removed. Ultramafic soils, where phytomining would occur, usually contain a distinct flora, which is often protected by law. It is unlikely that local environmentalists would appreciate the removal of native vegetation to make way for a monoculture of a (possibly exotic) hyperaccumulator species.

When applied to a contaminated site, rather than a natural metalliferous soil, the economics of phytomining become more attractive, since any recovered TEs will offset the cost of site remediation. Phytomining might usefully be combined with conventional mining, where mine pits or tailings that contain a concentration of the target metal that is too low for conventional exploitation, but still elevated concentrations of TEs could be phytomined to reduce the risk of the residual TEs causing environmental harm, whilst producing an economic return.

1.5 Phytostabilization

The environmental and human heath risk posed by TEs in contaminated soil is lower if the TEs are immobile. Some regulators are now adopting a risk-based approach for assessing soil quality that incorporates the influence of TE solubility and mobility (Fernandez *et al.*, 2005; Swartjes, 1999), as well as its total concentration in the soil.

Phytomanagement to reduce TE fluxes (phytostabilization) exploits plant transpiration and root growth to reduce leaching and control erosion. Roots help maintain an aerobic environment in the vadose zone, which helps prevent the formation of reduced TE species that are often more toxic and more mobile than oxidized species. Roots also add organic matter to the substrate that binds the TEs. Phytostabilization involves the establishment of vegetation on the contaminated site that enhances the value of the land. Value may come in the form of ecological benefits, or the production of nonedible commercial products such as bioenergy or timber.

The effectiveness of the vegetation in controlling leaching is climate dependent. Meteorological conditions set an upper limit on evapotranspiration (ET). Biological and soil parameters determine the actual ET of various vegetation types, which may be much less than the theoretical upper limit. In dryer climates, evapotranspiration is usually greater from deep-rooted species because shallow rooted species have less access to water during periods of drought and are, therefore, more likely to suffer from dieback or reduced transpiration and growth (Vogeler *et al.*, 2001). The aerial portions of trees canopies act as umbrellas, re-evaporating some 15% of rainfall before it reaches the ground (McNaughton and Jarvis, 1983). In addition to intercepting precipitation, canopies also reduce evaporation from below and thus keep the forest floor moist.

Vegetated land is porous. During periods of high-intensity rainfall, leaching may occur through root-induced macropores. Similarly, high intensity rainfall is likely to induce surface runoff. Leaching is inevitable in climates where rainfall is greater than evapotranspiration. Figure 4 shows the modelled drainage from a (contaminated) site in the absence and presence of poplar trees in three climates (Table 2). In all three

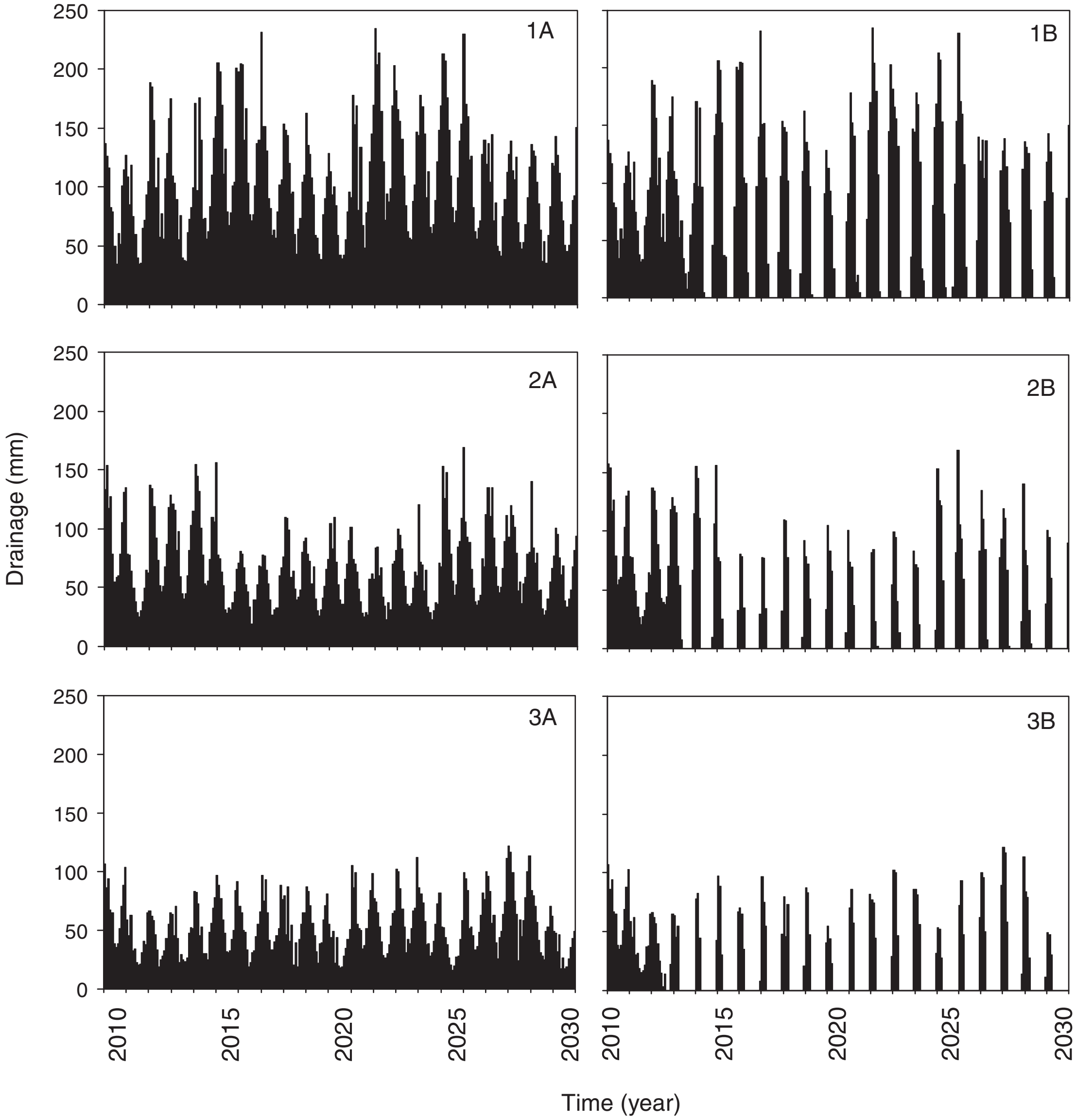

FIG. 4. Phyto-DSS (Robinson *et al.*, 2003) simulations of drainage volumes in wet (1), moderate (2), and dry (3) climates, from bare soils (A) and under poplar trees (B). Table 2 shows the average rainfall and potential evaportranspiration of the three climates.

scenarios, drainage occurs throughout the year for the first 4–5 years as the trees establish. Thereafter, in all climates, drainage is eliminated during the summer months, but occurs during the winter. Trapping the leachate and circulating it back onto the vegetation would further reduce the drainage, with the drawback of increasing costs. Recirculating the leachate from the site would hydraulically isolate the site in the moderate and dry climates (Fig. 4) because potential evapotranspiration is greater than rainfall (Table 2). Leachate circulation may be a necessary requirement for complete hydraulic isolation, because the lack of transpiration during the winter months and preferential flow processes would always produce some drainage. Leachate recirculation could occur *ad infinitum*: each pass through the root zone further modifies the leachate. An increase in the level of solutes, especially Na^+ and Cl^-, may be of concern during leachate reapplication, resulting in soil impartibility or salt damage to the plants. However, depending on the composition of the leachate, reapplication may have beneficial effects on plant growth compared to un-irrigated vegetation (Nixon *et al.*, 2001). Table 2 shows that the biomass production in the medium and dry climates is lower than in the wet climate, indicating the trees suffer from drought stress during the summer, and thus leachate circulation would improve growth. Leachate irrigation via overhead sprinklers increases total evaporation, but may reduce plant growth if the leachate contains high contaminant or salt concentrations. Surface irrigation may avoid this problem.

TABLE 2
Climatic Conditions and Phyto-DSS (Robinson *et al.*, 2003) Simulations of Average Potential Evapotranspiration (ETo), Actual Evapotranspiration (ET) and Biomass Production of a Stand of Poplars over a 20-year Period in Temperate Wet, Moderate and Dry Climates. Figure 4 shows the Drainage as a Function of Time

	Wet climate	Moderate climate	Dry climate
Rainfall (mm yr^{-1})	1271	835	653
ETo (mm yr^{-1})	759	1052	1227
Average ET (mm) (% of rainfall)	437 (33 %)	390 (45 %)	385 (56 %)
Average drainage (mm)	899	486	391
Wood production (t ha^{-1} 20 yr^{-1})	90	85	80

Drainage is inevitable when rainfall is greater than evapotranspiration. However, vegetative caps may eliminate drainage during low-rainfall periods. Depending on the TE, the high flow rates of receiving waters may dilute any leachate to the point that they do not pose an environmental risk, although such dilution-solutions to pollution are unacceptable in many regulatory environments.

Irrigation may be necessary in the establishment or maintenance of vegetation in phytostabilization. Irrigation may increase macropore-facilitated leaching, particularly when high volumes of water are applied over a short time.

Were net evapotranspiration the only criterion controlling TE leaching, then there would be more leaching under shallow-rooted species, such as most grasses, than under deep-rooted species such as trees (Vogeler *et al.*, 2001). However, deep roots may create macropores that facilitate the preferential transport of contaminants to groundwater (Roulier *et al.*, 2008). A fundamental research question is the role of roots in the preferential transport of contaminants, especially in soils where the contaminants occur heterogeneously.

The time required to implement phytostabilization, i.e., the time required for the roots to penetrate the contaminated material and evapotranspiration to reach a maximum, is dependent on the species composition of the vegetatation. Densely planted perennial trees require 2–5 years (Robinson *et al.*, 2006). Mendez and Maier (2008a) concluded that phytostabilization of mine tailings requires the identification of regional and climatic specific plants which show low metal uptake in shoots and determination of the minimum requirements in amendments (compost, fertilizer, irrigation) required for plant growth. Importantly, phytostabilization requires that the site be permanently vegetated, thus limiting future land use options. Nevertheless, periodically harvesting the vegetation could produce non-food products, such as bioenergy or timber. The timing and nature of the harvest is critical, since it will necessarily decrease evapotranspiration from the site and potentially increase contaminant leaching. Removing biomass from the site will also gradually reduce the soil's TE burden, because some TEs will be removed with the biomass. Partially harvesting alternate rows of trees may maintain hydraulic control while recovering valuable biomass and removing contaminating TEs (Robinson *et al.*, 2007). Whole system models could calculate the likely effect of harvesting on TE fluxes.

Vangronsveld *et al.* (1996) and Mendez and Maier (Mendez and Maier 2008a, 2008b) detailed how the use of vegetation and soil amendments could control erosion and leaching on metalliferous mine tailings. The vegetation reduces dust and leaching and improves the aesthetic and ecological value of the site. Phytomanagement to stabilize mine tailings may involve the establishment of vegetation directly in the tailings or it may follow capping, which can be costly as it may require reengineering of any tailings dam as well as a large earth-moving operation. Here, deep-rooted species may be less suitable as they may disrupt the integrity of the cap. Penetration of tree roots into the underlying tailings may not only provide a transport pathway for rainfall, but may also result in tree death, depending on the toxicity of the underlying material. The natural colonization of mine sites is slow since the physicochemical characteristics of these sites are not favourable to most plant species. Nevertheless, some of these tolerant plant species can spread easily in these environments due to the lack of competitors (Macnair, 1987). Weed species often have stress resistant properties, and can grow under poor fertilizer and drought conditions. Because of long-term natural selection, weed species have an extensive adaptive capacity (Wei *et al.*, 2005).

On disused tips and landfill sites, phytomanagement can prevent contaminants leaching to groundwater or local waterways (Robinson *et al.*, 2003b). Fast-growing and high-water use trees such as poplars (*Populus* spp.) and willows (*Salix* spp.) have been successfully employed in this role (Ferro *et al.*, 1997). These species are effective because they establish rapidly, have a high water-use, tolerate a wide range of environmental conditions, are easily of propagated (Quinn *et al.*, 2001), and take up high concentrations of some TEs, notably B, Cd and Zn (Robinson *et al.*, 2005).

In arid conditions where salinity is a problem, deep-rooting evergreen trees, such as *Eucalyptus* spp., can lower a saline water table thus reducing salt toxicity to crops. Bell *et al.* (1999) reported the effectiveness of this technology on some Australian soils.

The vegetation used for phytostabilization may enhance the ecological value of the site, by increasing biodiversity and providing a habitat for rare species. An excellent example of such an operation is the Guadiamar Phytomanagement Programme (Dominguez *et al.*, 2008). Here, a "green corridor" has been established on over 2000 ha of land that became contaminated with As, Cd, Pb, Zn, and other TEs following a mine tailings-dam failure at Aznalcóllar, Southern Spain, in 1998. Following removal of the most contaminated topsoil, the establishment of vegetation, combined with soil conditioners, reduced dust and TE leaching from the site, while providing an ecological connection between the Donaña World Heritage Park in the South and the Sierra Morena mountains in the North.

Low plant TE uptake is critical to the success of using the vegetation in phytostabilization to enhance biodiversity. Elevated concentrations of TEs in the aboveground tissues not only increase the likelihood of their entry into the food chain, but may also result in an accumulation of TEs on the soil surface, as TE-rich leaf litter is deposited (Robinson *et al.*, 2003a). Dominguez *et al.* (2008) showed that the TE concentrations in plants from the Guadiamar Phytomanagement Programme were within the normal range for plant tissues for all species except *Populus alba* (L.), which accumulated high concentrations of Cd and Zn. As a result, this species could be removed from the site or excluded from future plantings.

Phytostabilization is particularly suited for low value sites, where the land value is small compared to the cost of soil excavation and landfilling. Unlike phytoextraction, providers of phytostabilization technology can point to numerous examples of its successful application on areas as diverse as acidic mine tailings (Brown *et al.*, 2005; Mendez and Maier, 2008b), wood-waste piles (Robinson *et al.*, 2007) and disused sheep-dipping sites (Robinson and Anderson, 2007).

1.6 Biofortification

Biofortification aims to increase the concentration of essential TEs in crops to improve human health and agricultural productivity. The most commonly deficient elements in the diet of humans are Fe and Zn (Franca, 2002). Selenium and I are also deficient in many areas. Rice (*Oryza* spp.) and wheat (*Triticum* spp.) are the target crops for many biofortification programs, because they are the staple food for most of the earth's population.

Biofortification has several advantages over simply adding essential TEs to the final product, for example fortifying flour by directly adding Zn compounds, or taking dietary supplements. TEs that have been taken up by the plant via the roots provide a constant source of the TE with less risk of toxicity due to an overdose, or deficiency caused by gaps in supply of the TE-spiked product. Physiologically accumulated TEs in plant parts are also more bioavailable to humans (Storksdieck and Hurrell, 2007). However, biofortification may not provide source of TEs as reliably as traditional fortification or dietary supplements because the effectiveness of biofortification depends on soil properties, genotypes, agricultural management practices and climatic factors (Schulin *et al.*, 2009).

If the biofortification arises from genetic modification of the crop, via either traditional breeding or *in vitro* gene manipulation, then recurrent costs are low, and the germplasm could be shared internationally, reaching people with limited access to commercially fortified food or supplements (Nestel *et al.*, 2006). A potential drawback of biofortification is that any process used to increase the concentration of an essential TE in plant tissue may also increase the uptake of nonessential TEs or xenobiotic compounts. Thus, biofortified crops should be screened carefully for contaminants.

Biofortification is not restricted to humans. Livestock commonly suffer from TE deficiencies, especially in those regions where the TEs occur at low concentrations in the soil. Pasture species tend to take up low concentrations of TEs from soil compared to trees and shrubs (Robinson *et al.*, 2005). Robinson *et al.* (1999c) showed that the specimen trees *Nyssa* spp. accumulated Co from deficient soils to the extent where leaves from these trees could alleviate Co deficiency in livestock if it comprised just 2% of the animal's diet. The trees accumulated circa 7 mg Co kg^{-1} (dry matter) in the leaves, some 40 times more than the average pasture. Similarly, poplars and willows accumulate high concentrations of Zn and Co relative to pasture species and this may be linked to observed health benefits in sheep and cattle when the clones are used as stock fodder (Robinson *et al.*, 2005). A potential problem of using poplars and willows as stock fodder at some sites is their high ability to accumulate Cd (Granel *et al*., 2002); although varieties exist that accumulate high Zn and Co, but low Cd (Robinson *et al.*, 2005).

On contaminated sites where the contaminant is also an essential nutrient, food or fodder crops can be produced that contain high concentrations of an essential nutrient. Bañuelos (2006) demonstrated that broccoli and *Brassica juncea* (L.) could supplement Se to animal diets, when these plants were grown on Se-laden soil.

Plants biofortified with TEs could be used as an organic mulch to fertilize crops that are deficient in TEs. Robinson *et al.* (2007) showed that poplars grown on a B-contaminated site accumulated inordinate amounts of B in the leaves, while taking up only limited amounts of the other concurrent contaminants. Boron, the element of concern at this site, could be removed from the site via its accumulation poplar leaves, and used in nearby orchards to alleviate a B deficiency. Thus, combining biofortification with other remediation technologies may offset the cost of site remediation.

Bañuelos and Lin (2009) present some of the most recent research on the topic of biofortification. The feasibility of biofortification depends heavily on the concentrations of the target TE and any contaminants in the soil, as well as the cost of implementation. If the total concentration of the target TE is too low, it is ineffective to change the soil properties to increase the TE's bioavailability or introduce plant varieties with a high uptake of the target TE. On soils where nonessential TEs occur at elevated concentrations, the plant or soil treatment that enhances crop uptake of the essential TE may also increase the crop's contaminant uptake, thus endangering human health.

Since poor countries could achieve the greatest health benefit from biofortified foods, the cost of biofortification is of paramount importance. If the implementation costs are high, it may be cheaper to use other methods of supplying essential TEs to humans, such as TEs in tablet form. The development of genotypes that accumulate the target TE would cost little compared to shipping the already fortified food. This advantage would be greater if the crops produced seeds of the same genotype, thus rendering subsequent seed shipments unnecessary. However, political problems, including patent protection on plant varieties and public opposition to genetically modified food, may hinder the widespread use of such genetically modified organisms (Fox, 2001).

The idea of increasing essential TEs in crop plants through selective breeding and genetic modification is sound in principle, since there is a genetic variation among species and varieties that result in a range of TE concentrations in the edible parts (White and Broadley, 2005). Recent work has shown that Barley (*Hordeum vulgare* L.), modified to overexpress an *Arabidopsis* sp. Zn transporter, took up significantly more Zn than the unmodified variety (Ramesh *et al.*, 2004). To date, there are no full-scale field operations using this approach. In contrast, there are numerous field trials showing that on deficient soils, the addition of Zn to crops, via either soil or foliar application, enhances plant health and results in a higher shoot Zn concentration (Cakmak, 2008). A fertile area for future research is to investigate whether these methods result in increased grain Zn concentrations, or the extent to which any supplementary Zn, perhaps added as a chelate, is bioavailable to humans. Schulin *et al.* (2009) review the agronomic aspects of Zn biofortification.

A major disadvantage of agronomic measures to improve the concentrations of essential TEs in crop plants is that bad application practices result in reduced crop productivity or environmental pollution. Experience has shown that the inappropriate use of pesticides and fertilizers, especially in poor countries, has resulted in soil and water contamination and negative human health effects. Since essential TEs such as Zn and Se are toxic to both plants and animals at higher concentrations, overzealous or inhomogeneous application of the TE may lead to soil infertility or even present a human health risk. Similar environmental damage may occur if soil conditioners such as chelants are overapplied (Nowack *et al.*, 2006). These problems may be circumvented if the TE or other agent could be supplied as a seed coating. For example, seeds could be coated with a rhizobacteria that enhances plant uptake of the essential TE. There is a dearth of research demonstrating the feasibility of these possibilities.

Environmental conditions will affect agronomic and to a lesser extent genetic measures to biofortify food crops with essential TEs. Suitable soil conditioners and their application rates may vary greatly between sites. This increases the risk of inappropriate use and subsequent environmental degradation. Development of whole system phytomanagement models would alleviate problems arising from environmental heterogeneity.

1.7 Processes in Phytomanagement

The solubility and speciation of TEs in soil, as well as their tolerance and uptake by plants is critical for all forms of phytomanagement. Plants should tolerate soilborne TEs to produce maximal growth. In phytoextraction-related technologies (Fig. 3) high uptake of specific trace elements into the shoots is desirable, while successful phytostabilization requires that TEs be excluded from the aerial portions. In all phytomanagement applications, high uptake may present an environmental risk of toxic TEs entering the food chain.

We summarize the key process in TE interactions with plants as they relate to phytomanagement. We have divided these processes, arbitrarily, into three sections that describe (1) the phytoavailability of TEs in the soil, (2) the interactions in the soil: root interface, i.e., the rhizosphere, and (3) the uptake and tolerance of soluble TEs by plants. Finally, using processes 1–3, we describe the feasibility of phytomanagement and explore fertile areas of future research.

II. PROCESS 1: TRACE ELEMENT SOLUBILITY AND SPECIATION IN SOIL

The solubility and speciation of TEs in soil determines their toxicity, availability for plant uptake, and their downward mobility. These soil processes are thus critical for phytomanagement. Understanding the factors controlling TE solubility and speciation allows the selection of soil amendments that promote or reduce TE bioavailability and the selection plant species that are optimal for the desired goal of managing the TE flux in the soil – plant system.

The provenance of TEs in the environment may be natural, such as high concentrations of Ni and other siderophiles in ultramafic (serpentine) soils, or anthropogenic, such as high concentrations in base metals in lands surrounding industrial sites. The concentrations of TEs can vary considerably. In soil, the TE loading is a function of the parent material plus subsequent atmospheric or waterborne deposition.

Not all TEs in the environment interact with plants. Solubility is a prerequisite for plant uptake or toxicity. The interaction of the soluble TE with the plant is dependent on individual properties of the element, its chemical speciation, and the species of plant with which it is interacting. The solubility and speciation of TEs depend on a plethora of physical (e.g., water retention capacity) , chemical (e.g., pH), microbial (e.g., micorrhiza), and plant factors, as well as the properties of the TE.

The re-supply of TEs into the soluble phase is critical for phytomining, where high crop TE concentrations are required for as long as possible. Robinson *et al.* (1999b) showed, using sequential extractions that in ultramafic soils, between 13% and 80% of the total Ni could become soluble. The potentially available Ni was inversely proportional to the soil's Si concentration. The replenishment of Ni in soil solution occurred within 24 hours and cumulative extractions produced Langmuir-type curves, with the asymptote equal to the pool of potentially available Ni.

Such re-supply processes are, by definition, kinetically limited and may be slow. However, this slowness may not always be a disadvantage because it allows the plant time to absorb the mobile TE. Hamon and McLaughlin (1999) introduced the concept of "bioavailable contaminant stripping" for a phytoextraction procedure that aims to keep the bioavailable fraction of soil TEs low enough to be harmless.

2.1 Decreasing Trace Element Phytoavailability

In the phytostabilization of a contaminated site, naturally occurring or artificial soil amendments such as liming material, phosphate, zeolite, bentonite, clay, Fe metal, Fe and Mn oxides, and organic matter, may reduce the solubility of TEs (Cheng and Hseu, 2002). These amendments reduce TE solubility by promoting the formation of insoluble precipitates or by enhancing the soil's capacity to bind the TE. The latter can be achieved directly through the addition of adsorbent material or indirectly by adjusting the soil's pH–Eh conditions to promote TE absorption onto the soil's matrix. The effectiveness of these amendments in reducing TE phytoavailability has to be balanced with their potential negative effects on plant growth resulting from immobilization of essential nutrients, increases in salinity and any possible changes in the soils physical that inhibit root penetration, oxygenation, and water uptake.

Chemical immobilization using phosphate amendments, such as mineral apatite, synthetic hydroxyapatite, and phosphate salts, reduce cationic TE solubility by the formation of TE–phosphate precipitates (McGowen *et al.*, 2001) and by increasing the number of negatively charged exchange sites (Bolan *et al.*, 1999). The solubility of Pb in soil can be greatly reduced by the formation of chloropyromorphite [$Pb_5(PO_4)_3Cl$]. Several microcosm studies have shown that chloropyromorphite can be formed through the addition of hydroxyapatite [$Ca_{10}(PO_4)_6(OH)_2$] (Ma *et al.*, 1993; Ryan *et al.*, 2001). Brown *et al.* (2004) demonstrated that phosphate fertilizers could be combined with organic matter and Fe-rich material, as soil amendments to reduce the solubility of Cd, Zn, and Pb. Phosphate amendments such as hydroxyapatite are effective in reducing the solubility of Pb, Cd, Zn, Al, Ba, Co, Mn, Ni, and U. However, phosphate has been shown to promote the solubility of As and Cr (Seaman *et al.*, 2001), possibly through reduced sorption of the oxyanions due to an increase in pH and competition from PO_4^{3-}, HPO_4^{2-}, and $H_2PO_4^-$.

A variety of inorganic and organic amendments have been used to reduce Cr(VI) to the less soluble, less toxic Cr(III) species. Fe(II)-bearing minerals and surface-bound organic matter form effective reductants. The latter is catalyzed by soil mineral surfaces, and Cr(III) binds tightly to surface species or is precipitated as $Cr(OH)_3$ (Jardine *et al.*, 1999). Similarly, Bolan and Duraisamy (2003) showed that organic amendments, such as animal and poultry manures rich in dissolved organic carbon, are effective in reducing Cr(VI) to Cr(III).

Lombi *et al.* (2002) added 2% of the bauxite residue, "red mud" (ca. 60% Fe_2O_3,40% Al_2O_3) to various soils contaminated with heavy metals from smelter emissions and sewage sludge deposition. Red mud reduced the solubility of Cd, Pb, Ni, and Zn, but not Cu. The remedial action of this material was attributed to a rise in soil pH and adsorption of the metals onto oxides of Fe and Mn.

Liming has been demonstrated to be effective in reducing the solubility of TE cations in variable-charge soils by increasing the negative charge on oxides, clays, and organic matter (Kirkham, 2006). The effectiveness of raising the pH on metal immobilization also depends on the liming agent. Bolan and Duraisamy (2003) found that $Ca(OH)_2$ was less effective than KOH in immobilizing Cd^{2+} due to competition between Ca^{2+} and Cd^{2+} for adsorption sites.

2.2 Increasing Trace Element Phytoavailability

The simplest way of producing TE-biofortified plant products is to increase the TE concentration in the soil (Lyons *et al.*, 2004). However, this strategy does not guarantee success and carries risks. Simply increasing the total concentration of the TE in the soil does not necessarily enhance crop uptake, since the solubility and speciation of the TE in soil may render it unavailable to plants.

Phytoextraction requires plants, such as hyperaccumulators, that accumulate inordinate concentrations of TEs in their aerial portions. However, for some of the most common TE contaminants, such as Pb, there are no reliable reports of any hyperaccumulator species. Many authors (e.g., Huang and Cunningham, 1996)) have suggested that that high concentrations of the target TE(s) be brought into soil solution to increase plant uptake.

Chelants, such as ethylenediaminetetraacetic acid (EDTA), ethylenediamine-N,N′-disuccinic acid (EDDS), and nitrilotriacetic acid (NTA) effectively increase the solubility of Pb, Cd, Cu, Zn and other TE cations in soils (Blaylock *et al.*, 1997; Huang and Cunningham, 1996; Robinson *et al.*, 1999a; Tandy*et al.*, 2004; Thayalakumaran *et al.*, 2003). Thiosulphate and thiocyanate salts may be added to mine spoil to induce plants to accumulate Hg (Moreno *et al.*, 2005) and Au (Anderson *et al.*, 1998). High concentrations of chloride anions in soil solubilise Cd because of the formation of soluble and stable complexes [$CdCl^+$ & $CdCl_2$]. While chelants, lixivants, and soil acidification agents are effective in bringing many TEs into soil solution, the speciation of the dissolved TE may be such that the TEs are not phytoavailable and they may simply leach down the soil profile (Fig. 5).

The addition of EDTA to the Ni hyperaccumulator *Berkheya coddii* (Roessler) resulted in decreased Ni uptake, despite increasing the Ni solubility in the soil (Robinson *et al.*, 1999a). Chelants have also ben shown to decrease plant uptake of Cu and Zn uptake from hydroponic solution (Tandy *et al.*, 2004). The addition of chelants reduces plant accumulation of many

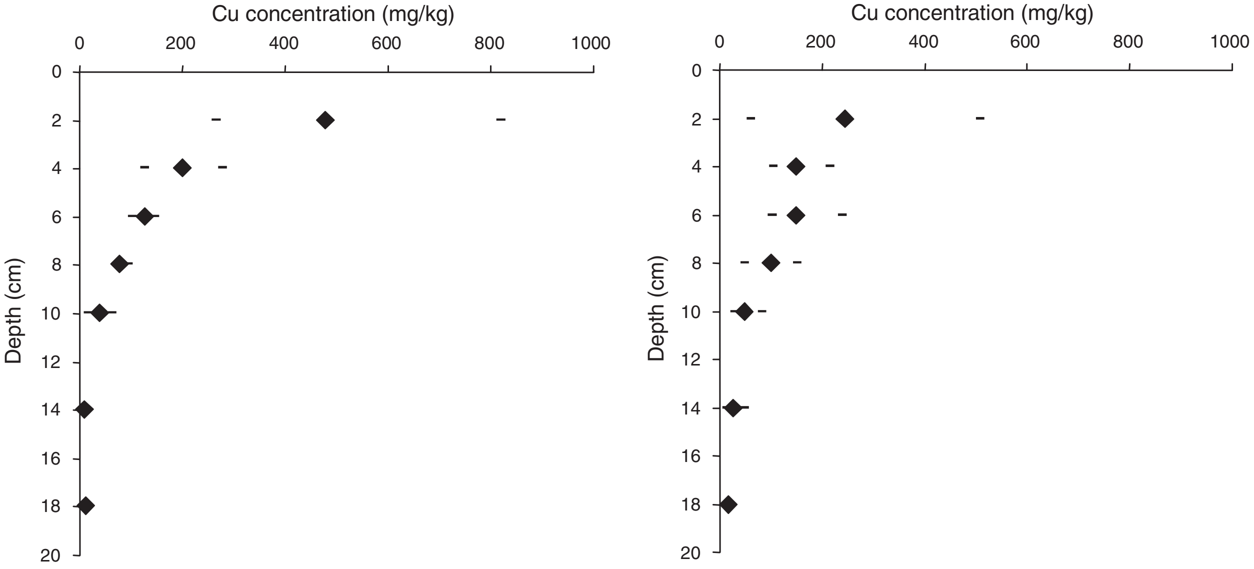

FIG. 5. Mean (blue diamonds), minimum and maximum (black dash) Cu concentrations (n=3) before and one month after the addition of 2g EDTA kg^{-1} soil in which 3-year-old poplars were growing. The Cu concentration in the poplar trees was unaffected, while there was a significant decrease in the Cu concentration in the top 4 cm. This indicates that Cu leaching, rather than phytoextraction caused the decrease.

biologically essential TEs because the TEs are rendered unavailable to the plant's TE transporters (see Section 4.2).

The use of chelants and other lixivants exacerbate the leaching of TEs through the soil profile (Lombi *et al.*, 2001; Nowack *et al.*, 2006). Preferential flow pathways in the soil exacerbate TE leaching to groundwater (Bundt *et al.*, 2000). The use of EDTA to induce plant TE accumulation is unacceptable because it persists in the environment as well as presenting a severe risk of leaching high concentrations of mobile TE-complexes to groundwater (Nowack, 2002). Thayalakumaran *et al.* (2003) demonstrated that, in an undisturbed soil profile containing 300 mg kg^{-1} Cu, plants removed just 5% of the Cu solubilized by EDTA. The remaining 95% leached below the root zone (Fig. 5). An alternative may be the use of NTA or EDDS, which degrade more rapidly than EDTA. However, rapid degradation may not prevent the complexed metal from leaching to groundwater via preferential flow processes, which occur in a matter of hours.

Chelants are usually added as Na salts because the free-acids are only sparingly soluble. Increased Na concentrations in soil can reduce plant growth and causes the dispersion of clay minerals, clogging pores and possibly resulting in increased preferential flow. Most chelants solubilize TEs other than the target element and these may be toxic to plants. Chelants redistribute surface contamination down the soil profile. This has the benefit of reducing the TE concentration near the soil surface, thereby reducing exposure pathways, however, this benefit is more than offset by the risk of TE leaching to groundwater.

Chelant-induced phytoextraction may therefore be limited to applications where the connection to receiving waters has been broken, or where leaching is unimportant (Nowack *et al.*, 2006). Phytoextraction could be conducted ex situ, where the TE-contaminated material would be placed on a liner whereby any leachate could be collected and recycled (Kos and Lestan, 2003). Such systems already exist for soil washing and the recovery of Au from low-grade ores. Potentially, phytomanagement could enhance this process by concentrating the target TE in their biomass. Anderson *et al.* (2005) demonstrated the economic feasibility of lixiviant-induced Au phytoextraction.

III. PROCESS 2: RHIZOSPHERE INTERACTIONS

Plant roots influence soil in their immediate vicinity, a zone known as the rhizosphere. The solubility and speciation of TEs in this zone may be distinct from the bulk soil. Roots improve soil aeration by extracting soil moisture and forming continuous channels for drainage and air exchange. However, increased metabolic activity can result in anaerobic conditions if more oxygen is consumed than can be re-supplied. Plant roots excrete H^+ ions that exchange with nutrient base cations (Salisbury and Ross, 1992). Grass species exude chelants, phytosiderophores, which mobilise Fe and perhaps other essential nutrients of low availability. These organic acids solubilize TEs by competing for cation binding sites. Root exudations may acidify the rhizosphere by up to 2 pH units (Salisbury and Ross, 1992). Such acidification invariably increases the solubility of nonessential TE cations such as Cd^{2+} (Naidu *et al.*, 1994). Root exudates may also render TEs unavailable for plant uptake, and thus enhance plant tolerance to high concentrations of TEs. Kramer *et al.* (2000) reported that Ni-citrate and Ni-histidine complexes accumulated in the roots of Ni-tolerant, but non-accumulator, species of*Thlaspi* spp. growing in an Ni-rich substrate. Wenzel *et al.* (2003) reported higher concentrations of DOC in the rhizosphere of the Ni-hyperaccumulator *Thlaspi goesingense* Halacsy compared to adjacent non-hyperaccumulator species.

3.1 Microflora

Plant root exudates and detritus provide a growth substrate for soil microflora, increasing microbial biomass in the rootzone. The stimulation of soil biological activity affects the speciation, and therefore mobility, of TEs (Pedersen and Albinsson, 1992). Some soil bacteria reduce Cr(VI) to Cr(III) (Pal and Paul, 2004). Soil fauna and microorganisms behave similarly to soil organic matter in that they possess binding sites for some TEs. Robinson *et al.* (2001) found that rhizobacteria such as *Pseudomonas fluorescens* (Flügge) from New Zealand pasturelands accumulated Cd to levels about 100 times that of the ambient solution in which they were grown. As with soil organic matter, the adsorption of Cd by these microorganisms decreased at lower solution pHs. The metabolites produced by root-promoted soil organisms may further change the solubility and speciation of TEs in the rhizosphere. The soil's microbiota plays a crucial role in plant TE tolerance and uptake (Whiting *et al.*, 2001).

With the notable exception of the Brassicaceae, the roots of most plants form symbiotic relationships with mycorrhizal fungi. These fungi solubilise and sequester nutrients and possibly TEs in the rhizosphere, and transport them toward, possibly into the root (Salisbury and Ross, 1992). Many authors have demonstrated that mycorrhizal fungi enhance plant metal tolerance (Marschner, 1995; Schutzendubel and Polle, 2002). Mycorrhizas may absorb TEs in their hyphal sheath and external mycelium. The fungal sheath may reduce access to the apoplast due to hydrophobicity and fungal chelants may complex toxic metals rendering then unavailable for plant uptake (Jentschke and Godbold, 2000). The mechanisms of TE uptake in the fungi and translocation subsequent into the root may be more specific than the corresponding plant uptake mechanisms, and thereby reducing the amount of analogous toxic TEs that enter the symplast. Some microorgaisms increase plant TE uptake. Chopra et al.(2007) showed that rhizosphere microbes from an As-contaminated cattle dip site increased the shoot As concentration of *Agrostis tenuis* (L.) by 45%.

Engineering the rhizobiota could be a tool to enhance or reduce plant TE uptake (Nie *et al.*, 2002). Establishing a healthy substrate microflora, especially mycorrhizal symbiots, can greatly enhance plant tolerance to TEs on contaminated sites (Vosatka, 2001). This may be particularly important in the phytostabilization of contaminated sites with tree species, where mycorhizae play a key role in the protection against TE toxicity (Schutzendubel and Polle, 2002). Ma *et al.* (2006) showed that earthworms and mycorrhizal fungi enhanced plant growth and foliar concentrations of N, P, and K. Certain strains of bacteria, known as Plant Growth Promoting Rhizobacteria (PGPR) enhance plant growth and metal tolerance by fixing atmospheric N, producing phytohormones, and protecting the roots against pathogens (Burd *et al.*, 2000; Kamnev and van der Lelie, 2000). The plants may be inoculated directly with mycorrhizae or PGPRs, or their growth may be promoted using conditioners (Khan, 2005). As yet, it is unclear what role these PGPRs have on plant TE uptake or tolerance. This is a fertile area for future research.

3.2 Root Effects on Trace Element Mobility in the Rhizosphere

Plants affect TE fluxes via the extraction of contaminating TEs into the aboveground biomass, or changing their mobility in the soil profile. The formation of root channels not only affects water flux via enhanced soil drainage and aeration, but also provides pathways for the rapid transport of solutes and suspended particles and colloids (Lesturgez *et al.*, 2004). These transport pathways can exacerbate the risk of groundwater contamination by reducing the contact time of the soil solution with the soil matrix and soil organisms that could otherwise retard the movement by sorption and transformation processes (Bundt *et al.*, 2000).

TE fluxes in soil are dependent on spatial concentration gradients driving diffusion and dispersion, and the mass flow of water (Vogeler *et al.*, 2001). Few studies have investigated the influence of plants on TE leaching from contaminated soils under controlled conditions. Banks *et al.* (1994) found that the Zn leaching from a mine-tailing contaminated soil increased in the order no plants < plants with microbes < plants without microbes. In a further study using contaminated mine tailings and clean subsoil and topsoil covering, the presence of plants increased Cu and Cd leaching in all columns, while Pb was unaffected (Zhu *et al.*, 1999). Turpeinen *et al.* (2000) demonstrated that pine seedlings reduced Pb solubility by up to 93%. Romkens *et al.* (1999) compared Cu speciation in soils with and without plants. Copper solubility was higher in planted pots, but the calculated free Cu^{2+} ion concentration was orders of magnitude lower than in soils without plants.

3.3 Trace Element Heterogeneity in Soil and Root-Trace Element Contact

Regardless of TE solubility and speciation, TEs are only phytoavailable if they come into physical contact with the plant roots. TEs occur heterogeneously in soil, both spatially and temporally. Their concentrations can vary at many scales, from the individual soil particle to the entire catchment. However, only heterogeneity that occurs on the scale of the plant's root zone affects root growth, plant uptake and TE leaching. There are three possible responses of plant roots to a patch of soil where TEs are enriched or depleted: inhibition or avoidance, indifference, and proliferation or foraging (Fig. 6).

Patches where the TE's concentration is not greatly different from the surrounding soil is unlikely to affect root development and hence response (B) in Figure 6 is likely. Unlike macronutrients, there are few studies on the response of roots to TE hotspots. The roots of some tree species such as (*Fagus sylvatica* L.) avoid TE-contaminated hotspots (Breckle and Kahle,

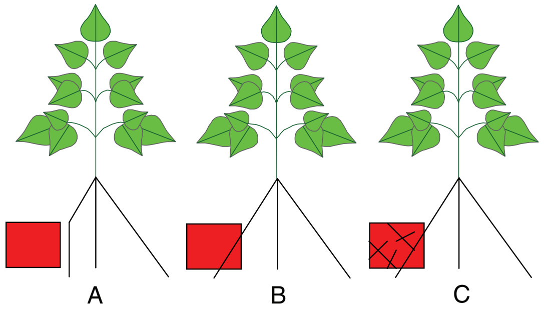

FIG. 6. Possible plant responses to an area of enrichment or depletion of trace elements in soil (red square). (A) avoidance or inhibition, (B) indifference, (C) proliferation or foraging.

1992; Dickinson *et al.*, 1991). Roots of *Lupinus albus* (L.), are inhibited by, or avoid, patches of high B concentration in soil (Menon *et al.*, 2007). Conversely, roots of the Zn hyperaccumulator plant *Thlaspi caerulescens* (J. & C. Presl.) actively forage Zn-rich hotspots in soils (Schwartz *et al.*, 1999; Whiting *et al.*, 2000).

It is unclear whether the promotion or reduction of growth in a TE patch arises from simple simulation or inhibition due to toxicity, or whether there is a root growth strategy. Any growth strategy would need a signalling mechanism, whereby roots near or in the TE patch communicate the location of the patch to the remainder of the root system. When the roots penetrate a patch that contains a high concentration of the TE(s), both plant uptake and leaching of the TE(s) should increase. As discussed above, root-induced changes in the rhizosphere tend to increase the solubility of TEs, making them more available for plant uptake and leaching. The physical structure of the roots may create macropores and provide a direct conduit for the TE to leach into groundwater, either in soil solution, or attached to mobile soil particles (Roulier *et al.*, 2007).

Ploughing may decrease TE heterogeneity by homogenising the soil. It may also bring TE-contaminated soil at depth to the surface, thus increasing the volume of material that can be treated using phytoextraction. However, ploughing may increase TE mobility by creating dust and enhancing TE solubility due to an increase in organic ligands caused by oxidation of the soil's organic matter.

IV. PROCESS 3: PLANT TRACE ELEMENT UPTAKE

All plants remove TEs from their surroundings. Some elements are essential for the plant to complete its life cycle. Plants take up others TEs incidentally, and at high concentrations, these elements may deleteriously affect growth. TEs in plant tissue have an enzyme-activation role, rather than a structural role (Salisbury and Ross, 1992). Depending on their concentration in plant tissue, essential TEs may occur at deficient, optimal or phytotoxic concentrations. Plants tolerate nonessential elements low concentrations, but higher concentrations these elements are invariably phytotoxic.

4.1 Root Uptake of Trace Elements

Plant roots are a sink for soil water. Roots draw soil solution from the rhizosphere to the plants root tissues and then via the stems to the leaves, where it is lost to the atmosphere via transpiration. Any TE in the soil solution that enters the roots will accumulate in either the roots or the shoots of the plant. High TE concentrations in the roots can result from the migration of the TEs, via transpiration-induced mass flow, to the root surface where they are precipitated (Zhao *et al.*, 2000).

The total amount of a TE that accumulates in the plant does not necessarily equal the cumulative product of the soil-solution TE concentration times the volume of water transpired by the plant, as one might predict for non diffusion-limited passive uptake. Deviations may arise where diffusion limits the root's TE supply, or when the rate of TE transport into the root occurs at a different rate from the root's water uptake.

4.1.1 Diffusion Limitation

For a sparingly soluble TE, and under conditions of low transpiration, the plant roots may remove the TE from soil solution at a greater rate than rate at which mass flow transports the element towards the root. Here the rate of diffusion of the TE from the soil matrix towards the root limits the plant's TE uptake (Chapin, 1980). Diffusion out of the soil matrix into the stagnant and mobile water phases may limit plant uptake in high transpiration conditions. Similarly, if the TE absorption and water uptake occur in different regions of the root, then mass flow caused by root water uptake may not transport TE to the zone where they are absorbed.

4.2 Trace Element Entry into the Root Xylem: Apoplastic and Symplastic Transport

The translocation of a TE to the aerial parts of a plant requires its entry into the root xylem, via either the apoplastic or the symplastic pathways (Marschner, 1995). The apoplastic pathway is discontinuous, being interrupted by the endodermis, where the cell walls contain hydrophobic incrustations of suberin, (Casparian strips). This obstructs the passive transfer of solutes into the stele and thence into the root xylem. There are small discontinuities in the Casparian strips at the junctions of lateral root branches (Crowdy and Tanton, 1970). Soil organisms, pathogens, or mechanical disturbance may create disruptions in the endodermis. The application of selected pesticides can disrupt root-membranes allowing chelated TEs to pass directly into the root xylem via the apoplastic pathway (Blaylock, 2000). Small amounts of some TEs may enter the root xylem directly via the apoplastic pathway at the root apices (Harrison-Murray and Clarkson, 1973).

Most TEs that enter the root xylem must, at some point, penetrate a cell membrane and move through the cytoplasm of the endodermis (Steudle and Peterson, 1998). Boron (B), in the form of boric acid (H_3BO_3), can pass directly through membranes (Dordas and Brown, 2000; Stangoulis *et al.*, 2001).

However, other TEs can only traverse the plasma membrane via embedded protein transporters. There are numerous transporters. For example, in *Arabidopsis thaliana* (L.), 4589 genes code for membrane-spanning proteins, representing some 18% of the protein-coding genome (Ward, 2001).

Passive transporters, or ion channels, permit ions with a specific size and charge to move across the cell membrane down their concentration gradient. Passive plant uptake requires that the target ion concentration in the cytoplasm be lower than the surroundings (Marschner, 1995). Active transporters, requiring metabolic energy, move TEs across membranes against their concentration gradient (Salisbury and Ross, 1992). There are multiple transporters for essential elements, being either constitutive or inducible (Reid and Hayes, 2003). Constitutive transporters are always operational, while a nutrient deficiency may induce the plant to activate additional transporters. Antiporters eject toxic elements from the cytoplasm. These may decrease plant trace uptake and increase the plant's tolerance to high concentrations of TEs in the environment (Martinoia *et al.*, 2002).

There are many known transporters for plant macronutrients, namely N, P, K, S, Mg, and Ca. However, less is known about TE transport across membranes, due to the difficulty of measuring their low concentrations. Transporters are known for Mn, Zn, Cu, Fe, Ni, Co, and Cd (Reid and Hayes, 2003; Reid *et al.*, 1996; Salt and Wagner, 1993). Most is known about the acquisition of Fe, which may be growth limiting at high soil pHs. Except for the Poaceae, roots reduce soluble, possibly chelated, Fe(III) to Fe(II) via ferric chelate-reductase enzymes. The reduced Fe(II) enters the cell via a ferrous transporter (strategy I). Members of the Poaceae (strategy II), release phytosiderophores that solubilize Fe(III), and the chelate enters the cell via a specialized transport system (Reid and Hayes, 2003). Similar phytometallophore transporter systems may exist for other TEs, notably Cu (Gries *et al.*, 1998) and Zn (Hopkins *et al.*, 1998). These systems become more active when the essential TE is deficient. This increased activity may also result in the transport of high amounts of nonessential TEs (Cohen *et al.*, 1998).

In their review of nutrient uptake by plants, Reid and Hayes (2003) concluded that most membrane-transport proteins, which mediate nutrient influx or efflux, lack specificity. Therefore, nonessential TE ions with a similar size to nutrients may be taken up into the symplast and ultimately be translocated to the shoots. Khattak *et al.* (1991) demonstrated that plants take up arsenate ($AsO_4{}^{3-}$) via the same physiological mechanism as phosphate ($PO_4{}^{3-}$). Similarly, Tl^+ may enter via the K^+ ion channel (Skulsky 1991), Cd^{2+} may enter via either Ca^{2+} or Zn^{2+} transport system, and Ni^{2+} may enter along with Mg^{2+}. Similarly, selenate ($SeO_4{}^{2-}$) and sulphate ($SO_4{}^{2-}$) may use the carrier-mediated process to enter the plant. Sulphate and selenate compete for common uptake in the roots and thus selenate uptake can be strongly decreased by high sulphur supply (Zayed and Terry, 1992) Small, uncharged, TE moieties, for example H_3BO_3, may also pass through aquaporins, which are passive protein gateways that permit the passage of water through cell membranes (Maurel, 1997).

Chaney *et al.* (2007) identified up regulation of transmembrane TE transport as the most fertile area of research to increase plant TE uptake. High biomass plants can be genetically altered to extract larger amounts of TE from soils (Rugh *et al.*, 1998). Dhankher *et al.* (2002) engineered *Arabidopsis thaliana* (L.) to accumulate As by inserting two bacterial genes that imparted tolerance and the ability to translocate As to the aerial portions. There are potential dangers in using this approach for phytomanagement. Increasing the uptake of TEs in high-biomass plants may facilitate the entry of toxic elements into the food chain via herbivore consumption. Worse, if the modified plant is also used in agriculture, then cross-pollination may create hybrids that result in food crops that are rich in unwanted TEs. Future research in this area should include producing such plants with sterile pollen granules and/or with reduced flower production.

4.3 Trace Element Translocation and Storage within the Plant

Plant water uptake drives TE translocation from the roots to the shoots via the xylem (Salt *et al.*, 1995). In the aboveground portions, the highest concentrations are often found in the leaves as they are the major water sink prior to evaporation of the water. However, phloem transport may redistribute some TEs within the plant, notably Fe, Mn, Zn, Cu (Pate *et al.*, 1975), Ni, Co, and Cd (Riesen and Feller, 2005). Boron, Sr, and Ba are generally phloem immobile (Kochian, 1991). Nicotianamine, which is ubiquitous in vascular plants, transports Fe in the phloem (Stephan and Scholz, 1993). The authors noted that most other divalent cations have higher constants of complex formation with nicotianamine than does Fe. This indicates that the same process may translocate other, possible nonessential, TEs. Essential TEs that are phloem mobile are translocated to developing tissues, a phenomenon that may be controlled by the phytohormones, especially cytokines (Salisbury and Ross, 1992). Phloem transport of TEs increases when the TEs are deficient in plant tissue (Gupta and Gupta, 2005).

The Ni hyperaccumulator *Berkheya coddii* (Roessler) translocates Ni to areas of new growth after tissue is excised (Robinson *et al.*, 1997a). This is consistent with the hypothesis that the Ni affords herbivore protection to the plant, and it is therefore translocated to organs that are vulnerable to herbivory. Other studies have shown that hyperaccumulator plants store TEs in the epidermis (Kupper *et al.*, 2000), the tissue first encountered by herbivores.

4.3.1 *The Importance of Trace Element Translocation and Storage in Biofortification*

Phloem transport is of particular importance when the goal of phytomanagement is to biofortify desirable TEs, such as Se, in food crops, especially when the edible part is the seed. There is a physiological barrier to TE translocation into the seed during maturation (Ernst *et al.*, 1992). Seeds and fruits generally have low transpiration rates. Therefore, xylem transport delivers only minimal amounts of TEs. Soil amendments that solubilize TEs in the soil and increase root uptake may not necessarily result in an increased TE concentration in the seeds. Synthetic chelants, such as EDTA, may bind TEs so strongly, that they become unavailable for complexation with plant transporter chelants, such as nicotianamine, which may be required for phloem transport into the seeds.

The speciation of TEs and the presence of phytoligands in the edible portions of the plant are critical for biofortification, because this affects the TE's absorption by humans or animals. Seeds store much of there phosphate as phytate ($C_6H_{18}O_{24}P_6$), a polydentate chelate that can complex essential TE cations, notably Fe and Zn, rendering them unavailable for absorption by the human gut (Zhou and Erdman, 1995). The effect of phytate on ruminants is smaller due to the presence of the phytate-degrading enzyme phytase in the gut (Lei *et al.*, 1993). Therefore, phytomanagement with the goal of biofortification, should limit plants' supply of phosphate. Polyphenolys, another phytochemical, may also reduce TE bioavailability to humans. A possible solution to the phytate problem is the genetic engineering of plants that overproduce phytase, and enzyme that degrades phytate when the food is prepared (Storksdieck and Hurrell, 2007). The benefits of phytate degradation for trace element nutrition need to be balanced against the beneficial effects, namely antioxidant activity (Vucenik and Shamsuddin, 2003), of phytate itself.

Plants also contain "promoter" compounds, such as inuline, palmitic acid, riboflavin and ascorbate, that stimulate the absorption of essential TEs by the gut (White and Broadley, 2005). Enhancing plant production of these compounds may be as important as increasing the concentration of the target TE in the plant.

4.4 Plant Tolerance of Trace Elements

In plant tissues, nonessential TEs, and essential TEs that occur at concentrations above there metabolic requirement, are potentially toxic. High concentrations of TEs may damage plant tissues via oxidative stress, which causes damage to biomolecules and depletion of adenosine triphosphate (ATP). High TEs also block essential functional groups in biomolecules and displace essential nutrients in biomolecules (Schutzendubel and Polle, 2002). Autooxidation of variable charge ions, such as Fe^{2+} (Fenton's reaction) and Cu^+ can produce free radicals and free radical precursors (H_2O_2) that damage cellular structures. Many TEs, especially Pb, Cd, and Hg, bind irreversibly to S, N, or O in biomolecules, possibly displacing essential TE ions, resulting in a loss of function.

All plants have basic TE tolerance mechanisms. Some species and varieties can survive in soil with inordinately high concentrations of TEs. Mostly, such plants are only tolerant to the TEs that occur in the soil in which they grow (Schat and Vooijs, 1997), indicating that there tolerance mechanisms are element specific. There are nine groups of tolerance mechanisms:

- Root avoidance
- External sequestration by bacteria, mycorrhizal fungi
- Exuding chelants that render the TEs unavailable for plant uptake
- Restriction of transport across plasmalemma into the cell
- Active efflux into the apoplast (pumping)
- Chelation of the TEs in the cytosol by phytochelatins, metallothioneins or organic acids
- Production of heat-shock proteins that repair cellular damage
- Storage of chelated TEs in the vacuole
- Abscission of organs, such as leaves, with a high TE load.

The toxic effects of TEs are larger when they are present as free ions. Therefore, most TEs found in plant tissues are complexed with organic ligands (Hall, 2002). In addition to low molecular weight organic acids, plants produce two classes of metal binding proteins, phytochelatins and metallothioneins (Cobbett and Goldsbrough, 2002). Metallothioneins are low molecular weight, cystine-rich, metal binding proteins (Kagi, 1991). Phytochelatins have the general structure (γ-Glu Cys)$_n$-Gly where n = 2–11 and are rapidly induced in plants that become exposed to high TE concentrations (Rauser, 1995; Zenk, 1996). TE cations that enter the cytoplasm are immediately chelated by "chaperone" ligands (Clemens, 2001) and transported to TE requiring cytosolic proteins or to organelles. The chaperone ligands are then freed to complex other TE cations. TEs that are in excess to metabolic requirements are usually transported to the vacuole, where they are often complexed with phytochelatins and stored (Hall, 2002). Hyperaccumulator plants often have the highest TE concentrations in the vacuoles of the epidermis (Frey *et al.*, 2000; Kupper *et al.*, 1999). Toxic concentrations of TEs in cells may induce the production of heat-shock proteins (Lewis *et al.*, 1999). Heat-shock proteins function in the production and repair of proteins under stress conditions (Hall, 2002).

4.4.1 Establishing Vegetation in Phytomanagement

Plant tolerance to TEs is an essential component of the phytomanagement of contaminated sites. When planting on a contaminated site, species or varieties are required that tolerate the contaminants and any nutrient imbalances in the substrate. Caution is required when introducing exotic species or genetically-modified organisms that they do not establish themselves as weeds (Wolfe and Bjornstad, 2002). However, exotic species have the advantage that they are less likely to suffer from native herbivores, thus increasing growth and reducing the amount of TE that enters, uncontrolled, into the food chain.

FIG. 7. A plant growth trial, comprising poplar, willow and *Eucalyptus nitens* (H. Deane & Maiden), on B-contaminated wood waste, Kopu, New Zealand. As well as interspecific differences, there was large variation in B-tolerance between poplar clones.

The soil contaminants may not be the main cause of infertility. Dickinson (2000) pointed out that on many contaminated sites weed competition and inadequate soils were more significant to plant growth than soil contaminants.

Since no contaminated sites are alike, choosing the most suitable species requires a short planting trial that tests several varieties on a small area of the site, particularly for non-soil media such as sewage sludge or mine tailings. Figures 7 and 8 show planting trials on both a B-contaminated sawdust pile and a B and salty saleniferous soil.

Substrate amendments and a succession of plant species may be required to establish the desired climax vegetation (Fig. 9). Phytomanagement systems that use several species or varieties overcome the risk that a new pest or climatic event destroys all the plants. Low-growing species may be combined with deciduous tree species to provide a transpiring green surface during the winter months. Legumes enhance fertility in nitrogen-deficient substrates.

Before planting, capping contaminated sites with fertile soil provides a better substrate for plant growth and a buffer zone that stores water from heavy rainfall events. Although more expensive, such capping systems reduce leaching by providing a deeper root zone, thus providing more time for the vegetation to extract and transpire the infiltrated rain water. The cost of earthmoving and reengineering the site offsets the advantages of a soil cap.

Given the large genetic variability of tolerance between varieties and ecotypes of a species, there is scope for improving plant tolerance by selective breeding or genetic engineering. Genetic engineering for plant tolerance does not face the same potential problem as engineering for accumulation, namely facilitating the entry of toxic TEs into the food chain, since tolerance may be achieved by exclusion of contaminants in the root zone. Local ecotypes that are adapted to the climatic and edaphic conditions of the site would be the best candidates for such genetic manipulation.

FIG. 8. Field-grown Brassica juncea planted on seleniferous soils high in boron and salinity in the western side of central California.

FIG. 9. Vegetation succession on mine tailings. *Alyssum bertolonii* (Desv.) establishes on the Ni and Mg rich tailings, which have a low organic matter content. *A.bertolonii* adds organic matter to the soil, allowing the establishment of a native grass *Cortaderia toetoe* (Zotov).

4.5 Plant Trace Element Uptake as Affected by the Soil's Trace Element Concentration

Baker (1981) divided plant species into "hyperaccumulators", "indicators" and "excluders" according to their aboveground TE concentrations in relation to the TE concentration in the soil. Figure 10 shows an adaptation of Baker's original version of how plants respond to the soluble concentration of TEs in soil.

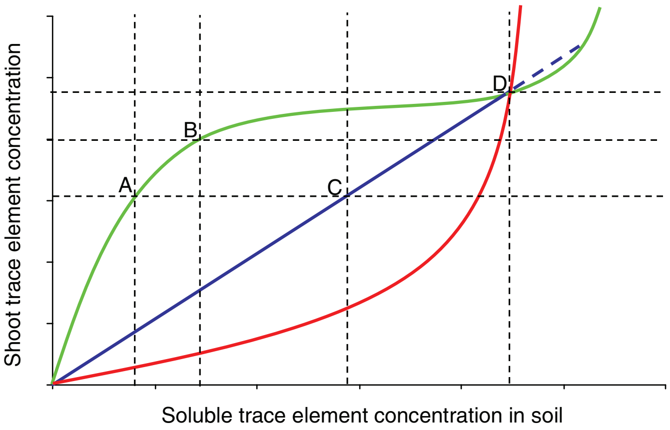

FIG. 10. Plant uptake of trace elements as a function of soluble trace elements in the soil (adapted from Baker, 1981). The green, blue and red lines indicate accumulators, indicators and excluders. Points A and C are sufficient concentrations for accumulators and indicators. Point B is the optimal concentration for accumulators. Concentrations above point D result in phytotoxicity.

4.5.1 Accumulators and Hyperaccumulators

Accumulator and hyperaccumulator plants (Fig. 10, green line), actively accumulate TEs into the shoots when the shoots are below a sufficient concentration (Fig. 10, point A). Above this concentration, luxury uptake (Salisbury and Ross, 1992) occurs until the shoot concentration reaches an optimal level (Fig. 10, point B). At higher concentrations, roots may restrict TE uptake until the threshold concentration (Fig. 10, point D) is reached, where the mechanisms of homeostasis break down and phytotoxicity symptoms appear.

Hyperaccumulator plants (Brooks *et al*., 1977) take up TEs, even those that are non-essential, to similar concentrations as macronutrients. Brooks *et al*. (1977) used hyperaccumulation to describe plants that take up Ni to concentrations greater than 1000 mg kg^{-1} on a dry matter basis on Ni-rich ultramafic (serpentine) soil. This concentration is at least an order of magnitude greater than concentrations found in other plants growing in the same environment. There are 440 species of known hyperaccumulator species, 75% of which hyperaccumulate Ni (Reeves, 2006). The remainder hyperaccumulate As, Cd, Mn, Na, Tl, and Zn (Brooks, 1998b). This list is growing as new hyperaccumulator species are discovered.

While hyperaccumulator plants can achieve a high TE concentration in their shoots, their biomass production is usually inferior to non-hyperaccumulator plants. Notable exceptions are Ni hyperaccumulators of the genera *Alyssum* (Robinson *et al*., 1997b) and *Berkheya* (Robinson *et al*., 1997a) (Fig. 11). These plants can achieve shoot Ni concentrations of >10000 mg kg^{-1} (1%) on a dry matter basis, while producing more than fifteen tonnes of dry matter per hectare per year. Improved agronomic

FIG. 11. Professor Robert Brooks, standing in a field of the high biomass hyperaccumulator *Berkheya coddii* (Roessler), Rustenberg, South Africa, 1997.

practices and genetic modification can increase the biomass production of smaller varieties of hyperaccumulator plants (Ow *et al*., 1998).

4.5.2 Indicators

Indicator plants (Fig. 10, blue line) take up TEs in proportion to their soluble concentration in the soil, thus the shoot concentration indicates the soil concentration. Such an uptake response implies there is no specific mechanism for homeostasis in the plant. Many plants follow the "indicator" pattern of accumulation for non essential TEs. However, some essential TEs, notably B, are taken up in this manner. Here passive uptake occurs both above and below the sufficient tissue concentration (Fig. 10, point C). Plants that do not occur naturally on metalliferous soils usually behave as 'indicators' when grown in the presence of nonessential elements. Madejon *et al*. (2004) showed that the leaves of *Populus alba* (L.) indicated the soluble Cd and Zn concentration in soils of the Guadiamar floodplain that became contaminated following a mine tailings dam collapse at Aznalcóllar in 1998. Cadmium uptake by this species may facilitate the entry of this toxic element into the food chain (Dominguez *et al*., 2008).

4.5.3 Excluders

For nonessential elements, such as Cd, Ni, and As, as well as essential elements that occur at high concentrations in soil, some plants have active exclusion mechanisms (Fig. 10, red line). Most plants that occur naturally on metalliferous soils are recognised as being "excluders." Here, the nonessential, possibly toxic, TE are maintained at a low tissue concentration, until the regulatory mechanisms are overloaded, or there is a disruption of the plasma membrane at the apoplast/symplast interface (Fig. 10, point D). When this occurs, TEs flood into the plant resulting in reduced growth accompanied by chlorosis or necrosis.

Excluder plants are ideal candidates for the phytostabilization of mine wastes or soils with a TE concentration so high that they are phytotoxic for other species. Excluders can stabilize the soil surface, reducing erosion and leaching, while minimizing the risk that the contaminating TEs enter the food chain via herbivores.

4.6 Calculating Trace Element Uptake into the Shoots

As discussed previously, the amount of TE accumulated in the shoots of the plant is a function of the soluble concentration of TE in the soil that is in contact with the plant's roots, the transpiration of the plant, and the rate of TE transfer between the soil solution and the root xylem. Thus

$$M(t) = \int_0^{z_R} \int_0^{t} R(t', z)\, T(t')\, C(t'z)\, \phi\left(C\left(t'z\right)\right) dt dz \quad [3]$$

where M is plant metal uptake in the time interval (0, t; days). R is the fraction of the total root water uptake at depth z (meters), T is the transpiration rate (L day^{-1}), C is the TE concentration (mg L^{-1}) in the soil solution, and ϕ is the root absorption factor.

R is dependent on the plant species and the soil in which it is growing; R can be estimated for most species. T can be calculated using environmental variables and a crop coefficient (Allen *et al*., 1998). C can be measured using soil extractions, or direct measurements of soil solution (Ernst, 1996).

The root absorption factor (ϕ) is a dimensionless parameter that represents the root xylem / soil solution metal concentration quotient (Robinson *et al.*, 2003a).

$$\phi = \frac{C_r}{C} \quad [4]$$

where C_r is the soluble metal concentration (mg L^{-1}) in the root xylem and C is the soluble metal concentration (mg L^{-1}) in the soil solution.

The parameter ϕ is a simple lumped parameter intended to remove the need to measure the aforementioned complex and often poorly understood factors that influence the passage of TEs from the soil into the roots.

The value of ϕ can be approximated using the plant's total water use, above-ground dry biomass, and the metal concentration in soil solution thus:

$$\phi \cong \frac{PW}{1000C} \quad [5]$$

ϕ equals the Root Absorption Factor for the TE species (dimensionless), P equals the TE concentration in the above-ground dry biomass (mg kg^{-1}), W equals the plant's water use efficiency (kg t^{-1}), and C equals the concentration of metal in soil solution (mg L^{-1}).

The issue is further complicated by the fact that ϕ changes depending on C. Therefore, several measurements are needed to determine the plant's response over a range of soil concentrations. For non-phytotoxic concentrations of C, the change in ϕ over a soil concentration range can be modelled by adding a constant K,that describes the decrease in ϕ, as Cincreases, thus:

$$\phi(C) = \frac{\phi_1 C_1}{C_1 + K(C - C_1)} \quad [6]$$

where ϕ (C) equals root adsorption factor at soil solution concentration C (mg L^{-1}), ϕ_1 equals the measured root adsorption factor at concentration C_1 (mg L^{-1}), and K ($0 \leq K < 1$) is the decay constant.

This model has been validated using a lysimeter experiment, where measured values were compared with those calculated using independent parameters (Robinson *et al.*, 2003b). The model performs well for predominately xylem-transported TEs, such as B, Cd, Mn, and Zn (Fig. 12), but poorly for Cu. The poor performance of the model to predict leaf Cu concentrations may be due to redistribution of this element in the plant via phloem transport.

4.7 Physicochemical Adsorption of Trace Elements onto Plant Surfaces

Plant cell walls carry a negative charge and thus effectively adsorb cationic TEs. Cation adsorption is important in the roots, where the plant's apoplast may adsorb both essential and nonessential TEs. Plant roots may accumulate Pb to concentrations many times higher than that of the soil solution (Huang and Cunningham, 1996) or even the bulk soil (Kahle, 1993). Deposition of TEs onto plant shoots may occur via dust, soil particles contained within rain splash, and industrial emissions such as smoke. Such surface-borne TEs may simply settle on the plant's surface and be removed in the next rainfall. Some may entangle themselves in structures such as leaf hairs, or they may become incorporated into waxy layer or stomata. There, some fraction of the TEs may dissolve and adhere to plant tissue.

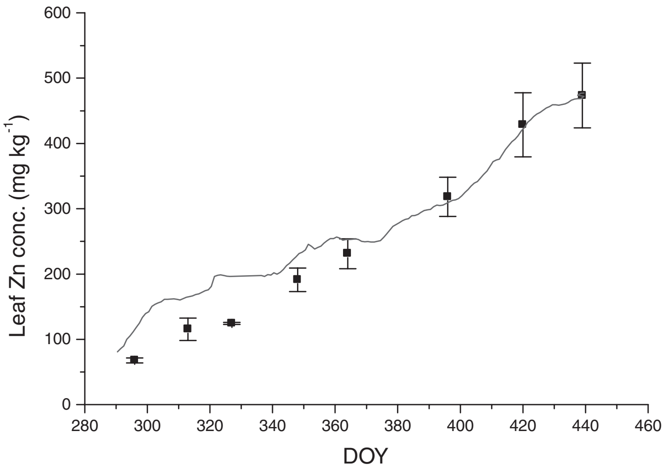

FIG. 12. Leaf Zn concentration in 'Toa' (*Populus euramericana* × *yunnanensis*) as a function of Day Of Experiment (DOY) growing in lysimeters filled with sawdust (Robinson *et al.*, 2005). The error bars represent the standard error of the mean. The blue line is the Zn concentration calculated using Eq. [4].

4.7.1 *Calculating the Fraction of Trace Elements Originating from Soil Particles*

Distinguishing between surface-deposited TEs in plant shoots and those that the plant has translocated from the roots is important for the assessment and design of phytomanagement systems. TE adsorption affects the fate of TEs in the environment. Even careful washing does not remove all surface-deposited TEs. Therefore, some fraction of a plant sample may actually consist of soil particles or dissolved TEs that have adhered to the leaves and stems. To determine the mass of soil and dust deposition on leaf samples, one can measure an indicator element that is indicative of the level of soil deposition (Hinton *et al.*, 1995). The indicator element should not be of interest in the study and should occur at high concentrations in the soil. Its physiological uptake by the plant via the roots should be either negligible or maintained at a constant tissue concentration over a wide range of soil concentrations. It is advantageous if it correlates with the TEs of interest. This is because the calculation uses the indicator element concentration in the soil where the plant is growing; however, wind may have deposited soil from some distance onto the plant shoots. Titanium, Ba, and Cr(III) make good indicator elements, since the plants do not take up significant concentrations of these elements (Brooks, 1998b). Therefore, the measured concentrations are directly proportional to surface deposited dust. However, these elements may not occur at high concentrations in the soil, leading to measurement difficulties and calculation errors. By contrast, Fe occurs at concentrations $>1\%$ in many soils, and is easy to measure. Iron is an essential nutrient that plants take up to concentrations of around 100 mg kg^{-1} (Marschner, 1995). Therefore, when using Fe to calculate surface deposition, one must first subtract a baseline value, namely the Fe that the plants have accumulated through root uptake. The mass fraction of soil on the plant sample, F (kg kg^{-1}) is therefore

$$F = \frac{I_{measured} - I_{baseline}}{S_I} \quad [7]$$

where $I_{measured}$ is the measured indicator element concentration in the plant tissue (mg kg^{-1}), $I_{baseline}$ is the baseline concentration of the indicator element that the plant has accumulated through the roots and translocated to the shoots (mg kg^{-1}), and S_I is the concentration of the indicator element in the soil (mg kg^{-1}). The corrected plant concentration of the target element, P^* (mg kg^{-1}), is:

$$P^* = P - FS \quad [8]$$

where P and S are the measured concentrations (mg kg^{-1}) of the target element in the plant and soil.

Errors will arise in the corrected values using Fe as an indicator element when the amount of Fe that the plant has taken up through the roots differs from the baseline value used in the calculation. Similarly, surface-deposited dust from soil that is chemically distinct from the soil used in Eq. [8] soil will result in errors.

The quality of the calculations can be validated by comparing independent tracers on the same sample set. Following the calculation, the plant macronutrient concentrations usually do not change greatly, whereas immobile, non-essential elements, such as Pb and Sb usually decrease. Robinson *et al.* (2008) used Eq. [7] with Fe (baseline value = 27.3 mg kg^{-1}) to calculate the dust deposition on the surface of plants collected from a shooting range. Separate calculations using Cu (baseline value = 2.3 mg kg^{-1}) and Al (baseline value = 22 mg kg^{-1}) were in reasonable agreement ($r = 0.96$ and $r = 0.92$) with the values calculated using Fe. Upon correction of the plant concentrations using Fe as an indicator element, the concentrations of the plant macronutrients, namely K, Ca, Mg, and P, stayed within 98% of their original value. However, the concentrations of Pb and Sb, the major soil contaminants, dropped to 45% and $<20\%$ of their original values.

V. FEASIBILITY OF PHYTOMANAGEMENT AND FERTILE AREAS FOR FUTURE RESEARCH

Phytomanagement is only feasible if it satisfies environmental regulation and either returns a profit or costs less than alternative technologies or inaction (Robinson *et al.*, 2003a). This is challenging, because unlike other systems to manage TE fluxes, such as capping and soil removal, phytomanagement systems are site dependent.

It is impractical to conduct long-term field trials to optimize the phytomanagement for every site. Therefore, whole system models that calculate TE flux are an essential component to test the feasibility of phytomanagement and design phytomanagement systems. Such models would remove the need for field trials by revealing where phytoextraction or phytostabilization will not meet regulations under a risk-based regime. Potentially, validated models could be used to gain regulatory approval for phytomanagement without the need for lengthy demonstration trials. Such models could also be used to determine the best management practices for each site.

A suite of ‘whole system’ have been developed to calculate the leaching of agrichemicals. These include the Soil Plant Atmosphere System Model (SPASMO), Leaching Estimation and Chemistry Model (LEACHM), HYDRUS – 1D, Water and Agrochemicals in soil, crop and Vadose Environment (WAVE) (Sarmah *et al.*, 2005). These models calculate water and solute transport in the root-zone using Richards’ and the Convection – Dispersion Equations, or simplified ‘tipping bucket’ algorithms (Green *et al.*, 1999). Evapotranspiration from the planted soil is usually calculated using the Penman-Monteith equation combined with a crop coefficient (Allen *et al.*, 1998). These models could be adapted to calculate TE movement in the soil-plant system. This would necessitate the incorporation of specific root-TE interactions and the changes in TE mobility over the long term induced by the vegetation. A fertile area for future phytomanagement research is the integration of the biogeochemical and economic factors that affect phytomanagement into existing whole system models that calculate water and solute transport.

5.1 Concluding Summary

Phytomanagement is a long-term technology. To compete successfully with alternative remediation technologies, contaminated site phytomanagement requires the production of valuable biomass as well as reducing environmental risk. Similarly, valuable biomass production is the most important criterion for biofortification and phytomining.

In this light, contaminated land is an undervalued resource. Bioenergy and timber production on contaminated land has several advantages over production on non-contaminated fertile soils. Most importantly, productive agricultural land will not be taken out of food production, which, in the case of bioenergy production, has led to increased food costs (*Economist*, 2007). Since the biomass produced either fixes carbon (timber) or produces CO_2-neutral energy (bioenergy), carbon credits may add further value to the operation. In addition to timber or bioenergy, the biomass of vegetation from a phytomanagement program may have other uses, where the possibly elevated concentrations of TEs in the plant tissues are either unimportant (because they will not be consumed) or beneficial essential nutrients (such as Se or Zn). This may include cut flowers, cotton, and stock fodder. In the latter case, the biomass should be carefully monitored for excessive concentrations or the presence of other contaminants.

Hitherto, narrow definitions have handicapped phytomanagement. For example, phytoextraction as a stand-alone technology is unlikely to find widespread use for the cleanup of contaminated sites and thus less likely to receive positive attention from scientists and regulators. In contrast, phytomanagement that combines phytoextraction, phytostabilization and the production of valuable biomass could be used extensively worldwide. Phytomanagement does not proscribe the use of other remediation technologies, but can form part of an environmental toolkit to achieve the best outcome. In particular, sequestration agents that immobilise toxic TEs (Smith *et al.*, 2008), clay liners, geotextiles, capping with uncontaminated topsoil, and reactive barriers will enhance the effectiveness of phytomanagement to reduce the risks posed by contaminating TEs.

Given the penchant for producing valuable crops on contaminated land, how does TE phytomanagement differ from simply growing normal crops of maize or trees? Firstly, soil conditioners may be used, if needed, to alter TE mobility. Secondly, the plants may be genetically distinct from standard crop or forestry species, particularly concerning TE tolerance and accumulation. Thirdly, a thorough assessment of the TE fluxes in the system and the need for a crop management program that minimizes the risk posed by the contaminating TEs. This final point exposes, yet again, a critical knowledge gap in phytomanagement research, namely the need for basic research on the integration of the many, individually well understood, processes that affect plant–TE interactions, including good agronomic practices.

Successful phytomanagement requires a multidisciplinary approach. Much work has focused, at the microcosm scale, on TE chemistry in soil, and molecular mechanisms of TE uptake plants. There is a lacuna of information on the biophysical processes affecting TE fluxes in the vadose zone. In particular, the role of roots on contaminant fluxes is poorly understood. Roots absorb water and TEs, and release organic matter, promoting the growth of microflora, and creating channels down through which soil particles, with TEs attached, can travel. Roots occur heterogeneously in soil, as do TEs. Can we model their interactions? Innovative techniques for rhizosphere research such as the specialized rhizobox described by Wenzel *et al.* (2001) can be used to measure chemical and microbiological processes with minimal disturbance to roots. Neutron radiography can reveal dynamic rhizosphere processes, including root development and water flux, with minimal disturbance to the system (Menon *et al.*, 2007; Tullis and Wright, 2007).

The potential application of phytomanagement is large, given the number of people affected by soilborne TEs. The growth, or lack thereof, of profitable private companies that provide this technology will indicate the true value of phytomanagement.

REFERENCES

Allen, R. G., Pererira, L. S., Raes, D., and Smith, M. 1998. Crop Evapotranspiration. Guidelines for Computing Crop Water Requirements. **In**: *FAO Irrigation and Drainage Paper No. 56*. FAO, Rome.

Anderson, C., Moreno, F. N., and Meech, J. A. 2005. A field demonstration of gold phytoextraction technology. *Mineral Engineering* **18**: 385–392.

Anderson, C. W. N., Brooks, R. R., Chiarucci, A., LaCoste, C. J., Leblanc, M., Robinson, B. H., Simcock, R. and Stewart, R. B. 1999. Phytomining for nickel, thallium and gold. *J.Geochem. Explor.* **67**: 407–415.

Anderson, C. W. N., Brooks, R. R., Stewart, R. B,. and Simcock, R. 1998. Harvesting a crop of gold in plants. *Nature* **395**: 553–554.

Baker, A. J. M. 1981. Accumulators and excluders—strategies in the response of plants to heavy-metals. *J. Plant Nutr.* **3**: 643–654.

Baker, A. J. M., McGrath, S. P., Sidoli, C. M. D., and Reeves, R. D. 1994. The possibility of in-situ heavy-metal decontamination of polluted soils using crops of metal-accumulating plants. *Resour. Conserv. Recy.* **11**: 41–49.

Banks, M. K., Schwab, A. P., Fleming, G. R., and Hetrick, B. A. 1994. Effects of plants and soil microflora on leaching of zinc from mine tailings. *Chemosphere.* **29**: 1691–1699.

Bañuelos, G., Terry, N., Leduc, D. L., Pilon-Smits, E. A. H., and Mackey, B. 2005. Field trial of transgenic Indian mustard plants shows enhanced phytoremediation of selenium-contaminated sediment. *Environ. Sci. Technol.* **39**: 1771–1777.

Bañuelos, G. S. 2006. Phyto-products may be essential for sustainability and implementation of phytoremediation. *Environ. Pollut.* **144**: 19–23.

Bañuelos, G. S. 2009. *Phytoremediation of selenium contaminated soil and water produces biofortified products and new agricultural byproducts.* CRC Press, Boca Raton, Fl.

Bañuelos, G. S., Ajwa, H. A., Wu, L., and Zambrzuski, S. 1998. Selenium accumulation by Brassica napus grown in Se-laden soil from different depths of Kesterson Reservoir. *J. Soil Contamination* **7**: 481–494.

Bañuelos, G. S., and Lin, Z. Q. 2009. *Development and Uses of Biofortified Agricultural Products.* CRC Press, Boca Raton, FL.

Bell, D. T. 1999. Australian trees for the rehabilitation of waterlogged and salinity-damaged landscapes. *Aust. J. Bot.* **47**: 697–716.

Blaylock, M. J. 2000. Field demonstrations of phytoremediation of Pb contaminated soils. In *Phytoremediation of contaminated soil and water*. Edited by Terry, N. and Bañuelos, G. pp. 1–12. Lewis Publishers, Boca Raton, FL.

Blaylock, M. J., Salt, D. E., Dushenkov, S., Zakharova, O., Gussman, C., Kapulnik, Y., Ensley, B. D., and Raskin, I. 1997. Enhanced accumulation of Pb in Indian mustard by soil-applied chelating agents. *Environ. Sci. Technol.* **31**: 860–865.

Bolan, N. S. and Duraisamy, V. P. 2003. Role of inorganic and organic soil amendments on immobilisation and phytoavailability of heavy metals: a review involving specific case studies. *Aust. J. Plant Res.* **41**: 533–555.

Bolan, N. S., Naidu, R., Syers, J. K., and Tillman, R. W. 1999. Surface charge and solute interactions in soils. *Adv. Agron*, **67**: 87–140.

Boominathan, R., Saha-Chaudhury, N. M., Sahajwalla, V., and Doran, P. M. 2004. Production of nickel bio-ore from hyperaccumulator plant biomass: Applications in phytomining. *Biotechnol. Bioeng.* **86**: 243–250.

Breckle, S. W., and Kahle, H. 1992. Effects of toxic heavy-metals (Cd, Pb) on growth and mineral-nutrition of beech (FAGUS-SYLVATICA L). *Vegetation* **101**: 43–53.

Brooks, R. R. 1998a. Phytoremediation by volatilisation. **In**: *Plants That Hyperaccumulate Heavy Metals: Their Role in Phytoremediation, Microbiology, Archaeology, Mineral Exploration and Phytomining*. Brooks, R. R. , Ed., pp. 289–312. CAB International, Wallingford, UK.

Brooks, R. R. 1998b. *Plants That Hyperaccumulate Heavy Metals: Their Role in Phytoremediation, Microbiology, Archaeology, Mineral Exploration and Phytomining*. CAB International, Wallingford, UK.

Brooks, R. R., Chambers, M. F., Nicks, L. J., and Robinson, B. H. 1998. Phytomining. *Trends Plant Sci.* **3**: 359–362.

Brooks, R. R., Lee, J., Reeves, R. D., and Jaffre, T. 1977. Detection of nickeliferous rocks by analysis of herbarium specimens of indicator plants. *J. Geochem. Explor.* **7**: 49–57.

Brown, S., Chaney, R., Hallfrisch, J., Ryan, J. A., and Berti, W. R. 2004. In situ soil treatments to reduce the phyto- and bioavailability of lead, zinc, and cadmium. *J. Environ. Qual.* **33**: 522–531.

Brown, S., Sprenger, M., Maxemchuk, A., and Compton, H. 2005. Ecosystem function in alluvial tailings after biosolids and lime addition. *J. Environ. Qual.* **34**: 139–148.

Bundt, M., Albrecht, A., Froidevaux, P., Blaser, P., and Fluhler, H. 2000. Impact of preferential flow on radionuclide distribution in soil. *Environ. Sci. Technol.* **34**: 3895–3899.

Burd, G. I., Dixon, D. G., and Glick, B. R. 2000. Plant growth-promoting bacteria that decrease heavy metal toxicity in plants. *Can. J. Microbiol.* **46**: 237–245.

Cakmak, I. 2008. Enrichment of cereal grains with zinc: Agronomic or genetic biofortification? *Plant Soil* **302**: 1–17.

Chaney, R. L. 1983. Plant uptake of inorganic waste constituents. **In**: *Land Treatment of Hazardous Wastes*. J. F. Parr, P. B. Marsh and J. M. Kla Eds., pp. 50–76. Noyes Data Corps, Park Ridge, NJ.

Chaney, R. L., Angle, J. S., Broadhurst, C. L., Peters, C. A., Tappero, R. V., and Sparks, D. L. 2007. Improved understanding of hyperaccumulation yields commercial phytoextraction and phytomining technologies. *J. Environ. Qual.* **36**: 1429–1443.

Chaney, R. L., Angle, J. S., McLntosh, M. S., Reeves, R. D., Li, Y. M., et al. 2005. Using hyperaccumulator plants to phytoextract soil Ni and Cd. *Zeitschrift Fur Naturforschung C-a J. Biosciences* **60**: 190–198.

Chapin, F. S. 1980. The mineral-nutrition of wild plants. *Annu. Rev. Ecol. Syst.* **11**: 233–260.

Cheng, S. F., and Hseu, Z. Y. 2002. In-situ immobilization of cadmium and lead by different amendments in two contaminated soils. *Water Air Soil Pollut.* **140**: 73–84.

Chopra, B. K., Bhat, S., Mikheenko, I. P., Xu, Z., Yang, Y., Luo, X., Chen, H., van Zwieten, L., Lilley, R. M., and Zhang, R. 2007. The characteristics of rhizosphere microbes associated with plants in arsenic-contaminated soils from cattle dip sites. *Sci. Total. Environ.* **378**: 331–342.

Clemens, S. 2001. Molecular mechanisms of plant metal tolerance and homeostasis. *Planta* **212**: 475–486.

Clothier, B. E., and Green, S. R. 1997. Roots: The big movers or water and chemical in soil. *Soil Sci.* **162**: 534–543.

Cobbett, C., and Goldsbrough, P. 2002. Phytochelatins and metallothioneins: Roles in heavy metal detoxification and homeostasis. *Ann. Rev. Plant Biol.* **53**: 159–182.

Cohen, C. K., Fox, T. C., Garvin, D. F., and Kochian, L. V. 1998. The role of iron-deficiency stress responses in stimulating heavy-metal transport in plants. *Physiol.* **116**: 1063–1072.

Crowdy, S. H., and Tanton, T. W. 1970. Water pathways in higher plants. 1. Free space in wheat leaves. *J. Exp. Bot.* **21**: 102–111.

Dhankher, O. P., Li, Y. J., Rosen, B. P., Shi, J., Salt, D., Senecoff, J. F., Sashti, N. A., and Meagher, R. B. 2002. Engineering tolerance and hyperaccumulation of arsenic in plants by combining arsenate reductase and gamma-glutamylcysteine synthetase expression. *Nature Biotechnol.* **20**: 1140–1145.

Dickinson, N. M. 2000. Strategies for sustainable woodland on contaminated soils. *Chemosphere.* **41**: 259–263.

Dickinson, N. M., Turner, A. P., and Lepp, N. W. 1991. How do trees and other long-lived plants survive in polluted environments? *Funct. Ecol.* **5**: 5–11.

Dominguez, M. T., Maranon, T., Murillo, J. M., Schulin, R., and Robinson, B. H. 2008. Trace element accumulation in woody plants of the Guadiamar Valley, SW Spain: A large-scale phytomanagement case study. *Environ. Pollut.* **152**: 50–59.

Dordas, C. and Brown, P. H. 2000. Permeability of boric acid across lipid bilayers and factors affecting it. *J. Membr. Biol.* **175**: 95–105.

Economist, April 4, 2007. Castro was right. In *The Economist*.

Ernst, W. H. O. 1996. Bioavailability of heavy metals and decontamination of soils by plants. *Appl. Geochem.* **11**: 163–167.

Ernst, W. H. O., Verkleij, J. A. C., and Schat, H. 1992. Metal tolerance in plants. *Acta Bot. Neerl.* **41**: 229–248.

Fernandez, M. D., Cagigal, E., Vega, M. M., Urzelai, A., Babin, M., Pro, J., and Tarazona, J. V. 2005. Ecological risk assessment of contaminated soils through direct toxicity assessment. *Ecotoxicol. Environ. Saf.* **62**: 174–184.

Ferro, A. M., Rieder, J. P., Kennedy, J., and Kjelgren, R. 1997. Phytoremediation of groundwater using poplar trees. **In**: *Phytoremediation*. Thibeault, C. A. and Savage, L. M., Eds., pp. 201–212. International Business Communications Inc., Southborough, MA.

Fox, J. L. 2001. Canadian farmer found guilty of Monsanto canola patent infringement. *Nature Biotechnol.* **19**: 396–397.

Franca, F., and Ferrari, M. 2002. Impact of micronutrient deficiencies on growth: The stunting syndrome. *Ann. Nut. Metab.* **46**: 8–17.

Frey, B., Keller, C., Zierold, K,. and Schulin, R. 2000. Distribution of Zn in functionally different leaf epidermal cells of the hyperaccumulator Thlaspi caerulescens. *Plant Cell Environ.* **23**: 675–687.

GACGC 1994. World in Transition: The Threat to Soils. **In**: *Annual Report*. German Advisory Council on Global Change, Bonn.

Granel, T., Robinson, B., Mills, T., Clothier, B., Green, S., and Fung, L. 2002. Cadmium accumulation by willow clones used for soil conservation, stock fodder, and phytoremediation. *Aust. J. Plant Res.* **40**: 1331–1337.

Green, S. R., Clothier, B. E., Mills, T. M,. and Millar, A. 1999. Risk assessment of irrigation requirements of field crops in a maritime climate. *J. Crop Prod.* **2**: 353–377.

Gries, D., Klatt, S., and Runge, M. 1998. Copper-deficiency-induced phytosiderophore release in the calcicole grass Hordelymus europaeus. *New Phytol.* **140**: 95–101.

Gupta, U. C., and Gupta, S. C. 2005. Future trends and requirements in micronutrient research. *Commun. Soil Sci. Plant Anal.* **36**: 33–45.

Hall, J. L. 2002. Cellular mechanisms for heavy metal detoxification and tolerance. *J. Exp. Bot.* **53**: 1–11.

Hamon, R. E., and McLaughlin, M. J. 1999. Use of the hyperaccumulator Thlaspi cearulescens for bioavailable contaminant stripping. **In**: *5th International Conference on the Biogeochemistry of Trace Elements*. Wenzel, W. W., Adriano, D. C., Alloway, B., Doner, H., Keller, C., Lepp, N. W., Mench, M., Naidu, R., and Pierzynski, G. M., Eds., pp. 908–909, Vienna.

Harrison-Murray, R. and Clarkson, D. 1973. Relationships between structural development and absorption of ions by the root system of Cucurbita pepo. *Planta* **114**: 1–16.

Hinton, T .G., Kopp, P., Ibrahim, S., Bubryak, I., Syomov, A., Tobler, L, and Bell, C. 1995. A Comparison of techniques used to estimate the amount of resuspended soil on plant-surfaces. *Health Phys.* **68**: 523–531.

Hopkins, B. G., Whitney, D. A., Lamond, R. E., and Jolley, V. D. 1998. Phytosiderophore release by sorghum, wheat, and corn under zinc deficiency. *J. Plant Nutr.* **21**: 2623–2637.

Huang, J. W., and Cunningham, S. D. 1996. Lead phytoextraction: Species variation in lead uptake and translocation. *New Phytol.* **134**: 75–84.

Jardine, P. M., Fendorf, S. E., Mayes, M. A., Larsen, I. L., Brooks, S. C,. and Bailey, W. B. 1999. Fate and transport of hexavalent chromium in undisturbed heterogeneous soil. *Environ. Sci. Technol.* **33**: 2939–2944.

Jentschke, G. and Godbold, D. L. 2000. Metal toxicity and ectomycorrhizas. *Physiol. Plant.* **109**(2): 107–116.

Kagi, J. H. R. 1991. Overview of metallothionein. *Methods. Enzymol.* **205**: 613–626.

Kahle, H. 1993. Response of roots of trees to heavy-metals. *Environ. Exper. Bot.* **33**: 99–119.

Kamnev, A. A., and van der Lelie, D. 2000. Chemical and biological parameters as tools to evaluate and improve heavy metal phytoremediation. *Biosci. Rep.* **20**: 239–258.

Keller, C., Ludwig, C., Davoli, F., and Wochele, J. 2005. Thermal treatment of metal-enriched biomass produced from heavy metal phytoextraction. *Environ. Sci. Technol.* **39**: 3359–3367.

Khan, A. G. 2005. Rote of soil microbes in the rhizospheres of plants growing on trace metal contaminated soils in phytoremediation. *J. Trace Elem. Med. Biol.* **18**: 355–364.

Khattak, R. A., Page, A. L., Parker, D. R. and Bakhtar, D. 1991. Accumulation and interactions of arsenic, selenium, molybedenium and phosphrous in alfalfa. *J. Environ. Qual.* **20**: 165–168.

King, J. C. 2002. Evaluating the impact of plant biofortification on human nutrition. *J. Nutr.* **132**: 511S–513S.

Kirkham, M. B. 2006. Cadmium in plants on polluted soils: Effects of soil factors, hyperaccumulation, and amendments. *Geoderma* **137**: 19–32.

Kochian, L. V. 1991. Mechanisms of micronutrient uptake and translocation in plants. **In**: *Micronutrients in Agriculture*. Mortvedt, J. J., Cox, F. R., Schumin, L. M., and Welch, R. M., Eds., pp. 229–296. Soil Science Society of America, Madison, WI.

Koppolu, L., Prasad, R., and Clements, L. D. 2004. Pyrolysis as a technique for separating heavy metals from hyperaccumulators. Part III: pilot-scale pyrolysis of synthetic hyperaccumulator biomass. *Biomass Bioenerg.* **26**: 463–472.

Kos, B., and Lestan, D. 2003. Induced phytoextraction/soil washing of lead using biodegradahle chelate and permeahle barriers. *Environ. Sci. Technol.* **37**: 624–629.

Kramer, U., Pickering, I. J., Prince, R. C., Raskin, I., and Salt, D. E. 2000. Subcellular localization and speciation of nickel in hyperaccumulator and non-accumulator Thlaspi species. *Physiol.* **122**: 1343–1353.

Kupper, H., Lombi, E., Zhao, F. J., and McGrath, S. P. 2000. Cellular compartmentation of cadmium and zinc in relation to other elements in the hyperaccumulator Arabidopsis halleri. *Planta* **212**: 75–84.

Kupper, H., Zhao, F. J., and McGrath, S. P. 1999. Cellular compartmentation of zinc in leaves of the hyperaccumulator Thlaspi caerulescens. *Physiol.* **119**: 305–311.

Leblanc, M., Petit, D., Deram, A., Robinson, B. H., and Brooks, R. R. 1999. The phytomining and environmental significance of hyperaccumulation of thallium by Iberis intermedia from Southern France. *Econ. Geol. Bull. Soc.* **94**: 109–113.

Lei, X. G., Ku, P. K., Miller, E. R., Ullrey, D. E., and Yokoyama, M. T. 1993. Supplemental microbial phytase improves bioavailability of dietary zinc to weanling pigs. *J. Nutr.* **123**: 1117–1123.

Lesturgez, G., Poss, R., Hartmann, C., Bourdon, E., Noble, A., and Ratana-Anupap, S. 2004. Roots of Stylosanthes hamata create macropores in the compact layer of a sandy soil. *Plant Soil* **260**: 101–109.

Lewis, S., Handy, R. D., Cordi, B., Billinghurst, Z., and Depledge, M. H. 1999. Stress proteins (HSP's): Methods of detection and their use as an environmental biomarker. *Ecotoxicol.* **8**: 351–368.

Li, Y. M., Chaney, R., Brewer, E., Roseberg, R., Angle, J. S., Baker, A., Reeves, R., and Nelkin, J. 2003. Development of a technology for commercial phytoextraction of nickel: economic and technical considerations. *Plant Soil* **249**: 107–115.

Licht, L. A. and Isebrands, J .G. 2005. Linking phytoremediated pollutant removal to biomass economic opportunities. *Biomass Bioenerg.* **28**: 203–218.

Lombi, E., Zhao, F. J., Dunham, S. J. and McGrath, S. P. 2001. Phytoremediation of heavy metal-contaminated soils: Natural hyperaccumulation versus chemically enhanced phytoextraction. *J. Environ. Qual.* **30**: 1919-1926.

Lombi, E., Zhao, F. J., Zhang, G. Y., Sun, B., Fitz, W., Zhang, H., and McGrath, S. P. 2002. In situ fixation of metals in soils using bauxite residue: chemical assessment. *Environ. Pollut.* **118**: 435–443.

Lyons, G. H., Stangoulis, J. C. R., and Graham, R. D. 2004. Exploiting micronutrient interaction to optimize biofortification programs: The case for inclusion of selenium and iodine in the HarvestPlus program. *Nutr. Rev* **62**: 247–252.

Ma, Q. Y., Traina, S .J., Logan, T. J., and Ryan, J. A. 1993. In-situ lead immobilization by apatite. *Environ. Sci. Technol.* **27**: 1803–1810.

Ma, Y., Dickinson, N. M., and Wong, M. H. 2006. Beneficial effects of earthworms and arbuscular mycorrhizal fungi on establishment of leguminous trees on Pb/Zn mine tailings. *Plant Biol. Biochem.* **38**: 1403–412.

Macnair, M. R. 1987. Heavy-Metal Tolerance in Plants - a Model Evolutionary System. *Trends Ecol. Evol.* **2**: 354–359.

Madejon, P., Maranon, T., Murillo, J. M., and Robinson, B. 2004. White poplar (Populus alba) as a biomonitor of trace elements in contaminated riparian forests. *Environ. Pollut.* **132**: 145–155.

Margni, M. B., Lothenbach, B., and Schulin, R. 1997. Entsorgungsverfahren für schwermetall-belastetes Pflanzenmaterial. *TerraTech* **2**: 53–57.

Marschner, H. 1995. *Mineral nutrition of higher plants*. Mineral nutrition of higher plants, Academic Press, London.

Martinoia, E., Klein, M., Geisler, M., Bovet, L., Forestier, C., Kolukisaoglu, U., Muller-Rober, B., and Schulz, B. 2002. Multifunctionality of plant ABC transporters - more than just detoxifiers. *Planta* **214**: 345–355.

Maurel, C. 1997. Aquaporins and water permeability of plant membranes. *Annual Rev. Physiol. Plant Molecul. Biol.* **48**: 399–429.

McGowen, S. L., Basta, N. T., and Brown, G. O. 2001. Use of diammonium phosphate to reduce heavy metal solubility and transport in smelter-contaminated soil. *J. Environ. Qual.* **30**: 493–500.

McGrath, S. P., Sidoli, C. M. D., Baker, A. J. M., and Reeves, R. D. 1993. The potential for the use of metal-accumulating plants for the in situ decontamination of metal-polluted soils. **In**: *Integrated Soil and Sediment Research: A Basis for Proper Protection*. Eijsackers, H. J. P. and Hamers, T., Eds., pp. 673–676. Kluwer Academic Publishers, Dordrecht.

McLaughlin, M. J., Tiller, K. G., Naidu, R., and Stevens, D. P. 1996. Review: The behaviour and environmental impact of contaminants in fertilizers. *Aust. J. Plant Res.* **34**: 1–54.

McNaughton, K. G., and Jarvis, P .G. 1983. Effects of vegetation on transpiration and evaporation. **In**: *Water Deficits and Plant Growth*. Kozlowski, T. T., Ed., pp. 1–47. Academic Press, New York.

Mendez, M. O., and Maier, R. M. 2008a. Phytoremediation of mine tailings in temperate and arid environments. *Rev. Environ. Sci. Bio/Technol.* **7**: 47–59.

Mendez, M. O., and Maier, R. M. 2008b. Phytostabilization of mine tailings in arid and semiarid environments - An emerging remediation technology. *Environ. Health. Perspect.* **116**: 278–283.

Menon, M., Robinson, B., Oswald, S. E., Kaestner, A., Abbaspour, K. C., Lehmann, E., and Schulin, R. 2007. Visualization of root growth in heterogeneously contaminated soil using neutron radiography. *Euro. J. Soil Sci.* **58**: 802–810.

Mills, T. M., Robinson, B. H., Sivakumaran, S., Arnold, B., Clothier, B. E., and Kim, N. 2005. Current practice and future land-use: the sustainability of productive sector environments. *Acta Hort. (ISHS)* **694**: 159–164.

Moreno, F. N., Anderson, C. W. N., Stewart, R. B., Robinson, B. H., Nomura, R., Ghomshei, M., and Meech, J. A. 2005. Effect of thioligands on plant-Hg accumulation and volatilisation from mercury-contaminated mine tailings. *Plant Soil* **275**: 233–246.

Naidu, R., Bolan, N. S., Kookana, R. S., and Tiller, K. G. 1994. Ionic-strength and pH effects on the sorption of cadmium and the surface-charge of soils. *Euro. J. Soil Sci.* **45**: 419–429.

Nestel, P., Bouis, H. E., Meenakshi, J. V., and Pfeiffer, W. 2006. Biofortification of staple food crops. *J. Nutr.* **136**: 1064–1067.

Nicks, L. J., and Chambers, M. F. 1994. Nickel farming. *Discover*, Mag. 19 September.

Nicks, L. J., and Chambers, M. F. 1995. Farming for metals. *Mining Environ. Mgt.* 15–18 September.

Nie, L., Shah, S., Rashid, A., Burd, G. I., Dixon, D. G. and Glick, B. R. 2002. Phytoremediation of arsenate contaminated soil by transgenic canola and the plant growth-promoting bacterium Enterobacter cloacae CAL2. *Physiol. Biochem.* **40**: 355–361.

Nixon, D. J., Stephens, W., Tyrrel, S. F., and Brierley, E. D. R. 2001. The potential for short rotation energy forestry on restored landfill caps. *Bioresour. Technol.* **77**: 237–245.

Nowack, B. 2002. Environmental chemistry of aminopolycarboxylate chelating agents. *Environ. Sci. Technol.* **36**: 4009–4016.

Nowack, B., Schulin, R., and Robinson, B. H. 2006. Critical assessment of chelant-enhanced metal phytoextraction. *Environ. Sci. Technol.* **40**: 5225–5232.

Oliver, M. A. 1997. Soil and human health: a review. *Euro. J Soil Sci.* **48**: 573–592.

Ow, D. W., Shewry, P. R., Napier, J. A., and Davis, P .J. 1998. Prospects of engineering heavy metal detoxification genes in plants. **In**: *Symposium of the Industrial Biochemistry and Biotechnology Group of the Biochemical Society*, pp. 111–124, IACR-Long Ashton Research Station, Long Ashton, Bristol, UK.

Pal, A., and Paul, A. K. 2004. Aerobic chromate reduction by chromium-resistant bacteria isolated from serpentine soil. *Microbiol. Res.* **159**: 347–354.

Pate, J. S., Sharkey, P. J., and Lewis, O. A. M. 1975. Xylem to phloem transfer of solutes in fruiting shoots of legumes, studied by a phloem bleeding technique. *Planta* **122**: 11–26.

Pedersen, K., and Albinsson, Y. 1992. Possible effects of bacteria on trace-element migration in crystalline bed-rock. *Radiochim. Acta* **58-9**: 365–369.

Pulford, I. D., McGregor, S. D., Duncan, H. J, and Wheeler, C. T. 1995. Uptake of Heavy Metals From Contaminated Soil by Trees. **In**: *Fourteenth Annual Symposium. Current Topics in Plant Biochemistry, Physiology and Molecular Biology. Will Plants have a role in Bioremediation?* pp. 49–50, Interdisciplinary Plant Group, University of Missouri, Columbia, MO.

Quinn, J. J., Negri, C. M., Hinchman, R. R., Moos, L. P., Wozniak, J. B., and Gatliff, E. 2001. Predicting the effect of deep-rooted hybrid poplars on the groundwater flow system at a large scale phytoremediation site. *Int J. Phytoremediation* **3**: 41–60.

Ramesh, S. A., Choimes, S., and Schachtman, D. P. 2004. Over-expression of an Arabidopsis zinc transporter in Hordeum vulgare increases short-term zinc uptake after zinc deprivation and seed zinc content. *Plant Mol. Biol.* **54**: 373–385.

Rauser, W. E. 1995. Phytochelatins and related peptides - structure, biosynthesis, and function. *Physiol.* **109**: 1141–1149.

Reeves, R. 2006. Hyperaccumulation of trace elements by plants. **In**: *Phytoremediation of Metal-Contaminated Soils*. Morel, J. L., Echevarria, G., and Goncharova, N., Eds., pp. 25–52.

Reid, R., and Hayes, J. 2003. Mechanisms and control of nutrient uptake in plants. **In**: *International Review of Cytology - a Survey of Cell Biology, Vol 229*, pp. 73–114.

Reid, R. J., Brookes, J. D., Tester, M. A., and Smith, F. A. 1996. The mechanism of zinc uptake in plants—Characterisation of the low-affinity system. *Planta* **198**: 39–45.

Riesen, O., and Feller, U. 2005. Redistribution of nickel, cobalt, manganese, zinc, and cadmium via the phloem in young and maturing wheat. *J. Plant Nutr.* **28**: 421–430.

Robinson, B., and Anderson, C. W. N. 2007. Phytoremediation in New Zealand / Australia. **In**: *Phytoremediation Methods and Reviews*. Willey, N., Ed., pp. 455–468. Humana Press, Totowa, NJ.

Robinson, B., Fernandez, J. E., Madejon, P., Maranon, T., Murillo, J. M., Green, S., and Clothier, B. 2003a. Phytoextraction: an assessment of biogeochemical and economic viability. *Plant Soil* **249**: 117–125.

Robinson, B., Green, S., Mills, T., Clothier, B., van der Velde, M., Laplane, R., Fung, L., Deurer, M., Hurst, S., Thayalakumaran, T., and van den Dijssel, C. 2003b. Phytoremediation: using plants as biopumps to improve degraded environments. *Aust. J. PlantRes.* **41**: 599–611.

Robinson, B., Mills, T., Green, S., Chancerel, B., Clothier, B., Fung, L., Hurst, S., and McIvor, I. 2005. Trace element accumulation by poplars and willows used for stock fodder. *N. Z. J. Agric. Res.* **48**: 489–497.

Robinson, B., Russell, C., Hedley, M., and Clothier, B. 2001. Cadmium adsorption by rhizobacteria: implications for New Zealand pastureland. *Agric. Ecosyst. Environ.* **87**: 315–321.

Robinson, B. H., Bischofberger, S., Stoll, A., Schroer, D., Furrer, G., Roulier, S., Gruenwald, A., Attinger, W., and Schulin, R. 2008. Plant uptake of trace elements on a Swiss military shooting range: Uptake pathways and land management implications. *Environ. Pollut.* **153**: 668–676.

Robinson, B. H., Brooks, R. R., and Clothier, B. E. 1999a. Soil amendments affecting nickel and cobalt uptake by Berkheya coddii: Potential use for phytomining and phytoremediation. *Ann. Bot.* **84**: 689–694.

Robinson, B. H., Brooks, R. R., Gregg, P. E. H., and Kirkman, J. H. 1999b. The nickel phytoextraction potential of some ultramafic soils as determined by sequential extraction. *Geoderma* **87**: 293–304.

Robinson, B. H., Brooks, R. R., and Hedley, M. J. 1999c. Cobalt and nickel accumulation in Nyssa (tupelo) species and its significance for New Zealand agriculture. *N. Z. J. Agric. Res.* **42**: 235–240.

Robinson, B. H., Brooks, R. R., Howes, A. W., Kirkman, J. H., and Gregg, P. E. H. 1997a. The potential of the high-biomass nickel hyperaccumulator Berkheya coddii for phytoremediation and phytomining. *J. Geochem. Explor.* **60**: 115–126.

Robinson, B. H., Chiarucci, A., Brooks, R. R., Petit, D., Kirkman, J. H., Gregg, P. E. H., and DeDominicis, V. 1997b. The nickel hyperaccumulator plant Alyssum bertolonii as a potential agent for phytoremediation and phytomining of nickel. *J. Geochem. Explor.* **59**: 75–86.

Robinson, B. H., Green, S. R., Chancerel, B., Mills, T. M., and Clothier, B. 2007. Poplar for the phytomanagement of boron contaminated sites. *Environ. Pollut.* **150**: 225–233.

Robinson, B. H., Schulin, R., Nowack, B., Roulier, S., Menon, M., Clothier, B. E., Green, S. R., and Mills, T. M. 2006. Phytoremediation for the management of metal flux in contaminated sites. *Forest Snow Landscape Res.* **80**: 221–234.

Romkens, P., Bouwman, L. A., and Boon, G. T. 1999. Effect of plant growth on copper solubility and speciation in soil solution samples. *Environ. Pollut.* **106**: 315–321.

Roulier, S., Robinson, B., Kuster, E., and Schulin, R. 2008. Analysing the preferential transport of lead in a vegetated roadside soil using lysimeter experiments and a dual-porosity model. *Eur. J. Soil Sci.* **59**: 61–70.

Rugh, C. L., Senecoff, J. F., Meagher, R. B., and Merkle, S. A. 1998. Development of transgenic yellow poplar for mercury phytoremediation. *Natur. Biotechnol.* **16**: 925–928.

Ryan, J. A., Zhang, P. C., Hesterberg, D., Chou, J., and Sayers, D. E. 2001. Formation of chloropyromorphite in a lead-contaminated soil amended with hydroxyapatite. *Environ. Sci. Technol.* **35**: 3798–3803.

Salisbury, F. B. and Ross, C. W. 1992. *Physiol.* Wadsworth, Belmont, CA.

Salt, D. E., Blaylock, M., Kumar, N., Dushenkov, V., Ensley, B. D., Chet, I., and Raskin, I. 1995. Phytoremediation—a novel strategy for the removal of toxic metals from the environment using plants. *Bio-Technol.* **13**: 468–474.

Salt, D. E., and Wagner, G. J. 1993. Cadmium transport across tonoplast of vesicles from oat roots - evidence for a Cd2+/H+ antiport activity. *J. Biol. Chem.* **268**: 12297–12302.

Sarmah, A. K., Close, M. E., Pang, L. P., Lee, R., and Green, S. R. 2005. Field study of pesticide leaching in a Himatangi sand (Manawatu) and a Kiripaka bouldery clay loam (Northland). 2. Simulation using LEACHM, HYDRUS-1D, GLEAMS, and SPASMO models. *Aust. J. Plant Res.* **43**: 471–489.

Schat, H., and Vooijs, R. 1997. Multiple tolerance and co-tolerance to heavy metals in Silene vulgaris: a co-segregation analysis. *New. Phytol.* **136**: 489–496.

Schulin, R., Khoschgoftarmanesh, A., Afyuni, M., Nowack, B., and Frossard, E. 2009. Effects of soil management on zinc uptake and its bioavailability in plants. **In**: *Development and Use of Biofortified Agricultural Products*. Bañuelos, G. S. and Lin, Z. Q., Eds., pp. 95–114. CRC Press, Boca Raton, FL.

Schutzendubel, A., and Polle, A. 2002. Plant responses to abiotic stresses: heavy metal-induced oxidative stress and protection by mycorrhization. *J. Exp. Bot.* **53**: 1351–1365.

Schwartz, C., Morel, J. L., Saumier, S., Whiting, S. N., and Baker, A. J. M. 1999. Root development of the Zinc-hyperaccumulator plant Thlaspi caerulescens as affected by metal origin, content and localization in soil. *Plant Soil* **208**: 103–115.

Seaman, J. C., Arey, J. S., and Bertsch, P. M. 2001. Immobilization of nickel and other metals in contaminated sediments by hydroxyapatite addition. *J. Environ. Qual.* **30**: 460–469.

Sellers, W. D. 1965. *Physical Climatology*. University of Chicago Press, Chicago.

Skulsky, I. A. 1991. Isomorphism of Tl+ and K+ in membrane-transport processes. *Tsitologiya.* **33**: 118–129.

Smith, E., Naidu, R., Weber, J., and Juhasz, A. L. 2008. The impact of sequestration on the bioaccessibility of arsenic in long-term contaminated soils. *Chemosphere.* **71**: 773–780.

Stangoulis, J. C. R., Reid, R. J., Brown, P. H., and Graham, R. D. 2001. Kinetic analysis of boron transport in Chara. *Planta* **213**: 142–146.

Stephan, U. W., and Scholz, G. 1993. Nicotinamine - mediator of transport of iron and heavy-metals in the phloem. *Physiol. Plant* **88**: 522–529.

Steudle, E., and Peterson, C. A. 1998. How does water get through roots? *J. Exp. Bot.* **49**: 775–788.

Storksdieck, D., and Hurrell, R. 2007. The impact of trace elements from plants on human nutrition: a case study for biofortification. **In**: *9th International Conference on the Biogeochemistry of Trace Elements (ICOBTE)*, Bejing, China.

Swartjes, F. A. 1999. Risk-based assessment of soil and groundwater quality in the Netherlands: Standards and remediation urgency. *Risk Anal.* **19**: 1235–1249.

Tandy, S., Bossart, K., Mueller, R., Ritschel, J., Hauser, L., Schulin, R., and Nowack, B. 2004. Extraction of heavy metals from soils using biodegradable chelating agents. *Environ. Sci. Technol.* **38**: 937–944.

Thayalakumaran, T., Robinson, B. H., Vogeler, I., Scotter, D. R., Clothier, B. E., and Percival, H. J. 2003. Plant uptake and leaching of copper during EDTA-enhanced phytoremediation of repacked and undisturbed soil. *Plant Soil* **254**: 415–423.

Tullis, B. P., and Wright, S. J. 2007. Wetting front instabilities: a three-dimensional experimental investigation. *Transport Porous Med.* **70**: 335–353.

Turpeinen, R., Salminen, J., and Kairesalo, T. 2000. Mobility and bioavailability of lead in contaminated boreal forest soil. *Environ. Sci. Technol.* **34**: 5152–5156.

Vangronsveld, J., Colpaert, J. V., and VanTichelen, K. K. 1996. Reclamation of a bare industrial area contaminated by non-ferrous metals: Physicochemical and biological evaluation of the durability of soil treatment and revegetation. *Environ. Pollut.* **94**: 131–140.

Vogeler, I., Green, S. R., Clothier, B. E., Kirkham, M. B., and Robinson, B. H. 2001. Contaminant transport in the root zone. **In**: *Trace Elements in the Soil, Bioavailability, Flux and Transfer*. Iskandar, I. K. and Kirkham, M. B., Eds., pp. 175–198. Lewis Publishers, Boca Raton, FL.

Vosatka, M. 2001. A future role for the use of arbuscular mycorrhizal fungi in soil remediation: a chance for small-medium enterprises? *Minerva Biotecnol.* **13**: 69–72.

Vucenik, I., and Shamsuddin, A. M. 2003. Cancer inhibition by inositol hexaphosphate (IP6) and inositol: From laboratory to clinic. *J. Nutr.* **133**: 3778S–3784S.

Ward, J. M. 2001. Identification of novel families of membrane proteins from the model plant Arabidopsis thaliana. *Bioinformat.* **17**: 560–563.

Wei, S. H., Zhou, Q. X., and Wang, X. 2005. Identification of weed plants excluding the uptake of heavy metals. *Environ. Int.* **31**: 829–834.

Wenzel, W. W., Bunkowski, M., Puschenreiter, M., and Horak, O. 2003. Rhizosphere characteristics of indigenously growing nickel hyperaccumulator and excluder plants on serpentine soil. *Environ. Pollut.* **123**: 131–138.

Wenzel, W. W., Wieshammer, G., Fitz, W. J., and Puschenreiter, M. 2001. Novel rhizobox design to assess rhizosphere characteristics at high spatial resolution. *Plant Soil* **237**: 37–45.

White, P. J. and Broadley, M. R. 2005. Biofortifying crops with essential mineral elements. *Trends Plant Sci.* **10**: 586–593.

Whiting, S. N., De Souza, M. P., and Terry, N. 2001. Rhizosphere bacteria mobilize Zn for hyperaccumulation by Thlaspi caerulescens. *Environ. Sci. Technol.* **35**: 3144–3150.

Whiting, S. N., Leake, J. R., McGrath, S. P., and Baker, A. J. M. 2000. Positive responses to Zn and Cd by roots of the Zn and Cd hyperaccumulator Thlaspi caerulescens. *New. Phytol.* **145**: 199–210.

Wolfe, A. K., and Bjornstad, D. J. 2002. Why would anyone object? An exploration of social aspects of phytoremediation acceptability. *Crit. Rev. Plant Sci.* **21**: 429–438.

Yang, X. E., Chen, W. R., and Feng, Y. 2007. Improving human micronutrient nutrition through biofortification in the soil-plant system: China as a case study. *Environ. Geochem. Health.* **29**: 413–428.

Zayed, A., Pilon-Smits, E., Desouza, M., Lin, Z. Q., and Terry, N. 2000. Remediation of selenium polluted soils and waters by phytovolatalisation. In: Phytoremediation of Contaminated Soil and Water. Terry, N. and Bañuelos, G., Eds., pp. 1–12. Lewis Publishers, Boca Raton, FL.

Zayed, A. M., and Terry, N. 1992. Selenium volatilisation in broccoli as influenced by sulphate supply *J. Plant Physiol.* **140**: 646–652.

Zenk, M. H. 1996. Heavy metal detoxification in higher plants—A review. *Gene* **179**: 21–30.

Zhao, F. J., Lombi, E., Breedon, T., and McGrath, S. P. 2000. Zinc hyperaccumulation and cellular distribution in Arabidopsis halleri. *Plant Cell Environ.* **23**: 507–514.

Zhou, J. R., and Erdman, J. W. 1995. Phytic acid in health and disease. *Crit. Rev Food Sci. Nutr.* **35**: 495–508.

Zhu, D., Schwab, A. P., and Banks, M. K. 1999. Heavy metal leaching from mine tailings as affected by plants. *J. Environ. Qual.* **28**: 1727–1732.