Vegetable species for phytoextraction of boron, copper, lead, manganese and zinc from contaminated soil

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ABSTRACT: Phytoremediation is an attractive option to remove metal from contaminated soil since it is a simple, low-cost, and environmentally friendly procedure. To better examine the phytoremediation potential of kenaf (*Hybiscus canabinnus)*, mustard (*Brassica juncea*), turnip (*Raphunus sativus*) and amaranth (*Amaranthus crentus*) plants, a greenhouse experiment was performed in which these species were grown on a soil contaminated with Zn, Cu, Mn, Pb and B. The translocation, the bioconcentration and the removal index, the transference factor and the time to reach 50% of element removal from soil, among other indicators, were used in order to identify a hyperaccumulator. Kenaf plants were more tolerant to the conditions tested, with the highest dry matter production and no visual toxicity symptoms. Amaranth would be the species chosen to remediate the soil under field conditions as it presented the higher indexes for decontamination of Zn and Mn and was also able to remove B. Turnip showed the best results for Pb removal. All species tested were able to remove B from soil. In spite, none of the plant species tested could be characterized as a hyperaccumulator.

Key words: phytorremediation, tropical soil, heavy metal

Espécies vegetais na fitoextração de boro, cobre, chumbo, manganês e zinco de solo contaminado

RESUMO: A fitorremediação é uma opção atraente na remoção de solos contaminados com metais por ser uma técnica simples, de baixo custo e ambientalmente aceitável. O potencial de fitorremediação da kenaf (*Hybiscus canabinnus)*, mostarda (*Brassica juncea*), rabanete (*Raphunus sativus*) and amaranto (*Amaranthus crentus*) foram examinadas num experimento usando solo contaminado por Zn, Cu, Mn, Pb e B. O experimento foi conduzido em vasos, cultivando as quatro espécies até o florescimento. Os índices de translocação, bioconcentração e remoção, o fator de transferência e o tempo necessário para atingir a remoção de 50% do elemento do solo, entre outros indicadores, foram empregados na tentativa de identificar uma espécie hiperacumuladora. A kenaf foi a espécie mais tolerante nas condições empregadas, com a maior produção de matéria seca e ausência de sintomas visuais de toxidez. O amaranto foi a espécie mais indicada na remediação do solo testado em condições de campo por apresentar os melhores índices de descontaminação para Zn e Mn entre as espécies testadas, além de também apresentar valores satisfatórios para remoção do B. Todas as espécies testadas foram capazes de remover B do solo. Apesar destes resultados, não foi possível identificar uma espécie hiperacumuladora.

Palavras-chave: fitorremediação, solo tropical, metal pesado

Introduction

In the State of São Paulo, Brazil, 2,500 contaminated locations have been registered, at least 15% of the land-contaminated sites are due exclusively to heavy metal addition to the soils (CETESB, 2009). From the resulting 375 areas, the remediation of only 7% have been concluded, while 37% are under some kind of remediation process but the remaining 56% have not been treated yet (CETESB, 2009).

Metals such as Cu (copper), Pb (lead) and Zn (zinc) are important since high quantities of them can decrease crop production due to the risk of biomagnification and bioaccumulation in the food chain. There is also the risk of underground and surface water contamination (Schmidt, 2003; Nowack et al., 2006). Other trace elements, such as B (boron), can be extremely toxic to some plants at concentrations only slightly above optimum for others (Gupta, 1993). Although the requirement for B by plants is small, the concentration range between deficiency and toxicity is narrow. In Arid and semi-arid areas, B toxicity results from high levels of B in soils and from additions of B via irrigation water (Akar, 2007; Gemici and Tarcan, 2002; Ryan et al., 1998).

Due to the potential toxicity and high persistence of metals, soils polluted with these elements are an environmental problem that requires an effective and affordable solution (Nascimento and Xing, 2006). Phytoextraction was developed in the framework of an intense research effort for more efficient, cheaper and less hazardous techniques to remediate contaminated soils. It consists in the removal of metals by plants through uptake and accumulation into biomass (Nascimento and Xing, 2006). Interestingly, phytoremediation was recognized and documented by humans more than 300 years ago, however the scientific study and development of suitable plants was not conducted until the early 1980's (Lasat, 2000).

Hyperaccumulators are conventionally defined as species capable of accumulating metals at levels 100-fold greater than those typically measured in common nonaccumulator plants. Thus, a hyperaccumulator will concentrate more than 10 mg kg^{-1} Hg (mercury); 100 mg kg^{-1} Cd; 1,000 mg kg^{-1} Co (cobalt), Cr (cromium), Cu and Pb; and 10,000 mg kg^{-1} Ni and Zn. The capacity to hyperaccumulate metals is a relatively rare phenomenon in the plant kingdom, occurring in approximately 400 species of vascular plants total (Reeves and Baker, 1999). The vast majority of the hyperaccumulator species discovered so far are Ni hyperaccumulator (Nascimento and Xing, 2006). Plant species that can accumulate Cd, Pb, Zn, Co, As and Cu are much less numerous (McGrath et al., 2001). Recently, Babaoglu et al. (2004) reported that *Gypsophila sphaerocephala Fenzi ex Tchihat* var. *G. sphaerocephala* contained considerably higher B concentrations in its above-ground parts (2093 mg kg^{-1} seeds; 3345 mg kg^{-1}, leaves).

A large number of studies to identify hyperaccumulators have been conducted in temperate conditions. Among the few studies available in Brazil, many have been restricted to academics (Carneiro et al., 2001; Coscione and Berton, 2009; Gabos et al., 2009; Marques et al., 2000; Pereira et al., 2007; Romeiro et al., 2006; Santos et al., 2007; Zeittouni et al., 2007). In spite of that, promising species well known in Brazilian agriculture due to its robustness, good climate adaptation, deep root system and good biomass production have not been evaluated. In this study, kenaf, mustard, turnip and amaranth plants with phytoremediation potential were evaluated for the clean-up of a soil contaminated with B, Cu, Pb, Mn (manganese) and Zn.

Material and Methods

The experiments were conducted in a greenhouse, in Piracicaba, SP, Brazil from June, 2004 to September, 2004. The plant species used were mustard (*Brassica juncea,* cv. Florida Broad Leaf), turnip (*Raphanus sativus* L.*)*, amaranth (*Amaranthus crentus,* cv**.** BRS Alegria) and kenaf (*Hybiscus cannabinus*). Rhodic Hapludox (Soil Taxonomy, 1996) soil samples were collected from the 0-20 cm depth layer, at Paulínia, state of São Paulo, Brazil (22°45' S, 47°09' W), from a site contaminated with Zn, Cu, Mn, Pb and B. This area was contaminated more than ten years ago, during an accident with fertilizer's raw material. The soil sample was submitted to physical and chemical characterization (Table 1) after air drying and sieving through a 2 mm-mesh screen. Soil samples were placed in 3 dm^{-3} capacity pots. Plants were watered to 80% of water holding capacity on a daily basis by weighing the pots and adding water to compensate for any weight loss.

The experiment was carried out in a Completely Randomized Design, with three replicates. The treatments were constituted by the four plant species evaluated: Mustard (*Brassica juncea,* cv. Florida Broad Leaf), Turnip (*Raphanus sativus* L.*)*, Amaranth (*Amaranthus crentus,* cv. BRS Alegria) and Kenaf (*Hybiscus cannabinus*)**.** Seedlings were prepared under greenhouse conditions, grown in a mixture of contaminated soil and coconut fiber in the proportion 1:1 (soil:coconut fiber). The mixture were fertilized with N, P, K, Ca and S. Nitrogen was supplied by adding 21.5 mg of this nutrient as potassium nitrate (KNO_3) and 20.0 mg N as calcium nitrate ($CaNO_3$). Phosphate was supplied by adding 1,728 mg of P_2O_5 of this nutrient as triple superphosphate

Table 1 – Chemical and physical attributes of the Rhodic Hapludox soil.

Chemical												
Available Content[1]												
O.M.	P	pH	C.E.C	K	Ca	Mg	Base Saturation	B	Cu	Mn	Pb	Zn
g kg^{-1}	mg kg^{-1}		----------- mmol$_c$ kg^{-1} -----------				%	---------------------- mg kg^{-1} ----------------------				
48	209	5.4	120	1.7	60	16	65	3.8	156	117	18	490
			Soluble Content[2]					Total Content[3]				
			B	Cu	Mn	Pb	Zn	B	Cu	Mn	Pb	Zn
			---------------------------- mg kg^{-1} ----------------------------					---------------------- mg kg^{-1} ----------------------				
			3.8	1.67	127	0.18	204	6.4	354	482	104	747
Granulometric Analysis[4]												
		Clay	Silt	Fine Sand			Coarse sand	Total Sand		Soil Texture		
		-- g kg^{-1} --										
		320	250	260			170	430		Clay Loam		

[1]Available Content = DTPA / hot water extractable concentrations (Raij et al., 2001); [2]Soluble Content = $CaCl_2$ 0.05 mol L^{-1} (Lebourg et al., 1996); [3]Total Content = *Aqua Regia* (3:1, v/v, HCl to HNO_3); [4]Pipet method (Embrapa, 1997).

(ST). Potassium was supplied by adding 0.267 g of this nutrient as potassium sulphate (K_2SO_4) and 0.155 g as potassium nitrate (KNO_3) per 3 dm^{-3} of the mixture. After 15 days of germination, three seedlings were transferred to pots.

Only nitrogen and calcium were applied to the pots, weekly, at the rate of 30 mg N dm^{-3}, by alternation of ammonium nitrate and calcium nitrate. Shoots were harvested 40 days (turnip) or 60 days (mustard, amaranth, kenaf) after seeding corresponding to blossom of the species.

After harvest, plant shoots were first rinsed in tap water, then in 1% HCl solution and finally in distilled water. After excess water flowed off, each sample was put in paper bags and dried in a forced air oven at 70°C until constant weight, and then, weighed and ground in a *Wiley* type grinder. Roots were sieved to separate them from soil, rinsed in tap water, immersed in a 0.02 mmol L^{-1} disodium EDTA solution during 90 minutes, thoroughly rinsed in distilled water, bagged, dried at 70°C and ground as described for plant shoots. The rinsed EDTA solution was reserved in order to determine the content of elements adsorbed to roots (Yang et al., 1996). All ground vegetal samples were submitted to HNO_3/H_2O_2 (1:2 v/v) digestion in a microwave (United States Environmental Protection Agency - USEPA, 2009). After plant material collection, soil samples were also collected from pots and submitted to chemical analysis for B, Cu, Mn, Zn and Pb availability. Boron was determined by hot water extraction and the metals were determined by DTPA-TEA solution at pH 7.3 as described by Lindsay and Norvell (1978). This is the method used by some Brazilian laboratories, adopting the "IAC System of Soil Analysis" (Abreu et al., 1998).

The extraction of the total concentration of B and heavy metals from the soil samples was done with Aqua regia (ISO Standard 11466). The concentrations of B, Cu, Pb, Mn and Zn in the extracts were determined by inductively coupled plasma optical emission spectrometry (ICP-OES) (Varian Vista MPX). Memory effects on ICP readings were minimized by rinsing with water among samples.

Results were submitted to analysis of variance and comparison of treatment means by Tukey's test ($p < 0.05$). The B, Cu, Pb, Mn and Zn transport from soil to the shoots was evaluated using the transfer factor (F) = SC (mg kg^{-1}) + RC (mg kg^{-1}) / TC (mg kg^{-1}) where, SC = elements shoots concentration (Table 2); RC = elements root concentration (Table 2); TC = elements soil concentration obtained with Aqua Regia method (Table 1) (Lubben and Sauerbeck, 1991). The species ability in B, Cu, Pb, Mn and Zn translocation from roots to shoots was calculated by the translocation index (TI) suggested by Bichequer and Bohrlen cited by Paiva et al. (2002): TI (%) = SQ (mg per pot)/ WPQ (mg per pot) × 100, where, SQ = element accumulation in the shoots (Table 2); WPQ = element accumulation in the whole plant (shoots + roots) (Table 2). The plant metal accumulation capacity was calculated by the bioconcentration index (BI) suggested by Gosh and Singh (2005): TC (mg kg^{-1}) / SM (mg kg^{-1}) where, TC = elements soil concentration obtained with Aqua Regia method (Table 1); SM= soluble metals content obtained with CaCl2 0.05 mol L^{-1} (Table 2). The plant efficiency of element's removal was calculated using the removal factor (R), suggested by Lubben and Saerbeck (1991): R (%) = SQ (mg per pot) / RQ (mg per pot) × 100, where, SQ = element accumulation in the shoots (Table 2); RQ = metal quantity to be removed from soil (Table 2). For that, it was considered as target 50% of available elements content as measured by DTPA (Cu, Pb, Mn and Zn) or water (B), as shown in Table 1. It corresponded to, in mg per pot: B = 5.73; Zn = 735.6; Cu = 234.5; Mn = 175.5 and Pb = 27.9. These indicators allowed estimating also the period of time necessary to remove all soil contamination, considering four annual cultivation cycles. Soil decontamination is considered successful when the plants are able to extract up to 1 – 2% of the metal from soil (Lasat, 2000) or up to 1% of Zn and Cu, 0.6% of Pb total content from soil (Garbisu and Alkorta, 2001).

Results and Discussion

A comparison of the metal content of the studied soil with others reported in the literature confirmed that they should be considered as contaminated ones (Table 1). Amounts of Cu and Zn are close to or above of the maximum content commonly found in soils of São Paulo State and when it is considered the local environmental agency recommended limits (Cetesb, 2009). The reference limit for total metal content in São Paulo State soils are (in mg kg^{-1}) Cu - 35 and 60, Pb - 17 and 72, Zn - 60 and 300. No reference value is provided for B or Mn.

The DTPA method for quantifying heavy metals in soil samples (Lindsay and Norvell, 1978) can be helpful in monitoring soil contamination with heavy metals (Vidal-Vasquez et al., 2005) in addition to the fact that this method is also useful to evaluate micronutrient availability for agricultural purposes (Abreu et al., 2005). According to Abreu et al. (2005), Brazil's soils range for micronutrients is as follows (in mg dm^{-3}): B-0.01-10.6; Cu-0.1-56; Mn-1-325; Pb-0.00-63.9 and Zn-1-453, while the respective average values for São Paulo state were: B-0.32; Cu-2.5; Mn-16; Pb-0.85 and Zn-4.8. The higher values would be indicative of anthropogenic inputs, either due to excess application of fertilizers or to industrial or mining activities. Thus, the levels of B and heavy metals (Table 1) should be of substantial concern due to their high availability to plants and the potential for entering the food chain.

The visual symptoms of toxicity varied depending on the element and plant species tested. In general kenaf showed to be more tolerant to the presence of excess metals and B in the soil and did not show any sign of toxicity. For the other species, shoots presented generalized interveinal chlorosis, purple spots and browning, shriveling, decrease in development and in some cases necrosis and leaf death. All the symptoms described have

Table 2 – Elements content in soil and plant tissues.

Plant species	Dry matter(DW)	Zn	Cu	Mn	Pb	B
	g per pot	Shoots (SC), mg kg^{-1}				
Kenaf	32 a	264 c	3.44 b	70.0 b	8.90 b	92.6 a
Mustard	16 b	962 a	21.7 a	88.5 b	18.8 a	84.8 a
Turnip	15 b	573 b	12.3 ab	48.9 b	15.4 a	62.9 b
Amaranth	15 b	1,107 a	22.0 a	251 a	16.9 a	82.7 ab
		Roots (RC), mg kg^{-1}				
Kenaf	1.0 b	233 b	8.62 b	7.17 b	2.43 b	18.3 a
Mustard	2.5 b	343 ab	4.73 b	7.94 b	0.15 b	40.9 b
Turnip	5.5 a	205 b	28.2 a	37.0 a	15.5 a	20.7 c
Amaranth	1.1 b	558 a	11.1 b	43.8 a	5.2 b	21.6 c
		Shoots accumulate content (SQ), mg per pot				
Kenaf		8.53	0.11	2.27	0.29	3.01
Mustard		15.1	0.33	1.38	0.29	1.33
Turnip		8.36	0.18	0.71	0.22	0.93
Amaranth		16.5	0.35	3.92	0.26	1.24
		Roots accumulate content (RQ), mg per pot				
Kenaf		0.24	0.01	0.01	< 0.01	0.02
Mustard		0.87	0.01	0.02	< 0.01	0.10
Turnip		1.13	0.16	0.20	0.09	0.11
Amaranth		0.60	0.01	0.05	0.01	0.02
		Whole plant accumulate content (WPQ), mg per pot				
Kenaf		8.77	0.12	2.28	0.29	3.03
Mustard		15.9	0.35	1.40	0.29	1.43
Turnip		9.50	0.34	0.91	0.31	1.04
Amaranth		17.2	0.37	3.96	0.26	1.26
		SC/RC ratio				
Kenaf		1.1	0.4	9.8	3.7	5.3
Mustard		2.8	4.6	11.1	95	1.9
Turnip		2.8	0.4	1.3	1.0	2.6
Amaranth		2.0	2.0	5.7	3.2	4.2
		Root's adsorbed elements, mg kg^{-1}				
Kenaf		412 a	8.1 b	22 ab	8.2 a	17 a
Mustard		250 a	3.3 b	6.5 b	3.9 a	11 b
Turnip		293 a	45 a	37 ab	6.5 a	5 c
Amaranth		393 a	9.9 b	56 a	4.7 a	12 ab
		Soluble Metals Content, mg kg^{-1}				
Kenaf		191	1.37	108	0.12	3.82
Mustard		213	1.81	135	0.24	3.75
Turnip		205	1.50	118	0.15	3.71
Amaranth		206	1.61	146	0.21	3.83

*Means followed by different letters in columns differ (Tukey test, $p < 0.05$).

been reported in literature for eucalyptus cultivated in heavy metals contaminated soil (Accioly et al., 2009). However, among the species tested some have been reported as heavy metal hyperaccumulator such as the indian mustard *(Brassica juncea* L. Czern and Coss) (Schmidt, 2003). *Lotus corniculatus* L. (birdsfoot trefoil), *Festuca arundinacea* Schreb cv. Fawn (tall fescue) and *Hibiscus cannibinus* L. (kenaf) have been tested for B phytoextraction in soils containing from 1 to 10 mg kg^{-1} of B (water extracts) and were able to reduce up to 24% of B content in the soil (Banuelos et al., 1993) in sixty months. The mean shoot tissue concentrations of B ranged from a low of 96 mg B kg^{-1} DM in tall fescue to a high of 684 mg B kg^{-1} DM in leaves from kenaf. Also, *Amaranthus retroflexus* L. when tested in soils containing from 1 to 30 mg kg^{-1} of B presented severe toxicity effects, but reached up to 323 mg kg^{-1} of B in shoots (Aydin and Çakir, 2009).

In the present study, the shoots and roots dry matter yield, the B and heavy metals' concentration in the plants and the total content of the element extracted varied depending on the plant species (Table 2). Kenaf exhibited the highest shoots dry matter yield, while the other species did not differ. The concentration of elements in shoots, per element, varied as follows (mg kg^{-1}): from 264 (kenaf) to 1,107 (amaranth) for Zn; from 3.44 (kenaf) to 22 (amaranth) for Cu; from 49 (Turnip) to 251 (amaranth) for Mn; from 9 (kenaf) to 19 (Mustard) for Pb; and from 63 (Turnip) to 93 (kenaf) for B.

The Shoots/Roots (SC/RC) ratio was calculated in order to evaluate the translocation of the element inside the plant, from the roots to the shoots, and its potential accumulation in the biomass (Table 2). For most cases relatively more metal was accumulated in the shoots than in the roots, with the exception of Cu extracted by kenaf and turnip (SC/RC around 0.4) and Pb extracted by turnip (SC/RC = 1.0).

One of the selection criteria for hyperaccumulator's identification is the leaf's metal concentration. According to that a hyperaccumulator should concentrated more than 10,000 mg kg^{-1} of Zn or Mn, 1,000 mg kg^{-1} of Ni, Pb or Cu and more than 100 mg kg^{-1} of Cd (DW) (Brown et al., 1995). The mustard and amaranth exhibited the highest concentration of metals in shoots, while kenaf was the lowest for all the metals considered and the highest of boron (Table 2). However, according to the criteria already presented, none of the plants species tested would be classified as hyperaccumulator for Cu, Zn, Mn or Pb. No concentration target has been established for B hyperaccumulators, although plants with more than 800 mg kg^{-1} (DW) have been considered as promising (Marins and Oron, 2007; Robinson et al., 2007). Furthermore, all the plant species tested accumulated Zn and B in toxic levels, while mustard and amaranth also reached toxic levels for Cu (Table 2). Toxicity in plants is reported in literature to occur when Zn is found in the range of 100 - 400 mg kg^{-1}, for Cu from 20 - 100 mg kg^{-1}, for Mn from 300 - 500 mg kg^{-1}, for Pb from 30 - 300 mg kg^{-1} and for B from 50 - 200 mg kg^{-1} (Kabata-Pendias and Pendias, 2001; Pais and Jones Jr., 2000). In addition to this, when grown in soils with several levels of contamination the toxicity effects caused by Zn seemed to occur at lower concentrations, in the range of 50 - 270 mg kg^{-1} depending on the species (Marques et al., 2000).

The heavy metals and B content in shoots reported in literature are generally higher than the ones obtained in the present study. For B, concentrations in mustard and kenaf leaves up to 224 and 685 mg kg^{-1}, respectively, have been reported under field conditions (Bañuelos et al., 1993). One explanation may be the use of a pot experiment, which may have limited the plant development due to the small volume of soil to explore and the prolonged contact with contaminated soil. Also, the immobilization of elements by root adsorption may be of some importance. This mechanism avoids element's translocation from roots to shoots in some species working as a defense barrier, decreasing the phytoextraction potential of such species since only a small part of ions associated to the roots are effectively absorbed (Lasat, 2000).

Silva et al. (2007) used soybean and rice to evaluate the absorption and capacity of translocation of heavy metals in the same soil studied here and observed that the roots limited the translocation of copper and lead to the aerial parts of both cultures. Although the translocation of lead was also limited for rice, lead was found in soybean grains as zinc and manganese, which could reach the food chain. The immobilization of elements by root adsorption is not rare when plants are not hyperaccumulators and it is reflect by the plant efficiency of heavy metal transference from roots to shoots as indicated by the translocation index (TI) and in SC/RC ratio. In addition it is also possible to verify high ion retention in the roots and a limited phytoextraction by some plants (Table 2).

Considering Zn, Mn and B, the higher TI were obtained, in decreasing order, for kenaf, amaranth, mustard and turnip (Table 3). For Pb, the highest TI was obtained for mustard and kenaf. In spite of the high TI observed for B with all the species tested, the transference factor (F), which reflects the plant's capacity to transport metals from soil to shoots, was much higher for B than for Zn, Cu, Mn or Pb. Such behavior can be attributed to high B mobility in soil. Furthermore, the lower the TI the fewer symptoms were observed for the plants tested. The exception to that was kenaf, with the highest TI for B and no visual symptoms.

The bioconcentration index (BI) is also considered effective in the evaluation of the plant specie's element accumulation potential and its ability to absorb selectively elements form the soil solution (Zhu et al., 1999). The highest BI values for Zn, Cu and Mn were obtained with amaranth while Kenaf showed the highest values for Pb and B (Table 3). Usually, plants with high BI values are the best choices for phytoextraction (Ghosh and Singh, 2005).

The target value for B, Cu, Mn and Pb to be removed from soil, in order to evaluate its phytoextraction potential, was set as 50% of the available element content in soil measured in hot water extracts for B and by the DTPA method for metals. Further information of those metals availability was provided by Coscione et al. (2009) confirming that Cu, and Pb were found in the forms highly available to plants, among other metals

Table 3 – Indexes used to evaluate the species phytoremediation potential.

Species	Translocation Index (TI)				
	Zn	Cu	Mn	Pb	B
Kenaf	97	93	100	99	99
Mustard	94	96	99	100	93
Turnip	88	54	78	73	88
Amaranth	96	96	99	98	98
Bioconcentration Index (BI)					
Kenaf	1.4	2.5	0.6	139	25
Mustard	4.5	12.5	0.6	79	23
Turnip	2.8	8.3	0.4	115	17
Amaranth	5.4	14	1.7	88	22
Transfer Factor (F)					
Kenaf	0.7	0.03	0.2	0.1	17.3
Mustard	1.8	0.07	0.2	0.2	19.6
Turnip	1.0	0.11	0.2	0.3	13.1
Amaranth	2.2	0.09	0.6	0.2	16.3
Removal Factor (R)					
Kenaf	1.2	0.05	1.3	1.0	52.8
Mustard	2.2	0.15	0.8	1.0	25.0
Turnip	1.3	0.14	0.5	1.1	18.2
Amaranth	2.3	0.16	2.3	0.9	22.0
Years to target value[1]					
Kenaf	21	491	19	24	< 1
Mustard	12	171	32	24	< 1
Turnip	19	176	49	23	< 1
Amaranth	11	190	12	28	< 1

[1] Four cycles per year.

found in the soil. When compared to total metal amounts found in the soil samples up to 33.5% of Cu and 60.5% of Pb were found in soluble + exchangeable and organic matter fractions of the soil studied. Considering the target values set and four cycles per year, it was possible to estimate the years needed to clean up the site (Table 3).The best results for metals were reached with amaranth for the clean up of Zn and Mn (11 and 12 years). According to the literature criteria described above, the decontamination is reflected by means of the removal index (R) and all the species would be successful in clean-up of Zn and Pb, with only kenaf and amaranth for Mn. No plant species would be considered successful for the decontamination of Cu but according to both the criteria discussed all the species could be used for B clean up. The large time range observed in Table 3 (11 to 491 years) emphasizes the importance of the adequate plant selection for the clean-up of each element. Besides of the plant species and the element to be removed, soil properties, such as its pH and organic matter content, can also affect the indexes used for phytoremediation evaluation (Lübben and Sauerbeck, 1991).

Conclusions

Amaranth would be the species chosen to remediate the soil used under field conditions as it presented the higher decontamination indexes for Zn and Mn and was also able to remove B. The turnip presented the best results for Pb removal. All species tested were able to remove B from soil.

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