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Accumulation and translocation of heavy metals in soil and plants from fly ash contaminated area

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Abstract: *The present investigation deals with the accumulation of heavy metals in fields contaminated with fly ash from a thermal power plant and subsequent uptake in different parts of naturally grown plants. Results revealed that in the contaminated site, the mean level of all the metals (Cd, Zn, Cr, Pb, Cu, Ni, Mn and Fe) in soil and different parts (root and shoots) of plant species were found to be significantly ($p<0.01$) higher than the uncontaminated site. The enrichment factor (EF) of these metals in contaminated soil was found to be in the sequence of Cd (2.33) > Fe (1.88) > Ni (1.58) > Pb (1.42) > Zn (1.31) > Mn (1.27) > Cr (1.11) > Cu (1.10). Whereas, enrichment factor of metals in root and shoot parts, were found to be in the order of Cd (7.56) > Fe (4.75) > Zn (2.79) > Ni (2.22) > Cu (1.69) > Mn (1.53) > Pb (1.31) > Cr (1.02) and Cd (6.06) ≃ Fe (6.06) > Zn (2.65) > Ni (2.57) > Mn (2.19) > Cu (1.58) > Pb (1.37) > Cr (1.01) respectively. In contaminated site, translocation factor (TF) of metals from root to shoot was found to be in the order of Mn (1.38) > Fe (1.27) > Pb (1.03) > Ni (0.94) > Zn (0.85) > Cd (0.82) > Cr (0.73) and that of the metals Cd with Cr, Cu, Mn, Fe; Cr with Pb, Mn, Fe and Pb with Fe were found to be significantly correlated. The present findings provide us a clue for the selection of plant species, which show natural resistance against toxic metals and are efficient metal accumulators.*

Key words: *Accumulation, Fly ash, Heavy metals, Accumulator species, Enrichment factor, Translocation factor*
PDF of full length paper is available online

Introduction

Production of electricity in India is mainly dependent on coal fired thermal power plants. Fly ash, a by-product of coal fired thermal power industries, amounts to about 35-40% of the coal used by the thermal power plant. In other words, generation of one MW of electricity from coal requires about one acre of land for disposal of fly ash (Sahu *et al.,* 1994). In India Fly ash production was about 112 million tones during 2005-06 and it is expected to be about 150-170 million tones per annum by the year end of 2012 (MOEF, 2007; Pandey *et al.,* 2009). However, such large scale production of fly ash by thermal power industries would pose a formidable environmental challenge regarding its disposal and overall impact on environment.

Besides many essential macronutrients (P, K, Ca and S) and micronutrients (Fe, Mn, Ni, Cu, Co, B, and Mo), fly ash also contains a number of toxic heavy metals such as Cd, Pb and Se (Rautaray *et al.,* 2003; Adriano *et al.,* 1980). Sometimes, the concentration of trace metals in fly ash exceeds the levels of these metals found in normal soil (Kalra *et al.,* 1996). Various studies concerning the impact of fly ash on soil or plant productivity have been mostly carried out under laboratory conditions (Garg *et al.,* 1996; Kalra *et al.,*1997; Mishra and Shukla,1986; Sikka and Kansal, 1994; Sinha and Gupta, 2005).

There is an inherent tendency of plants to take up toxic substances including the heavy metals, that are subsequently transferred along the food chain. Use of polluted land or water for cultivation of crops mainly accounts for decrease in the overall productivity and results in contaminated food grains and vegetables which adversely affects human health. The main advantage associated with study of plants including crops, is their ability to accumulate metals, if grown on metal polluted land or irrigated with polluted water. Thus, plants serve as a good tool for phytoremediation. However, determination of the nature of toxicity, distribution of toxicants and level of accumulation in different plant parts would be essential before selection and cultivation of plants for phytoremediation (Barman and Lal, 1994; Barman and Bhargava, 1997; Barman and Ray, 1999; Barman *et al.,* 1999, 2000, 2001). Several studies have shown that plants can automatically acquire characteristic resistance against toxicants including heavy metals, depending upon the various eco-physiological factors in time and space (Gregory and Bradsaw, 1965; Antonovics *et al.,* 1967; Porter and Peterson, 1977; Ray *et al.,* 1988). However, all plants are not equally resistant to all types of pollutants in the environment. It appears that the plant resistance against a particular toxicant is also dependent on the cyto-genetic makeup of the particular species.

The present investigation relates to the study of levels of metal accumulation in different parts of eleven plant species, which are growing naturally in fly ash contaminated soil. This study is expected to provide us clues for selection of accumulator/resistant plant species towards metals found in the fly ash, contaminated site.

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Materials and Methods

Study area and sample collection: The study area selected for the present study falls in the vicinity of a coal based thermal power plant, situated at an elevation of 271 m above mean sea level in the Sonebhadra district of South- Eastern Uttar Pradesh, and dumping its fly ash in an ash-pond as slurry since 1965. The thermal power plant lies between latitude 24°10' 24°12' N and longitude 82°46' 82°48' E and is located in a semi industrialised area near Northern Coal Field Limited (NCL) coalmines on Varanasi-Singrauli Highway. The area of ash-pond is nearly about 79.67 ha almost over flooded with slurry.

Eleven plant species were collected from the vicinity of the ash-pond during March 2004 and their scientific and common/local name (within bracket) are *Datura stramonium* (Datura), *Typha spp* (weed), *Triticum aestivum* (Wheat*), Solanum xanthocarpum (*Katai, Kateli, Ringani), *Dolichos lablab* (Bean), *Lycopersicum esculentum* (Tomato), *Parthenium hysterophorus* (Congress grass), *Ricinus communi* (Castor oil plant), *Croton bonplandianum (*Ban Tulsi, Kala Bhangra)*, Solanum nigrum*(Kali Makoy) and *Brassica campestries* (Yellow sorson, Mustard). Out of the eleven plant species, seven were naturally growing weeds and rest of them were a cereal (wheat), vegetables (bean and tomato) and an oil seed (mustard). Similar plant species growing on an unpolluted site (about 2 km away from the ash-pond) served as control. All the plant samples were uprooted at maturity and separated into root and shoot parts for estimation of metal content.

Ten to fifteen plants of each species were collected randomly from fly ash contaminated soil as well as from the control site. Ten soil samples were drawn from contaminated and control site with an average depth of 20 cm. During plant sampling, it was ensured that different plant samples of each species had the same physiological age, identical size and appearance. The plant samples were first washed with running tap water followed by distilled water to remove extraneous matter. After washing, the plant material was oven dried at 65°C for 24 hr and chopped. The soil samples taken from both contaminated as well as control sites were air dried and sieved before analysis individually.

Metal analysis: One gram of each plant and soil sample (each in triplicate) was digested overnight with a mixture of HNO_3: $HClO_4$ (4: 1, v/v). Samples were slowly digested on the hot plate until a clear solution was obtained (Barman *et al*., 2000; Kisku *et al*., 2000). It was then filtered and assayed by AAS (Varian Spectra AA-250 Plus) for Cd, Zn, Cr, Pb, Cu, Ni, Mn and Fe. The AAS value of blank (without sample) of each metal was deducted from the sample value for final calculations.

Enrichment factor (EF) and Translocation factor (TF): The enrichment factor (EF) has been calculated to derive the degree of soil contamination and heavy metal accumulation in soil and in plants growing on contaminated site with respect to soil and plants growing on uncontaminated soil (Kisku *et al.,* 2000).

$$EF = \frac{\text{Concentration of metals in soil or plant parts at contaminated site}}{\text{Concentration of metals in soil or plant parts at uncontaminated site}}$$

Translocation factor (TF) or mobilisation ratio (Barman *et al.,* 2000; Gupta *et al.,* 2008) was calculated to determine relative translocation of metals from soil to other parts (root and shoot) of the plant species.

$$TF = \frac{\text{Concentration of metal in plant tissue (parts)}}{\text{Concentration of metal in corresponding soil or root}}$$

Statistical analysis: Concentrations (μg g^{-1}) of eight metals in three parts of 11 species of two sites were assessed together with main effect four factor analysis of variance (ANOVA) and groups mean within factors were compared by Newman Keuls post hoc test (Zar, 1974). A two-tailed probability value less than 0.05 was considered to be statistically significant. Analysis was performed on STATISTICA (Version 7) software.

Results and Discussion

Accumulation of metals: The average metal content of contaminated soil was found in the order of Fe (816.41) > Zn (117.61) > Mn (59.14) > Cu (30.14)> Pb (26.48) > Ni (8.96)> Cr(6.41) > Cd (2.98) μg g^{-1} d.w. whereas, in case of control soil it was found almost in the same sequence of Fe (435.23) > Zn (89.65) > Mn (46.75) > Cu(27.41) > Pb(18.64) > Cr(5.78) > Ni(5.68)> Cd (1.28) μg g^{-1} d.w. (Table 1) but in contaminated soil the mean metals levels were significantly ($p<0.01$) higher than the uncontaminated site.

Among the eleven plant species, the mean concentration of each metal in the vegetative part above the ground (shoot) was found in the order of Fe (499.10) > Mn (69.05) > Zn (67.10) > Cu (26.44) > Pb (15.76) > Ni (8.12) > Cr (2.05) > Cd (1.22) μg g^{-1} d.w. whereas in uncontaminated site the respective metal levels were found in the order of Fe (82.36)>Mn (31.54)> Zn (25.30) > Cu (16.72) > Pb (11.51) > Ni (3.16) > Cr (2.04) > Cd (0.20) μg g^{-1} d.w.(Table 1). The maximum and minimum concentration (μg g^{-1} d.w.) of metals in the shoot part of plant grown in contaminated site was found as 2.92 in *Datura stramonium* and 0.05 in *Typhus* spp. for Cd, 160.33 in *Datura stramonium* and 31. 0 in *Brassica campestries* for Zn, 4.87 in *Parthenium hysterophorus,* and 0.14 in *Triticum aestivum* for Cr, 39.08 in *Parthenium hysterophorus*, and 3.15 in *Typha* spp. for Pb, 53.58 in *Croton bonplandianum* and 3.15 in *Brassica campestries* for Cu, 20.17 in *Datura stramonium* and 1.58 in *Triticum aestivum* for Ni, 139.0 in *Typha* spp. and 11.38 in *Triticum aestivum* for Mn and 925.0 in *Parthenium hysterophorus* and 129.57 in *Typha* spp for Fe respectively. The maximum and minimum values of each metal were found to be comparatively higher in contaminated soil than in uncontaminated soil (Fig. 1).

In the root part, the mean concentration of metals (μg g^{-1} d.w.) in plants species grown on contaminated site were found in the order of Fe (393.70) > Zn (78.94) > Mn (50.03) > Cu (30.95) > Pb (15.37) > Ni (8.60) > Cr (2.83) > Cd (1.48) μg g^{-1} d.w. whereas, in uncontaminated site it was in the order of Fe (82.97) > Mn (32.59) > Zn (28.26) > Cu (18.26) > Pb (11.75) > Ni (3.87) > Cr (2.77) > Cd (0.20). The maximum and minimum concentration (μg g^{-1} d.w.) of metals in the root part of plant grown in contaminated soil was found as 2.94 in *Ricinus communis and* 0.25 in *Typha* spp for Cd, 165.92 in *Datura stramonium* and 37.58 *in Brassica campestries* for Zn, 5.61 in *Parthenium hysterophorus and 1.1 in Typha* spp for Cr, 36.87 in *Parthenium hysterophorus* and 4.67 in *Brassica campestries* for Pb, 47.33 in *Solanum xanthocarpum* and 12.94 in *Ricinus communis* for Cu, 14.7 in *Solanum nigrum* and 1.79 in *Brassica campestries* for Ni, 86.94 in *Ricinus communis* and 3.08 in *Brassica campestries* for Mn, and 712.44 in *Ricinus communis* and 141.69 in *Brassica campestries for Fe* respectively (Fig. 1).

Among the eleven plant species, trace metals in edible part was estimated only in *Triticum aestivum* and *Brassica campestris.* The concentration (μg g^{-1} d.w.) of metals in contaminated site was found to be in the order of Fe (38.58) > Zn (80.23)> Mn> (33.5) > Cu (15.92) >Pb (7.5) > Cr (1.04) > Ni (0.92) > Cd (0.75) for *Triticum aestivum* and Fe (143.43) > Zn (80.5)> Mn (44.5)> Cu (9.98) > Ni (3.02) > Pb (2.58)> Cr(1.25)> Cd (0.28) for *Brassica campestris.* The concentration of these metals were higher than the control site except for Cu and Ni for *Triticum aestivum* and Zn Cr and Pb for *Brassica campestries.* The variation and heterogeneous accumulation in the edible part of these two species may be due to genetic difference.

The concentrations of eight metals in three parts (soil, root and shoot) of 11 species at two sites were analysed statistically and summarised in Fig. 2. ANOVA revealed that the concentrations of metals were similar among plants (F=0.47, p>0.05) while it differed significantly (p<0.01) between metals (F=120.70, p<0.01), locations (F=18.80, p<0.01) and sites (F=42.85, p<0.01). Comparing mean, the concentration of Fe was found to be significantly (p<0.01) different and higher than the other metals. The concentration of Zn was also found to be significantly (p<0.01) higher than Cd, Cr, Pb, Cu and Ni while the concentrations of Cd, Cr, Pb, Cu, Ni, and Mn did not differ significantly (p > 0.05). Similarly, the metal concentrations in soil were significantly (p<0.01) higher than the roots and the shoots and the concentrations in shoots were also found to be significantly (p<0.01) higher than in the roots. The metal concentrations in contaminated site was found to be significantly (p<0.01) higher than the uncontaminated site for all parts (soil, root and shoot) and showed heterogeneous accumulation or did not follow any specific pattern.

Enrichment factor (EF) and Translocation factor (TF): The enrichment factor (EF) in contaminated soil, root and shoot parts of the plant species were found in the following sequence.

Soil: Cd (2.33) >Fe (1.88) >Ni (1.58)>Pb(1.42)> Zn (1.31)>Mn(1.27)>Cr(1.11)>Cu(1.10)

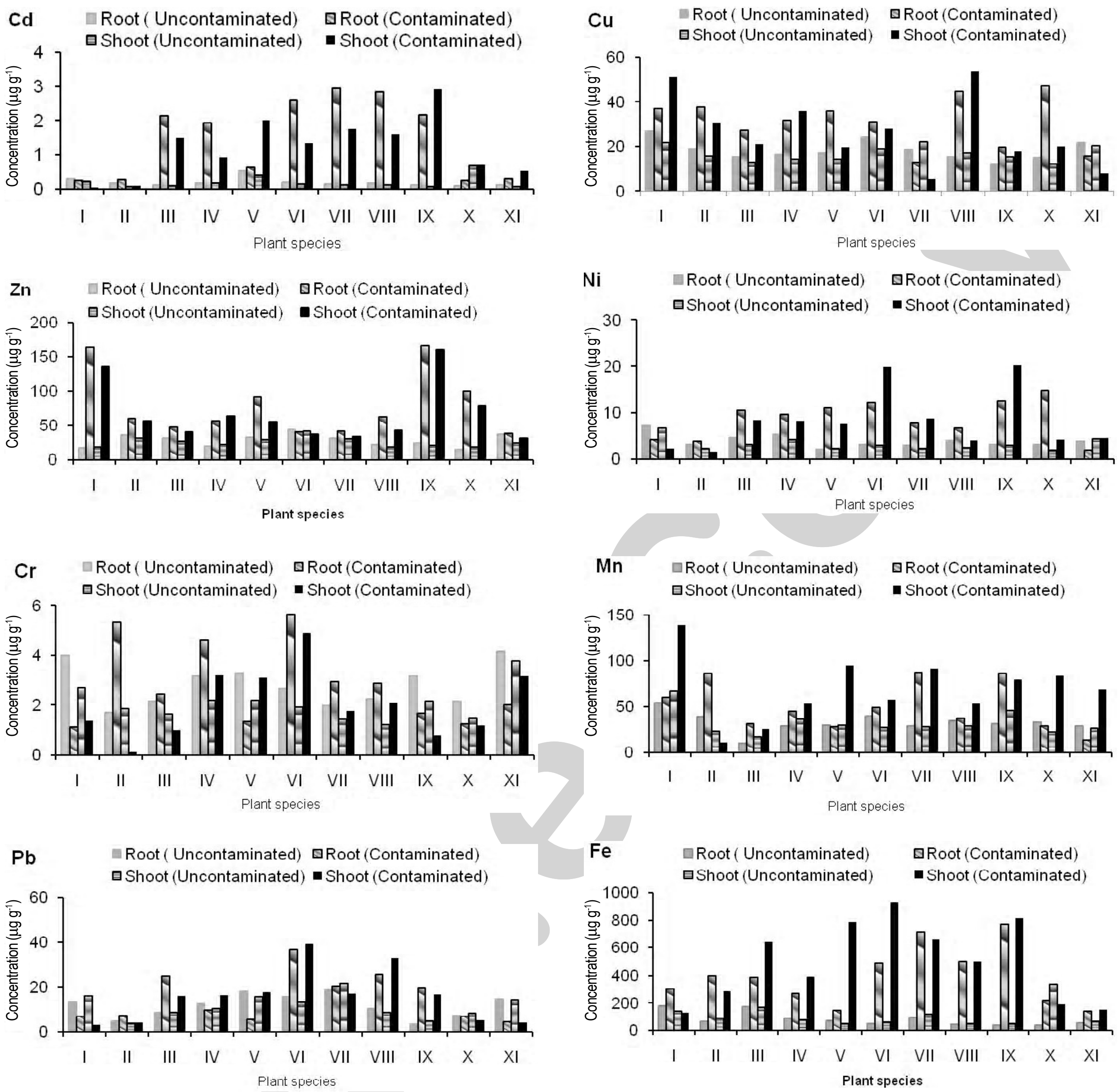
Root: Cd (7.56) >Fe (4.75) >Zn(2.79)>Ni(2.22)>Cu (1.69)>Mn(1.53)> Pb(1.31) >Cr(1.02)

Shoot: Cd (6.06) ≃ Fe (6.06)>Zn(2.65)>Ni(2.57)> Mn(2.19)> Cu(1.58) >Pb(1.37)>Cr(1.01)

All the values of enrichment factor was greater than one which indicates higher availability and distribution of metals in soil contaminated with fly ash and thereby increasing the metal accumulation in plants species grown on the contaminated soil (Kisku *et al.,* 2000; Gupta *et al.,* 2008). Among the eight metals estimated, the maximum enrichment was found in case of Cd followed by Fe for soil, root and shoot part but in overall, the sequence did not follow any specific pattern. In case of individual species, the EF in shoot parts of the plant species grown in contaminated soil showed different range values Cd : 36.60-0.22, Zn : 7.85-0.91, Cr : 2.54-0.08, Pb : 3.7-0.19, Cu : 3.16-0.25, Ni : 6.98-0.33, Mn : 3.85- 0.50, Fe : 16.14-0.93. Among the plant species, *Datura stramonium* showed exceptionally higher enrichment factor (EF) for Cd, Zn, Pb, Ni and Fe. Some other species showed comparative higher enrichment of metals such as in case of like *Solanum xanthocarpum for Cd* and *Parthenium hysterophorus* for Cd, Ni and Fe. (Fig. 3).

The enrichment factor in the edible part is an important criterion for the selection of suitable crop species which can be selected for cultivation in a field having higher level of metal contamination or receiving industrial effluent (Barman and Bhargava, 1997). In the present study, the enrichment of metals were found in the order of Fe (3.40) ≃ Mn (3.40) > Cd (3.07) > Zn (1.39) ≳ (1.39) > Pb (0.85) > Cu (0.43) for *Triticum aestivum* and Cr (2.60) > Zn (2.49) > Pb (0.82) > Ni (0.47) > Cu (0.35) > Mn (0.30) ≃ Fe (0.30) > Cd (0.21) for *Brassica campestries*. The enrichment values indicate ability to higher accumulation of metals like Fe, Mn, Cd and Zn for *Triticum aestivum* and Cr and Zn for *Brassica campestries* from fly ash contaminated soil.

Translocation factor (TF): The translocation factor (TF) / mobilisation ratio of metals from soil to root (TF_S) and root to shoot (RF_R) have been estimated (Table 1). The average translocation of metals from soil to root was found to be in the order of Cu (1.03) > Ni (0.96) > Mn (0.85) > Zn (0.67) >Pb (0.58) > Cd (0.50) > Fe (0.48) and when this value was compared with control value it was observed to be higher in the contaminated site for Cd, Zn, Cu, Ni, Mn and Fe. In case of shoot (root to shoot) TF_R was found in the order of Mn (1.38) > Fe (1.27) > Pb (1.03) > Ni (0.94) > Zn (0.85) > Cd (0.82) > Cr (0.73) and among the metals Mn, Fe, Ni and Pb TF_R was found to be higher than the control value (Table 1). Comparatively the translocation values from soil to root and root to shoot showed lower values than the enrichment factors and did not follow the similar pattern indicating that distribution of metals in contaminated soil is quite high and their translocation from soil to root and root to shoot is somehow restricted. Regarding translocation of individual metals from root to shoot it has been found that among metals Cd with Cr (r = 0.67, p <0.05), Cu (r = -0.65, p <0.05), Mn



Plant species: I = *Typha* sp., II = *Triticum aestivum,* III = *Solanum xanthocarpum,* IV = *Dolichos lablab,* V = *Lycopersicum esculentum,* VI = *Parthenium hysterophorus,* VII = *Ricinus communis,* VIII = *Croton bonplandianum,* IX = *Datura stramonium,* X = *Solanum nigram* and XI = *Brassica compestries*

Fig. 1: Accumulation of metals in different parts of the plant species grown in contaminated and uncontaminated soil

(r = 0.66, p <0.05) and Fe (r = 0.60, p <0.05); Zn with Cr (r = -0.60, p <0.05), Cr with Pb (r = 0.67, p <0.05), Mn (r =0.82, p <0.01) and Fe (r = 0.68, p <0.05) and Pb with Fe (r = 0.92, p <0.01) correlated significantly with each other in contaminated site, whereas in uncontaminated site only Cd is found to be correlated significantly with Zn (r =0.60, p <0.05) (Table 2).

The translocation factor (Fig. 4) revealed that some species showed higher biomagnification of metals in contaminated soil whereas the same species showed less accumulation of a particular metal in uncontaminated soil such as the species *Lycopersicom esculentum* showed >1 translocation value only in case of Ni and Cu, and this specific value is lesser when the species is grown in fly ash contaminated soil but the same species (*Lycopersicom esculentum)* showed accumulation of maximum number of metals (Cd, Cr, Pb, Mn, and Fe) with >1 TF_S when grown in contaminated soil. Other species are *Triticum aestivum* which showed lesser translocation for Cd, Cr Pb, Ni in contaminated soil than the control soil. *Brassica campestris,* showed higher level of accumulation of Cd, Zn, Cr, Ni and Mn in contaminated soil than uncontaminated

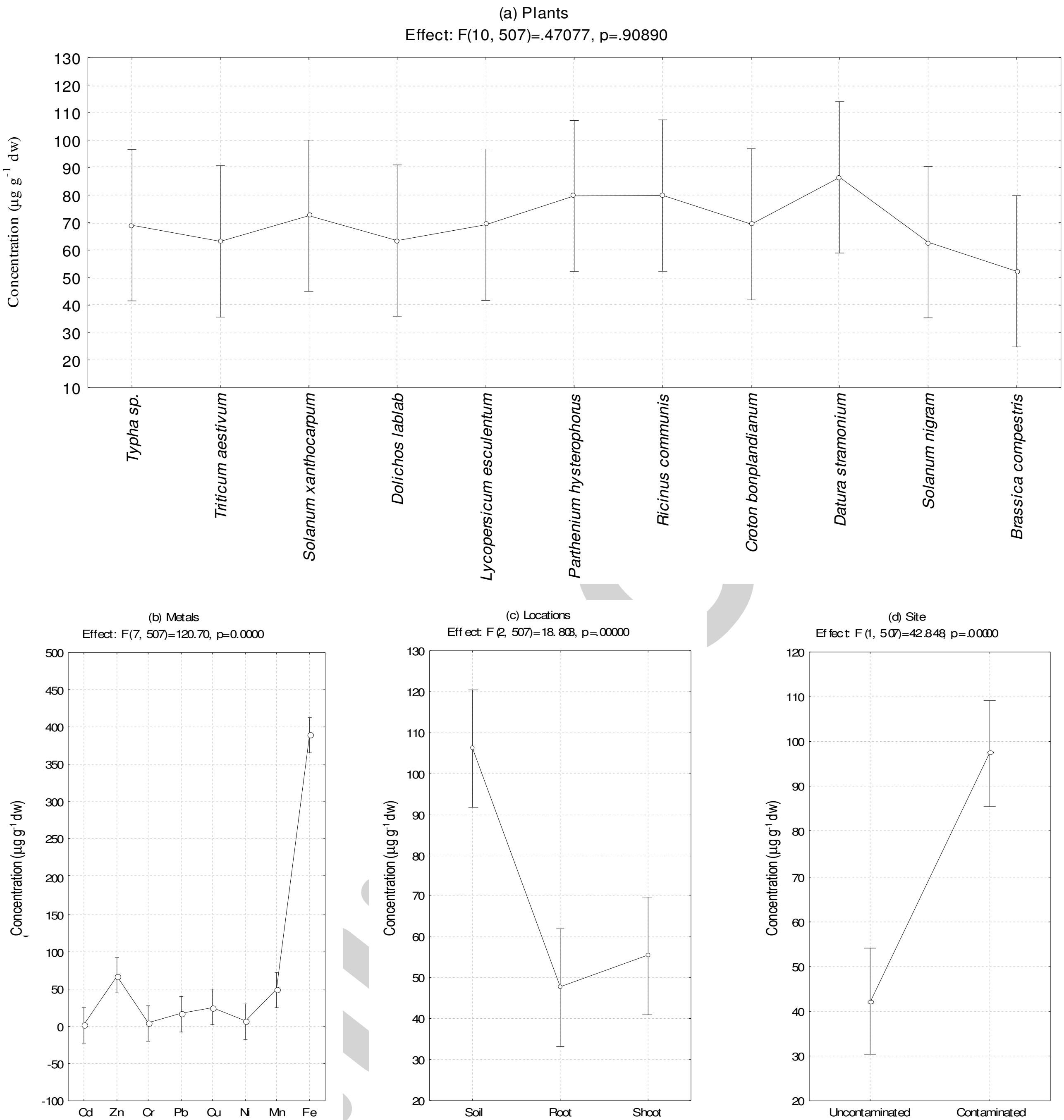


Fig. 2: Effect of plants (a), metals (b), locations and sites (c) on concentration (µg g-1 dw) with 95% confidence intervals (vertical line)

soil. Comparative results revealed that some metals were also less translocated in the contaminated soil than in the control soil (Fig. 4), which indicated that cytogenetic make-up as well as other unknown factors are responsible for different patterns of translocation of metals to the upper part of the plant.

Furthermore, establishing a pattern of translocation of metals from root to other parts of a plant species can be very useful in biological monitoring of heavy metal contamination as well as selection of metal accumulator or tolerant species. The metal translocation process in plant species is a crucial factor in determining the metal distribution in different plant tissues (Xiong 1998). A number of factors including anatomical, biochemical and physiological factors (Salt *et al.,* 1995) contribute to heavy metal accumulation and distribution in the upper vegetative parts.

Metals are mobilised and taken by root cells from soil, bound by cell wall and then transported across the plasma membrane,

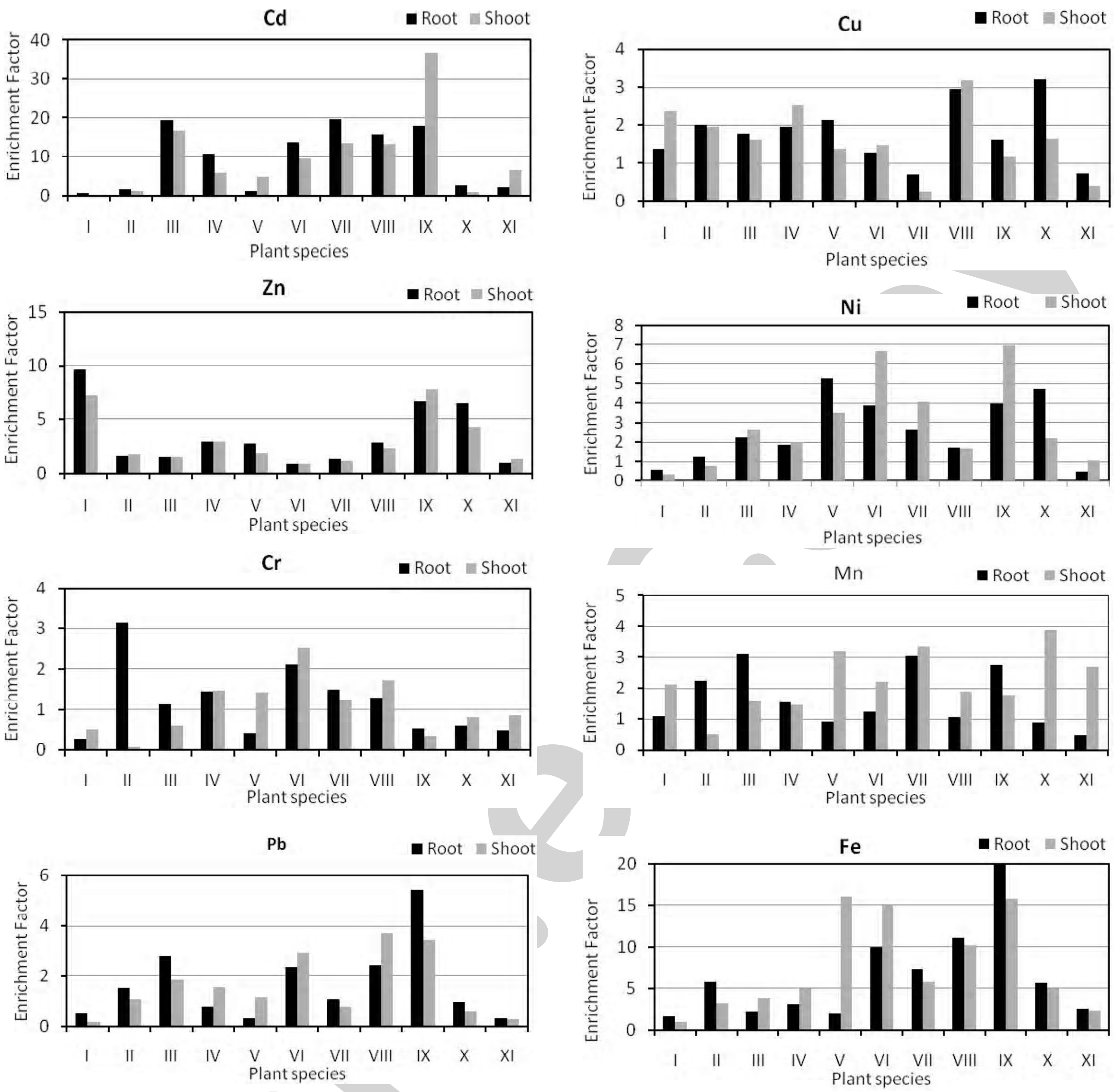
Table - 1: Transfer Factor (TF) or mobilisation of different metals from soil to root, root to shoot in contaminated and uncontaminated site

Metals	Location	Contaminated soil			Uncontaminated soil			Enrichment factor (EF)
		Metal level ($\mu g\ g^{-1}$)	Translocation factor (TF)		Metal level ($\mu g\ g^{-1}$)	Translocation factor (TF)		
			Soil to root	Root to shoot		Soil to root	Root to shoot	
Cd	Shoot	1.22 ± 0.86	0.50	0.82	0.20 ± 0.19	0.15	1.02	6.06
	Root	1.48 ± 1.14			0.20 ± 0.12			7.56
	Soil	2.98 ± 1.17			1.28 ± 0.28			2.33
Zn	Shoot	67.10 ± 42.91	0.67	0.85	25.30 ± 7.62	0.32	0.90	2.65
	Root	78.94 ± 46.92			28.26 ± 9.23			2.79
	Soil	117.26 ± 10.7			89.65 ± 22.35			1.31
Cr	Shoot	2.05 ± 1.39	0.44	0.73	2.04 ± 0.71	0.48	0.73	1.01
	Root	2.83 ± 1.65			2.77 ± 0.83			1.02
	Soil	6.41 ± 1.50			5.78 ± 1.74			1.11
Pb	Shoot	15.76 ± 11.80	0.58	1.03	11.51 ± 5.29	0.63	0.98	1.37
	Root	15.37 ± 10.71			11.75 ± 5.15			1.31
	Soil	26.48 ± 4.64			18.64 ± 4.84			1.42
Cu	Shoot	26.44 ± 15.63	1.03	0.85	16.72 ± 3.82	0.67	0.92	1.58
	Root	30.95 ±11.26			18.26 ± 4.42			1.69
	Soil	30.14 ± 7.99			27.41 ± 3.47			1.10
Ni	Shoot	8.12 ± 6.40	0.96	0.94	3.16 ± 1.472	0.68	0.82	2.57
	Root	8.60 + 4.11			3.87 ± 1.42			2.22
	Soil	8.96 ± 2.56			5.68 ± 2.13			1.58
Mn	Shoot	69.05 ± 34.91	0.85	1.38	31.54 ± 13.88	0.70	0.97	2.19
	Root	50.03 ± 26.39			32.59 ± 10.67			1.53
	Soil	59.14 ± 10.92			46.75 ± 12.72			1.27
Fe	Shoot	499.10 ± 287.29	0.48	1.27	82.36 ± 42.34	0.19	0.99	6.06
	Root	393.70 ± 210.07			82.97 ± 51.19			4.75
	Soil	816.41 ± 128.50			435.23 ± 91.92			1.88

Table - 2: Correlation (N=11) of metals mobilization (from root to shoot) in control and contaminated site

Metals	Cd	Zn	Cr	Pb	Cu	Ni	Mn	Fe
				Uncontaminated site (control)				
Cd	1.00							
Zn	0.60*	1.00						
Cr	-0.16	-0.40	1.00					
Pb	0.30	0.17	-0.24	1.00				
Cu	-0.24	-0.35	-0.27	0.45	1.00			
Ni	-0.42	-0.39	0.07	0.06	-0.10	1.00		
Mn	-0.31	-0.07	-0.31	0.37	0.23	0.11	1.00	
Fe	-0.15	-0.40	0.38	0.13	0.50	-0.15	-0.26	1.00
				Contaminated site				
Cd	1.00							
Zn	-0.53	1.00						
Cr	0.67*	-0.60*	1.00					
Pb	0.55	-0.38	0.67*	1.00				
Cu	-0.65*	0.33	-0.21	-0.15	1.00			
Ni	0.03	0.17	0.18	-0.07	-0.22	1.00		
Mn	0.66*	-0.46	0.82**	0.28	-0.36	0.41	1.00	
Fe	0.60*	-0.46	0.68*	0.92**	-0.29	-0.04	0.27	1.00

* $p<0.05$; ** $p<0.01$



Plant species: I = *Typha* sp., II = *Triticum aestivum,* III = *Solanum xanthocarpum,* IV = *Dolichos lablab,* V = *Lycopersicum esculentum,* VI = *Parthenium hysterophorus,* VII = *Ricinus communis,* VIII = *Croton bonplandianum,* IX = *Datura stramonium,* X = *Solanum nigrum* and XI = *Brassica campestries*

Fig. 3: Enrichment of metals in root and shoot parts of the different plant species grown in contaminated soil

driven by ATP-depended proton pumps that catalyzes H^+ extrusion across the membrane. Along with cationic nutrients, plant transporters are also involved in shuttling potentially toxic cations across plant membranes (Maser *et al.,* 2001; Singh *et al.,* 2003). The tolerance of plants to increasing levels of toxic elements can result from the exclusion of toxic elements or their metabolic tolerance to specific elements. The major mechanism in tolerant species of plants appears to be compartmentalization of metal ions, *i.e.* sequestration in the vacuolar compartment, which excludes them from cellular sites where processes such as cell division and respiration occur, thus proving to be as effective protective mechanism (Chaney *et al.,* 1997; Hall 2002; Lee *et al.,* 1977). Higher Ni content in roots of a metal accumulator plant species is mainly dependent on at least two factors namely: sequestration and/or translocation. Ni could be transported as a nickel-citrate complex (Lee *et al* ., 1977) or as a nickel-peptide complex or as a nickel-histidine complex (Krämer *et al.,* 1996) to ensure high mobility of Ni within the plant. The Ni tolerant proteins TgMTPIs from the Ni hyperaccumulator species *T. goesingense* have been suggested to be responsible for metal ion accumulation in the shoot vacuoles of this plant (Persans *et al.,* 2001). Similarly the protein ZAT1 has been implicated in the vacuolar sequestration of Zn (Zaal *et al.,* 1999). Memon and Yatazawa (1984) suggested

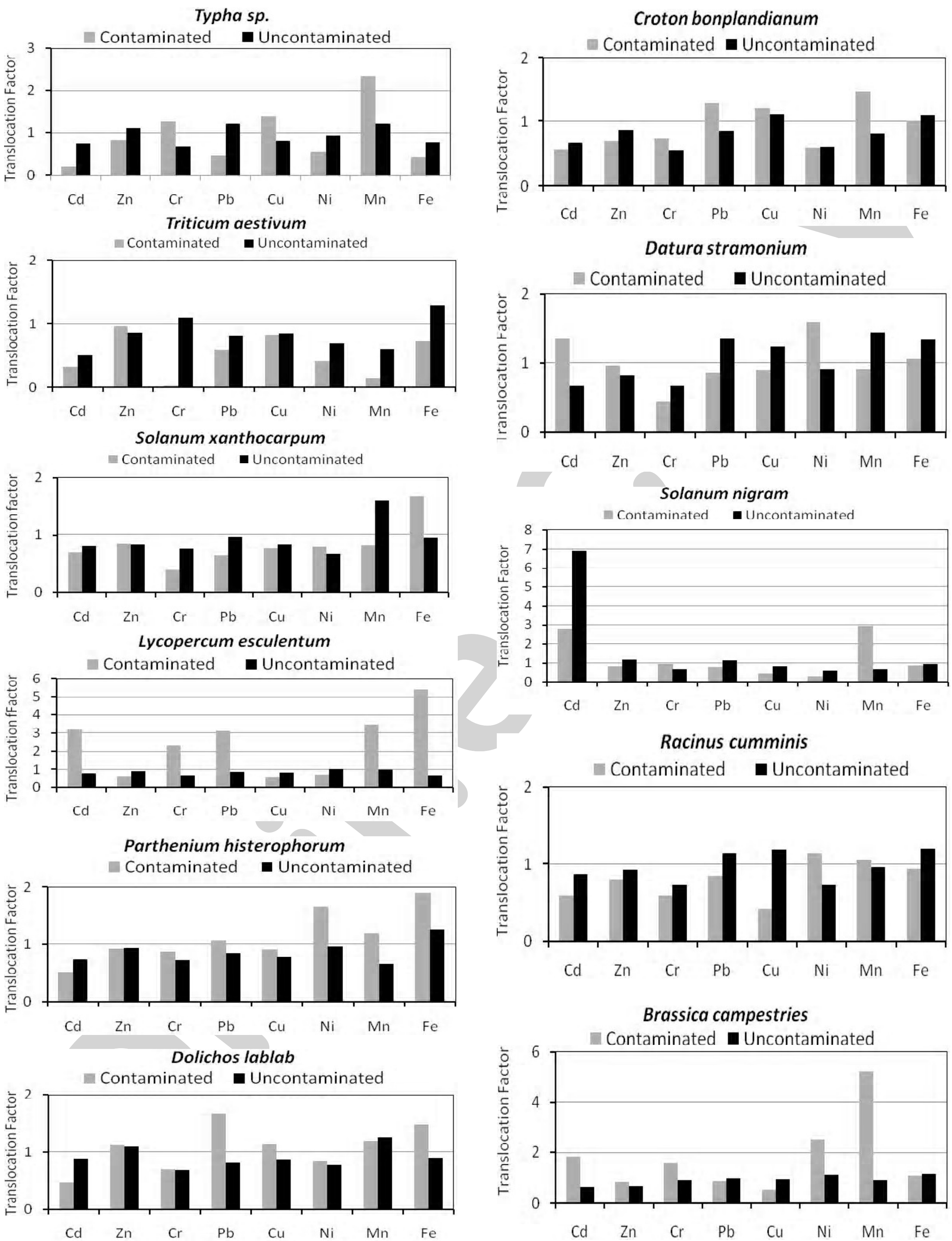


Fig. 4: Translocation of different metals from root to shoot of the different plant species grown in contaminated soil

from their observation for Mn distribution in plant species of *Acanthopanz sciadophyllo*ides and *Thea sinensis*, that Mn^{+2} is taken up at the plasma membrane and binds with malate in the cytoplasm and this Mn-malate complex is transported through tonoplast membrane to the vacuole where Mn dissociates from malate and complexes with oxalate. Here malate functions as a "transport vehicle" through the cytoplasm and oxalate as the "terminal acceptor" in the vacuole. Several other mechanisms may contribute to heavy metal tolerance depending on the type of metal and plant species (Memon and Yatazawa, 1984; Memon *et al*., 2001). Complex with metal binding peptide, metallothioneins and phytochelatins may also serve to alleviate the toxicity of heavy metals in plants. Metallothioneins such as cysteine-rich proteins have high affinity for binding metal cations such as Cd, Cu and Zn (Singh *et al.,* 2003). Dan *et al.* (2000) suggested that lignifications of cell wall and formation of metal-lignin complex might be one of the primary mechanisms of Pb tolerance in the roots of scented *Geranium* plant. In the present study, Pb accumulation/translocation is higher (*i.e.* ratio >1) in *Parthenium hysterophorus, Croton bonplandianum* and *Lycopersicum esculentum.* The uptake and translocation of Pb may also be dependent on the mobility of Pb as well as its competition with other metals within plants.

The study revealed that fly ash contributed a high level of Fe, Mn, Cu, Zn, Cr, Ni, Pb, and Cd in the contaminated soil and subsequent higher accumulation in plant parts. Higher level of metal accumulation in plant parts especially in upper parts of *Typha* sp. *Datura stramonium, Parthenium hysterophorus, Lycopersicum esculentum, Brassica campestris* and *Croton bonplandianum* showed biomagnification of metals and thus these plants can be considered as accumulator species.

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