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Decontamination and/or revegetation of fly ash dykes through naturally growing plants

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Abstract

Present study is focused on the decontamination and/or revegetation of fly ash dykes through naturally growing plants, namely *Calotropis procera*, *Cassia tora*, *Chenopodium album*, *Sida cardifolia*, *Blumea lacera*. The results of sequential extraction study showed that maximum amount of metals (Na, K, Fe, Mn, Cr, Pb, Ni, Cd) were associated with residual and Fe–Mn fractions. Diethylenetriamine penta acetic acid (DTPA)–triethanolamine (TEA) extraction assessed the bioavailability of the metals. The total metal accumulation in tested plants was found in the order; *C. album* > *S. cardifolia* > *C. tora* > *C. procera* > *B. lacera*. The maximum bioconcentration factor (BCF) was recorded in *S. cardifolia* for the metals (Na, Fe, Zn, Cd), in *C. procera* for the metals (Mn, Cu, Ni, Cr) and in *C. album* for the metals (Co, Pb). However, the translocation factor (TF) of most of the metals was found more in *S. cardifolia* followed by *C. album* than other plants. Among all the plants, *C. album* have shown high BCF and low TF values for toxic metals (Pb, Cd) and suitable for phytostabilization of these metals. Principal component analysis was used to predict translocation behavior of the metals in different parts of the plants which was found similar for the metals (Cu, Zn, Mn, Cr). All examined plants are suitable for revegetation (naturally grows on fly ash dykes) and *S. cardifolia* and *C. album* may be used for decontamination purposes.

Keywords: Fly ash dykes; Decontamination; Sequential extraction; Principal component analysis; Accumulation

1. Introduction

The management of coal combustion residue, i.e., fly ash is becoming more challenging due to strict environmental regulations of its use and disposal, which may affect soil fertility and eventually ground water quality. Coal fly ash contains a number of heavy metals including toxic and essential elements to plant growth. To overcome this environmental menace, the revegetation of fly ash landfills with tolerant plants is one of the cheap alternatives. It may check the dispersal of fly ash arising into the atmosphere and develop a bioaesthetic environment for local inhabitants [1]. In addition, it may also check the leaching of toxic metals.

The use of plants including trees and grasses has been reported to remove, destroy, or sequester hazardous contaminants from media (soil, water, air) [2]. It offers the benefits of being *in situ*, low cost and environmentally sustainable solution. Plants have unique ability to tolerate essential and nonessential elements from soils basically through two strategies: exclusion and accumulation. In addition to hyper-accumulators [3], plants such as trees and grasses are now being actively evaluated for phytoremediation [2,4]. Though, their metal bioconcentration capability is well below than that of hyper-accumulator plants [2]. Contamination with multiple heavy metals also makes it a difficult challenge for phytoremediation. A potential strategy is required for the remediation of contaminated soil or sites having low fertility and poor soil structure. The trees have the most massive root systems, which penetrate the soil for several meters. In some tree species, above-ground biomass can be harvested, and trees will be uprooted. This attribute would be valuable if periodic removals of metals sequestered in plant tissue are desired [5]. Recently, author's laboratory have reported phytoextraction

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potential of four naturally growing plants (*Sida acuta*, *Ricinus communis*, *Calotropis procera*, *Cassia fistula*) on tannery sludge dumping site of Jajmau, Kanpur (UP, India) [6]. Similarly, the selection of the plants for decontamination of the wastes such as fly ash, is a challenging task, due to the presence of high level of pH, EC, essential, non-essential metals and absences of nitrogen, available phosphorus, and organic matter. In this connection, the use of naturally growing plants for the decontamination or revegetation of these dykes is vast focus areas of research.

In India, National Thermal Power Plant (NTPC) run more than 70 thermal power plant and subsequently generate about 110 million tonnes of coal-ash [7] and this is predicted to increase up to 170 million tonnes per annum by the year 2012. Huge amounts of fly ash are deposited in adjoining area of the thermal power plants. These barren fly ash dykes create varied kinds of environmental hazards due to erosion and leachate generation and health hazards to the nearby inhabitants. Therefore, various revegetation trials have been carried out using amendments of fly ash with organic material and N_2-fixing organisms to check fly ash entering into the environment [1,8].

In view of the above, the present study focused on phytoextraction capacity of the naturally growing plants collected from fly ash dykes. Emphasis was also put fourth on the accumulation of metals and its translocation pattern in the plants naturally growing on fly ash dykes. Single and sequential extractions of fly ash help to predict mobility and bioavailability of metals from fly ash to the plants. Since the data obtained in this study had multivariate nature and several of the variables were correlated, principal component analysis (PCA) were used for the interpretation of the data. This technique is an unbiased methods which can indicate natural associations between samples and/or variables. The multivariate analysis of the data is widely used to characterize and evaluate the behavior of the metals accumulated in the plants grown on fly ash dykes. Information obtained from this study should provide insight for using native plants to remediate metal contaminated sites.

2. Materials and methods

2.1. Study area and sampling

Firoz Gandhi National Thermal Power Plant (FGNTPC), Unchhahar, Raibarelli (UP), is one of the biggest thermal power plant of Uttar Pradesh (India). Five plants namely *C. procera* (Ait) R.Br., *Cassia tora* L., *Chenopodium album* L., *Sida cardifolia* L., *Blumea lacera* (Burm f.) DC, naturally growing on fly ash dykes during winter period (December 2004–January 2005). These plants (4–5 months old) were collected randomly, carefully packed into polythene bags and brought to the laboratory for the analysis. The plants were carefully washed with doubled distilled water to eliminate the adhering soil and other contaminants. Special attention was given to the roots, which were scrubbed free of soil. The dry weights (g) of the plants were as follows: *C. procera* (17.12), *C. tora* (13.32), *C. album* (11.51), *S. cardifolia* (16.9) and *B. lacera* (8.66). After dry weight determinations, the plants were separated, crushed and mixed thoroughly for the processing of metal analysis. The samples of fly ash were also collected randomly at the time of harvesting of the plants. These samples were oven dried, sieved by 2 mm stainless steel sieve and used for analysis. All the samples were analyzed in triplicates. The Analytical Grade reagents were used. Certified aqueous standards of the elements (SIGMA) were used to prepare standard curve for Atomic Absorption Spectrophotometer. All the standards, reagents solution and samples were stored in polyethylene containers previously cleaned with 4 M HNO_3 and rinsed with double distilled water.

2.2. Physico-chemical analysis of fly ash

The pH of the fly ash was measured with 1:2 (w/v) fly ash water suspension using Orion pH meter (Model 420); electrical conductivity (EC) was measured using Orion Conductivity meter (Model 150). Organic carbon (OC), organic matter (OM) and cation exchanges capacity (CEC) were measured using manual of Kalra and Maynard [9].

2.3. Total, DTPA extractable, sequential extraction of metals and metal accumulation in the plant

After homogenization, a small portion (ca. 0.10 g) of plant samples was digested in 5 ml of 70% HNO_3 using Microwave Digestion System (MDS 2000) in closed teflon vessels at 630 W, 40 PSI for 20 min. For total metal analysis, fly ash (ca. 0.50 g) was digested in 10 ml double distilled water +5 ml HNO_3 + 4 ml HF + 1 ml HCl using Microwave Digestion System (MDS 2000) at 630 W, 120 PSI for 120 min of time. Digested samples were filtered through Whatman filter paper (42 No.) prior to run in GBC Avanta Σ Atomic Absorption Spectrophotometer for metals analysis. For diethylenetriamine penta acetic acid (DTPA) extractable metal fraction, fly ash samples (10 g) was mechanically shaken with 20 ml of 0.005 M DTPA, 0.01 M $CaCl_2$, 0.1 M triethanolamine (TEA) buffered at pH 7.3 for 2 h [10]. The fly ash was also fractionated (five steps) by the method of Tessier et al. [11] and chemical reagent, extraction conditions and corresponding fractions are listed in Table 1.

Table 1
Sequential extraction scheme

Steps	Fractions	Extraction procedures
1	Exchangeable (EXC)	1 g of soil sample, 8 ml 1 mol l^{-1} $MgCl_2$, pH 7, shake 1 h, room temperature
2	Bound to carbonate (CAR)	8 ml 1 mol l^{-1} CH_3COONa, adjusted pH to 5.0 with CH_3COOH, shake 5 h, room temperature
3	Bound to Fe–Mn oxides (Fe–Mn)	20 ml 0.04 mol l^{-1} $NH_2OH{\cdot}HCl$ in 25% CH_3COOH, pH 2.0, water bath, 96 °C, 6 h, occasional shaking
4	Bound to organic matter (OM)	3 ml 0.02 mol l^{-1} HNO_3, 30% H_2O_2 (adjusted to pH 2.0), water bath, 85 °C, 5 h, 3.2 mol l^{-1} CH_3COONH_4 in 20% (v/v) HNO_3, shake 30 min
5	Residual (RES)	3 ml HNO_3 + $HClO_4$ + HF under high pressure, 170 °C

2.4. Bioconcentration factor (BCF) and transfer factor (TF)

Bioconcentration factor was calculated by dividing the trace element concentration in plant tissues at the time of harvest by the initial concentration of the DTPA extractable element in the fly ash/soil. Translocation factor is defined as the ratio of metal concentration in shoots to that in roots.

2.5. Statistical analysis

Principal component analysis was performed on experimental data standardized through *z-scale* transformation in order to avoid misclassification due to wide differences in data dimensionally. It was applied to the metals accumulation in the plants in order to examine the interactions between metals in the studied plant species. PCA was performed on a varimax normalized original matrix composed of 33 lines and eleven even columns (Na, K, Fe, Zn, Mn, Cu, Ni, Co, Pb, Cr and Cd). These columns can be considered as statistically dependent variables or factors. Student's *t*-test (two tailed) and Pearson's correlation coefficients analysis were also performed. All the mathematical and statistical computations were made using Excel 97 and STATISTICA 5.0 software.

2.6. Quality control and quality assurance

The standard reference material of metals (E-Merck, Germany) was used for the calibration and quality assurance for each analytical batch. Method validation (accuracy and repeatability) was performed by analyzing the certified materials reference solution (BND 1101.02) of multi-elements (Zn, Fe, Cu) and single element reference solution, BND 102.03 (Pb); BND 402.02 (Cr) and BND 1001.02 (Ni) provided by National Physical Laboratory (NPL), New Delhi, India and the results were found within ±1.81% of certified values ($n = 10$). For fly ash, analytical data quality of metals was ensured through repeated analysis of ash of Resource Technology Corporation (EPA Certified Reference material) ASH No. 3 (Catalog No. CRMO 19-050; Lot No. Y019a) and results were found to be within Prediction Intervals. The blanks were run in triplicate to check the precision of the method with each set of samples.

3. Results and discussion

3.1. Physico-chemical analysis of fly ash

The results of physico-chemical analysis of fly ash are presented in Table 2. The analysis of the data indicated that pH was alkaline, and EC was quite high in comparison to normal soil. High pH may also be a limiting factor for the growth of the plant on fly ash, particularly on unweathered deposits. As mentioned earlier by Carlson and Adriano [12], pH of the fly ash is basically dependent on the composition of parent coal. In India, most of the mines having bituminous coal, they have low sulphur content and high pH.

Table 2
Physico-chemical properties of fly ash

Parameters	Fly ash
pH (1:2)	8.12 ± 0.01
EC ($\mu S\,cm^{-1}$)	354.1 ± 3.5
Salinity (‰)	7.4 ± 0.00
CEC ($Cmol\,kg^{-1}$)	8.5 ± 1.01
OC (%)	1.7 ± 0.01
OM (%)	3.0 ± 0.02

Metals	DTPA extractable metals ($mg\,kg^{-1}$ dw)	Total metals ($mg\,kg^{-1}$ dw)
Na	386.48 ± 31.06[b] (0.935)	41,321.5 ± 2145
K	283.62 ± 17.91[b] (0.988)	28,706.09 ± 992
Fe	31.81 ± 3.2[a] (0.158)	20,054 ± 105
Zn	12.8 ± 0.2[a] (13.52)	94.67 ± 6.1
Mn	40.6 ± 2.1[a] (15.496)	262 ± 19.0
Cu	3.4 ± 0.1[a] (12.20)	27.85 ± 2.11
Ni	3.9 ± 0.0[a] (16.63)	23.44 ± 2.01
Co	2.89 ± 0.5[a] (10.77)	26.81 ± 1.24
Pb	3.8 ± 0.8[a] (11.83)	32.11 ± 2.55
Cd	5.25 ± 0.12[b] (16.81)	31.23 ± 1.23
Cr	1.22 ± 0.11[b] (8.01)	15.23 ± 0.21

All the values are mean of three replicates ± S.D. Student's *t*-test (two tailed as compared to total metal). Values in parenthesis are percent of total metals.
[a] $p < 0.01$.
[b] $p < 0.001$.

Soluble salt concentrations in unweathered fly ash were generally high, which may result in high EC values [13]. The level of soil EC values exceeding 13 $dS\,m^{-1}$ considered as threshold for causing adverse effects on most of the plant species, including agronomic and horticultural crops. Salinity problems usually do not occur in the plants until the soil EC values are greater than ∼1.5 $dS\,m^{-1}$ for salt-sensitive, 3.5 $dS\,m^{-1}$ for moderately salt-sensitive, or 6.5 $dS\,m^{-1}$ for moderately salt-tolerant species [14]. In addition to direct effects on the plant establishment by phytotoxic constituents in unweathered fly ash, indirect effects can also occur in the rhizosphere due to the inhibition of microbial activity by high pH and nutrient imbalances in the plants due to the presence of elevated levels of trace elements (e.g., Cd, Cr, Zn, etc.) and soluble salts (e.g., Ca, S) [15]. Adriano and Weber [16] also discussed several physical properties which can also limits the successful vegetation on fly ash dykes. In most of the cases, regular ash deposit develop cemented or compact layers that reduce the aeration, water infiltration, and penetration of the roots. Several organic amendments like press mud, cow dung [1] and sewage sludge [17] may help to break up cemented layers.

3.2. Total metal, DTPA extractable metals and sequential extraction of fly ash

In fly ash, total metal concentrations ($mg\,kg^{-1}$ dw) of Na (41,321) > K (28,706) > Fe (20,054) were found most abundant followed by Mn (262) > Zn (94.67) > Pb (32.11) > Cd (31.23) > Cu (27.85) > Co (26.81) > Ni (23.44) > Cr (15.23) (Table 2). The concentration of metals except Fe was below the toxicity threshold limit proposed by several researchers for soil materials [18]. However, the soil conditions, which influence the mobility and availability of the metals to the plant species, may modify the toxicity threshold. In case of alkaline (pH 8.3) fly ash; metal contents (mg kg^{-1} dw) were reported as Fe, 18,444; Mn, 242.2; Zn, 64.5; Ni, 17.7; Cu, 10.9; Pb, 112.3 which affect growth of the plant of *Brassica juncea* var vaibhav [7].

The total metal concentration of the soil includes all fractions of a metal, from the readily available to highly unavailable. Whereas, total metal provides the maximum pool of metal in the soil, other factors have a greater importance in determining how much of this soil pool will be available to the plants [19]. The estimation of bioavailable fraction of the metal is probably more significant rather than the analysis of total metal contents, because the former allows prediction of the risk of metal uptake by the plants and its mobility in the system. The mobility and availability of the metals are strongly influenced by pH of the substrate. Higher the pH, lower the availability of most of the trace elements except, As, Se and V as they are more soluble at higher pH [20]. Recently, author's lab reported that DTPA extraction procedure was found best among all the tested extraction (DTPA, $CaCl_2$, NH_4NO_3) procedures for fly ash amended soil to *B. juncea* grown in earthen pots [7]. On the basis of the above findings, we also used DTPA extraction procedures in the present study. Data showed that DTPA extraction have shown the following order: Na > K > Mn > Fe > Zn > Ni > Pb > Cu > Co > Cd > Cr. The maximum extraction was observed for Ni (16.81%) and minimum for Fe (0.158%) with respect to total metal content in fly ash. It is also evident from the metals partitioning data (Fig. 1) where 14.4% Ni and 0.03% Fe was associated with EXC fraction. The DTPA extracted percentage of toxic metals, Pb, Cd and Cr were 11.83, 16.81 and 8.01%, respectively, with respect to total metal content in the fly ash, whereas, percentage of essential metals (DTPA extracted) were as follows: Zn (13.52), Mn (15.49) and Cu (12.20) with respect to total metal. These variations largely depend on their association with EXC, CAR and OM fractions which was clearly evident from sequential extraction analysis of fly ash.

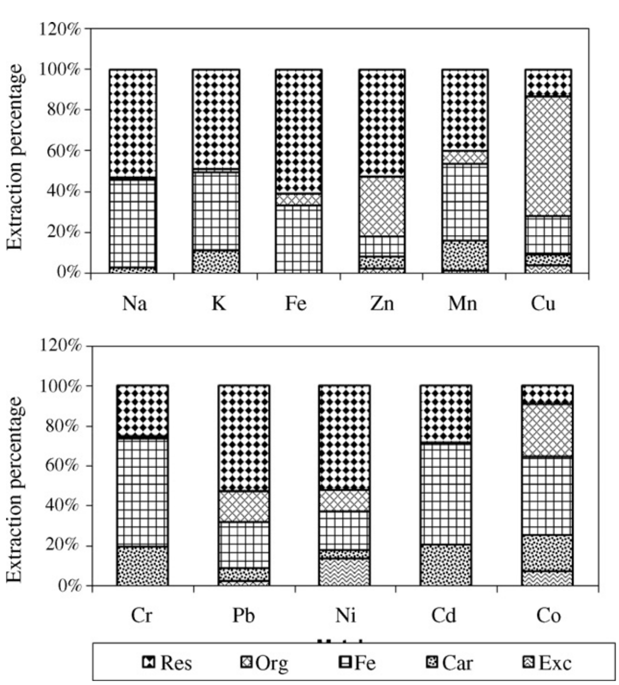

Fig. 1. Distribution of metals in fly ash among different phases obtained by the five-step procedure.

Tessier's classified five chemical types of ions to bind on the surface of an inorganic matrix: EXC, CAR, Fe–Mn, OM and RES (silicates) phase. The results of sequential fractionation (Fig. 1) of fly ash showed that the levels of K, Fe and Ni were found maximum in RES fraction, while Zn, Mn, Cr, Pb, Cd and Co were mostly associated with Fe–Mn oxide fraction. However, majority of Cu and Na are associated with OM and CAR fractions, respectively. Fly ash usually contains large fraction of carbonates and ferro-alumino silicate which explains that most of the metals fraction was bound with these fractions. Such information is potentially valuable for predicting bioavailability, metal leaching rates and transformations between chemical forms in agricultural and contaminated soils.

3.3. Metals accumulation

The general trend of metal accumulation in the all the studied plants were found in the order; Na > K > Fe > Mn > Zn > Co > Ni > Cd > Pb > Cu > Cr (Fig. 2A–C). Among all the studied plants, the accumulation of the metals was found in the order: *C. album* > *S. cardifolia* > *C. tora* > *C. procera* > *B. lacera*. Interestingly, the total accumulation of toxic metals particularly, Pb and Cd was recorded quite high in most of the plants. The accumulation of essential metals was found maximum in the plants of *C. procera* followed by *C. album*, however, the accumulation of tested toxic metals (Pb, Cd, Cr) was found maximum in the plant of *C. album* followed by *S. cardifolia*.

The ability of the plants to accumulate metals from soil can be estimated using bioconcentration factor, which is based on the ratio of metal concentration in the roots to that in soil [21]. The BCF values (Table 3) greater than 1 are an indication of potential heavy metal phytoremediator species [22,23]. However, the metal moved from root to shoot indicates that they are transported in to the transpiration stream via the xylem. The rate

Table 3
Bioconcentration factor (BCF) from soil (DTPA extractable metals) to the tested plants collected from fly ash dykes

Metals	Bioconcentration factor				
	C. procera	*C. tora*	*C. album*	*S. cardifolia*	*B. lacera*
Na	6.178	7.760	6.187	**9.015**	5.831
K	20.697	**27.392**	15.478	4.922	26.088
Fe	26.366	11.514	29.209	**36.995**	26.269
Zn	1.660	1.546	1.441	**3.348**	1.105
Mn	**1.196**	0.560	0.384	0.625	0.142
Cu	**12.066**	1.878	2.706	3.994	1.448
Ni	**1.600**	1.108	1.481	0.439	0.538
Co	1.397	1.212	**1.772**	0.577	0.707
Cr	**9.545**	4.235	5.236	4.886	4.074
Pb	9.126	17.991	**26.123**	7.315	2.023
Cd	9.316	27.901	30.895	**33.136**	18.296

Values presented as bold showed maximum BCF values.

of transportation was not identical for all the heavy metals in the plant. Translocation factor was calculated to represent the ratio of translocation of metals from root to shoot of the plants.

Bioconcentration factor showed maximum values (Table 3) in the plants of *S. cardifolia* for Fe (36.99) and Cd (33.14) followed by *C. album* for Pb (26.12), Cd (30.89) and Fe (29.20). It is clearly evident from the above analysis that these plants may be suitable to phytoextract these metals (Fe, Pb, Cd) from the fly ash dykes. Kabata-Pendias and Pendias [18] shared the opinion that BCF of Cd is normally 1–10, however, Dudka et al. [24] indicated that this index for mature plants grown over contaminated soil was well below 1. Recently, Gupta and Sinha [26] also reported high transfer factor values for all the tested metals in the plants of *C. album* grown on soil amended with 10% tannery sludge.

The maximum root-to-shoot transfer of the metal was observed with Mn (9.488 for *S. cardifolia*) followed by Zn (3.61 for *C. album*), whereas, minimum translocation was observed for Co (0.444 for *C. procera*) followed by Na and K (Table 4). The values of transfer factor were considerably low in *B. lacera* than in *C. procera*. It might be suggested that the requirement of an element in the rapidly growing tissues of the plant may explain the different movement of the heavy metal transfer from roots to the shoot. Among all the tested plants, the highest translocation was recorded in the plants of *S. cardifolia* for the metals (Fe, Mn, Cu, Cr and Cd) and in *C. album* for the metals (Zn, Ni, Co and Pb). On the basis of above findings, the plants of *S. cardifolia* and *C. album* may be suitable for phytoextraction of the metals from fly ash dykes. Cadmium and Cr may be recognized as toxic compound by the roots of both *S. cardifolia* and *C. album*, thus leading to the activation of mechanisms such as sequestration in the vacuole or in the cell walls, especially in *C. album* in order to avoid an accumulation of cadmium in the shoot. In addition, very high affinity of metals, e.g. Cr and Pb, to the root cell walls may hamper their translocation into the shoot [25]. Recently, Gupta and Sinha [26] also reported that the high accumulation metals (Fe, Mn, Zn, Cr) in the plant of *C. album* grown on soil amended with tannery sludge.

Several studies indicated that the partitioning of heavy metals in the whole plant could broadly be divided into three cate-

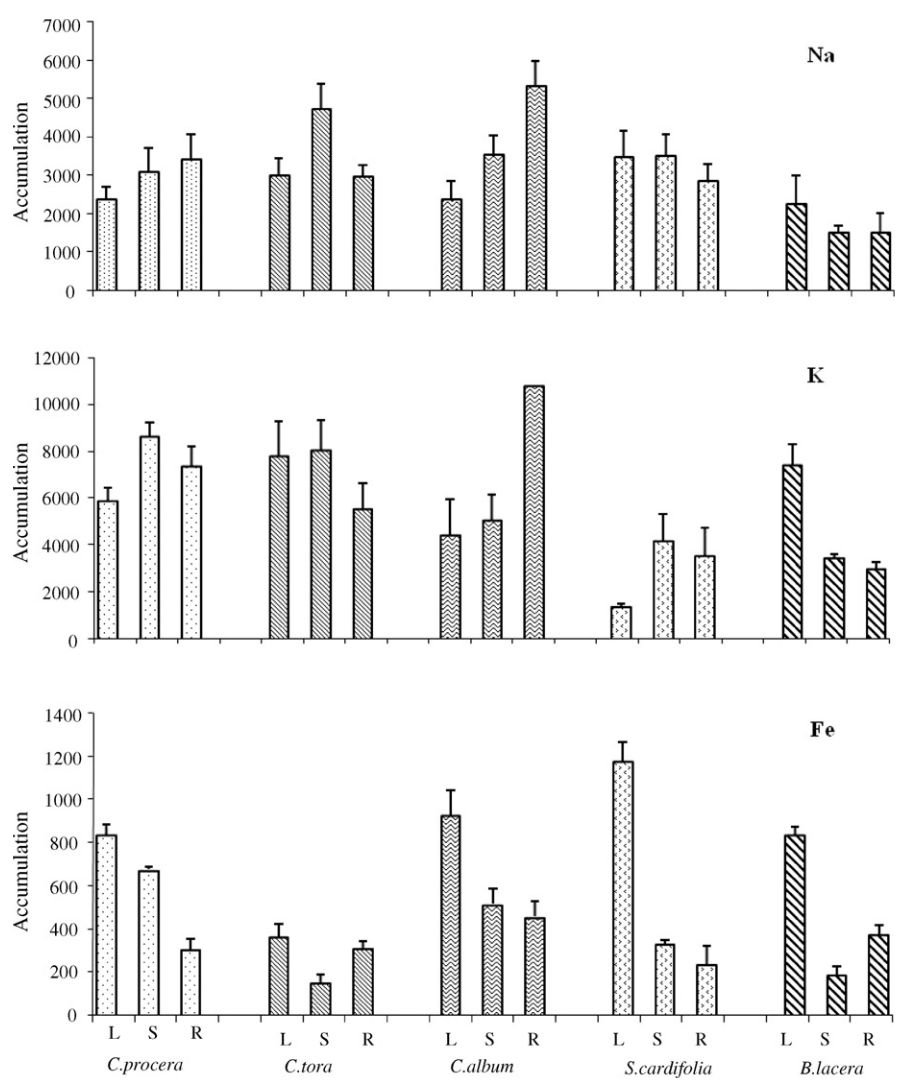

Fig. 2. Metal accumulation (mg kg^{-1} dw) in the leaves (L), stem (S) and roots (R) of the plant collected from fly ash dykes.

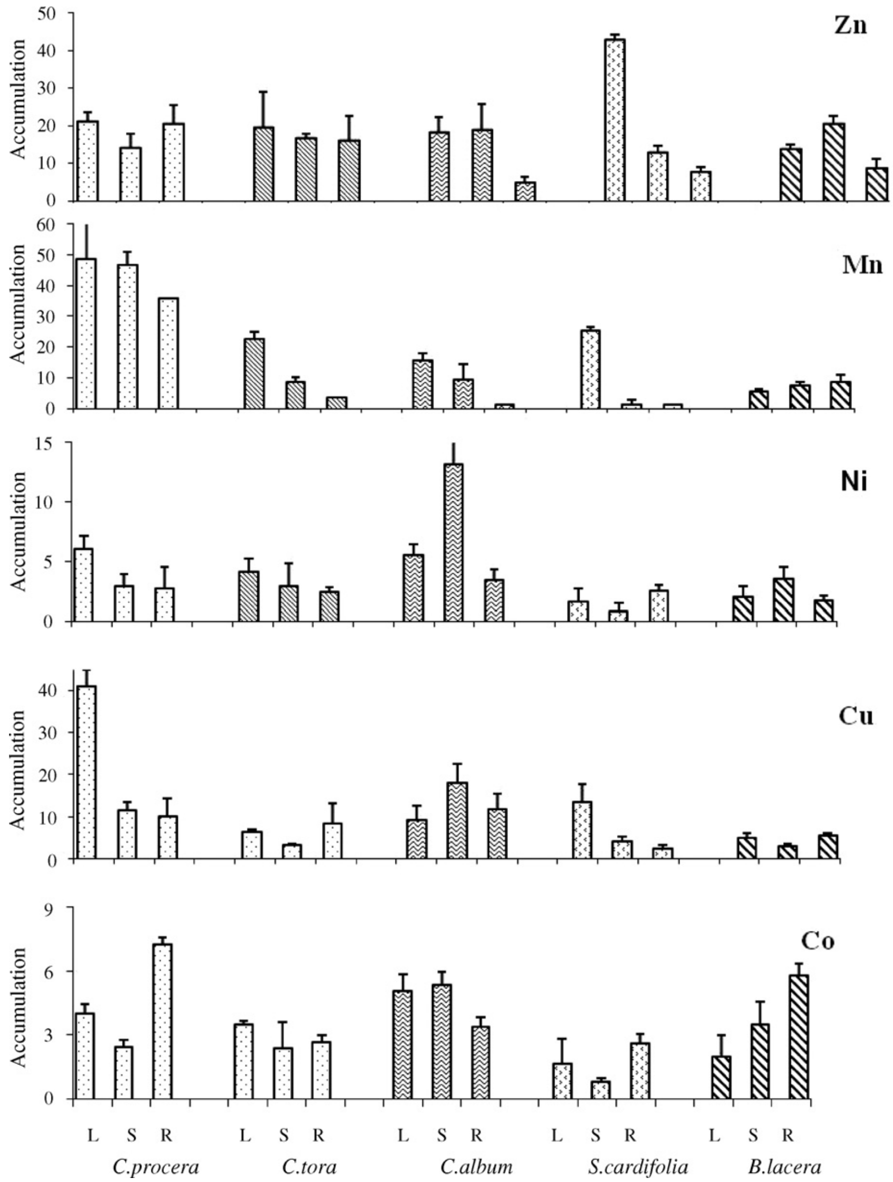

Fig. 2. (*Continued*)

gories. For instance, Alloway [27] classified Mn, Zn, Cd, B, Mo and Se as elements, which were readily translocated to the plant shoots; Ni, Co and Cu, were intermediate, and Cr, Pb and Hg were translocated to the lowest extent. Above classification strengthen the present finding, where, maximum translocation factor was found with Mn in all the tested plants followed by Zn and Cd. It is well known that Zn is a competing ion for Cd [28] and depresses Cd uptake [29]. Uptake of Cd depends upon the content of Zn in the soil, and plant generally takes up more Cd if the Zn content is low. This was clearly visible in case of *C. album*, where, translocation was found better with Zn and poor with Cd. Hart et al. [30] reported the reason of competition among these two metals. They emphasized that Zn and Cd are transported by a common carrier at the root plasma membrane, which has a higher affinity for Cd than for Zn. Therefore, Cd and Zn ions should experience competitive inhibition. Recently, Yang et al. [31] also suggested that Cd and Zn showed synergistic interaction for both absorption and transport in the plant.

In case of *S. cardifolia* and *B. lacera*, the concentrations of Pb in the roots were much greater than those of the shoot, indicating its low mobility from the roots to the shoots and immobilization in to roots. In contrast, the plants of *C. tora* showed maximum translocation factor (557.8%) for Pb followed by *C. procera* (262.3%). Translocation of Cd was found high in most of the tested plants except, *C. album.* The maximum translocation of Cd was noticed in case of *S. cardifolia* (264.8%) followed by *C. tora* (238.9%). In contrast, Gupta and Sinha [26] have also emphasized that the translocation of Cr and Cd was better in the leaves of the *C. album* grown on tannery waste amended soil.

Metal accumulation and its distribution in the plant tissue are important aspects to evaluate the role of plants to remediate the contaminated sites. The success of phytoremediation

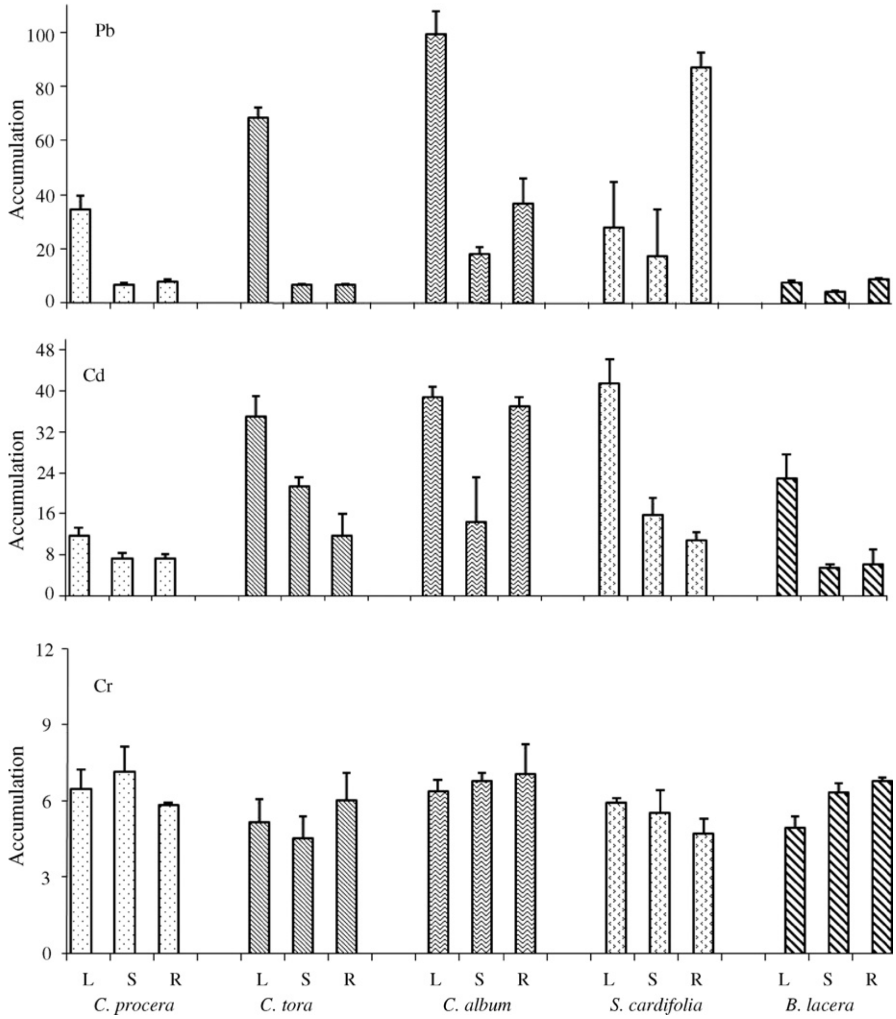

Fig. 2. (*Continued*).

process depends on adequate plant yield and hyperaccumulator plants possess an ability to take up abnormally high amounts of heavy metals in their above ground parts. Plants must have the ability to translocate metals from the root to the shoot, or to compartmentalize it, in order to continue absorption of metals from the substrate. Better translocation is advantageous for phytoextraction; it can reduce metal concentration and thus reduce toxicity potential to the root, and translocation to the shoot from the roots is one of the mechanisms of resistance to high metal concentration.

Table 4
Translocation factor (TF) from roots to shoots of the all tested plants

Metals	Translocation factor				
	C. procera	*C. tora*	*C. album*	*S. cardifolia*	*B. lacera*
Na	0.799 (79.9)	**1.298 (129.8)**	0.555 (55.5)	1.227 (122.7)	1.248 (124.8)
K	0.985 (98.5)	1.429 (142.9)	0.439 (43.9)	0.789 (78.9)	**1.823 (182.3)**
Fe	2.472 (247.2)	0.830 (83.0)	1.579 (157.9)	**3.175 (317.5)**	1.361 (136.1)
Zn	0.858 (85.8)	1.135 (113.5)	**3.610 (361.0)**	3.541 (354.1)	1.964 (196.4)
Mn	1.331 (133.1)	4.274 (427.4)	8.573 (857.3)	**9.488 (948.8)**	0.744 (74.4)
Cu	2.589 (258.9)	0.567 (56.7)	1.143 (114.3)	**3.566 (356.6)**	0.714 (71.4)
Ni	1.624 (162.4)	1.456 (145.6)	**2.734 (273.4)**	0.482 (48.2)	1.525 (152.5)
Co	0.444 (44.4)	1.094 (109.4)	**1.541 (154.1)**	0.482 (48.2)	0.481 (48.1)
Cr	1.169 (116.9)	0.807 (80.7)	0.935 (93.5)	**1.215 (121.5)**	0.834 (83.4)
Pb	2.623 (262.3)	**5.578 (557.8)**	1.454 (145.4)	0.258 (25.8)	0.86 (86.0)
Cd	1.304 (130.4)	2.389 (238.9)	0.720 (72.0)	**2.648 (264.8)**	2.252 (225.2)

Values in parenthesis are (percent) TF values. Values presented as bold showed maximum TF values.

Number of reports emphasized that the metal accumulation depends on the concentration of available metals in the soils, their mobility and the plant species growing on [6,17]. During this study, the accumulation of metals (Zn, Pb and Cd) was recorded more in above ground parts of some of the plants. These findings strengthen with earlier findings of Sinha et al. [32] as they have reported high translocation of metals from roots to shoots in the plants of *Prosopis juliflora* L. grown on fly ash amended soil. Significantly high accumulation of essential metals (Fe, Mn, Zn, Cu) has been reported in different parts of the plants namely *Cassia siamea* Lamk. [33] and *Sesbania cannabina* L. [34] grown on different amendments of fly ash. Recently, Gupta and Sinha [6] also reported that the accumulation of metals (Na, K, Fe, Mn, Cu, Zn, Ni, Cr, Pb) in the plant of *C. procera* was found high among all the collected plants (*S. acuta*, *R. communis*, *C. procera*, *C. fistula*) grown on tannery sludge dumping sites.

Zayed et al. [22] mentioned that bio-concentration factor was a better indicator to classify a particular plant as a hyper-accumulator. Transfer factor can be used to evaluate the capacity of a plant to transfer metals from roots to shoots (TF is usually >1) in (hyper) accumulators and <1 in excluders [35]. Although, no hyper-accumulator plants were found in this study, however, among all the collected plants, *C. album* have shown high BCF and low TF values for toxic metals (Pb and Cd), it restricts the mobility of metals in the soil (phytostabilization), and leaching

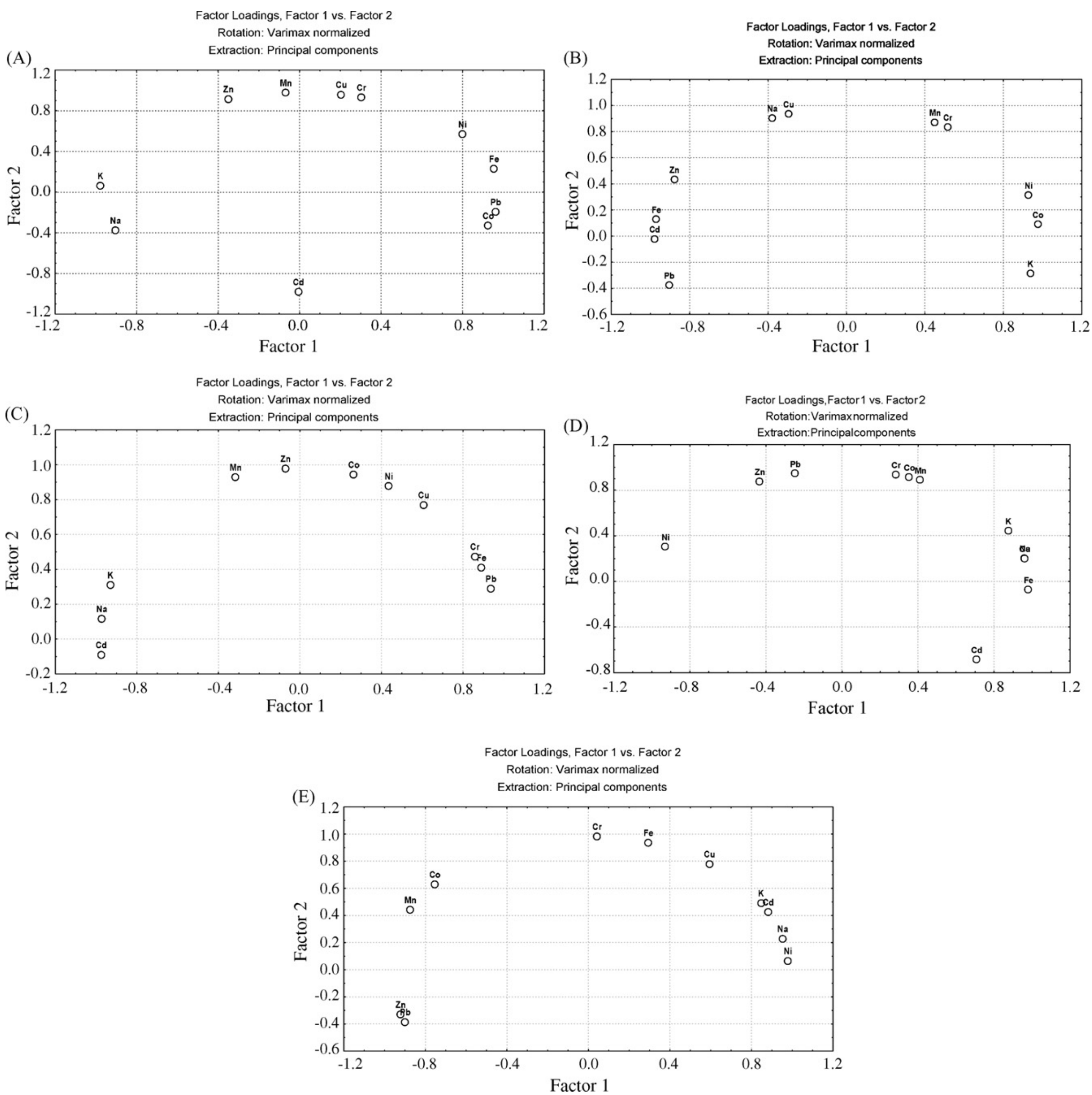

Fig. 3. PCA applied to the metals accumulated in different parts of the *C. procera* (A), *C. tora* (B), *C. album* (C), *S. cardifolia* (D) and *B. lacera* (E) collected from fly ash dykes.

into ground water. By using such metal-tolerant plant species for stabilizing contaminants in soil, it could also provide improved conditions for natural attenuation or stabilization of contaminants in the soil. Metals accumulation in the roots is considered relatively stable as far as release of the metals in the environment is concerned.

3.4. Principal component analysis

Principal component analysis is a unsupervised multivariate technique in which new variables are calculated as linear combinations of the old ones (metal concentrations in lower and upper parts the plants); the new variables, called principal components (PC), have two main features: (i) they are uncorrelated between themselves; (ii) the first PCs keep the main part of the variance of the original data set. In this way, it is possible to show a great part of information by plotting the first two or three PCs (in this case, the first three PCs were always computed and the two most meaningful ones, usually PC_1 and PC_2, were considered to represent all examined objects and variables). The combined plot of scores (coordinates of the objects on the new variables) and loadings (weights of original variables on the linear combination, PCs are built from) allow us to recognize groups of samples with similar behavior and the existing correlation among the original variables.

The results of PCA analysis based on the metal accumulation in all the tested plants are presented in Fig. 3A–E. According to these results, the eigen values of the first extracted components are greater than rest of the components. As a consequence, heavy metals could be grouped into a four-component model that accounts for 36.46% of all the data variation. Spatial representation of the three rotated components is shown in Fig. 3A. Factor 1, which accounts for 36.46% of total variance of Fe and Cd and have shown significant positive loading, whereas, Co and Ni have shown significant negative loadings. Bioconcentration factor data also showed that the values of Fe and Cd were higher in most of the tested plants, whereas, Co and Ni were lowest. Copper and Pb showed significant negative and Zn has shown significant positive loading accounting for 22.72% of total variance and associated with PC_2. Macro-elements Na and K factor explain 14.32% of the total variance and associated with PC_3, however, Mn showed negative loading and explain 9.79% of the total variance with PC_4. These results may suggest distribution pattern of all the tested metals accumulated in the plants grown on fly ash dykes.

The loadings and scores of the first two PCs (PC_1, PC_2) were plotted in Fig. 3. The loadings plot (Fig. 3A) showed distribution of all the metals in the first (upper right) and fourth (lower right) quadrants. The lines joining the variables and passing through origin in the plot of the factor loadings are indicative of the contribution of the variables to the samples. Closeness of lines for two variables signifies strength of their mutual correlation [36]. Grouping of metals (Fe and Mn; Ni and Cr; Cd and Pb) in the loadings plot suggests their significant mutual positive correlation. The PCs score plots describe the characteristics of the samples and help to understand their spatial distribution. The PCs scores plot (PC_1 and PC_2) (Fig. 3B) showed the spatial distribution of the samples. It showed clustering of site-specific samples in space. From Fig. 3A–E, it is evident that samples distributed in upper right quadrant are more enriched with Fe, Mn, Zn and Ni, than those in lower right quadrant with Cr, Cu, Pb and Cd. The samples distributed in other two quadrants (upper left and lower left) are enriched with metals to a lesser extent.

4. Conclusion

The total metal accumulation was found high in all the plants and followed the order; *C. album* > *S. cardifolia* > *C. tora* > *C. procera* > *B. lacera*. However, the translocation factor of most of the tested metals was found better in *S. cardifolia* followed by *C. album.* Bioconcentration factor analysis indicated that the plant of *S. cardifolia* have shown maximum value for the metals (Na, Fe, Zn, Cd), in *C. procera* for the metals (Mn, Cu, Ni, Cr) and in *C. album* for the metals (Co and Pb). Among all the plants, *C. album* has shown high BCF and low TF values for toxic metals (Pb and Cd) and may be used for phytostabilization of fly ash dykes. Principal component analysis showed that translocation behavior of the metals (Cu, Zn, Mn and Cr) was found similar in the plants. Among all the naturally growing plants, the plants of *S. cardifolia* and *C. album* may be used for the decontamination of most of the metals from fly ash dykes, however, rest of the plants may be suitable for revegetation.

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