METAL ACCUMULATION IN 5 NATIVE PLANTS GROWING ON ABANDONED CU-TAILINGS PONDS

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Abstract: Tailings and plants were sampled from the abandoned Cu-tailing ponds of Rakha mines, Jharkhand, India. Tailings have high concentration of Cu, Ni and characterized by moderately acid environment and low nutrient contents. Plants belonging to 5 genera and 4 families were collected and analysed for metals in their above and underground tissues. Plant communities respond differently depending on their ability to uptake or exclude a variety of metals. Accumulated metals were mostly retained in root tissue indicating that an exclusion mechanism for metal tolerance widely exists in them. Retention of some metals more than toxic level in the above ground tissues of some plants suggests the presence of internal metal detoxification and tolerance mechanisms in them.

Keywords: Cu-tailings, heavy metal, metal tolerance, natural vegetation.

Introduction

Mill tailings are generally characterised by high metal content, low fertility and obviously low water holding capacity. A vegetative cover on such tailings may play an important role in metal removal via filtration, adsorption and cation exchange, and through plant induced chemical changes in the rhizosphere. Additionally, it helps to avoid dispersion of contaminants through wind erosion and by reducing water percolating through the soil. This may keep contaminants away from underlying ground water by stabilising them in the soil profile [27]. Some plants phytostabilise heavy metals in the rhizosphere through root exudates immobilisation [4] whilst other species incorporate them into root tissues [16]. Some plant species also transfer metals to their aboveground tissues, potentially allowing the soil to be decontaminated by harvesting the aboveground parts of the plants. The success of any phyto remediation technique depends upon the identification of suitable species that hyperaccumulate trace elements and produce large amount of biomass using established crop production and management practices [3,8,9]. Thus collecting plant species on contaminated sites prior to evaluation of plant metal concentration can be used to get information about specific plant behavior in this environment, which may be effective for selecting potential plants to be used in mine remediation and also to complete data about metal dispersion, with reference to their mobility to biomass [12,14]. There are many evidences that plants grown on metal enriched soil can accumulate high amount of heavy metals in their tissues such as Silene armeria [12], Typha latifolia [29], Festuca sp. [2], Salix viminalis [20], Vetiveria zizanioides [28], Juncus conglomerates [14]. In Rakha mines area, Jharkhand there are several abandoned Cu-tailings ponds that caused severe metal pollution to the nearby areas. Our investigation in Cu-tailings ponds revealed that many plant species could thrive in heavily metal enriched tailings. Henceforth, the concentration of Cu, Ni, Mn, Zn, Pb and Cd were examined in both the above and
underground tissues of 5 naturally growing plant species. The main objectives of our study were:

- to evaluate the different form of toxic metals present in Cu tailings and their bioavailability;
- to identify plant species grown naturally on abandoned Cu-tailings and to determine the ability of these species to accumulate and tolerate Cu, Ni, Mn and Zn in their tissue.
- to examine the extent of metal translocation from underground tissue to aboveground tissue in different plants for different metals.

The above information could be used for selection of appropriate plant species in metal enriched Cu-tailing ponds to exploit removal potential of heavy metals by naturally growing vegetation combined with other chemical and biological process.

Materials and methods

Sampling location and plant species

The Rakha Cu mines located at about 50 km away from Tatanagar by road and at a distance of 7 km from Rakha mines railway station, East Singhbhum, Jharkhand. According to Dunn and Dey [13] and Deb and Sarkar [10] the mineral distribution of Rakha ore follow the order as silicious gangue (85.12%) > chalcopyrite (8.48%) > magnetite (3.82%) > pyrite and marcasite (2.37%) > covellite, vellerite and cubanite (0.12%) > molybdenite (0.06%) > violarite, linnacite and tellurites (0.03%). The climate is humid tropical, temperature variation lies between 39°C (max) and 11°C (min). Rainy season experiences during June, July, August and September and average rainfall is 1800mm per annum. The tailing ponds studied had been abandoned from 2001, remained mostly dry, barren and growth of some metal and draught tolerant species had been observed. Five plants were collected from the selected site during winter season (December 2004) which belonged to five genera and four families (Table 1).

Plant sampling and analysis

Plant samples were collected in at least three replicates and each replicate consisted of subsamples. The herbaria were prepared for all the species and identified at Botanical Survey of India (BSI, Howrah). Individual plants were divided into two components root and shoot, carefully washed in deionized water and attention was given to washing, until visual inspection revealed that no solid particles remained adhering to the roots or shoots. Plant samples were then oven dried at 80°C overnight and ground into powder for metal concentration analysis. Metal analysis of the plant samples was carried out by acid digestion (5: 1 conc. HNO₃ and conc. HClO₄, v/v) of accurately 0.5 g of plant tissue [28] followed by measurement of metal concentration using Atomic Absorption Spectrometry (AAS).

Tailing sampling and analysis

The tailings loosely adhered to roots were gently shaken off and the rhizosphere tailings adhering to roots were separated by hand. Rhizosphere soils were then air-dried and sieved through 500-micron nylon mesh. Samples were then analysed for total metals, DTPA and CaCl₂ extractable fractions, pH, electrical conductivity (EC), organic carbon (OC), available N, exchangeable cations (Ca, Mg, Na and K) and cation exchange capacity (CEC). The pH was measured using 1:2.5 (w/v) soil/deionised water
ratio by a pH meter and the same was also used for conductivity measurement in an EC meter. OC was determined by the Walkley-Black procedure [18], CEC by ammonium acetate method, available N by the alkaline permanganate method [23]. Exchangeable cations were extracted with 1N neutral NH4OAc (pH 7) in 1:5 (w/v) soil: extracting solution ratio prior to analysis of Ca and Mg in AAS and Na and K in flame photometer. For total metal content, accurately 0.5g oven dried (80°C for overnight) tailings was digested twice with 5ml of 3:1:1 HCl: HNO3: HF and once with 5ml aqua regia in a covered teflon-beaker. Digested mass was then boiled with 1% HNO3 solution, filtered in a volumetric flask followed by addition of 1% HNO3 solution to make a final volume of 100 ml. For DTPA and CaCl2 extractable fractions oven dried tailings were extracted with 0.005 M DTPA (20g tailings extracted by 40ml solution) and 0.01005 M CaCl2 (5g tailings extracted by 50ml solution) respectively [17]. Total and extractable metal contents were measured by using Atomic Absorption Spectrometry (AAS) for Cu, Ni, Mn, Zn, Pb and Cd.

**Translocation factor**

The translocation factor for metals within a plant was determined as the ratio of [Metal]_shoot/[Metal]_root to evaluate the extent of metal translocation from roots to shoots [22,11].

**Results**

**Characteristics of tailings**

The tailings at the abandoned Cu-tailings ponds are slightly acidic with an average pH of 6.3 and EC 0.32 dSm⁻¹ (Table 2). Organic carbon content varied widely (0.12 - 1.3%) with an average value of 0.5%. The available nitrogen in tailings was very low ranging from 41 to 219 ppm with an average of 103 ppm. This macronutrient most often limits plant establishment, either because of its low content or because of a lack of the microorganisms which make it bioavailable [6]. Regarding exchangeable cations, Ca was the most abundant among alkaline-earth cations followed by Mg, K and Na. K contents in almost all the samples were lower than 0.2 cmol (+) kg⁻¹ revealing a deficiency of this element [21]. The average cation exchange capacity observed as 4.39 cmol (+) kg⁻¹. The heavy metal content in tailings was very high and varied widely (Table 3). With regard to total concentration tailings was characterised by high Cu (1008-1803 mg kg⁻¹) followed by Ni (323-656 mg kg⁻¹), Mn (142-179 mg kg⁻¹), Zn (66-82 mg kg⁻¹), Pb (26-51 mg kg⁻¹) and Cd (7-11 mg kg⁻¹). However, in the DTPA and CaCl2 fraction Cu, Ni, Mn and Zn concentration attained slightly different order. Pb and Cd in CaCl2 extracts were below the detection limit.

**Table 1: General characteristics of plant species identified at abandoned Rakha Cu-tailings pond.**

<table>
<thead>
<tr>
<th>Scientific name</th>
<th>Common name</th>
<th>Family</th>
<th>Characteristics</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ammania baccifera Linn</td>
<td>Blistering ammania</td>
<td>Lytheraceae</td>
<td>Herbaceous plant acid and vesicant, common in rice fields and has irritant properties.</td>
</tr>
<tr>
<td>Fimbristylis dichotoma (L) Vahl</td>
<td>Tall-fringe rush</td>
<td>Cyperaceae</td>
<td>Annual sedge, culms, slender, tufted, 20-50 cm tall, leaves, well developed, shorter than the stem, prefers wet conditions.</td>
</tr>
<tr>
<td>Pycreus flavidus (Retz) Koyama</td>
<td>Yellow Flat-sedge</td>
<td>Poaceae</td>
<td>Tufted, small annual, or sometimes caespitose perennial, 15-50 cm, stem 1-2 mm diam., grows along riversides and brooklets, rice fields.</td>
</tr>
</tbody>
</table>
Das – Maiti: Metal accumulation in 5 native plants growing on abandoned CU-tailings ponds

Typha latifolia Linn.
Cattail
Typhaceae
Perennial plants mostly 1.5-3 m tall, leaves erect, green, mostly 3-10 mm wide, the auricles of the leaf sheath rounded and surpassing the base of the blade, grow in marshes, shores, stream banks, ditches and margins of lakes and ponds, usually in shallow water, more tolerant of brackish or saline conditions.

Echinochola colona (L) Link
Jungle rice
Poaceae
Tufted annual 20-90cm tall, flowers in summer and winter, grows in loams, silts and clays in low places.

**Table 2. General properties of tailings.**

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Mean ± sd (min-max)</th>
</tr>
</thead>
<tbody>
<tr>
<td>pH (1: 2.5)</td>
<td>6.29 ± 0.6 (4.8 – 7.2)</td>
</tr>
<tr>
<td>EC (dS m⁻¹)</td>
<td>0.32 ± 0.14 (0.17 – 0.68)</td>
</tr>
<tr>
<td>Organic carbon (%)</td>
<td>0.5 ± 0.4 (0.12 – 1.3)</td>
</tr>
<tr>
<td>Available nitrogen (ppm)</td>
<td>103 ± 68 (41 – 219)</td>
</tr>
<tr>
<td>Ca</td>
<td>2.0 ± 1.0 (0.8 – 4.1)</td>
</tr>
<tr>
<td>Mg</td>
<td>0.8 ± 0.6 (0.14 – 2.34)</td>
</tr>
<tr>
<td>Na</td>
<td>0.09 ± 0.04 (0.02 – 0.16)</td>
</tr>
<tr>
<td>K</td>
<td>0.11 ± 0.09 (0.03 – 0.3)</td>
</tr>
<tr>
<td>CEC [cmol(+) kg⁻¹]</td>
<td>4.39 ± 2.5 (1.75 – 9.43)</td>
</tr>
</tbody>
</table>

**Table 3: The total, DTPA and CaCl₂ metal contents in tailings (mg kg⁻¹, dw).**

<table>
<thead>
<tr>
<th>Total metal</th>
<th>DTPA (0.005 M) extractable metal</th>
<th>CaCl₂ (0.01 M) extractable metal</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cu</td>
<td>1008 - 1803</td>
<td>11 – 199</td>
</tr>
<tr>
<td>Ni</td>
<td>323 – 656</td>
<td>0.3 – 25</td>
</tr>
<tr>
<td>Mn</td>
<td>142 – 179</td>
<td>0.5 – 20</td>
</tr>
<tr>
<td>Zn</td>
<td>66 – 82</td>
<td>0.3 – 1.3</td>
</tr>
<tr>
<td>Pb</td>
<td>26 – 51</td>
<td>0.7 – 4.3</td>
</tr>
<tr>
<td>Cd</td>
<td>7 - 11</td>
<td>0.3 – 0.6</td>
</tr>
</tbody>
</table>

bdl = below detection limit

**Table 4: Translocation factor (TF [Metal]_{Shoot}/[Metal]_{Root}) in plant species grown on Cu-tailings pond of Rakha mines.**

<table>
<thead>
<tr>
<th>Plant species</th>
<th>Cu</th>
<th>Ni</th>
<th>Mn</th>
<th>Zn</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ammania baccifera</td>
<td>0.06</td>
<td>0.37</td>
<td>1.70</td>
<td>0.20</td>
</tr>
<tr>
<td>Fimbristylis dichotoma</td>
<td>0.20</td>
<td>0.36</td>
<td>1.45</td>
<td>1.00</td>
</tr>
<tr>
<td>Pycreus flavidus</td>
<td>0.09</td>
<td>0.09</td>
<td>2.71</td>
<td>2.46</td>
</tr>
<tr>
<td>Typha latifolia</td>
<td>0.02</td>
<td>0.04</td>
<td>2.71</td>
<td>0.16</td>
</tr>
<tr>
<td>Echinocola cholona</td>
<td>0.12</td>
<td>0.46</td>
<td>1.30</td>
<td>0.96</td>
</tr>
</tbody>
</table>

Metal accumulation and translocation by plants

Metal accumulation in plant tissue collected from the tailing pond is shown in Figure 1. The data show that metal contents in the plant tissues varied widely among species, indicating their different capacities for metal uptake. *T. latifolia* accumulated significantly higher Ni (129 mg kg⁻¹) and Zn (125 mg kg⁻¹) in its root and higher Mn in...
both above- and underground tissues than the other species. *A. baccifera* attained an average concentration of 1613 mg kg$^{-1}$ Cu in the underground tissues, which was much higher than that of the other four species, while *F. dichotoma* accumulated significantly higher Cu and Ni in its above ground tissues than the other species. On the other hand *P. flavidus* and *E. colona* accumulated higher Zn in their aboveground tissues than others.

The translocation factor (TF), the ratio of shoot to root metals indicates internal metal transportation. Present study indicate that Cu, Ni, Zn accumulated by the plants growing on Cu-tailings, were largely retained in roots as shown by general TF values < 1 (Table 4). Exceptions occurred in *F. dichotoma* (1.0) and *P. flavidus* (2.46) for Zn. However, in all the plants TF for Mn was greater than 1 and varied with in the range of 1.3 to 2.7. Among Cu, Ni and Zn, TF for Cu was the lowest while Zn was highest in the species studied whereas in *A. baccifera* TF value for Ni was found higher than Zn. The TF values also varied among the plant species. For instance, TF values for Zn in *A. baccifera* and *F. dichotoma* were 0.2 and 1.0 respectively, although both are collected from the same tailings pond.

![Metal concentrations in roots and shoots of plants collected from abandoned Cu tailing pond of Rakha mines area.](image)

*Figure 1:* Metal concentrations in roots and shoots of plants collected from abandoned Cu tailing pond of Rakha mines area. 
AB: Ammania baccifera; FD: Fimbristylis dichotoma; PF: Prcreus flavidus; TL: Typha latifolia; EC: Echinochola colona.
Discussion

The present study reveals that plants like *A. baccifera*, *F. dichotoma*, *P. flavidus*, *T. latifolia* and *E. colona* can colonize sites with low organic carbon and macronutrients but a wide range of metal concentrations (Table 3). According to Tietjen [26] and Kabata-Pendias and Pendias [15] 100 mg kg⁻¹ Cu, 100 mg kg⁻¹ Ni based on total fraction in soil, would be considered toxic to plants. Concentration of Cu, Ni in the tailings greatly exceeds these values; therefore the five plants grown in the study area have exhibited high metal tolerance.

The general trend (Figure 1) shows that the root tissue accumulate significantly greater concentration of metals than shoots indicating high plant availability of the substrate metals as well as its limited mobility once inside the plant. Except Mn, concentration of other metals in aboveground tissues were maintained at low levels and varied in a short range compared to underground tissues (Figure 1). Such, exclusions of metals from above ground tissues has been suggested as a metal tolerant strategy adopted by different plants [24,25]. Therefore, the elevated metal concentrations in the root tissue and low translocation to the aboveground tissue in the five plants examined might suggest that they are capable of rather well balanced uptake and translocation of metals under heavily metal-enriched substrate. Cu is an essential element to plant growth but will cause toxic effects when shoots or leaves accumulate more than 20 mg kg⁻¹ [5]. In almost all the plant species examined, Cu concentration in aboveground tissues were found significantly higher than the above-mentioned value. Compared to shoots, Cu concentration in the under ground tissues were maintained at higher level and varied wide in the range of 532 -1613 mg kg⁻¹. The concentration of Ni in plants generally reflects the concentration of the element in the soil and the mobility of Ni in soil increases as the pH and CEC decreases [1]. Except *F. dichotoma* and *T. latifolia*, Ni concentration in above- and underground tissue were below or within the toxicity range of 10-100 mg kg⁻¹ [15]. The average Ni concentration in the underground tissue of the above two plants was 117 and 129 mg kg⁻¹ respectively. However, Ni in the aboveground tissue of *T. latifolia* was found less than the toxic level indicating restricted translocation of Ni from root to shoot. Existence of such a mechanism is the result of evolution rather than an innate physiological tolerance [7]. Uptake of Mn by plants is a function of the concentration of this element present in ionic form in the soil solution and the concentration present on the exchange sites of the cation exchange complex, i.e. the available or labile pool. The highest concentration of Mn was found in the aboveground tissue of *T. latifolia* whereas lowest value corresponded to *E. colona*. Although in *T. latifolia*, Mn accumulation was within the range considered as toxic by Kabata-Pendias and Pendias [15], in the other species the Mn concentrations were in the range of (20-300mg kg⁻¹) considered as normal by these authors. In comparison with the tailings and the underground tissues, the concentration of Mn in shoots were maintained at higher level and varied within range 49-341 mg kg⁻¹ in the plants investigated. It is to be pointed out that the limits of Mn toxicity were also never reached in the total Mn concentration in tailings. Zn is an essential element to all plants. Plants predominantly absorb Zn as a divalent cation which act either as a metal component of enzymes or as a functional, structural or regulatory co-factor of a large number of enzymes [1]. The concentrations of Zn in the examined plants were within the normal values (20-400 mg kg⁻¹) reported by Reeves and Baker [19]. The highest concentration of Zn (121mg kg⁻¹) in the aboveground tissue was found in *P. flavidus*. Plants like *A. baccifera* and *T. latifolia* were found to accumulate more Zn in their underground tissue than compare to shoot whereas in *F. dichotoma* and *E. colona*, Zn attained nearly equal distribution.
Present investigation reveals that all the plant species studied could tolerate excess Cu both in their root as well as shoot. A. baccifera, F. dichotoma and T. latifolia were the species accumulating Ni higher than toxic levels (100 mg kg\(^{-1}\)) in their underground tissues. The higher than toxic levels of metal concentration in some species indicate that internal metal detoxification tolerance mechanisms might exists in these naturally colonising plants, in addition to their exclusion strategies [24,25].

**Conclusions**

The present study shows that tailings in abandoned Cu-tailings ponds are slightly acidic, contain low N, Ca, Mg, K and are heavily enriched with toxic metals. Concentration of Cu, Ni and Cd in total fraction in most cases exceeded the toxicity threshold limits. The most abandoned metal in total fraction of tailings was Cu followed by Ni with comparatively lower amount of Mn, Zn, Pb and Cd. Despite the chemical limitation many plants can colonize this heavy metal enriched tailings pond and are found to accumulate a wide range of toxic metals. Low metal translocation into shoots indicates that metal tolerance in natural vegetation grown on tailings may mainly depend on the metal exclusion ability of underground tissue. Metal accumulation higher than toxic level in some plants indicates that they might have developed some internal detoxification metal tolerance mechanisms. These species therefore, may be considered as tolerant to some elements and hence can be used for detoxification of metal enriched Cu-tailings.

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