

Phytotoxicity and remediation of heavy metals by fibrous root grass (sorghum)

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ABSTRACT

Objectives: To determine the uptake and effects of heavy metals (Cd, Cu, Ni, Pb and Zn) on seed germination, growth of fibrous roots, root/shoot growth and biomass of grass (sorghum) grown in soil - vermicompost media (3:1).

Methodology and results: Vermicompost developed using vermiculture biotechnology from vegetable market waste was characterized and found to have high concentrations of the nutrient elements Ca, Zn, Cu, Mg, Fe and Mn. The vermicompost was used as a natural fertilizer for phytoremediation studies of heavy metals. The selected heavy metals were dosed at concentrations from 0, 5, 10, 20, 40 and 50 ppm separately in the soil - vermicompost media (3:1) in pot experiments. The phytotoxic effect of heavy metals on the growth of sorghum and physicochemical parameters of soil-vermicompost media were measured. The uptake of heavy metals in plant samples were analyzed by atomic absorption spectrophotometer. Plant growth was adversely affected by heavy metals at the higher concentration of 40 and 50 ppm, while lower concentrations (5 to 20 ppm) stimulated shoot growth and increased plant biomass. Heavy metals were efficiently taken up mainly by roots of sorghum plant at all the evaluated concentrations of 5, 10, 20, 40 and 50 ppm.

Conclusions and application of findings: Vermicompost from vegetable waste has high nutrient contents and therefore it can be used as a natural fertilizer to increase growth of plants that play a role in phytoremediation. Although heavy metals at 40 and 50 ppm reduced seed germination, plants germinated and grew efficiently when Zn was available. Vermicompost application as a natural fertilizer or soil amendment would enhance sorghum plant growth and enhance phytoremediation of heavy metals from contaminated environments.

Key words: heavy metals, sorghum, phytoremediation, vermicompost.

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INTRODUCTION

In the environment, metals come from the natural weathering processes of the Earth's crust, soil erosion, mining and industrial discharge, fertilizers, urban runoff, sewage effluent, air pollution, pest and disease control agents and other sources (Fargasova, 1994). Many metals found in the environment have no nutritional value. The contamination of agricultural environment by trace metals is of growing worldwide concern because of the transportation of trace metals in the food chain (Peng *et al.*, 2006).

Some of the currently employed processes for cleaning up heavy metal pollution are expensive and environmentally destructive. Unlike organic compounds, metals cannot degrade, and therefore effective cleanup requires their immobilization to reduce or remove toxicity. In recent years, scientists and engineers have started to generate cost effective technologies that include use of microorganisms/ biomass or live plants to clean polluted areas (Qui *et al.*, 2006).

Phytoremediation is an emerging technology for cleaning up contaminated sites, which is cost effective, and has aesthetic advantages and long term applicability (Su & Wong, 2004; Boonyapookana et al., 2005). The technology involves efficient use of plants to remove, detoxify or immobilize environmental contaminants in a growth matrix (soil, water or sediments) through the natural, biological, chemical or physical activities and processes of the plants (Ciura et al., 2005). It is best applied at sites with shallow contamination of organic, nutrient or metal pollutants that are amenable to one of five applications, i.e. phytotransformation, rhizosphere bioremediation, phytostabilization, phytoextraction, rhizofiltration (Schnoor, 1997).

Monocotyledonous plant species are usually more tolerant to metals than dicotyledonous species (Lombi et al., 2001). Several plants, e.g. Indian mustard (Brassica juncea), corn (Zea mays L.), sunflower (Helianthus annuus L.) and fescue grass show high tolerance to heavy metals and are therefore used in phytoremediation studies (Schmidt, 2003: Charlier al., 2005). et For grasses are the phytoremediation, most commonly evaluated plants (Ebbs & Kochian, 1998; Shu et al., 2002). The large surface area of their fibrous roots and their intensive penetration of soil reduces leaching, runoff, and erosion via stabilization of soil, and offers advantages for phytoremediation. Some of the plant material may be used for non-food purposes or it can be ashed for recycling of the metals or to be disposed in landfills (Angel & Linacre, 2005).

Uptake of contaminants from the soil by plants occurs primarily through the root system in which the principle mechanisms of preventing contaminant toxicity are found. The root system provides an enormous surface area that absorbs and accumulates the water and nutrients that are essential for growth, but also absorbs other nonessential contaminants (Arthur et al., 2005). One of the mechanisms by which uptake of metal occurs in the roots may include binding of the positively charged toxic metal ions to negative charges in the cell wall (Gothberg et al., 2004). Plants that accumulate metals are suitable for extracting environmentally important toxic metals, e.g Pb, Cd, Cu, and Ni from the soil. When such plants are harvested and removed from the contaminated sites, the toxic metals are removed at reduced costs and with little loss of top soil (Nandakumar et al., 1995).

It has been observed that prolonged and lavish application of chemical fertilizers reduces productivity of land and crops due to dependence on periodic inputs. Factories that manufacture fertilizers also contribute to pollution of the air, land and water. Natural fertilizers, e.g. vermicompost can provide superior nutrient value to soils, and thereby improve yields and quality of crop produce.

Vermicomposting of vegetable waste with earthworm (Eisenia foetida) develops the waste into a natural fertilizer (Maharashtra Nature Park Bulletin, 2003). The vermicompost has a high nutrient value, increases fertility of soil and maintains soil health (Suthar et al., 2005). Application of normal compost in contaminated areas improves soil fertility and physical properties (Zheljazkov & Warman, 2004). Due to increased growth of plants with more biomass attained, more metals can potentially be taken up from the contaminated soils. The use of vermicompost developed from vegetable waste using vermiculture biotechnology enhances the conditions for phytoremediation (Elcock & Martens, 1995).

In the work presented here we evaluated the application of vermicompost developed from vegetable waste in soil contaminated heavy with metals for phytoremediation studies. The study determined the effects of heavy metals on seed germination, plant arowth. biomass accumulation and examined the uptake of metals by sorghum.

MATERIALS AND METHODS

Soil sampling and characterization: Soil was collected from a depth of about 0-15 cm along the banks of Surva River, Palghar (located 100 km away from Mumbai). Stones and plant tissues were carefully removed from the soil prior to drying in the laboratory. The soil was screened through 2 mm stainless steel sieve, and stored in a plastic bag at room temperature (28-30°C) until use. Concentrations of Pb, Zn, Cu, Ni, and Cd were determined by atomic absorption spectrophotometer (APHA, 1998) while the physicochemical parameters were measured by standard methods (Table1). Soil texture was determined by the Bouyoucos hydrometer method and the moisture content calculated by the weight difference before and after drying to a constant weight at 105°C. The pH and electrical conductivity (EC) were measured after 20 min of vigorous mixing samples in 1: 2.5 (solid:deionized water), using digital meters [Elico, Model LI-120] with a combination pH electrode and a 1-cm platinum conductivity cell, respectively. Total nitrogen and total phosphorus were determined according to the standard methods of the American Public Health Association (1998). Cation exchange capacity was determined after extraction with ammonium acetate at pH 7.0, and the organic carbon was determined using the Walkley-Black method (Jackson, 1973).

Vermicompost production: The vermicompost was produced from vegetable waste (cabbage, French bean, cauliflower, lady finger, spinach, carrot and radish) collected from local market. Vermicomposting was carried out in rectangular perforated (sides and bottom) plastic boxes of dimensions [2(l) x 1 (b) x 1 (h)] ft, with working volume of 15-kgs. Each box was equipped with aeration and stirrer system for turning of composting materials. The turning of composting materials was done twice a day for a period of 15 days. The moisture content, temperature and pH of composting materials were maintained at 55 to 65%, 30 to 33°C and 6.8 to 7.2, respectively. Exotic varieties of earthworms (Eisenia foetida) were obtained from Maharashtra Nature Park, Mahim Dharavi (Mumbai) and about 1/2 Kg (200 to 250 earthworms) was used in vermicomposting.

The physicochemical parameters were measured during vermicomposting as described above for soil analysis. After 2.5 months, vermicompost was collected, air dried, sieved (2-mm) and a portion of it was taken for nutrient analysis in order to determine its suitability as biofertilizer. The nutrients in dried vermicompost sample were digested with concentrated nitric acid and 30% hydrogen peroxide and then quantified by an atomic absorption spectrophotometer [AAS, Perkin Elmer] (APHA, 1998). Nutrient and trace elements content in the vermicompost are presented in table 2.

Phytoremediation experiments: Pot culture experiments were conducted in the green house to study the effect of heavy metals on seed germination, root growth, shoot growth and phytoremediation (metal uptake) by fibrous root grass (Sorghum). The growth medium in the pots consisted of alluvial soil and vermicompost (3:1). The growth medium was amended with the heavy metals: Cd as Cd $(NO_3).4H_2O$; Cu as CuSO₄.5H₂O; Ni as Ni $(NO_3)_2$; Pb as Pb (NO₃) ₂ and Zn as Zn (NO₃) ₂.6H₂O. Each heavy metal was applied at 0, 5, 10, 20, 40 and 50 ppm. Sorghum seeds were obtained from Ratanshi Agro-Hortitech (Byculla, Mumbai) and immersed in 3% (v/v) formaldehyde solution for 5 minutes, followed by washing several times with distilled water to remove fungal contaminants.

To determine the effect of heavy metals on seed germination 30 seeds of uniform size were placed in each Petri dish containing soilvermicompost media amended with heavy metals (Cd, Cu, Ni, Pb and Zn) at 0, 5, 10, 20, 40 and 50 ppm. The Petri dish lids were left partially open and kept in the dark to observe germination. The seeds were considered germinated when the radicles emerged. Ten plants were grown in each small plastic pots containing 250 g soil-vermicompost media amended with heavy metals (Cd, Cu, Ni, Pb and Zn) at 0, 5, 10, 20, 40 and 50 ppm for studying effect of metals on shoot and root growth. Similarly, in a separate set ten plants were grown in each 2 kg capacity plastic pots for phytoremediation study.

Soil moisture content was adjusted regularly using deionized water to about 60% of water-holding capacity. To prevent loss of nutrients and trace elements out of the pots, plastic trays were placed under each pot and the leachates collected were put back in the respective pots. Each treatment was replicated thrice. The seeds were set under 12/12 hrs light/dark cycle and temperatures of 30°C during the day and 27°C during the night, at 75% average relative humidity. The seedlings were removed after four days for determination of germination rate and shoot/root length was evaluated after two weeks.

For the phytoremediation study plants were harvested after 10 weeks and each plant was separated into roots and shoots. The plant samples were washed with distilled water and dried in an oven at 70°C for 3 days, and weighed to determine the dry weight of biomass. The samples were stored in

brown paper bags until digestion with concentrated nitric acid and 30% hydrogen peroxide and determination of heavy metal content using an atomic absorption spectrophotometer [AAS, Perkin Elmer]

RESULTS AND DISCUSSION

The sandy loam texture affected soil properties including its water supplying power, rate of water intake, aeration, fertility and ease of tillage (Table 1). The soil pH of 7.2 is within the recommended value for proper growth and efficient uptake of nutrients and compounds from soil. Macronutrients including metals were also present in substantial amount but there was no history of heavy metal (Cd, Ni, and Pb) contamination.

Table 1:	Physicochemical properties of experimental
soil.	

Soil parameter	Value
Clay %	25.9 <u>+</u> 1.8
Silt %	21.7 <u>+</u> 2.5
Sand %	50.4 + 2.8
рH	7.20 + 0.1
Organic matter %	0.80 + 0.045
Nitrogen %	0.05 + 0.01
CEC [*] c mol/ 100 gm soil	11.27 + 0.76
EC [‡] dS ⁻¹	1.10 + 0.1
Potassium mg/kg	22.7 3 + 2.63
WHC** %	62 + 4.0
Moisture Content %	34 <u>+</u> 1.8
Heavy metal (ppm)	
Cu	3.6 <u>+</u> 0.5
Cd, Ni, Pb	ND
7n	12 + 1 5

† Values are averages of three replicates ± S.D.; *CEC= Cation exchange capacity; [‡] EC= Electrical onductivity; ^{**}WHC= Water Holding Capacity, ND= Not Detected.

Upon characterization, the vermicompost was found to have high concentration of nutrients such as Ca, Zn, Cu, Mg, Fe, and Mn (Table 2). The vermicompost was thus ascertained to be a rich source of beneficial microorganisms and nutrients, as reported by Paul (2000) and and can also be used as a soil conditioner (Hattenschwile & Gaser, 2005). (APHA, 1998). Data were analyzed for mean and standard deviation (X±S.D.) using standard statistical methods (Mahajan, 1997).

Germination of sorghum seeds decreased with increase in heavy metal concentrations (Table 3). The metals Cd, Ni and Pb at 5 and 10 ppm had very low toxicity on seeds, and even copper at the same doses increased seed germination. However, seed germination decreased significantly when exposed to Cu, Ni, Pb and Cd at 20, 40 and 50 ppm. Delayed germination was also observed with all metals above 40 ppm concentration. In this study, Zn was the only metal that caused a significant increase in seed germination even at 50 ppm (compared to the control).

Table 2: Chemical properties and nutrient content of
vermicompost from vegetable waste.

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Parameter	Value	
рН	6.8 <u>+</u> 0.173	
EC [‡] dS m ⁻¹	10.55 <u>+</u> 0.01	
Total C %	13.5 <u>+</u> 0.7	
Total N %	1.33 <u>+</u> 0.015	
Available P %	0.47 <u>+</u> 0.09	
Sodium mg /100gm	54.68 <u>+</u> 9.44	
Magnesium mg/100gm	832.48 <u>+</u> 22.48	
Iron mg/100gm	746.26 <u>+</u> 23.39	
Zinc mg/100gm	16.19 <u>+</u> 0.55	
Manganese mg/100gm	53.86 <u>+</u> 2.84	
Copper mg/100gm	5.16 <u>+</u> 0.36	

† Values are averages of three replicates ± S.D.; ‡ EC= Electrical conductivity.

In phytoremediation, metal removal from contaminated media starts at the first stage of the germination process when the seed absorbs water along with large quantities of metal ions. Once accumulated, ions enter the root where they can be stored or exported to the shoot via the transpiration stream (Ximenez-Embun *et al.*, 2001). In this study, it is likely that the absorbed heavy metal ions inhibited the germination and growth of sorghum seedlings, as also observed by Ganesh *et al.* (2006). Claire *et al.* (1991) obtained similar results in a study using Nickle and other heavy metals on cabbage, lettuce, millet, radish, turnip and wheat.

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Table 3: Seed germination and root and shoot length of fibrous root grass (Sorghum) after two weeks of exposure to heavy metals.

Metal	Dose (ppm)	Germination rate (%)	Root length (cm)	Shoot length (cm)
	0	75 <u>+</u> 3	6.50 <u>+</u> 0.10	13.18 <u>+</u> 1.86
Cd	5	73 <u>+</u> 4	5.27 <u>+</u> 0.294	14.40 <u>+</u> 1.70
	10	68 <u>+</u> 2.5	5.00 <u>+</u> 0.70	12.81 <u>+</u> 1.69
	20	61 <u>+</u> 3	4.24 <u>+</u> 0.74	11.20 <u>+</u> 1.01
	40	50 <u>+</u> 3	3.15 <u>+</u> 0.35	9.100 <u>+</u> 0.85
	50	43 <u>+</u> 4.6	2.10 <u>+</u> 0.25	7.200 <u>+</u> 1.35
	5	77 <u>+</u> 4.7	6.82 <u>+</u> 0.71	14.92 <u>+</u> 1.74
Cu	10	80 <u>+</u> 6	7.13 <u>+</u> 0.67	16.42 <u>+</u> 1.25
	20	70 <u>+</u> 2.4	4.44 <u>+</u> 0.34	14.75 <u>+</u> 0.75
	40	66 <u>+</u> 4	3.63 <u>+</u> 0.53	12.12 <u>+</u> 1.62
	50	60 <u>+</u> 3	3.05 <u>+</u> 0.75	9.930 <u>+</u> 1.13
	5	74 <u>+</u> 3	6.70 <u>+</u> 0.60	14.90 <u>+</u> 1.65
Ni	10	70 <u>+</u> 3	6.90 <u>+</u> 0.70	14.48 <u>+</u> 1.38
	20	63 <u>+</u> 5	5.10 <u>+</u> 0.80	13.70 <u>+</u> 2.05
	40	56 <u>+</u> 3	4.20 <u>+</u> 0.50	10.20 <u>+</u> 1.70
	50	50 <u>+</u> 4	3.10 <u>+</u> 0.80	9.500 <u>+</u> 1.55
	5	72 <u>+</u> 4	6.60 <u>+</u> 1.15	16.70 <u>+</u> 1.75
Pb	10	69 <u>+</u> 3	6.80 <u>+</u> 0.65	15.60 <u>+</u> 1.25
	20	65 <u>+</u> 4	6.10 <u>+</u> 0.60	15.20 <u>+</u> 1.23
	40	60 <u>+</u> 4	4.10 <u>+</u> 0.55	10.60 <u>+</u> 1.45
	50	58 <u>+</u> 6	3.80 <u>+</u> 0.65	9.200 <u>+</u> 1.65
	5	76 <u>+</u> 1	7.00 <u>+</u> 0.80	16.40 <u>+</u> 1.22
Zn	10	80 <u>+</u> 4	7.80 <u>+</u> 0.63	16.90 <u>+</u> 1.80
	20	86 <u>+</u> 3	9.40 <u>+</u> 0.55	17.44 <u>+</u> 1.29
	40	92 <u>+</u> 5	10.9 <u>+</u> 0.80	19.80 <u>+</u> 1.85
	50	97 <u>+</u> 2	12.2 <u>+</u> 1.08	21.00 <u>+</u> 1.40

 \dagger Values are averages of three replicates \pm S.D.

Presence of heavy metals in the soil-vermicompost medium decreased plant root length and caused stunted growth of roots (Table 3). Metal toxicity symptoms on roots included browning, reduced number of roots hair, and reduced growth. In comparison to the control treatment (without heavy metals Cd, Pb, Cu, and Ni), plant roots were healthy and normal. The color of the roots receiving higher heavy metals treatment (40 and 50 ppm), except Zn, changed gradually over time from creamy white to dark brown, which indicates intense suberification.

There was a reduction in the formation of secondary roots and the number of root hairs by Cadmium metal at 40 and 50 ppm. Plants treated at lower concentrations were not significantly affected by the metals. All Zinc concentrations increased root growth.

Lateral roots were observed in plants grown in all treatments. Except Zn, all the tested metals

demonstrated concentration dependent inhibition of root growth at higher concentrations.

One of the possible explanations for observed effect of toxic metals on roots might be that roots are the specialized absorptive organs and thus they are affected more due to earlier and more intense accumulation of heavy metals than any of the other organs This could be one of the reasons for using root length as a measure for determining heavy metal-tolerance ability of plants (Xiong, 1998). Oncel *et al.* (2000) obtained results that are similar to those of this study using cadmium in wheat seedlings. According to Chaignon and Hinsinger (2003), higher concentrations of copper can inhibit root growth before shoot growth and can accumulate in the roots without any significant increase in its content in the aerial parts.

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Table 4: Biomass of sorghum after 10 weeks of growth in soil-vermicompost artificially contaminated with heavy metals.

Metal	Dose (ppm)	Root Dry Weight (g)	Shoot Dry Weight (g)
	0	0.625 <u>+</u> 0.025	0.910 <u>+</u> 0.080
Cd	5	0.575 <u>+</u> 0.063	0.897 <u>+</u> 0.085
	10	0.807 <u>+</u> 0.086	1.005 <u>+</u> 0.096
	20	0.937 <u>+</u> 0.098	1.220 <u>+</u> 0.090
	40	0.458 <u>+</u> 0.087	0.799 <u>+</u> 0.087
	50	0.319 <u>+</u> 0.022	0.498 <u>+</u> 0.071
	5	0.638 <u>+</u> 0.066	1.220 <u>+</u> 0.040
Cu	10	0.726 <u>+</u> 0.086	1.174 <u>+</u> 0.072
	20	0.974 <u>+</u> 0.064	1.376 <u>+</u> 0.078
	40	0.798 <u>+</u> 0.075	1.132 <u>+</u> 0.120
	50	0.703 <u>+</u> 0.079	1.067 <u>+</u> 0.042
	5	0.670 <u>+</u> 0.090	0.968 <u>+</u> 0.056
Ni	10	0.723 <u>+</u> 0.035	1.037 <u>+</u> 0.064
	20	0.879 <u>+</u> 0.058	1.352 <u>+</u> 0.083
	40	0.501 <u>+</u> 0.067	0.829 <u>+</u> 0.031
	50	0.431 <u>+</u> 0.064	0.651 <u>+</u> 0.064
	5	0.619 <u>+</u> 0.063	1.111 <u>+</u> 0.039
Pb	10	0.834 <u>+</u> 0.066	1.123 <u>+</u> 0.062
	20	0.961 <u>+</u> 0.070	1.276 <u>+</u> 0.075
	40	0.749 <u>+</u> 0.040	1.183 <u>+</u> 1.071
	50	0.663 <u>+</u> 0.051	0.939 <u>+</u> 0.050
	5	0.567 <u>+</u> 0.055	0.938 <u>+</u> 0.056
Zn	10	0.860 <u>+</u> 0.048	0.997 <u>+</u> 0.085
	20	1.140 <u>+</u> 0.580	1.380 <u>+</u> 0.080
	40	1.298 <u>+</u> 0.156	1.779 <u>+</u> 0.058
	50	1.437 <u>+</u> 0.105	2.017 <u>+</u> 0.095

 \dagger Values are averages of three replicates \pm S.D.

All the plants appeared to be healthy and plant shoot growth was stimulated by concentrations of 5, 10 or 20 ppm of the heavy metals (Table 3). These results indicate that low concentrations of Cd, Cu, Ni, and Pb have micronutrient-like effects on the sorghum plants. However, at 40 and 50 ppm gradual reduction of shoot growth was observed as compared to the control. These symptoms were more common in Cadmium treatments. No signs of toxicity were observed on the shoot with high levels of Zn. Ormrod et al. (1986) reported that Nickel caused stunted and deformed growth of shoot with symptoms of chlorosis. Wenger et al. (2003) reported that the critical toxicity level of Cu in the shoots of crop plants is greater than 20 to 30 mg kg⁻¹. These data corresponded with those of Oncel et al. (2000), who found that Cd reduces chlorophyll a and b in wheat. High amounts of Cu, Zn, Pb, Ni, Cr and Cd in wheat *cv.* Vergina resulted in depressed shoot growth (Athar & Ahmad, 2002).

At 10 weeks after germination plant biomass showed an increasing trend as the concentration of heavy metals increased from 5, 10, and 20 ppm levels (Table 4). However, plant biomass decreased as the concentration of Cd, Cu, Pb and Ni increased to 40 ppm and above. Zn had significant positive effect on plant biomass gain as compared to the control. Nwosu *et al.* (1995) reported that the mean plant biomass decreased in both lettuce and radish, as the concentration of Cd and Pb in soil increased. Grifferty and Barrington (2000) showed that the increased Zn concentration from 25 to 50 mg/kg had a significantly positive effect on dry biomass yield of wheat plants (*Triticum aestivum L.*).

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Table 5: Concentration of heavy metals in roots and shoots of sorghum plants grown in artificially contaminated soil-vermicompost medium.

Metal	Dose (ppm)	Roots Metal uptake (ppm)	Shoots Metal uptake (ppm)
Cd	5	0.915 <u>+</u> 0.065	0.197 <u>+</u> 0.076
	10	0.927 + 0.060	0.237 + 0.048
	20	2.782 <u>+</u> 0.087	0.796 <u>+</u> 0.071
	40	4.510 <u>+</u> 0.070	2.014 <u>+</u> 0.104
	50	7.108 <u>+</u> 0.093	3.260 <u>+</u> 0.148
Cu	5	1.669 <u>+</u> 0.140	0.539 <u>+</u> 0.052
	10	2.507 <u>+</u> 0.127	1.063 <u>+</u> 0.058
	20	2.590 <u>+</u> 0.120	1.590 <u>+</u> 0.119
	40	7.350 <u>+</u> 0.149	1.828 <u>+</u> 0.137
	50	12.66 <u>+</u> 0.140	3.450 <u>+</u> 0.131
Ni	5	1.230 <u>+</u> 0.080	0.603 <u>+</u> 0.086
	10	1.813 <u>+</u> 0.198	0.817 <u>+</u> 0.090
	20	5.147 <u>+</u> 0.147	1.003 <u>+</u> 0.086
	40	5.790 <u>+</u> 0.176	1.319 <u>+</u> 0.083
	50	6.274 <u>+</u> 0.151	1.982 <u>+</u> 0.112
Pb	5	0.603 <u>+</u> 0.024	0.105 <u>+</u> 0.020
	10	0.918 <u>+</u> 0.054	0.237 <u>+</u> 0.016
	20	2.206 <u>+</u> 0.081	0.471 <u>+</u> 0.023
	40	3.914 <u>+</u> 0.114	0.534 <u>+</u> 0.034
	50	6.078 <u>+</u> 0.134	0.812 <u>+</u> 0.086
Zn	5	3.100 <u>+</u> 0.089	1.820 <u>+</u> 0.055
	10	4.812 <u>+</u> 0.127	2.488 <u>+</u> 0.063
	20	9.454 <u>+</u> 0.137	3.012 <u>+</u> 0.120
	40	12.18 <u>+</u> 0.187	5.693 <u>+</u> 0.075
	50	12.61 <u>+</u> 0.138	5.864 <u>+</u> 0.092

† Values are averages of three replicates ± S.D.

The metal concentration in the tissues of the sorghum plants increased as the concentration of metals in the soil- vermicompos medium increased (Table 5). Zn and Cu accumulated in the largest proportions in root tissue. The heavy metals were taken up by the sorghum plants in the following order: Zn>Cu>Cd>Ni>Pb. Several studies have demonstrated that the concentration of metals in plant tissue is a function of the metal content in the growing environment (Grifferty & Barrington, 2000). Bennett et al. (2003) observed that the Indian mustard translocates heavy metals to the shoot, while grasses tend to accumulate them in the root. The uptake of 8.2mg of Pb by root of sorghum sp has been reported (Nandkumar et al., 1995)., indicating that lead might be found to the outer surface of plant roots, as crystalline or amorphous deposits, and could also be deposited in the cell walls or in might explain vescicles. This the hiaher concentrations of Pb in roots of sorghum plants in our study.

A ultrastructural study using transmission electron microscopy revealed the retention of unchelated Pb mainly in cell wall of roots, particularly around intercellular spaces (Wenger *et al.*, 2003). Grasses are known to concentrate heavy metals in the roots, with only very low translocation of heavy metals to the shoot (Speir *et al.*, 2003; Bennett *et al.*, 2003). Alloway (1995) reported that Alyssum species, which are naturally adapted to serpentine soils, can accumulate over 2% Ni. The application of peat and manure in contaminated soil increased Cu, Zn, and Ni accumulation by wheat (Schmidt, 2003). Organic matter in soil could effectively increase the activity of metals in soil and improve metal mobility and distribution in soil.

In the present research work, sorghum plants efficiently took up five different heavy metals from soil mainly by roots. The order of uptake of heavy metals was: Zn>Cu>Cd>Ni>Pb. The large surface area of fibrous roots of sorghum and intensive penetration of roots into the soil reduces leaching via stabilization of soil. The plant is further

capable of immobilizing and concentrating heavy metals in the roots. Sorghum grass, with its distinctive characteristics like higher biomass, fast growth and strong fibrous root system is thus proven to be an ideal plant for phytoremediation, which is an economical, effective, pleasing, and environmentally

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