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## Full Length Research Paper

# Bioaccumulation of heavy metals by *Dyera costulata* cultivated in sewage sludge contaminated soil

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High concentrations of heavy metals are harmful to plants, animals and humans and their potential accumulation in human tissues and bio-magnification through the food chain cause serious health hazards. An experiment was conducted in the glasshouse to evaluate the potential of *Dyera costulata* as a bioaccumulator to absorb heavy metals from sewage sludge contaminated soils. *D. costulata* seedlings were planted in the following growth media: T<sub>0</sub> (control soil), T<sub>1</sub> (100% sludge), T<sub>2</sub> (80% sludge + 20% soil), T<sub>3</sub> (60% sludge + 40% soil), T<sub>4</sub> (40% sludge + 60% soil) and T<sub>5</sub> (20% sludge + 80% soil). T<sub>4</sub> showed the best growth performance in terms of height, basal diameter and number of leaves. The maximum reduction of Cd, Cr and Pb was found in the 100% sludge treatment. Zn, Cd, Ni and Cr were highly concentrated in the leaves, while Pb accumulated mainly in the stems. *D. costulata* showed high potential to retain high amounts of Zn, Ni and Cr in the leaves and Pb in the stems. The species had high translocation factor (TF) and low bioconcentration factor (BCF) values in the soil at higher metal concentrations as well as it was able to tolerate and accumulate high concentrations of Zn, Cd, Ni, Cr and Pb. It means that, this species is a good accumulator of heavy metals and can be considered as a potential bioaccumulator species.

**Key words:** Phytoremediation, *Dyera costulata*, heavy metals, sewage sludge.

## INTRODUCTION

Heavy metal pollution has become one of the most widespread and serious environmental problems nowadays. In addition, soil and water contamination with toxic metal pose a major environmental and human health problem and has been drawing considerable public attention over the last decades. Human activities have introduced numerous potential hazardous trace elements into the environment since the industrial growth. Other sources of heavy metal contamination associated with agricultural soil are sewage sludge, fertilizer and pesticides (Alloway and Ayres, 1993).

The concentrations of heavy metals in soils are related with the biological and geochemical cycles and are influenced by anthropogenic activities, such as agricultural practices, transport, industrial activities and waste disposal (Lund, 1990). However, it is a well known fact that, metals are present in soils in different chemical

forms, which influence their reactivity and hence, their mobility and bioavailability.

Heavy metals, unlike organic contaminants, are generally immutable, not degradable and persistent in soil (Adriano et al., 2004). Soils have a natural capacity to attenuate the bioavailability and the movement of metals ions by different mechanisms including precipitation, adsorption process and redox reactions. When the concentrations of heavy metals become too high to allow the soil to limit their potential effects contaminants cannot be removed, resulting in serious contamination of agricultural products or ground water.

Sewage sludge from municipal wastewater treatment plants contains high amount of nutrients like N, P, Cu, Zn, Mo, B, Fe, Mg and Ca and organic matter which are beneficial to soil fertility and productivity. In contrast, it also contains high concentration of hazardous heavy metals like Pb, Cr, Cu, Zn and Ni. In Malaysia, sewage sludge is mainly produced from domestic and light industrial areas. It has been estimated that about 3 million metric tons (wet basis) of sewage sludge is produced annually. The total cost of managing sewage

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sludge is estimated at RM 1 billion per year (Kadir and Mohd, 1998). Sewage sludge contains heavy metals and restricts the use of sludge recycling to agricultural land. Heavy metals pose threats to soil quality and human health and high concentrations of heavy metal are harmful to plants, animals and humans. The frequent environmental problems are related with plant productivity, food quality and human health (Alloway, 1990). Their potential accumulation in human tissues and bio-magnification through the food-chain can cause DNA damage and carcinogenic effects. Therefore, studies of effective methods for heavy metal removal from sludge are very important in order to minimize future health risk during application (Lasheen et al., 2000).

Currently, remediation methods for heavy metal removal from soil and sludge are expensive and difficult. Recently, efforts have been directed toward finding remediation strategies that are less expensive and less damaging to soil properties than current approaches. One of those strategies is phytoextraction in which plants absorb heavy metals from the soil or sludge, followed by harvesting the aboveground biomass. Harvested material is then disposed in a landfill or treated to recover the metals (Cooper et al., 1999).

Plants have many properties that make them ideally suited to clean polluted soil, water and air, in a process call phytoremediation. Malaysia is considered one of the world's 12 leading mega biodiversity countries with a high diversity of flora and fauna. There are several natural products such as medicinal plants that can be used as phytoremediators. Phytoremediation has been recognized as an alternative method for the removal of organic pollutants from soil in comparison to other physicochemical remediation technologies due to its low cost and suitability for applications that require sustenance and low maintenance.

Phytoremediation is defined as the use of green plants to remove pollutants from the environment or to render them harmless (Cunningham et al., 1995). Trees, for instance, absorb mobile metals and other contaminants up into their aboveground parts, which can be removed by harvesting/coppicing (phytoextraction). At the same time, trees can decrease metal mobility, toxicity and dispersion by root growth (phytostabilisation). Some works of phytoremediation on polluted soils have been done but phytoremediation with *Dyera costulata* has not been reported yet. Therefore, this study was initiated with the following objectives: (1) to determine heavy metal concentrations in *costulata* plant parts and (2) to quantify the heavy metal concentrations in the growth medium before planting and after harvesting period.

## MATERIALS AND METHODS

### Site description and planting materials

The experiment was conducted at the glasshouse, Universiti Putra

Malaysia, at 4°06' N latitude and 101°16' E longitude. The average temperature in the greenhouse was 27, 36 and 32°C for morning, afternoon and evening, respectively. The growth media prepared using soil mixing with different levels of sludge was: T<sub>0</sub> (control, soil), T<sub>1</sub> (100% sludge), T<sub>2</sub> (80% sludge + 20% soil), T<sub>3</sub> (60% sludge + 40% soil), T<sub>4</sub> (40% sludge + 60% soil) and T<sub>5</sub> (20% sludge + 80% soil). A completely randomized design (CRD) was used with four replications. *D. costulata* was used as the test plant. Healthy saplings of three months old and similar in form were selected for this study. Before planting, a seedling was tested for toxicity in 100% sewage sludge without soil for about one week. 24 plants were used to measure growth parameters including basal diameter, number of leaves and height at certain interval of time. The height was taken by using a ruler. Basal diameter was measured by using caliper. The growth parameters were measured twice in a month.

### Plant and soil sampling and chemical analysis

Soil samples were collected from each pot before planting and after harvest and kept in a standard plastic container and air-dried before physico-chemical analysis. For analysis of heavy metals, 1.0 g dried plant sample and 20 ml aqua regia solution (mixture of concentrated HNO<sub>3</sub> and HCl in a ratio of 3:1) was taken into the digestion tube and digestion was completed with 80 to 120°C for 3 h. After filtering the digestion into 100 ml beaker, the solution was ready for analysis and ICP-MS (inductively couple plasma mass spectrometry) method (Sahoo et al., 2009) was applied for analyzing the concentrations of heavy metals in the planting medium, plant parts and sample solutions. Particle size distribution was analyzed by pipette gravimetric method and the texture was determined using USDA textural triangle. Soil pH and total carbon were determined by using glass electrode pH meter and loss on ignition method, respectively.

### Plant biomass measurement

Plant biomass was measured separately according to leaves, stems and roots and calculated. The loss in weight upon drying is the weight originally present. The moisture content of the sample was calculated using the following equation:

$$\% \text{ Moisture} = \frac{\text{Wt. Wet sample} - \text{wt. dry sample}}{\text{wt. dry sample}} \times 100 \quad (1)$$

### Determination of bioconcentration factor and translocation factor

The plant's ability to accumulate metals from soils and translocate metals from roots to shoots can be estimated using the bioconcentration factor (BCF) and translocation factor (TF), respectively. BCF and TF factors can be calculated as follows:

$$\text{BCF} = \frac{\text{Metal concentration in root}}{\text{Metal concentration in soil}} \times 100 \quad (2)$$

$$\text{TF} = \frac{\text{Metal concentration in aerial part}}{\text{Metal concentration in root}} \times 100 \quad (3)$$

**Table 1.** pH and total-C (%) in growth media as influenced by different treatments.

Treat	pH		Total-C (%)	
	Before planting	After harvest	Before planting	After harvest
T0	4.23	5.01	1.79	4.46
T1	3.76	5.10	16.4	14.1
T2	4.43	5.54	2.16	6.64
T3	4.36	5.17	1.84	8.28
T4	4.23	5.39	1.84	5.77
T5	4.38	5.01	2.01	1.60
CV (%)	5.33	4.00	11.0	9.11
LSD (0.05)	0.34	0.30	0.76	0.93

T<sub>0</sub> = Control; T<sub>1</sub> = 100% sludge; T<sub>2</sub> = 80% sludge + 20% soil; T<sub>3</sub> = 60% sludge + 40% soil; T<sub>4</sub> = 40% sludge + 60% soil; T<sub>5</sub> = 20% sludge + 80% soil.

### Statistical analysis

The analysis of variance for growth and heavy metals concentrations (in soil, sludge and plant parts) were done following the ANOVA test and the mean values were adjusted by DMRT (P= 0.05) method (Steel and Torrie, 1960). Comparison using t-test was also done to detect any significant differences between before planting and after harvest. Computation and preparation of graphs were done by using Microsoft excel 2003 program.

## RESULTS AND DISCUSSION

### Characteristics of the growth media

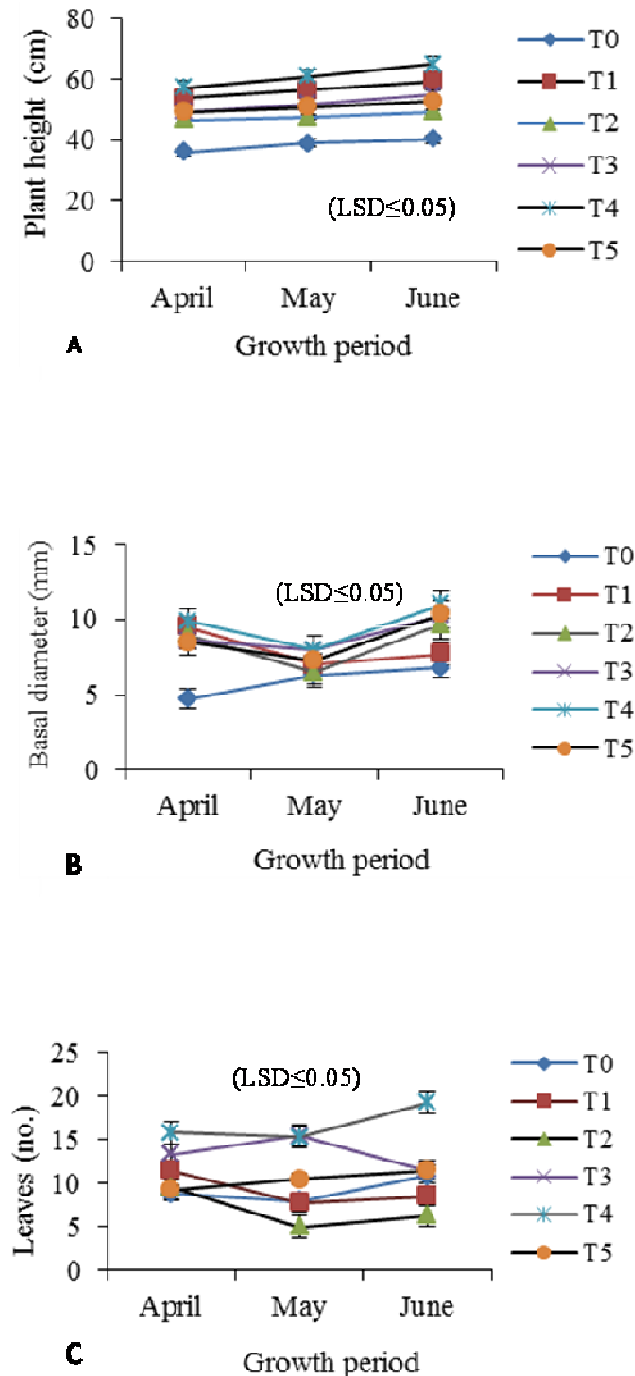
Texture is an important soil characteristic that affects soil management and crop production. Most of the treatments presented a sandy clay loam texture except T1 and T2 which had a clay texture. Clay has high values of water holding capacity, plasticity and cation exchange capacity (CEC) and the pH varies from neutral to slightly acidic (Singh et al., 2001). Clay is usually nutrient rich, the nutrients are too tightly bound to be easily released and absorbed by plant roots (Kerrigan and Nagel, 1998). On the other hand, sandy soils warm up faster than clay ones and can be tilled more easily. It also dries out more quickly and it is not as rich in nutrients (Kerrigan and Nagel, 1998). pH values varied from 4.23 to 4.43 having the highest value (4.43) in T4 and the lowest value (4.23) was in T1 and T4 (Table 1). All the treatments showed low pH values which means the medium was acidic. After harvest, pH increased and ranged from 5.01 to 5.54. Treatment T2 showed the maximum pH value (5.54) followed by T4 (5.39) and the minimum (5.01) was in the control and T5 (Table 1). The pH increase after harvest might be due to reduction of acidic elements such as Al, Fe, Mn and other heavy metals from the growth media. Knight et al. (1997) also found significant increment of soil pH after *Thlaspi caerulescens* was grown in contaminated soils which corroborated the findings of our results. Soil pH affects all the chemical, physical and biological properties of the soil (Brady and Weil, 2002).

Chemical element accumulation in plants depends not only on their absolute content in soil but also on the level of soil acidic-alkaline and reductive oxidative conditions and content of organic matter (Lorenz et al., 1994; Golovatyj, 2002). Soil pH affects the solubility of trace elements and bioavailability of elements in the soil for plant uptake. Most of the plant species survive in a relatively narrow pH range ( $\geq 4.5$ ) (Salisbury and Ross, 1978). Before planting, the highest total carbon (16.15%) was found in T1 (100% sludge) and the lowest (1.79%) was in the control (Table 1). All the treatments except T1, showed similar carbon content. After harvest, total carbon increased in all the treatments except in T1 and T5. The maximum total-C content (6.44%) was found in T3 followed by T2 (4.48%) with the minimum content in the control (2.67%) (Table 1). It was observed that the total C (%) was proportional to the clay and silt content in soil but inversely proportional to the sand content. Similar result was also observed by Hassink (1997). An increase in soil organic matter increases cation exchange capacity (CEC) and nutrient content which improves soil fertility (Rice, 2009). Organic matter also can improve water holding capacity, thus, increasing the plants ability to withstand short droughts.

### Growth performance

The growth parameters measured in this study were height, basal diameter and number of leaves for each level of treatment. The data was taken once a month for four months. There was a significant difference ( $p \leq 0.05$ ) in plant height, basal diameter and number of leaves among treatments in the four months of the study. Treatment T4 (40% sludge and 60% soil) showed the maximum height (65.0 cm) followed by T1. The minimum height (35.88 cm) was noted in the control (Figure 1a).

The monthly total height also showed an increment for each treatment. The average total height on April, May and June was 48.64, 51.06 and 53.58 cm, respectively.



**Figure 1.** Increment of plant height, basal diameter and number of leaves at different treatments. T<sub>0</sub> = Control; T<sub>1</sub> = 100% sludge; T<sub>2</sub> = 80% sludge + 20% soil; T<sub>3</sub> = 60% sludge + 40% soil; T<sub>4</sub> = 40% sludge + 60% soil; T<sub>5</sub> = 20% sludge + 80% soil.

This trend showed that plant height increased with age, which was expected as *D. costulata* is known to be a fast-growing species (Paval et al., 2009).

The highest basal diameter (11.0 mm) was found in T<sub>4</sub> (40% sludge + 60% soil) which was followed by T<sub>5</sub> (10.4

mm) and T<sub>3</sub> (10.3 mm). The lowest basal diameter (4.7 mm) was recorded in the control in April (Figure 1b). The average basal diameter was 8.40, 7.18 and 9.32 mm for April, May and June, respectively. In May, basal diameter decreased in all the treatments except the control and this might be due to heavy metal toxicity. Treatment T<sub>4</sub> also produced the highest number of leaves (19.3) after 3 months (Figure 1c). The second highest number of leaves (11.5) was noted in T<sub>3</sub> and T<sub>5</sub> and the minimum (8.8) was in the control. The height, basal diameter and number of leaves were highest in June (Figure 1). It was because the soil conditions became more stable to plant growth after a certain period. In this situation, plants can produce more leaves, increase height and basal diameter much better than other months.

### Plant biomass

Treatment T<sub>4</sub> produced the highest root biomass (56.86 g) followed by T<sub>3</sub> (53.8 g) and T<sub>2</sub> (51.9 g) and the lowest was in the control (33.59 g). The highest leaf biomass (71.43 g) was also found in T<sub>4</sub> (Table 2). The second highest biomass (65.6 g) was recorded in T<sub>5</sub> which was followed by T<sub>2</sub> (62.4 g) and T<sub>3</sub> (61.8 g). The lowest biomass (30.54 g) was noted in the control (without sludge). Treatment T<sub>5</sub> (20% sludge + 80% soil) produced the maximum stem biomass (71.19 g) and the minimum was in the control (21.0 g). 20 to 40% sludge in combination with soil produced the highest leaves and stems biomass so this plant can be grown for remediation of sludge contaminated soils.

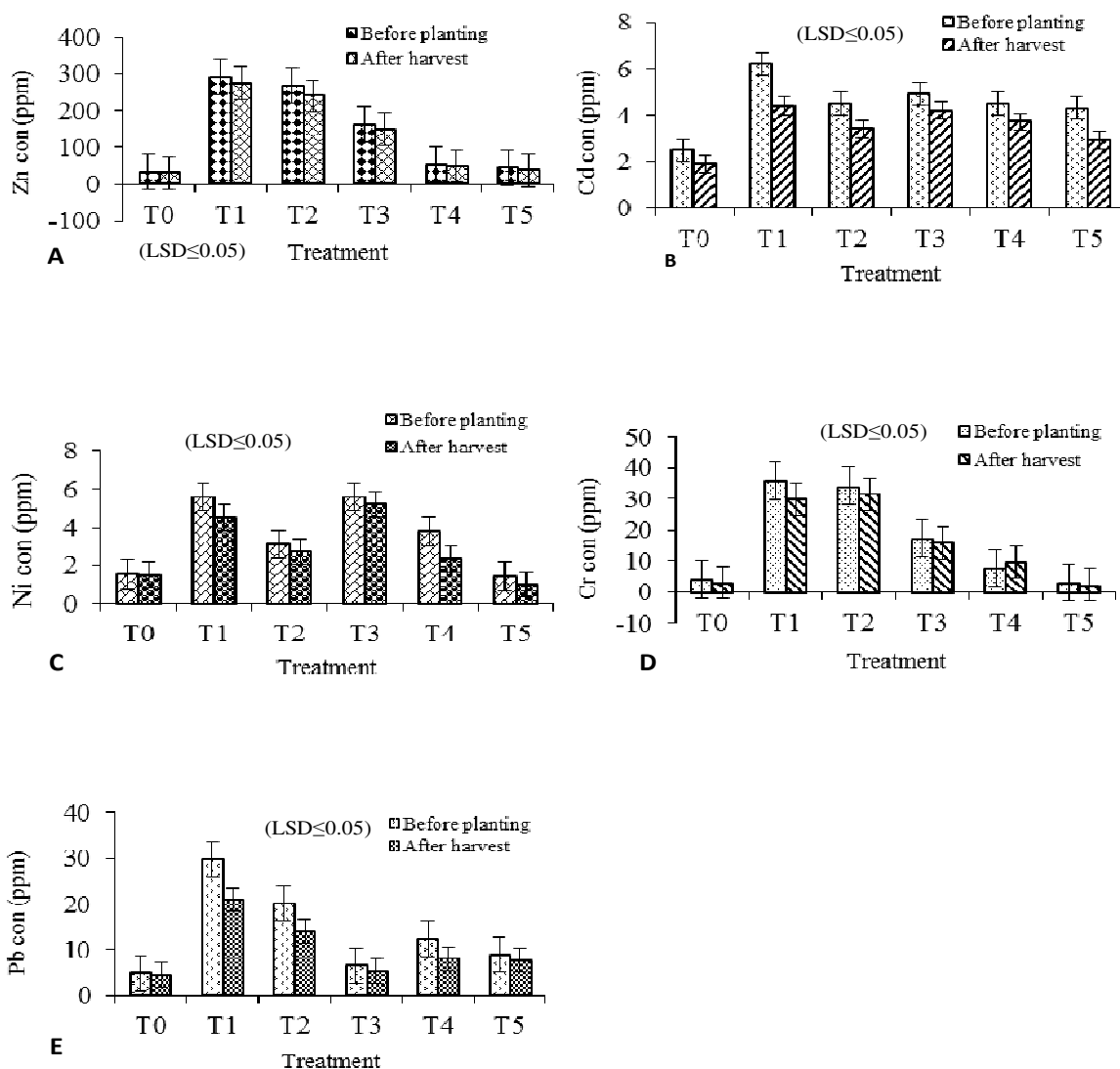
### Heavy metals in the growth medium

Five elements (Zn, Cd, Ni, Cr and Pb) were selected for this study since they are commonly found in sewage sludge. Zn, Cd and Cr concentrations in the growth media were significantly different ( $p \leq 0.05$ ). After harvest, Zn concentration decreased in the growth media having the highest reduction (27.3 ppm) in T<sub>2</sub> followed by T<sub>1</sub> (15.3 ppm) and T<sub>3</sub> (14.4 ppm). The lowest (1.6 ppm) reduction was noted in the control (Figure 2a). The decrease in Zn concentration at the growth media after harvest may be due to uptake by the plants. Zinc is an essential element in the growth of all animals and plants. If the concentration in soil exceeds 30 ppm, the soil is considered contaminated and toxic to several species of microorganisms (Perk, 2006). Concentration of Cd decreased after harvest having the highest in T<sub>1</sub> followed by T<sub>5</sub> (1.4 ppm) and the minimum (0.60 ppm) was in the control (Figure 2b). Cd is non-essential and potentially toxic for higher plants, animals and humans. In normal soil it ranged from 0.01 to 2.0 ppm and the critical concentration varied from 3 to 8 ppm (Alloway, 1995). Treatment T<sub>4</sub> showed the highest reduction of Ni (1.45

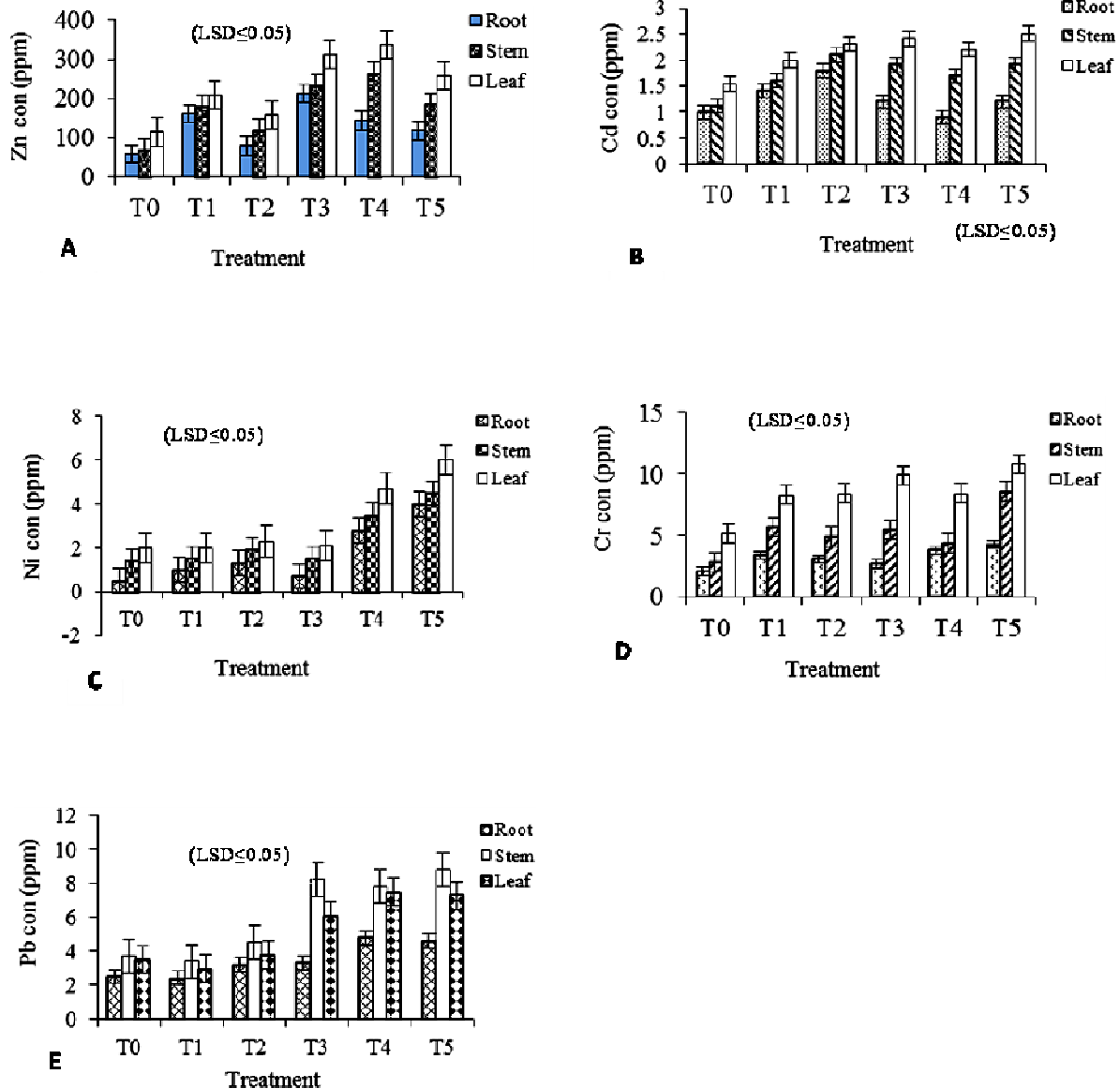
**Table 2.** Weight of plant biomass as influenced by different treatments.

Treatment	Plant biomass (g)			
	Root	Stem	Leaf	Total
T0	33.6	21	30.5	85.1
T1	52.1	55.7	71.4	179.2
T2	43.9	46	62.4	152.3
T3	53.8	53.2	61.9	168.9
T4	56.9	65.9	62.1	184.9
T5	47.2	71.2	65.6	184
CV (%)	9.44	9.86	11.4	9.42
LSD (0.05)	6.82	7.75	10.2	9.09

T<sub>0</sub> = Control; T<sub>1</sub> = 100% sludge; T<sub>2</sub> = 80% sludge + 20% soil; T<sub>3</sub> = 60% sludge + 40% soil; T<sub>4</sub> = 40% sludge + 60% soil; T<sub>5</sub> = 20% sludge + 80% soil.



**Figure 2.** Zn (A), Cd (B), Ni (C) and Cr (D) concentrations in the growth media at different treatments; Pb (E) concentration in the growth media at different treatments. T<sub>0</sub> = Control; T<sub>1</sub> = 100% sludge; T<sub>2</sub> = 80% sludge + 20% soil; T<sub>3</sub> = 60% sludge + 40% soil; T<sub>4</sub> = 40% sludge + 60% soil; T<sub>5</sub> = 20% sludge + 80% soil; con = concentration.



**Figure 3.** Heavy metals accumulation in different plant parts as influenced by different treatments. T<sub>0</sub> = Control; T<sub>1</sub> = 100% sludge; T<sub>2</sub> = 80% sludge + 20% soil; T<sub>3</sub> = 60% sludge + 40% soil; T<sub>4</sub> = 40% sludge + 60% soil; T<sub>5</sub> = 20% sludge + 80% soil; con = concentration.

ppm) followed by T<sub>1</sub> (1.10 ppm). The minimum reduction (0.20 ppm) was found in the control (Figure 2c).

After harvest, Cr concentration in the growth media decreased and the maximum reduction (5.00 ppm) was observed in T<sub>1</sub> followed by T<sub>2</sub> (2.60 ppm). Treatment T<sub>5</sub> showed minimum reduction (0.70 ppm), whereas increment (2.00 ppm) was found in T<sub>4</sub> (Figure 2d). Pb content was also decreased in the growth media. The maximum reduction (8.80 ppm) was noted in T<sub>1</sub> followed by T<sub>2</sub> (4.30 ppm) and T<sub>4</sub> (4.30 ppm) and the minimum (0.5 ppm) was in the control (Figure 2e). Pb is one of the most frequently inorganic pollutants in the soils (Alkorta et al., 2004). It is potentially toxic even at low

concentrations and above 400 mg Pb kg<sup>-1</sup>, the soil is considered hazardous to human health (US-EPA, 2001).

### Heavy metal concentrations in plant tissues

The highest Zn concentration (230.73 ppm) was observed in the leaves followed by stems (173.41 ppm) and the lowest (128.54 ppm) was in the roots. Among the treatments, T<sub>4</sub> showed maximum Zn accumulation (337.40 ppm) in the leaves which was followed by T<sub>3</sub> (311.35 ppm). Treatments T<sub>4</sub> and T<sub>3</sub> also exhibited high accumulation of Zn (262.45 and 229.43 ppm, respectively) in the stems (Figure 3a). The lowest Zn

**Table 3.** Bioconcentration factor of heavy metals as influenced by different treatments.

Treatment	Bioconcentration factor of heavy metal				
	Cd	Pb	Cr	Ni	Zn
T0	0.53	0.56	0.70	0.33	1.83
T1	0.24	0.11	0.11	0.22	0.58
T2	0.53	0.23	0.10	0.48	0.33
T3	0.29	0.62	0.17	0.13	1.43
T4	0.32	0.6	0.39	1.19	2.94
T5	0.41	0.60	1.80	4.00	3.10
CV (%)	7.91	7.51	5.77	6.14	6.71
LSD (0.05)	0.05	0.06	0.06	0.12	0.21

T<sub>0</sub> = Control; T<sub>1</sub> = 100% sludge; T<sub>2</sub> = 80% sludge + 20% soil; T<sub>3</sub> = 60% sludge + 40% soil; T<sub>4</sub> = 40% sludge + 60% soil; T<sub>5</sub> = 20% sludge + 80% soil.

concentration (114.10, 67.60 and 57.25 ppm for leaves, stems and roots, respectively) was detected in the control. The average highest Cd concentration (2.16 ppm) was found in leaves followed by stems (1.72 ppm) and the lowest was in the roots (1.25 ppm) (Figure 3b). Among the treatments, the highest Cd accumulation (2.50 ppm) was noted in T5, while roots (1.8 ppm) and stems (2.1 ppm) showed the highest absorption in T2. The minimum Cd concentration (0.88 ppm) was observed in the T4 for root, while T0 showed the lowest Cd accumulation of 1.54 and 1.12 ppm for leaves and stems, respectively (Figure 3b). Cadmium is not essential for plant growth and can cause various phytotoxic symptoms including leaf chlorosis, root putrescence and growth inhibition. It can be transported over great distances when it is absorbed by sludge. The cadmium-rich sludge can pollute surface water as well as soils and it can enter into food chain. Plants can absorb more cadmium from soil when Zn is insufficient (Cornelis et al., 2009).

The highest Ni concentration (5.95, 4.50 and 4.03 ppm for leaves, stems and roots, respectively) was also obtained in T5 followed by T4 and the lowest was in the control (2.00, 1.40 and 0.50 ppm Ni in leaves, stems and roots, respectively) (Figure 3c). The leaves showed the highest concentration (5.95 ppm Ni) followed by the stems (4.50 ppm Ni) and the roots (4.03 ppm Ni) (Figure 3c). Treatment T5 showed the highest concentration of Cr (10.78, 8.63 and 4.30 ppm for leaves, stems and roots, respectively) and the lowest concentration (5.18, 2.83 and 2.13 ppm in leaves, stems and roots, respectively) was in the control (Figure 3d). Among the plant parts, leaves had the highest accumulation of Cr (10.78 ppm) followed by stems (8.63 ppm) and roots (4.30 ppm) (Figure 3d). Chromium compounds are highly toxic to plants and are detrimental to their growth and development. Chromium is unstable in the presence of oxygen and produces a thin oxide layer protecting the metal from new oxidation processes. In leaves, treatment T5 showed the maximum Pb accumulation (8.80 ppm) followed by T3 (8.18 ppm) and the minimum (3.41 ppm)

was in the T1 (Figure 3e). In the case of the leaves, the highest concentration (7.46 ppm) was found in T4 followed by T5 (7.34 ppm) and the lowest in the T1 (2.98 ppm). T4 also showed the highest accumulation of Pb (4.83 ppm) in the roots and the lowest was in the T1 (2.38 ppm). The ranking of the plant parts in respect of Pb concentration was in the order of stems>leaves>roots (Figure 3e) which was opposite that of Zn, Ni, Cd and Cr (leaves > stem > root). Wozny et al. (1995) reported that Pb is more accumulated in roots than in leaves which are in agreement with the findings of our results. The normal Pb concentration in the plant lies between 0.1 to 5 ppm (Reeves and Baker, 2000). All the treatments (except T1 and T2) exceeded the normal Pb range. So, before remediation, it is wise not to use sewage sludge for food crops cultivated soils.

#### Bioconcentration factor and translocation factor of heavy metals

Treatment T5 showed the highest BCF (1.80, 4.00 and 3.10 for Cr, Ni and Zn, respectively) (Table 3). The lowest BCF of 0.10 and 0.33 for Cr and Zn respectively was observed in T2 while T3 showed the minimum BCF (0.13) of Ni. In the case of Pb and Cd, maximum BCF value (0.62 and 0.53 for Pb and Cd, respectively) was found in T3 and T2, respectively (Table 3). The minimum values of 0.11 and 0.24 for Pb and Cd respectively were noted in the T1, which may imply the restriction in soil-root transfer at higher metal concentrations in the soil. Similar results were found by Yoon et al. (2006). Niu et al. (2007) reported the highest BCF in a hydroponic culture in sunflower (BCF = 11). Ho et al. (2008) also observed 1.92 to 3.31 BCF values in Pb treated kenaf (*Hibiscus cannabinus* L.) which are partly in agreement with the findings of our results.

*D. costulata* roots showed the ability to absorb heavy metals from sludge contaminated soils. The maximum translocation (4.18 and 4.33 for Zn and Cd, respectively)

**Table 4.** Translocation factor of heavy metals as influenced by different treatments.

Treatment	Translocation factor of heavy metal				
	Cd	Pb	Cr	Ni	Zn
T0	2.66	2.88	3.37	6.8	3.17
T1	2.44	2.59	4.12	3.5	2.4
T2	2.57	2.67	4.29	3.23	3.46
T3	3.58	4.33	5.7	5.14	2.54
T4	4.33	3.19	3.81	2.93	4.18
T5	3.67	3.5	4.5	2.63	3.75
CV (%)	8.70	4.44	5.77	5.44	4.36
LSD (0.05)	0.51	0.26	0.45	0.40	0.26

T<sub>0</sub> = Control; T<sub>1</sub> = 100% sludge; T<sub>2</sub> = 80% sludge + 20% soil; T<sub>3</sub> = 60% sludge + 40% soil; T<sub>4</sub> = 40% sludge + 60% soil; T<sub>5</sub> = 20% sludge + 80% soil.

was observed in T4 followed by T5 (3.75 and 3.67 for Zn and Cd, respectively) and the minimum was in the T1 (2.4 and 2.44), while the control showed the highest translocation of Ni (6.80) (Table 4). For Cr and Pb, treatment T3 showed the highest translocation (5.7 and 4.33 for Cr and Pb, respectively) and the lowest was in the control (3.37) and T1 (2.59) for Cr and Pb, respectively (Table 4). Translocation was pronounced in all the treatments. Metal translocation reduction in T1 might be due to high concentration of heavy metals. Similar results were also observed by Yoon et al. (2006) and Ho et al. (2008). TF of metal excluder species is < 1, whereas, metal accumulator species has TF > 1 (Baker, 1981). It was observed that, all the treatments exhibited higher TF values (> 1). *D. costulata* has high TF and low BCF in soil at higher metal concentrations. Heavy metal tolerance with high TF and low BCF value was suggested for phytoaccumulator of contaminated soils (Yoon et al., 2006).

## Conclusion

The study showed that, *D. costulata* can be used as a potential phytoremediator for contaminated soils and to mitigate soil pollution. The plant was able to remove zinc, cadmium, lead, nickel and chromium from growth media. Treatment T1 (100% sludge) presented the highest concentration of heavy metals. The heavy metal concentrations in the growth media were higher before planting but decreased at harvest. The highest concentration reduction for Cd, Cr and Pb was found in T1, while Zn was in T2 and Ni in T4. The later composition (40% sludge + 60% soil) gave the best growth performance in terms of height and basal diameter.

*D. costulata* showed high absorbing capacity of heavy metals. Zn, Cd, Ni and Cr were highly concentrated in the leaves while Pb was accumulated in the stems. The species also showed high TF value and metal tolerance

ability. The use of this species in metal extraction (phytoremediation) appeared as a promising alternative for heavy metal removal from soil and water via extraction. However, a longer term experiment is needed to confirm if this species can be use as a good accumulator for heavy metals. It is also suggested that, a field study should be conducted to verify the findings obtained from the glasshouse study.

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