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METAL ACCUMULATION BEHAVIOR OF THE WEED SPECIES GROWING UNDER SOIL CADMIUM STRESS

MD. DULAL SARKAR[1*], MD. JAHEDUR RAHMAN[1], JASIM UDDAIN[1], MD. QUAMRUZZAMAN[2], ROJOBI NAHAR ROJONI[3] AND SREERAMANAN SUBRAMANIAM[4]

[1]Department of Horticulture, Sher-e-Bangla Agricultural University, Dhaka-1207, Bangladesh
[2]Department of Agronomy, Sher-e-Bangla Agricultural University, Dhaka-1207, Bangladesh
[3]Directorate of National Consumer Rights Protection, Ministry of Commerce, Dhaka, Bangladesh
[4]School of Biological Sciences, Universiti Sains Malaysia, Penang-11800, Malaysia
[*]Corresponding author's email: dulalsau_121@yahoo.com

Abstract

Three weeds, *Enhydra flactuans*, *Amaranthus viridis* and *Chenopodium album* were considered as Cd accumulating plants. Accumulated Cd in *C. album* reached to 0.32 mg for each plant under soil Cd stress of 15 mg. Thus, Cd accumulation capability of weeds was decreased with the increased using rate of Cd and it was between 1.64% and 4.96%. After 45 days? of Cd stress, the redistribution of Cd content in the root and shoot was studied from underground parts to above ground parts. The outcome confirmed that weed species had sophisticated acceptance to Cd and could accumulate affectively. They did not show any abnormal growth appearance even in soils having 15 mg kg^{-1} Cd, and for *C. album*, the Cd content in the roots was up to 264.32 mg kg^{-1} dry biomass while in the shoots it was about 126.55 mg kg^{-1}. This signified that three weeds behaved as decent accumulating plant although they showed infirm ability in transporting Cd from below ground biomass to above ground biomass. Afterwards, shoot:soil and root:soil further proved that, studied weed species were likely to uptake more Cd as a way of diminution of the mobility of Cd. So, this research predicts that using green technology as a process of phyto-mitigation of trace elements from the defiled soils would be a good choice.

Key words: Bioremediation, Hyperaccumulator, Phytoremediation, Trace elements, Weed species.

Introduction

Contaminants absorption is happening gradually in the land and aquatic sources as a result of both natural and anthropogenic deeds (Hasanuzzaman & Fujita, 2012) and this is frightful to human world and environment (Grytsyuk *et al*., 2006). Cd, Pd and As are the most significant hazardous elements of agricultural soil causing fastest deterioration (Islam *et al*., 2015) and the groundwater also contain As significantly in Bangladesh (Rahman *et al*., 2015). Contaminants are immutable, released slowly and remain concealed in nature (Boularbah *et al*., 2005; Zhou & Qiu, 2005). They are effortlessly absorbed by underground biomass and transferred to above ground shoot biomass which are the causes of innumerable ailments (Hu *et al*., 2013). Many approaches such as, enrichment factor, contamination factor and geoaccumulation index (Rashed, 2010, Liu *et al*., 2014), soil flora, soil fauna and microorganisms (Beesley & Marmiroli, 2011; Huang *et al*., 2014), acidic and neutral biochars approach (Qi *et al*., 2018) are advantageous to remediate contaminants and some of them are perilous and costly at large scale (Naidu *et al*., 2008; Rakhshaee *et al*., 2009). Phytoremediation is an aesthetically pleasing method that remediate contaminants from polluted sites and has received enormous interest from the scientific community (Salt & Baker, 2008). Harvesting and processing simply can be done for plant parts that help in accumulating toxic metals resulting in comparatively less troublemaking to the contaminated sites (Salido *et al*., 2003). Hyperaccumulating plants are inadequate but they are very significant indeed to find out effective one (Wei *et al*., 2006, Meers *et al*., 2005). Thus, it is important to find more hyperaccumulating plants to remediate contaminants from polluted sites (Zhou & Song, 2004).

Weed species have capability of heavy metal uptake (Kaimi *et al*., 2006) while some herbaceous ornamental plants had been used in Cd and Pb contaminated soil (Wang, 2005). The ability for heavy metal accumulation depends on plant species (Inelova *et al*., 2018). Weeds might be the strongest species to adjust themselves under soil Cd adverse conditions and considered as potential hyper-accumulator for soil remediation. Previous studies showed that weed plants are effective to remediate metal contaminated soils and it is now considered as a valuable phytoremediation technique. Proper choice of plant species can bring fruitful outcome in accumulating toxic metals through producing higher fresh and dry biomass (Rodriguez *et al*., 2005). Thus, present study investigated the physiological response and Cd accumulation performance of the weed species.

Materials and Methods

Experimental site: The present study was carried out at the Sher-e-Bangla Agricultural University, Dhaka, Bangladesh in 24.09^0N and 90.26^0E longitude following plastic tunnels provision. This site is used to with strong and scant cloudburst from April to September and October to March, respectively.

Experimental design: Each treatment was repeated 4 times maintaining Randomized Complete Block Design for a period of 45 days. Four levels of Cd were studied as 0, 5, 10 and 15 mg kg^{-1} of soil respectively. Cd in the form of $CdCl_2{\cdot}2.5H_2O$ was practiced on 3 weed species namely Alligator weed (*Enhydra flactuans*), Green amaranth (*Amaranthus viridis*) and Chenopodium (*Chenopodium album*). Three trials were conducted separately at the same time over the same period.

Growing conditions: Sandy loam type soil was tested before experimentation which were considered as unpolluted containing pH 5.90 and Cd 0.11 mg kg^{-1}. Plastic pots (10″) were filled with soil that could pass through 4.0mm sieve. As per treatments, different amounts of $CdCl_2 \cdot 2.5H_2O$ were applied for maintaining 0, 5, 10 and 15 mg kg^{-1} Cd. Seeds of weed species were collected from nature and 5 seeds were sown in a pot. Distilled water was applied by hand sprayer to saturation. No fertilizer was used during conducting this experiment and 45 days old plants were harvested.

Soil sampling and Cd determination: Soil samples were air-dried, crushed, and passed through a 2-mm sieve. Soil pH was determined by pH meter with a glass electrode. Dried soil samples were digested with concentrated HNO_3 and $HClO_4$ mixture as described by Piper (1966) for determination of Cd. Soil samples weighing 0.5 g were taken into a 50 ml boiling flask where 5 ml of nitric-perchloric acid solution was added. The flask was placed on cool plate and then the temperature was turned slowly to about 375°F and the digestion was allowed for 2 hours. Then the flask was removed and 15 ml distilled water was added to the flask following the flask agitation and heating to dissolve. The content was filtered through a filter paper (Whatman no. 42) in a 100 ml volumetric flask and then distilled water was added to make the volume up to the mark (100 ml). Atomic Absorption Spectrophotometer (model no. 170-30, HITACHI, Japan) was calibrated with standard solution of Cd and calibration curve was prepared by the series of standard solution.

Physiological features and Cd determination in plant sample: Prior to harvesting of each plant, total plants in each pot were tallied and pursued by computing height from soil level to peak point. Thereafter, weed species were sensibly uprooted avoiding any damage to the plants and for assistance of humid condition as they were kept into a plastic bag. Each clean root was measured by a scale after drying out of adhesive water. Fresh biomass of shoots and roots were taken and then fresh plant parts were oven dried maintaining the temperature about 80°C for 48 hours. The dry biomass was measured by a sensitive balance. For Cd determination, previously described standard procedure by Piper (1966) was followed.

Statistical analysis

Mean value of each parameter from the three trials were taken to analysis. Tukey HSD test (Tukey, 1977) was used to determine variances among the treatments where $p<0.05$ was considered as significant.

Results and Discussion

Physiological response: Weed species grown up under Cd stress remained shorter than the controlled treatment. Weed species showed significant acceptance capability to Cd while among them *E. flactuans* had maximum acceptance followed by *A. viridis* and *C. album* (Table 1). In *C. album*, plant height was reduced through rising Cd level that restricted vegetative development moderately for 15 mg kg^{-1} whereas *A. viridis* differ in high Cd stress that is approximately comparable to *E. flactuans*. It appeared that plant height was steadily reduced in Cd treated soil that sustained to higher concentrations. Cd has inhibitory effect that is regulated by volume and bioavailability (Liu *et al*., 2008; Gisbert *et al*., 2006). Every plant was green under Cd stress that means, they behaved normal without showing any disorder at their different parts. Though root length (ranged from 6.25 to 12.88) was apparently analogous (Table 1), thereafter root growth decreasing trend was found under high level Cd stress.

E. flactuans and *A. viridis* showed seemingly comparable fresh biomass increasing trend while *C. album* exposed better trend (Table 1). Dry biomass of plant was non-significant (Fig. 1) and ranged from 2.15g to 3.59 g in the total plant body. *E. flactuans* and *A. viridis* had the capability to contribute dry biomass more even in presence of high Cd level soil culture (Fig. 1). Experimented weed species had privileged acceptance to Cd beside plant growth and behaved as a plant of hyperaccumulator (Liu *et al*., 2008; Gisbert *et al*., 2006).

Table 1. Physiological response of three weeds under soil Cd stress.

Weed species	Soil Cd concentrations (mg kg^{-1})	Plant height (cm)	Root length (cm)	Plant fresh biomass (g)
Enhydra flactuans	0	35.75 ± 0.24^{a}	12.88 ± 0.32^{a}	18.24 ± 0.26^{ab}
	5	32.29 ± 0.22^{bc}	10.20 ± 0.14^{ab}	14.29 ± 0.47^{def}
	10	29.50 ± 0.32^{c}	9.13 ± 0.21^{bc}	12.07 ± 0.22^{fg}
	15	25.75 ± 0.24d^{e}	8.88 ± 0.33^{bc}	10.15 ± 0.15^{g}
Amaranthus viridis	0	32.75 ± 0.24^{b}	10.00 ± 0.29^{ab}	16.87 ± 0.47^{abcd}
	5	26.50 ± 0.48^{d}	9.75 ± 0.31^{ab}	15.40 ± 0.31^{bcde}
	10	24.00 ± 0.20d^{ef}	8.75 ± 0.38^{bc}	14.45 ± 0.42^{def}
	15	23.50 ± 0.32^{ef}	7.75 ± 0.43^{bc}	12.65 ± 0.36^{efg}
Chenopodium album	0	22.50 ± 0.32^{f}	8.19 ± 0.19^{bc}	18.55 ± 0.26^{a}
	5	18.88 ± 0.30^{g}	8.00 ± 0.46^{bc}	17.98 ± 0.23^{abc}
	10	16.75 ± 0.31^{g}	7.00 ± 0.35^{bc}	15.02 ± 0.26^{cdef}
	15	13.25 ± 0.24^{h}	6.25 ± 0.38^{c}	13.48 ± 0.22^{ef}
F-value		2.85	1.11	2.63
P-value		0.0224	0.3758	0.0325

Different letters in the same column indicate significant differences between treatments ($p<0.05$). Values are mean ± SE

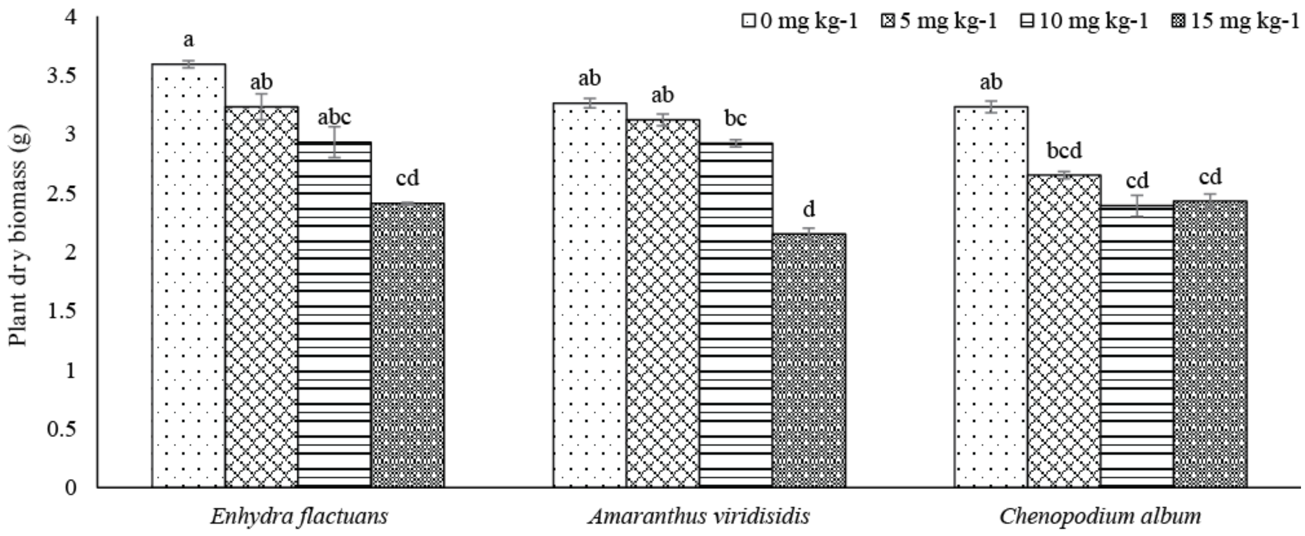

Fig. 1. Comparison of plant dry biomass in the three weeds growing under soil Cd stress values with different letter indicate significant differences between treatments at $p<0.05$. Vertical bars indicate standard errors of means.

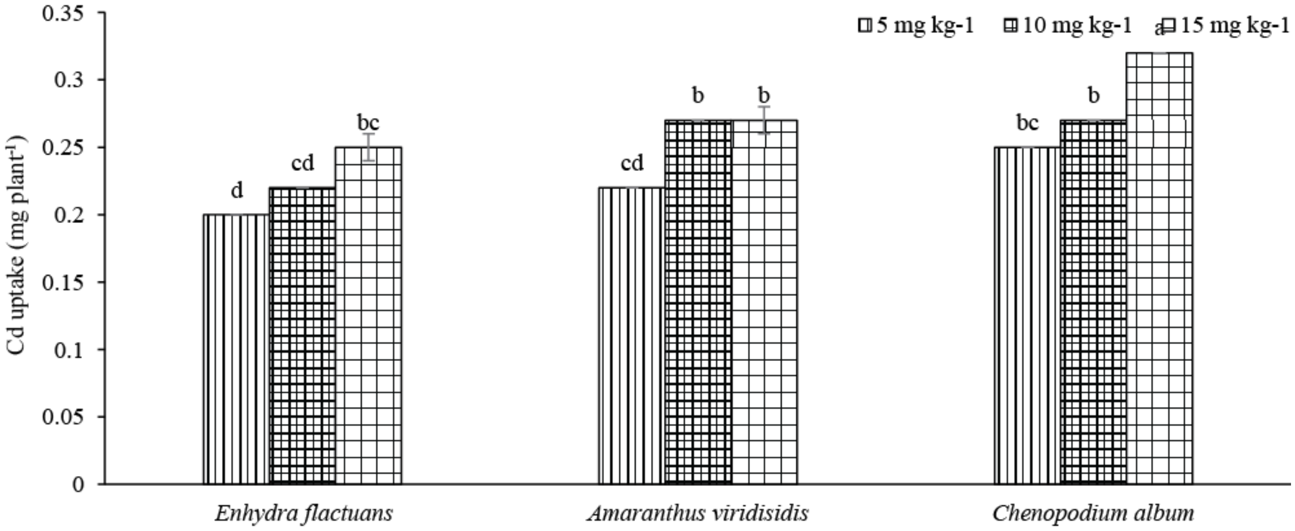

Fig. 2. Cd uptake comparison of the three weed species under soil Cd concentrations values with different letter indicate significant differences between treatments at $p<0.05$. Vertical bars indicate standard errors of means.

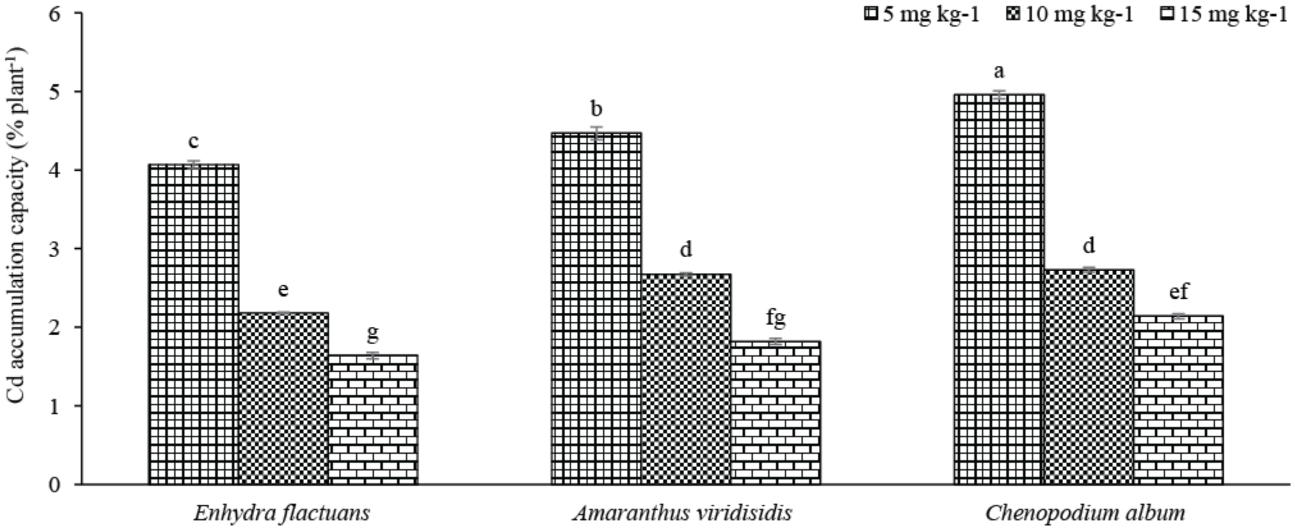

Fig. 3. Cd accumulating capacity of three weed species under Cd stress values with different letter indicate significant differences between treatments at $p<0.05$. Vertical bars indicate standard errors of means.

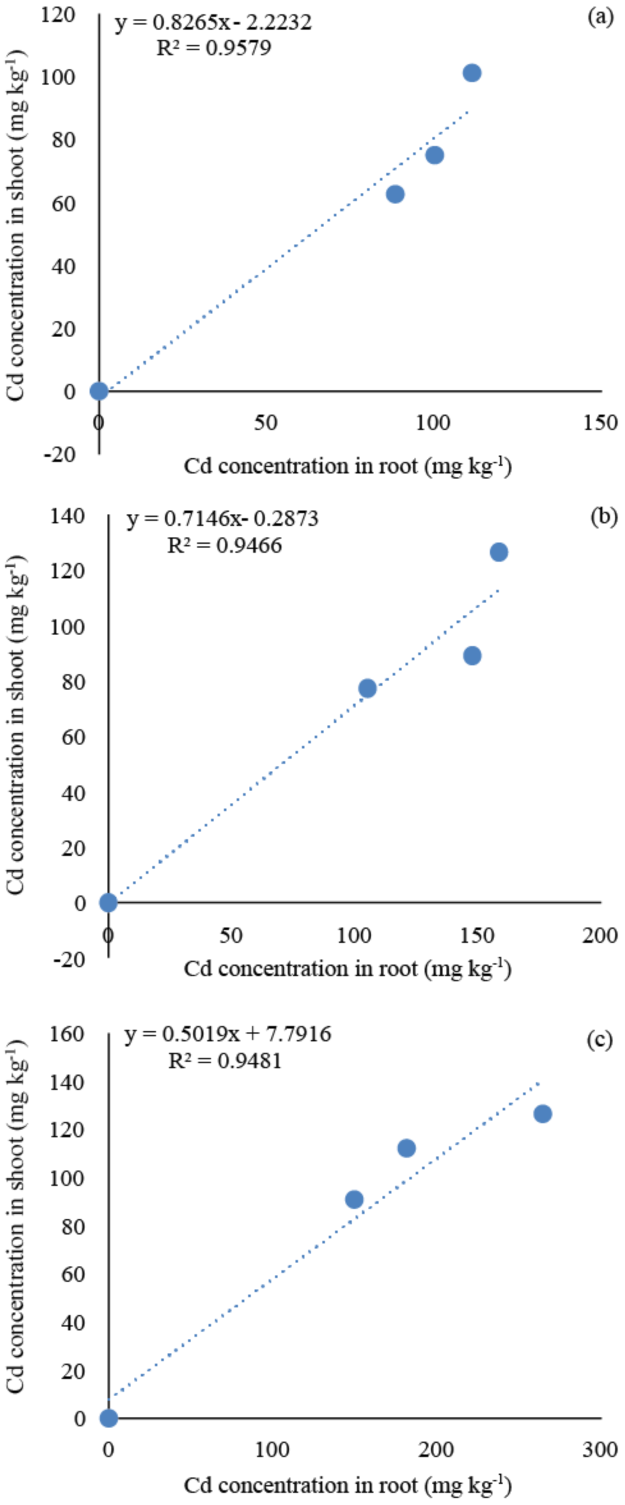

Fig. 4. Comparative relationship in the accumulation of Cd in shoot and roots of (a) *E. flactuans* (b) *A. viridis* and (c) *C. album* under soil Cd stress.

Cd accumulation: Accumulated Cd in *C. album* weeds body were 0.25, 0.27 and 0.32 mg kg $^{-1}$ having maximum acceptance behavior rather than other two under the Cd soil culture (Fig. 2). On the other hand, *E. flactuans* accumulated 0.20, 0.22 and 0.25 mg kg $^{-1}$ while *A. viridis* had an uptake of 0.22, 0.27 and 0.27 mg kg $^{-1}$ in their body mass. Weeds grown-up in presence of high Cd level soil culture resulted in more Cd gathered in their body. Cd accumulation capability by weed species reduced with increasing Cd level and it was minimum (1.64%) under 15 mg kg^{-1} Cd culture in *E. flactuans* and it reached up to 4.96% in *C. album* weeds body under 5 mg kg^{-1} Cd culture (Fig. 3). In fact, herbaceous plants were advantageous for substantial and practical purposes (Zhou, 2006). Cd accumulation might be reliant on Cd level in the defiled site. Soil solution and solid phase was responsible for Cd solubility and bioavailability (Liu *et al*., 2017; Qi *et al*., 2018).

Cd level in shoot was less than root in all weeds (Table 2). The highest degree Cd gathering inclined the capability of weeds where *C. album* was the most capable for translocating Cd in shoot and roots ranging from 62.69 mg kg^{-1} to 126.55 mg kg^{-1} and 88.40 mg kg^{-1} to 264.32 mg kg^{-1} respectively (Table 2). Cd level was increased in plant parts by increasing soil Cd level. Shoot: Soil and Root: Soil of Cd accumulation were more than 1.0 and was declined with increased soil Cd level while Shoot: Root was not significant and lower than 1.0 (Table 2).

Shoot of *A. viridis* also contributed to the lowest Cd accumulation than roots and it was decreased in both shoots and the roots with declining Cd level in soil where the accumulated Cd was 126.57 and 158.60 mg respectively in 15 mg Cd soil culture. The Shoot: Soil and Root: Soil of Cd accumulation were more than 1.0 and was the lowest with rising Cd level and Shoot: Root were also less than 1.0 (Table 2).

For *E. flactuans*, Cd accumulation trend both in the roots and shoots was similar and increased in both of parts with rising Cd level. Cd level reached up to 101.17 mg kg^{-1} in shoot and 111.26 mg kg^{-1} in root in high soil Cd stress. Shoot: Soil and Root: Soil values were more than 1.0 while Shoot: Root lesser than 1.0 (Table 2).

Variations in Cd accumulation by weed species were observed probably because of differences between the species. Cd accumulation by the shoots surpassed 100 mg kg^{-1} in all weed species, a criterion for being a hyperaccumulator. On the other hand, the total amount of accumulated Cd was lesser in the shoots than in roots signifying meagre ability in uptaking Cd by shoots that could not behave as hyperaccumulator.

These herbaceous weeds were green and grew without any disorders even in presence of high Cd soil culture, moreover, they showed the ability to accumulate Cd. Herbaceous ornamental plants are able to grow up, complete their development in the defiled soil and take out significant amount of pollutants (Liu *et al*., 2008; Dickinson & Pulford, 2005).

Cd accumulation relationship between root and shootl: Regression graph showed that, shoot and root of weeds had significant positive correlations between them in terms of Cd accumulation. Increasing trend of Cd in underground biomass simplified the Cd distribution to above ground biomass. Cd accumulating capability of weed species was about analogous (Fig. 4) which depended on plant types. Zhou *et al*., (2004) signified that contaminants accretion are linked to plant species as well as their different parts.

Table 2. Response of three weed species to accumulate Cd under different soil Cd concentrations.

Weed species	Soil Cd concentrations (mg kg^{-1})	Cd concentration (mg kg^{-1})		Cd concentration		
		Shoot	Root	Shoot: Soil	Root: Soil	Shoot: Root
Enhydra flactuans	0	-	-	-	-	-
	5	62.69 ± 2.08^{d}	88.40 ± 1.44^{d}	12.54 ± 0.42^{bc}	17.68 ± 0.29^{bc}	0.71 ± 0.02^{ab}
	10	75.06 ± 3.75^{cd}	100.18 ± 2.49^{cd}	7.51 ± 0.38^{e}	10.02 ± 0.25^{de}	0.75 ± 0.04^{ab}
	15	101.17 ± 2.64^{abc}	111.26 ± 1.63^{cd}	6.75 ± 0.18^{e}	7.42 ± 0.11^{e}	0.91 ± 0.03^{a}
Amaranthus viridis	0	-	-	-	-	-
	5	77.31 ± 3.57^{cd}	105.20 ± 5.00^{cd}	15.46 ± 0.71^{ab}	21.04 ± 1.00^{b}	0.76 ± 0.05^{ab}
	10	89.10 ± 1.14^{bcd}	147.82 ± 8.90^{bcd}	8.91 ± 0.11^{de}	14.78 ± 0.89^{bcd}	0.63 ± 0.04^{ab}
	15	126.57 ± 6.66^{a}	158.60 ± 13.36^{bc}	8.44 ± 0.44^{de}	10.57 ± 0.89^{cde}	0.870 ± 0.08^{a}
Chenopodium album	0	-	-	-	-	-
	5	90.97 ± 0.51^{bcd}	149.54 ± 8.57^{bc}	18.20 ± 0.10^{a}	29.91 ± 1.71^{a}	0.64 ± 0.05^{ab}
	10	112.29 ± 3.02^{ab}	181.22 ± 7.07^{b}	11.23 ± 0.30^{cd}	18.12 ± 0.71^{b}	0.63 ± 0.02^{ab}
	15	126.55 ± 3.16^{a}	264.32 ± 6.36^{a}	8.44 ± 0.21^{de}	17.62 ± 0.42^{bc}	0.49 ± 0.02^{b}
F-value		2.39	7.09	3.97	3.81	2.18
P-value		0.0482	0.0000	0.0038	0.0049	0.0682

Different letters in the same column indicate significant differences between treatments ($p<0.05$). Values are mean ± SE. Control treatment not tested

Conclusion

C. album had inordinate acceptance and uptaking capability of Cd together in their above ground and below ground parts. *A. viridis* and *E. flactuans* also showed higher acceptance to Cd while relocating Cd from below ground parts to above ground parts but they were not that effective. They grew without any disorder resulting sophisticated fresh and dry biomass accumulating higher Cd. On the other hand, Cd accumulation was less under increasing trend of Cd soil stress. Thus, they act as phytostabilizer in mitigating Cd from defiled soils. Although these three weeds may not be considered as a hyperaccumulator but they possibly will mitigate pollutants from defiled soils if they're grown before crop cultivation particularly in industrialized areas. *C. album* might be judged as a good plant planned for mitigating toxic metals because of metal accumulation behavior to Cd.

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