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Bioaccumulation of Toxic Metals (Cd and Cu) by *Groenlandia densa* (L.) Fourr.

Yesim Kara · Ali Zeytunluoglu

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Abstract In this study, *Groenlandia densa* (L.) Fourr. (opposite-leaved pondweed), was exposed to prepared stock solution of cadmium and copper with 1.0, 3.0, 5.0 and 7.0 mg L^{-1} concentration in certain periods (24, 48, 72 and 96 h) and changing amount of accumulation of plants in depending on time and concentration was measured by atomic absorption spectrophotometer. The results show that under experimental conditions, *G. densa* (L.) Fourr. proved to be a good accumulator of Cd and Cu. Removal of the metals from solution was fast in the first 4 days. The accumulation of Cd and Cu increased with the initial concentration and also with time. The highest concentrations of each trace element accumulated in opposite-leaved pondweed tissues were 1,955 µg Cd g^{-1}, 6,135 µg Cu g^{-1} after 4 days. The maximum values of bioconcentration factor (BCF) were found for Cd and Cu 724 and 1,669, respectively. BCF values for Cd and Cu increased with time.

Keywords Bioaccumulation · *Groenlandia densa* (L.) Fourr. · Cadmium · Copper

Heavy metals are important environmental pollutants and many of them are toxic even at very low concentrations (Memon et al. 2001). Heavy metal contamination can be separated from water using a variety of technologies, including chemical, physical, and biological (Axtell et al. 2003). Aquatic plants are known to accumulate metals from their environment (Soltan and Rashed 2003) and this is well documented (Gupta and Chandra 1998), wetland plants are being used successfully for the phytoremediation of trace elements in wastewaters. The use of metal accumulating plants to clean soil and water contaminated with toxic metals are the most rapidly developing component of this environmentally friendly and cost-effective technology.

The growth rate and yield of biomass are important factors in evaluating these plants for removal of metal ions, fuel, food and fertilizer especially for the needs of people of developing countries. Furthermore; aquatic biomass is also being used for production of materials such as paper, roofing and a variety of chemicals (Srivistav et al. 1994; Jain et al. 1990).

Cadmium and copper are reported in natural and wastewaters. Although cadmium does not have any metabolic use for plants, it has several industrial applications, viz. electroplating, pigments, stabilizers, batteries alloy, etc. However, Cu is essential for plant metabolism and also has a number of industrial applications (Prasad et al. 2001). Cd has been recognized for its negative effect on the environment where it accumulates throughout the food chain posing a serious threat to human health (Lu et al. 2004). Copper has been shown to be one of the most toxic heavy metals to human and animals when it was excess concentration in water.

In this study; heavy metal bioaccumulation by aquatic vascular plant *Groenlandia densa* (L.) Fourr. to elevated concentrations of cadmium and copper were investigated and the results were discussed in terms of the cleaning of wastewater treatments.

Y. Kara (✉) · A. Zeytunluoglu
Department of Biology, Faculty of Science and Arts, University of Pamukkale, Denizli 20017, Turkey
e-mail: eylul@pau.edu.tr

Materials and Methods

G. densa is a species of the family Potamogetonaceae (Pondweed family). There are about 80 species of pondweeds in the world. Pondweeds are very important as wildlife food. They occur in a variety of aquatic habitats. They are a perennial herbaceous and leaves are opposite. First flowering time is in May. Last flowering time is in September.

G. densa (L.) Fourr. was collected from Işıklı Lake (Fig. 1), Denizli City, Turkey in May 2006, and were transferred the laboratory in polyethylene bags. Plants of similar size, shape and height were selected and washed several times using tap and bi-distilled water.

All the chemicals were used of Analar grade (Merck). Working metal and standard solutions with different concentrations (1.0, 3.0, 5.0 and 7.0 mg L^{-1}) of each element was prepared by diluting the stock solution (1,000 mg L^{-1}) using de-ionized water. The experiments were carried out in 1 L glass jars containing 500 mL metal solution. The prewashed *G. densa* was inoculated with medium containing various concentrations of Cd and Cu (1.0, 3.0, 5.0, 7.0 mg L^{-1}) and exposured for 24, 48, 72 and 96 h. All experiments were carried out in a greenhouse under controlled temperature (23 (1°C) and light (3800 Lux). Digestion of plant samples in this study was performed as described by Kacar (1972). After the termination of each experiment, the plants were washed well using tap water and distilled water. The washed samples were carefully dried of adherent water using absorbent paper. Samples were dried to a constant mass in a fan-forced oven at 80°C (overnight). The oven-dried material was chopped finally then ground to ensure homogeneity and to facilitate organic matter digestion.

Approximately 1.0 g of grounded material from each sample was accurately weighed into 150 mL Erlenmeyer flasks. Concentrated nitric acid (5 mL) was added to each flask and the samples were placed on a water batch. The temperature of water batch was set at 40°C and the samples were kept on a water batch for about 1 h. Then the samples were heated on a heating block until the nitric acid solution completely evaporates. Then the samples were cooled, 10 mL of concentrated $HNO_3/H_2SO_4/HClO_4$ acid mixture (10:1:4 v/v) was added and the mixture was heated again at 150°C. Digestion was continued until the solution became clear. Then the samples were cooled again. The solutions were filtered and made up to 100 mL with de-ionized water.

Metal contents of the plant samples were determined using an Atomic Absorption Spectrophotometer (AAS 700, Perkin-Elmer). The detection limits were 20 µg/L for Cu and 4 µg/L for Cd were added in the Analyses section. The values of the metal content were calculated in µg g^{-1} dry wt. The bioconcentration factor (BCF) provides an index of the ability of the plant to accumulate the metal with respect to the metal concentration in the substrate. The BCF were calculated as described by Zayed et al. (1998) based on the initial concentration of the given element in the culture medium.

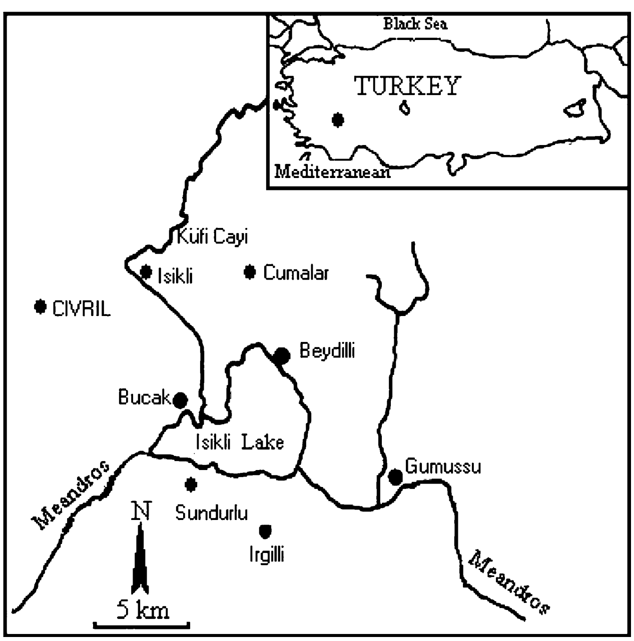

Fig. 1 Map of the study area

Table 1 The bioaccumulation amounts of heavy metals by *G. densa*

Time (h)	Metal concentration (mg L^{-1})	*G. densa* (µg g^{-1})	
		Cd^{++}	Cu^{++}
	Control	20 ± 0.00	26.0 ± 15.56
1	1	423 ± 7.07	581 ± 1.41
	3	649 ± 8.49	977 ± 7.78
	5	746 ± 19.80	1828 ± 7.07
	7	1150 ± 7.07	2538 ± 2.12
2	1	595 ± 16.26	1022 ± 5.66
	3	798 ± 2.83	1462 ± 7.78
	5	1166 ± 12.73	1950 ± 21.21
	7	1223 ± 15.56	2562 ± 10.61
3	1	604 ± 9.90	1596 ± 14.14
	3	893 ± 9.90	2274 ± 12.73
	5	1297 ± 4.24	3493 ± 13.44
	7	1631 ± 7.78	4583 ± 14.85
4	1	724 ± 9.19	1669 ± 5.66
	3	1101 ± 1.41	3122 ± 8.49
	5	1363 ± 18.38	4791 ± 20.51
	7	1955 ± 3.54	6135 ± 7.07

$$\mathrm{BCF} = \frac{\text{Trace element concentation in plant tissue } (\mu\,\mathrm{g\,g^{-1}}) \text{ at harvest}}{\text{Initial concentration of the element in the external nutrient solution } (\mathrm{mg\,L^{-1}})}$$

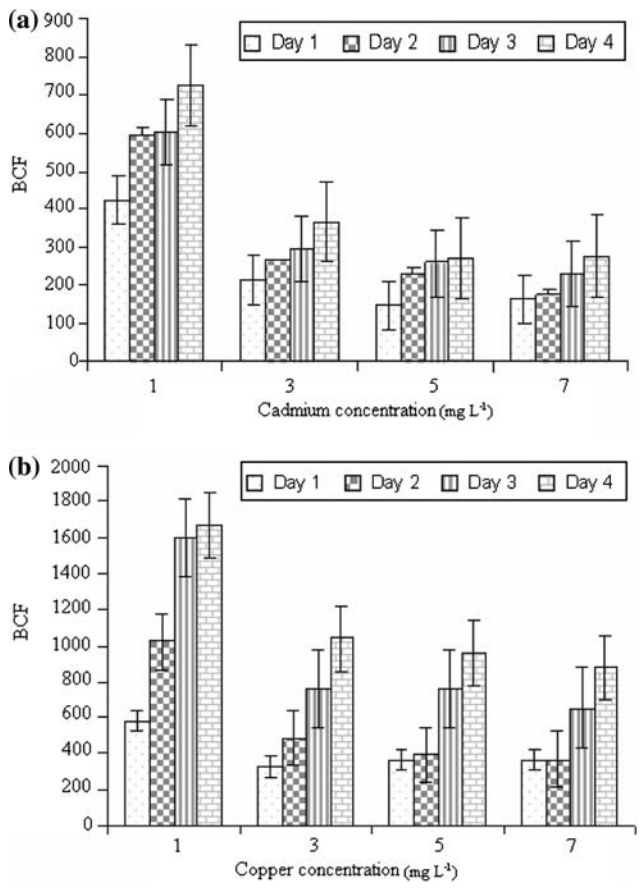

Fig. 2 The bioconcentration factor (BCF) values of (**a**) Cd and (**b**) Cu in *G. densa* at different metal concentrations and exposure times

All experiments were carried out in duplicate and the results were analyzed using the Minitab statistical package program 13.0.

Results and Discussion

The removal rates of heavy metals by *G. densa* are listed in Table 1. These data are indicative of two important findings. First; the uptake of heavy metals by *G. densa* generally increased with increasing metal concentrations and changing time ($p < 0.05$). Second; Cu is more accumulate than Cd by *G. densa.* These results are in accordance with other results as presented in Table 1.

A good heavy metal accumulator should have BCF of more than 1,000 (a 100-fold compared on a fresh weight or in vivo basis) (Zayed et al. 1998). BCF values of copper were higher when compared with that of cadmium in *G. densa* (Fig. 2a, b). The higher value of BCF indicates that *G. densa* is more effective for removal of copper. According to BCF values; the more accumulation has materialized in low concentrations and first day, then the accumulation has gradually decreased in following days. The maximum values of bioconcentration factor (BCF) were founded 724 for Cd and 1,669 for Cu. Bioconcetration factors reported for Cd and Cu in aquatic plants in Table 2. Especially wetland plants are being used successfully for the removal of metals in wastewaters. The use of *G. densa* to clean the contaminated environment with

Table 2 Bioconcentration factors (BCF) for Cd^{++} and Cu^{++} in various plants

Plant species	BCF (Cu^{++})	References	BCF (Cd^{++})	References
G. densa	724	Present study	1669	Present study
Eichhornia crassipes	622	(Lu et al. 2004)	595	(Zhu et al. 1999)
Lemna polyrrhiza	650	(Jain et al. 1990)	–	–
Lemna minor	12600	(Kwan and Smith 1991)	–	–
E. crassipes	2150	(Zhu et al. 1999)	–	–
Lemna minor	850	(Zayed et al. 1998)	–	–
H. reticulatum	6250	(Rai et al. 1995)	2481	(Rai et al. 1995)
S. polyrrhiza	5750	(Rai et al. 1995)	36500	(Rai et al. 1995)
C. demersum	3333	(Rai et al. 1995)	53333	(Rai et al. 1995)
V. spirali	2375	(Rai et al. 1995)	2009	(Rai et al. 1995)
B. monnieri	29000	(Rai et al. 1995)	18750	(Rai et al. 1995)
A. sessilis	23000	(Rai et al. 1995)	1051	(Rai et al. 1995)
H. aristata	4600	(Rai et al. 1995)	211	(Rai et al. 1995)
C. corollina	–	–	1103	(Rai et al. 1995)
C. demersum	3027	(Gupta and Chandra 1996)	–	–

metals are effective especially for cupper and cadmium as proved by this experimental study and can be applied in the field. This type of experimental studies on different plants for the removal of the heavy metals should be encouraged so that the cost effective method of cleaning the polluted environment around the world can be performed.

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References

Axtell NR, Sternberg SPK, Claussen K (2003) Lead and nickel removal using *Microspora* and *Lemna minor*. Bioresour Technol 89:41–48

Gupta P, Chandra P (1996) Presponce of cadmium to *Ceratophyllum demersum* L., A rootless submerged plant. Waste Manag 16(4):335–337

Gupta P, Chandra P (1998) Bioaccumulation and toxicty of mercury in rooted-Submerged macrophyte *Vallisneria spiralis*. Environ Pollut 103:327–332

Jain SK, Vasusevan P, Jha NK (1990) *Azolla pinnata* R.Br. and *Lemna minor* L. for removal of lead and zinc from polluted water. Water Res 24(2):177–183

Kacar B (1972) Plant and analyzis chemical of soil: II, practice book: 155, 646s. Ankara Univ, Agriculture Fac., Ankara, p 453

Kwan KHM, Smith S (1991) Some aspect of the kinetics of cadmium and thallium uptake by fronds of *Lemna minor* L. New Phytol 117:91–102

Lu X, Kruatrachue M, Pokethitiyook P, Homyok K (2004) Removal of cadmium and zinc by water hyacinth *Eichhornia crassipes*. Sci Asia 30:93–103

Memon AR, Ozdemir A, Aktoprakgil D (2001) Heavy metal accumulation in plants. J Biotechnol Biotech 15:44–48

Prasad MN, Malec P, Waloszek A, Bojke M, Strzalka K (2001) Physiological responses of *Lemna trisulca* L. (duckweed) to cadmium and copper bioaccumulation. Plant Sci 161:881–889

Rai UN, Sinha S, Tripathi RD, Chandra P (1995) Wastewater treatability potential of some aquatic macrophytes: removal of heavy metals. Ecol Eng 5:5–12

Soltan ME, Rashed MN (2003) Laboratory study on the survival of water hyacinth under several conditions of heavy metal concentrations. Adv Environ Res 7:321–334

Srivastav RK, Gupta SK, Nigam KDP, Vasudevan P (1994) Treatment of chromium and nickel in wastewater by using aquatic plants. Water Res 28(7):1631–1638

Zayed A, Gowthaman S, Terry N (1998) Phytoaccumulation of trace elements by wetlands. I. Duckweed. J Environ Qual 27:339–344

Zhu YL, Zayed AM, Qian JH, Souza M, Terry N (1999) Phytoaccumulation of trace elements by wetlands. II. Water hyacinth. J Environ Qual 28:1339–1344