Duckweed (Lemna minor) a Potent Iron Hyperaccumulator in Hokersar Wetland, an Important Ramsar Site of Kashmir Himalaya

Syed Shakeel Ahmad, Zafar A. Reshi,
Manzoor A. Shah and Irfan Rashid

Research Journal of Agricultural Sciences
An International Journal

P- ISSN: 0976-1675
E- ISSN: 2249-4538

Volume: 12
Issue: 06

Res. Jr. of Agril. Sci. (2021) 12: 2244–2247

CARAS

Res. Jr. of Agril. Sci. (Nov-Dec 2021) 12(6): 2244–2247
www.rjas.org

Full Length Research Article

Duckweed (*Lemna minor*) a Potent Iron Hyperaccumulator in Hokersar Wetland, an Important Ramsar Site of Kashmir Himalaya

Syed Shakeel Ahmad*[1], Zafar A. Reshi[2], Manzoor A. Shah[3] and Irfan Rashid[4]

Received: 04 Sep 2021 | Revised accepted: 21 Nov 2021 | Published online: 15 Dec 2021

ABSTRACT

Heavy metals are important class of contaminants having hazardous impact on plants animals and humans. Wetland macrophytes offer important, cheap and natural alternatives for the removal of heavy metals from the contaminated environment; however, macrophytes vary in their potential to sequester these heavy metals. In this context the present study was carried out to investigate the metal removal capability of *Lemna minor* growing densely in the Hokersar wetland of the Kashmir, an important Ramsar site of Kashmir Himalaya. The highest Bioconcentration factor BCF of *Lemna minor* also corresponded to Fe which also supports the good phytoremediation capability of this species. Our results suggest that *Lemna minor* is a potent wetland macrophyte that can be used for removal of Fe from the contaminated soils.

***Key words*:** *Lemna minor*, Heavy metal, Wetland, Removal, Avifauna

Fast urbanization, industrialization, fertilizer and pesticide use has resulted in heavy metal pollution of land and water resources. The increasing load of heavy metals has caused imbalance in aquatic ecosystems and the biota growing under such habitats accumulate high amounts of heavy metals (Cu, Zn, Cd, Cr and Ni etc.) which in turn, are being assimilated and transferred within food chains by the process of magnification [1]. Heavy metals are a serious problem because they cannot be degraded to organic material, thus they accumulate in water, soil, bottom sediment, and living organisms [2]. Although it may be possible to reduce the toxicity of certain metals by influencing their speciation, they do not degrade and are persistent in the environment [3]. Several technologies are available to remediate soils that are contaminated by heavy metals. However, many of these technologies are costly (e.g., excavation of contaminated material and chemical/physical treatment) or do not achieve a long-term nor aesthetic solution [4]. Phytoremediation can provide a cost-effective, long-lasting and aesthetic solution for remediation of contaminated sites. In the field of ecotoxicology, *L. minor* has been used for the removal of heavy metals from waste water and constructed wetlands [5-7]. This species presents the additional advantage of growing under varied environmental conditions with rapid growth rates. Species of *Lemna* are reported to accumulate toxic metals and therefore are being used as experimental model systems to investigate heavy metal induced responses [8-10]. Under experimental conditions, duckweed act as a good accumulator of Cd, Se and Cu, a moderate accumulator of Cr, and a poor accumulator of Ni and Pb [11]. The toxicity effect of each trace element on plant growth was in the order: Cu > Se > Pb > Cd > Ni > Cr. He also concluded that duckweed showed promise for the removal of Cd, Se and Cu from contaminated wastewater since it accumulated high concentrations of these elements. Not much work has been done on phytoremediation of heavy metals in Kashmir Himalayan wetlands except a few attempts in recent past [32-35]. In this context the present study was carried out to study the phytoremediation potential of a common macrophyte *Lemna minor* of Hokersar wetland of Kashmir Himalayas.

* **Syed Shakeel Ahmad**
ssahmad900@gmail.com

[1-4] Department of Botany, University of Kashmir, Nazratbal - 190 006, Srinagar, Jammu and Kashmir, India

MATERIALS AND METHODS

Study species

Lemna minor L. (common duckweed or lesser duckweed) belongs to the family Lemnaceae. It is a free-floating fresh water aquatic plant with one, two or three leaves each with a single root hanging in the water. As more leaves grow, the plants divide and become separate individuals. It grows in water with high nutrient levels, pH between 5 to 9, and temperature of 6 to 33°C.

Chemical analysis

Water samples (50mL) were digested with 2M HNO_3 at 95°C for 2hr and were made up to 100mL in volumetric flask with demineralized water. The digestion was done in glassware previously soaked in nitric acid and washed with demineralized water. The digested samples were analyzed for metals in duplicate using AAS Perkin Elmer, model Analyst 800. Sample blanks were also analyzed to correct for any contamination in the course of analysis. The plant samples were washed with distilled water in the laboratory. The washed plant samples were oven dried at 60°C till well dried. The dried samples were weighed and ground to pass a 40 mesh screen using a Wiley mill. The analysis of plants was carried out by acid digestion of dry samples with an acid mixture (9 parts nitric acid: 4 parts perchloric acid) at about 80°C. All the reagents that were used were of analytical grade and the reaction vessels were treated well to avoid external contributions of the metals. Sample blanks were analyzed to correct for possible external contributions of the metals while replicate samples were also evaluated and all the analyses were done in triplicate to ensure reproducibility of results. The digested samples were analyzed for ten metals (, Al, Cd, Co, Cr, Cu, Fe, Mn, Ni, Pb and Zn) using AAS Perkin Elmer, model Analyst 800.

Data analysis

The mean and standard error of mean of the metals in plant sample parts were calculated. Translocation factor which is a ratio of the concentration of metals (μg/g) in the plant shoot and leaves to that of the metals in root (μg/g) was also evaluated while the enrichment factor measuring the degree of enrichment and the transfer of metals into the plant sample by the water was evaluated by the ratio of the metal in the plant (μg/g) to the metal in the water (mg/L).

Enrichment factor (EF)

Enrichment factor is an index of the ability of the plant to accumulate metal ions with respect to the metal ion concentration in the growth environment. It is defined as the ratio of metal concentration in the dry biomass to the initial concentration of metal ion in the water.

$$\text{Enrichment factor} \quad \frac{\text{Concentration of metal in the plant tissue}}{\text{Concentration of the metal in the water}}$$

RESULTS AND DISCUSSION

The heavy metal concentration in *L. minor* was found in the order of Fe>Al>Mn>Pb>Cu>Zn>Co>Cr>Ni>Cd. The primary criteria for a plant to qualify as a good phytoremediation plant (hyperaccumulator) is that it should withstand very high concentrations of heavy metals (10,000 μg/g for Fe, Al, Zn and Mn; above 1000 μg/g dry mass for Pb, Cu, Ni and Co; and 100 μg/g for Cd [12-13]. The critical concentrations of the metals investigated in the present study were lower than the required values for all the studied heavy metals except Fe, thereby indicating that *Lemna minor* is a hyperaccumulator of Fe while an accumulator of other studied heavy metals. The lower values obtained for the metal concentrations in *Lemna minor* as against characteristic hyperaccumulators, could be due to lower concentration of heavy metals in the studied wetland waters (Table 1). EF is also an important index that determines the effectiveness of plant to be a good phytoremediation plant. EF greater than one is considered an important criterion of hyperaccumulators [14]. The order of enrichment factor (EF) of different heavy metals in *Lemna minor* was Fe>Cd>Mn>Co>Cr>Zn>Ni>Al>Cu. The highest EF was obtained for Fe metal (2390.78). This indicates that Fe is the most transferred metal into this species followed by Cd and Mn thus reducing the supply of Fe, Cd and Mn to marsh detrivores, avifauna, other bioaccumulators and surface waters. It has been hypothesised that high tissue iron concentrations carry an increased risk of neoplasia [15], atherosclerotic disorders [16-17], infection [17], neurodegenerative disorders [18], and inflammatory conditions [19]. Other conditions that have been associated with high iron intakes include symptoms similar to Alzheimer's disease, Parkinson's disease, arthritis, diabetes mellitus. Iron (II) sulfate heptahydrate, the most toxic form of the iron salts compounds, is moderately toxic to aquatic invertebrates and slightly toxic to fish [20]. Cd is another toxic metal that accumulates in kidneys and is usually associated with induction of hypertension, cirrhosis of the liver etc. [21]. The International Agency for Research on Cancer (IARC) has classified Cd as a human carcinogen (group I) on the basis of sufficient evidence in both humans and experimental animals. In addition to this Co can cause adverse health effects at high. The neurological effects of inhaled manganese have been well documented in humans chronically exposed to elevated levels in the workplace [22-25].

Table 1 Heavy metal accumulation (mg/kg) and EF in *Lemna minor* and water of Hokersar wetland of Kasmir

Heavy metal	Concentration in *Lemna minor*	Heavy metal in water	Heavy metal in sediment	Enrichment factor	Bioaccumulation factor
Pb	21.3 ± 1.05	0.06 ± 0.00	189.35 ± 13.10	343.55	0.11
Fe	14010 ± 212.13	5.85 ± 1.06	3250 ± 225.03	2390.78	4.31
Mn	166.6 ± 1.35	0.67 ± 1.06	1448 ± 6521.62	248.66	0.12
Al	502.7 ± 7.89	7.85 ± 0.78	9278.9 ± 383.79	64.04	0.05
Cr	4.58 ± 0.10	0.02 ± 0.00	109.02 ± 2.96	183.2	0.04
Co	5.15 ± 0.14	0.01 ± 0.00	125.9125 ± 3.155	206.00	0.04
Zn	9.96 ± 1.64	0.06 ± 0.00	177.45 ± 3.22	166.00	0.06
Cu	10.88 ± 2.71	0.30 ± 0.01	452.46 ± 48.19	36.27	0.02
Cd	3.41 ± 0.31	0.004 ± 0.00	24.835 ± 1.935	852.50	0.14
Ni	3.39 ± 0.04	0.03 ± 0.00	234.68 ± 31.31	113.00	0.01

BCF is the defining parameter in phytoremediation providing information on the uptake of metal, its mobilization into the plant tissues and storage in the plant parts. BCF greater than one is indicative of a potential HM-hyperaccumulator species [26]. The BCF of *L. minor* was found to be in the order of

Fe>Cd>Mn>Pb>Zn>Al>Co>Cr>Cu>Ni. The highest BCF was obtained for Fe (4.31). Duckweed is commonly found in wetlands, is fast growing, adapts easily to various aquatic conditions, and plays an important role in the extraction and accumulation of metals from waters [27]. *L. minor* has been found to be a potent wetland for the removal of heavy metals [7], [28] particularly Cd and Pb [29-30], and Fe [31]. Thus, the EF and BCF values of *L. minor* for different metals suggest that *L. minor* is a potent wetland plant for the removal of Fe from contaminated soils.

CONCLUSION

The results suggest that *L. minor* has the potential to remove Fe, Cd and Mn from the contaminated water and it lowers these heavy metals in the surrounding water and their availability to marsh detrivores, avifauna and other trophic levels including humans. The *L. minor* acts as a hyperaccumulator of Fe while an accumulator of other studied heavy metals. Thus *L. minor* is an efficient and a potent macrophyte for the removal of different heavy metals particularly Fe. It can be easily inoculated in constructed wetlands for the removal of different heavy metals particularly Fe.

Acknowledgements

We are thankful to Head, Department of Botany and Director, USIC, University of Kashmir, Jammu and Kashmir, India, for providing necessary facilities during the course of this study.

LITERATURE CITED

1. Pergent C, Pergent-Martini C. 1999. Mercury levels and fluxes in *Podosonia oceanica* meadows. *Environmental Pollution* 106: 33-37.
2. Brankovic S, Pavlovic-Muratspahic D, Topuzovic M, Glisic R, Milivojevic J, Dekic V. 2012. Metals concentration and accumulation in several aquatic macrophytes. *Biotechnol Biotechnological Equip*. 26: 2731-2736.
3. NRC. 1999. Metals and radionuclides: technologies for characterization, remediation, and containment. *In*: Groundwater and soil cleanup: improving management of persistent contaminants. Washington, DC, National Academy Press. pp 72-128.
4. Cao X, Ma LQ, Chen M, Singh SP, Harris WG. 2002. Impacts of phosphate amendments on lead biogeochemistry in a contaminated site. *Environ. Sci. Technology* 36: 5296-5304.
5. Maine M, Duarte M, Sufie N. 2001. Cadmium uptake by floating macrophytes. *Water Research* 35: 2629-2634.
6. Uysal Y, Taner F. 2007. The effect of cadmium ions on the growth rate of the freshwater macrophyte duckweed *Lemna minor*. *Ekoloji* 16: 9-15.
7. Rai PK. 2008. Heavy metal pollution in aquatic ecosystems and its phytoremediation using wetland plants: An eco-sustainable approach. *Int. Jr. Phytorem*. 10: 133-160.
8. Jafari N, Akhavan M. 2011. Effect of pH and heavy metal concentration on phytoaccumulation of zinc by three duckweed species. *American – Eur. Jr. Agr. Environ. Science* 10(1): 34-41.
9. Rahmani GNH, Sternberg SPK. 1999. Bioremoval of lead from water using *Lemna minor*. *Biores. Technology* 70: 225-230.
10. Severi A. 1997. Aluminum toxicity in *Lemna minor* L.: effects of citrate and kinetin. Environ Exp Bot. 37: 53–61
11. Zayed A, Gowthaman S, Terry N. 1998. Phytoaccumulation of trace elements by wetland plants: I. Duck weed. *Jr. Env. Qual*. 27: 715-721.
12. Baker AJM, Brooks RR. 1989. Terrestrial higher plants which hyperaccumulate metallic elements – a review of their distribution, ecology and phytochemistry. *Biorecovery* 1: 81-126.
13. Srivastava M, Ma LQ, Santos JAG. 2006. Three new arsenic hyperaccumulating ferns. *Sci. Total Eniron.* 364: 24-31.
14. Wei SH, Zhou, QX, Koval PV. 2006. Flowering stage characteristics of cadmium hyperaccumulator *Solanum nigrum* L. and their significance to phytoremediation. *Sci Total Environ*. 369: 441-446.
15. Nelson RL. 1992. Dietary iron and colorectal cancer risk. *Free Radic. Biol. Med*. 12: 161-168.
16. Sullivan JL. 1981. Iron and the sex difference in heart disease risk. *Lancet* 1: 1293-1294.
17. Kent S, Weinberg E. 1989. Hypoferremia: adaptation to disease? *N. Engl. Jr. Med*. 320: 672.
18. Thompson KJ, Shoham S. 2001. Iron and neurodegenerative disorders. *Brain Res. Bulletin* 55: 155-164.
19. Halliwell B. 1984. Oxygen toxicity, oxygen radicals, transition metals and disease. *Biochemistry Journal* 219: 1-14.
20. USEPA. 1993. *Iron Salts*. Office of Prevention, Pesticides and Toxic Substances (7508W). EPA-738-F-93-002.
21. Higgins IJ, Burns FG. 1975. *The Chemistry and Micro of Pollution*. London: Academic Press.
22. ATSDR. 2000. Toxicological profile for manganese. Atlanta, GA, United States Department of Health and Human Services, Public Health Service, Agency for Toxic Substances and Disease Registry.
23. Canavan MM, Cobb S, Srinker C. 1934. Chronic manganese poisoning. *Archives of Neurology and Psychiatry* 32: 501-512.
24. Cook DG, Fahn S, Brait KA. 1974. Chronic manganese intoxication. *Archives of Neurology* 30: 59-64.
25. Roels J, Huyghe G, Verstraete W. 2004. Microbially mediated phosphine emission. *Science of the Total Environment* 338: 253-265.
26. Zhang GP, Fukami M, Sekimoto H. 2002. Influence of cadmium on mineral concentrations and yield components in wheat genotypes differing in Cd tolerance at seedling stage. *Field Crops Research* 77: 93-98.

27. Prasad MNV, Freitas HMO. 2003. Metal hyperaccumulation in plants - Biodiversity prospecting for phytoremediation technology. *Elec. Jr. Biotechnology* 6(3): 225-321.
30. Kara Y, Basaran D, Kara I, Zeytunluoglu A, Genc H. 2003. Bioaccumulation of nickel by aquatic macrophyta *Lemna minor* (Duckweed). *Int. Jr. Agr. Biology* 5(3): 281-283.
29. Chaudri D, Majumdar A, Misra AK, Bandyopadyay K. 2013. Cadmium removal by *Lemna minor* and *Spirodela polyrhiza*. *Int. Jr. Phtorem*. DOI: 10.1080/15226514.821486
30. Wang H, Wong MH, Lan C, Baker AJM, Qin Y, Chen G, Shu W, Ye ZH. 2007. Uptake and accumulation of arsenic by 11 *Pteris* taxa from southern China. *Environ. Pollution* 145: 225-233.
31. Teixeira S, Vieira MN, Esphinha J, Marques, Pereira R. 2013. Bioremediation of iron rich mine effluent by *Lemna minor. Intern. Jr. Phtorem.* DOI: 10.1080/15226514.821454.
32. Ahmad SS, Reshi ZA, Shah MA, Rashid I, Ara R, Andrabi SMA. 2013. Phytoremediation potential of *Phragmites australis* in Hokersar wetland- A Ramsar site of Kashmir Himalaya. *Int. Jr. Phytorem*. DOI: 10. 1080/15226514.2013.821489
33. Ahmad SS, Reshi ZA, Shah MA, Rashid I. 2016. Constructed wetlands: role in phytoremediation of heavy metals. *In*: (Eds) Abid A. Ansari, Sarvajeet Singh Gill, Ritu Gill, Guy R. Lanza, and Lee Newman. *Phytoremediation*. Volume 3. Published by Springer Nature.
34. Ahmad SS, Reshi ZA, Shah MA, Rashid I, Ara R, Andrabi SMA. 2014. Phytoremediation potential of *Phragmites australis* in Hokersar wetland—a Ramsar site of Kashmir Himalaya. *Int. Jr. Phytorem*. 16: 1183-1191.
35. Ahmad SS, Reshi ZA, Shah MA, Rashid I, Ara R, Andrabi SMA. 2015. Heavy metal accumulation by *Potamogeton natans* and *Ceratophyllum demersum* in a Himalayan RAMSAR site: management implications. Wetlands Ecol. Manage. DOI 10.1007/s11273-015-9472-9.