Cu and Pb Accumulation and Removal From Aqueous Medium by *Enydra Fluctuans* DC. (Asteraceae) - A Medicinal Plant With Potential for Phytoremediation

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Abstract

Enydra fluctuans DC. (Asteraceae) is an edible semi-aquatic floating or trailing herbaceous plant widely distributed in tropical Africa, South and South East Asia, and Australia. Its leaves, which are consumed as a vegetable, are also used in traditional medicine to treat several diseases. The efficacy of this plant in removal of copper and lead from aqueous medium was tested in the present study. Accumulation of both Cu and Pb was significantly higher in root than that in leaf and stem. Though all the bioconcentration factor (BCF) values were greater than unity, none of the translocation factor (TF) values was greater than unity, indicating that this plant could not be considered a hyperaccumulator of these metals.

Nevertheless, *E. fluctuans* could remove Cu from aqueous medium at rates ranging from 98.8–99.7 %, with a mean reduction of 99.2 % after 96 h exposure at various concentrations. The removal of Pb ranged from 97.1–99.1 %, with a mean reduction of 98.2 %. Thus, *E. fluctuans* showed high potential in removal of Cu and Pb from aqueous medium, and has the prospect of being used in phytoremediation of these metals.

Introduction

Various human activities such as industrialization and urbanization accompanied by rapid population growth have added different types of pollutants into the freshwater ecosystems (CPCB 2008). Among these pollutants, heavy metals are of worldwide concern because of their toxic properties, tendency to accumulate in biota, and persistent nature (Rai et al. 1981; Lokeshwari and Chandrappa 2007; Mishra and Tripathi 2008; Chang et al. 2009; Sood et al. 2012; Verma and Suthar 2015). Heavy metals and metalloids such as arsenic (As), cadmium (Cd), chromium (Cr), copper (Cu), mercury (Hg), lead (Pb), zinc (Zn) and others can cause deleterious effects even at very low concentrations (Sood et al. 2012). Different sources such as smelters, tanneries, textile, fuel production, military activities, chemical industry, mining, application of fertilizer, urban sewage, waste incineration, and unsafe disposal of hazardous industrial wastes release various toxic heavy metals into the environment (Mishra and Tripathi 2008; Verma and Suthar 2015; Hargreaves et al. 2018; Ali et al. 2020). Some conventional techniques that are used for remediation of heavy metal pollutants include oxidation, chemical precipitation, adsorption, coagulation-flocculation, ion exchange, membrane filtration, ozonation, hydrogen peroxide-based methods, photocatalytic degradation, reverse osmosis, electrodialysis and electrochemical methods. However, these are mostly metal specific, expensive, energy-intensive, and often have adverse effects on the environment (Mishra and Tripathi 2008; Olguin and Sánchez-Galván 2012; Lara et al. 2014; Rezanian et al. 2016). Under these circumstances, it is necessary to identify broad-spectrum, simple and cost-effective technologies for removal of toxic metals (Zhang et al. 2020). The idea of using plants to cleanup pollutants from the environment was introduced in 1983, though this method was being applied for the last 300 years (Ali et al. 2020). These technologies are also named as green remediation, botanoremediation, vegetative remediation, and agro remediation (Sarwar et al. 2017; Kushwaha et al. 2018). Therefore, phytoremediation technologies to clean up environmental contaminants from water, soil and air using living plants (Raskin and Ensley 2000; Terry and Banuelos 2000; Maletić et al. 2019; Moya et al.

2019) comprise viable, innovative, non-intrusive and cost-effective alternatives to other conventional methods (Singh et al. 1996; Miretzky et al. 2004; Putra et al. 2015). Plants that are used in the process of phytoremediation take up heavy metals through their roots and later transfer these contaminants to the above-ground parts of their body (Ashraf et al. 2018).

Aquatic macrophytes are of structural and functional significance in aquatic ecosystems because they provide stable habitats, and are a source of food and oxygen to the macroinvertebrate and fish fauna. They contribute to nutrient cycling, improve water quality by regulating oxygen balance and also play an important role in heavy metal accumulation (Srivastava et al. 2008; Dhote and Dixit 2009). Their high biomass yield, fast growth rate, high tolerance, ability to accumulate heavy metals, and direct exposure to contaminated water, facilitate their remediation ability, which enables them to act as natural water filtration systems (Sood et al. 2012). The roots, shoots, and leaves of aquatic macrophytes have the capacity to absorb heavy metals from aquatic environments, while their roots have the potential to sequester selected heavy metals (Mishra et al. 2009; Paiva et al. 2009; Muffarrege et al. 2010). Heavy metal uptake by aquatic plants involve three broad mechanisms: (i) accumulation, whereby plants concentrate and transport metals from water into plant body such as roots; (ii) binding of metals to cell walls of roots, thereby restricting their entry into the plant; and (iii) confining to the roots the metals that are still able to enter the plant, thus restricting their transport to shoots (Mishra and Tripathi 2008).

Various aquatic plants like duckweed (*Lemna minor*) (Mo et al. 1989), pennywort (*Hydrocotyle umbellata*) (Dierberg et al. 1987), water velvet (*Azolla pinnata*) (Jain et al. 1989), water hyacinth (*Eichhornia crassipes*) (Mishra and Tripathi 2008), water lettuce (*Pistia stratiotes* L.) (Putra et al. 2015), *Ipomoea aquatica* (Khumanleima Chanu and Gupta 2014; Bedabati Chanu and Gupta 2016; Haokip and Gupta 2020), and several other species have been reported to remove various heavy metals from solution. *Enydra fluctuans* DC. (Asteraceae), which is a floating or trailing macrophyte has been selected in the present study for testing its efficacy in removal of copper and lead from aqueous medium. This plant, which is commonly available in the study area of Barak Valley, South Assam, is also widely distributed in tropical Africa, South and South East Asia, and Australia. It is an edible, semi-aquatic herbaceous vegetable plant with serrate leaves (Rahman et al. 2002; Sannigrahi et al. 2011; Dua et al. 2015). The slightly bitter leaves are extensively used in traditional medicine to treat small-pox, inflammation and skin diseases (Kirtikar and Basu 2002). It is also used as an antibilious agent for treating liver disorders and neural diseases (Chopra et al. 2000; Kuri et al. 2014); anthelmintic and anti-inflammatory (Ali et al. 1972); analgesic (Rahman et al. 2002); and hypotensive (Kuri et al. 2014). Furthermore, it is packed with vitamins and other nutrients (Dua et al. 2015). The plant grows abundantly in rice fields, ditches, drains, natural channels and edges of fish ponds, where they propagate by fragmentation and often choke the water courses (Ali et al. 2013). However, its potential for removal of heavy metals from aqueous medium has not been assessed to any great extent, though it is known to have the ability to remove the metalloids arsenic and boron from water (Shaheen et al. 2006, 2007).

The two heavy metals copper and lead were selected for several reasons. Copper (Cu) is an essential micronutrient for the growth of plants and a constituent trace nutrient of the protein component of

several enzymes, most of which are involved in catalyzing redox reactions, electron flow, etc. However, Cu can be toxic at high concentrations that are extremely harmful to both plants and animals (Devi and Prasad 1998; Wang and Dei 2001; Khumanleima Chanu and Gupta 2014). Due to direct exposure to its toxic effects, many aquatic plants are more sensitive to copper than their terrestrial counterparts (Fernandes and Henriques 1991). Cu is commonly used as a fungicide in tea plantations and different agricultural lands in the study area (Khumanleima Chanu and Gupta 2014). Lead at concentrations of more than 0.03 mg/100 gm (FAO/WHO 2001) can cause blood diseases and affect nervous system, teeth and gum, liver, pancreas, and bones in humans (Khan 2015). Lead is dense, soft, durable, and corrosion-resistant with a relatively low melting point, making it a major constituent of paint, ammunition, leaded glass, solder, storage batteries, etc. (Abadin et al. 2007). The principal consumption of lead (i.e., ~ 80% of the total use of lead) is for the lead-acid storage battery, and untreated wastewater containing lead used in recycling of batteries is released into the water bodies in the study area (Bedabati Chanu and Gupta 2016). In developing countries, there is a rapid increase in the demand for lead batteries due to an increase in the number of motorized vehicles, solar panels, telecommunication and computers (OK International 2021). A review of the available scientific literature reveals that the Cu and Pb accumulation and removal by *E. fluctuans* are not documented. Considering the above facts, the present paper is an attempt to critically evaluate the potential of this plant to accumulate Cu and Pb in its different tissues, and to remove these heavy metals from aqueous medium.

Materials And Methods

Plant material and culture of plants

Enydra fluctuans DC. was collected from water bodies of Irongmara area (latitude: 24.689 °N, longitude: 92.742 °E) in Cachar district, Assam, India. The plant material was brought to the laboratory in a plastic bag and washed with tap water. The plants were grown in hydroponic culture system till they developed new branches. These new branches were cut and grown in soil flooded with 50% modified Hoagland nutrient solution with its pH maintained at 5.8–6.2 (Göthberg et al. 2004; Bedabati Chanu and Gupta 2016). Hoagland nutrient medium comprised of 1 mM $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$, 2.5 mM $\text{Ca}(\text{NO}_3)_2 \cdot 4\text{H}_2\text{O}$, 0.5 mM KH_2PO_4 , 2.5 mM KNO_3 , 23 μM H_3BO_3 , 0.39 μM $\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$, 4.58 μM $\text{MnCl}_2 \cdot 4\text{H}_2\text{O}$, 0.16 μM $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$, 0.05 μM $\text{Na}_2\text{MoO}_4 \cdot 2\text{H}_2\text{O}$ and 50 μM Fe-EDTA (Hoagland and Arnon 1950). To avoid Pb precipitation, KH_2PO_4 was not added to the nutrient solution (Qiao et al. 2013, Bedabati Chanu and Gupta 2016). The control plants were also cultured in 50% Hoagland nutrient medium without KH_2PO_4 .

Chemicals

The metals used in the present study were copper as cupric chloride ($\text{CuCl}_2 \cdot 2\text{H}_2\text{O}$) and lead as lead nitrate ($\text{Pb}(\text{NO}_3)_2$). The metals were added to the 50 % Hoagland nutrient medium to obtain different graded concentrations for performing the tests. The nutrient solution without added Cu and Pb served as control.

Experimental Methods

Fully grown healthy shoots of similar height (20–25 cm long) were cut from the same mother plant, washed with tap water and placed in 50 % Hoagland nutrient media for 1 week (Temperature: 22–25°C; dissolved oxygen: 6.28–6.87 mg L⁻¹; electrical conductivity: 99.6–107 µS cm⁻¹; pH: 5.96–6.5) to produce new roots. After the acclimatization period, plants of similar shoot height were exposed to Cu and Pb in two sets of experiments. Plants were exposed for 96 h to graded concentrations of Cu (nominal concentrations: 1.05, 3.3, 6.2 and 11.2 mg L⁻¹; corresponding measured concentrations: 0.55, 2.9, 5.7 and 10.2 mg L⁻¹) and Pb (nominal concentrations: 12.5, 22.0, 37.2 and 52.5 mg L⁻¹; corresponding measured concentrations: 11.5, 20.1, 35.7 and 50.2 mg L⁻¹) in 50 % Hoagland medium contained in hydroponic microcosms consisting of 1000 ml conical flasks. Cu and Pb concentrations in Indian rivers ranged from 0.003–314.93 and 0.010–374.58 µg L⁻¹, respectively, while Cu and Pb concentrations in rivers of the study area could reach up to 134.25 and 19.76 µg L⁻¹, respectively (CWC 2014, 2018). Laboratory acclimatized plants without added Cu and Pb served as control in all the experiments. The plant parts were harvested after 3 h, 24 h, 48 h and 96 h of the experiment for determining uptake and accumulation of Cu and Pb.

Estimation of Cu and Pb

The Cu and Pb content in the plant samples were estimated using standard methods (Gupta 1996; Khumanleima Chanu and Gupta 2014). As soon as the experiments were completed, the leaf, stem and root were separated and cut into small pieces, and after drying to a constant weight at 60 ± 2°C, were ground and digested with concentrated HNO₃. The residues were dissolved in deionized water and the concentrations of Cu and Pb were estimated in a graphite furnace atomic absorption spectrophotometer (model-Analytic Jena Vario-6) at the Sophisticated Analytical Instrument Facility (SAIF), North Eastern Hill University (NEHU), Shillong. Cu and Pb concentrations were also determined in samples of test medium collected from the microcosms at 0, 3, 24, 48 and 96 h.

Bioconcentration and Translocation Factor

The ability of the plant to absorb Cu and Pb from an aqueous medium and its capacity to translocate them from the roots to different tissues of the plant are expressed by the Bioconcentration Factor (BCF) and the Translocation Factor (TF), respectively. BCF and TF were calculated by the following formulae:

$$BCF = \frac{\text{Metal concentration in } \mu\text{g g}^{-1} \text{ dry weight (dw) plant tissue}}{\text{Initial concentration of metal in the medium (mg L}^{-1}\text{)}}$$

(Ivanciuc et al. 2006; Ahmad et al. 2011), and

$$TF = \frac{\text{Metal concentration in stem or leaf}(\mu\text{g g}^{-1}\text{dw})}{\text{Metal concentration in root}(\mu\text{g g}^{-1}\text{dw})}$$

(Ahmad et al. 2011).

Statistical analysis

The data sets were subjected to One-way ANOVA to find the statistical significance of differences among them, with multiple comparisons made by Tukey tests. SPSS 20 software was used for performing all tests and wherever necessary log transformation of data was also done.

Results And Discussion

Accumulation of copper and lead in different tissues of *E. fluctuans*

The accumulation of Cu in leaf, stem and root of *E. fluctuans* after 3, 24, 48 and 96 h of exposure to graded concentrations of Cu are shown in Table 1. Cu was not detected in the tissues of the control plants. One-way ANOVA with multiple comparisons by Tukey tests revealed that there were significant differences ($p < 0.001$) in Cu accumulation among the three tissues of Cu-exposed plants at all concentrations and hours of exposure, in the order root > leaf > stem. There was a consistent increase in accumulation of Cu in the root with increasing concentration and duration of exposure, although this trend was not evident in the leaf and stem. The highest Cu concentration of $980.90 \pm 20.81 \mu\text{g g}^{-1}$ dry weight (DW) was observed in the root of plants exposed to $10.2 \text{ mg Cu L}^{-1}$ at 96 h. On the other hand, the lowest accumulation was $41.04 \text{ Cu } \mu\text{g g}^{-1} \text{ DW}$ in the stem exposed to $0.55 \text{ mg Cu L}^{-1}$ at 3 h. The highest Cu concentrations recorded in the leaf and stem were 129.01 ± 0.10 and $168.55 \pm 0.39 \mu\text{g g}^{-1} \text{ DW}$, respectively, when exposed to $10.2 \text{ mg Cu L}^{-1}$ at 24 and 3 h, respectively.

Table 1
Accumulation of Cu in different tissues of *E. fluctuans* at different Cu concentrations

Cu in mg L ⁻¹	Plant tissue	Cu accumulation (µg g ⁻¹ DW)			
		3h	24h	48h	96h
Control	leaf	ND	ND	ND	ND
0.55		52.78 ± 2.04 ^a	108.70 ± 1.41 ^a	83.00 ± 2.52 ^a	146.34 ± 1.00 ^a
	stem	41.04 ± 1.13 ^b	90.77 ± 1.01 ^b	45.95 ± 0.86 ^b	72.63 ± 1.17 ^b
	root	90.98 ± 0.76 ^c	151.26 ± 0.36 ^c	191.69 ± 0.65 ^c	232.00 ± 0.92 ^c
2.9	leaf	72.93 ± 0.11 ^a	125.84 ± 0.70 ^a	95.19 ± 0.88 ^a	151.56 ± 0.22 ^a
	stem	52.65 ± 0.69 ^b	92.58 ± 0.42 ^b	76.43 ± 1.43 ^b	73.12 ± 1.07 ^b
	root	189.44 ± 0.44 ^c	202.23 ± 0.31 ^c	255.25 ± 2.57 ^c	375.69 ± 0.42 ^c
5.7	leaf	84.82 ± 0.18 ^a	129.57 ± 0.36 ^a	110.77 ± 3.19 ^a	115.77 ± 0.33 ^a
	stem	57.27 ± 0.77 ^b	95.59 ± 0.44 ^b	92.72 ± 0.54 ^b	75.31 ± 0.72 ^b
	root	251.41 ± 0.31 ^c	238.43 ± 0.34 ^c	325.77 ± 0.32 ^c	450.98 ± 0.86 ^c
10.2	leaf	168.55 ± 0.39 ^a	131.51 ± 0.45 ^a	107.46 ± 2.26 ^a	164.84 ± 0.68 ^a
	stem	81.87 ± 0.50 ^b	129.01 ± 0.10 ^b	95.67 ± 1.12 ^b	115.47 ± 0.73 ^b
	root	501.95 ± 0.63 ^c	606.08 ± 4.65 ^c	965.48 ± 3.48 ^c	980.90 ± 20.81 ^c
Values are given as mean ± SD; values with different superscripts in the same column indicate significant differences at P < 0.05 among the different tissues of plants exposed to a given Cu concentration; DW: dry weight; ND: not detected					

Though copper acts as an essential nutrient by playing a prominent role in enhancing photosynthesis, and promoting development, metabolism and growth of plants, it can be highly toxic beyond certain threshold concentrations (Li and Xiong 2004). Copper affects several biochemical and physiological mechanisms in plants, inhibits growth, and imparts toxic effects due to accumulation in the tissues (Mishra and Tripathi 2008). Cu accumulation was highest in the roots due to its existence as cations i.e., Cu²⁺ and Cu⁺ (Krupanidhi et al. 2008) which got attached to negative charges present in the structure of cell wall in the roots (Göthberg 2008). Khumanleima Chanu and Gupta (2014) also found that roots of *Ipomoea aquatica* accumulated higher amount of copper than the apical parts of the plant.

In the case of Pb accumulation, a pattern similar to that observed for Cu could be observed (Table 2). Pb was not detected in the control plants, and its accumulation was highest in root, followed by that in leaf

and stem. One-way ANOVA with multiple comparisons by Tukey tests revealed that there were significant differences ($p < 0.001$) in Pb accumulation among the tissues at all concentrations and hours of exposure, in the order root > leaf > stem. The highest Pb accumulation of $1093.60 \pm 4.08 \mu\text{g g}^{-1}$ DW was found in the root after 96 h exposure to $50.2 \text{ mg Pb L}^{-1}$, while the lowest of $70.40 \pm 0.74 \mu\text{g g}^{-1}$ DW was found in the stem after 3 h exposure to $50.2 \text{ mg Pb L}^{-1}$. The Pb concentrations consistently increased with increasing exposure concentration and length of exposure time in root, though this pattern was not clear in leaf and stem. The highest Pb concentrations in leaf were $194.05 \pm 1.82 \mu\text{g g}^{-1}$ DW recorded at 24 h exposure to $50.2 \text{ mg Pb L}^{-1}$, while those in stem were $114.36 \pm 0.36 \mu\text{g g}^{-1}$ DW at 96 h exposure to $20.1 \text{ mg Pb L}^{-1}$.

Table 2
Accumulation of Pb in different tissues of *E. fluctuans* at different Pb concentrations

Pb in mg L^{-1}	Plant tissue	Pb accumulation ($\mu\text{g g}^{-1}$ DW)			
		3h	24h	48h	96h
Control 11.5	leaf	ND	ND	ND	ND
	stem	93.21 ± 1.18^a	102.31 ± 1.51^a	96.87 ± 0.73^a	108.13 ± 0.91^a
	root	74.76 ± 3.33^b	71.18 ± 0.21^b	90.98 ± 0.49^b	94.96 ± 0.72^b
20.1	leaf	257.36 ± 0.58^c	247.72 ± 0.79^c	328.07 ± 0.99^c	466.75 ± 0.77^c
	stem	107.51 ± 0.61^a	111.84 ± 0.96^a	124.50 ± 1.33^a	138.83 ± 0.32^a
	root	78.47 ± 0.40^b	83.73 ± 0.97^b	92.08 ± 1.36^b	114.36 ± 0.36^b
35.7	leaf	299.31 ± 0.67^c	421.02 ± 0.13^c	505.23 ± 0.40^c	544.05 ± 1.04^c
	stem	122.28 ± 0.89^a	113.34 ± 0.63^a	132.42 ± 0.61^a	143.98 ± 0.19^a
	root	92.17 ± 1.43^b	88.82 ± 0.71^b	99.64 ± 0.44^b	105.38 ± 0.34^b
50.2	leaf	325.95 ± 1.20^c	474.21 ± 0.19^c	653.92 ± 0.89^c	944.25 ± 2.47^c
	stem	139.11 ± 1.03^a	194.05 ± 1.82^a	189.20 ± 0.21^a	168.83 ± 1.10^a
	root	70.40 ± 0.74^b	104.74 ± 0.39^b	111.64 ± 0.60^b	113.91 ± 0.79^b
Values are given as mean $\pm$ SD; values with different superscripts in the same column indicate significant differences at $P < 0.05$ among the different tissues of plants exposed to a given Pb concentration; DW: dry weight; ND: not detected		587.72 ± 3.09^c	774.02 ± 2.58^c	877.13 ± 4.47^c	1093.60 ± 4.08^c

The significantly higher accumulation of Cu and Pb in root than that in leaf and stem at all exposure concentrations could be related to the plant's strategy to cope with the toxicity of heavy metals by immobilizing them in the root cell vacuoles as shown in the case of Cr (Shankar et al. 2005b). The study also revealed that the transportation of lead from the root to the upper portion of the plant was restricted due to the high capacity of Pb retention in the root tissues (Bedabati Chanu and Gupta 2016). Other studies also reported the preferential accumulation of Pb and other heavy metals in the root of many different species of macrophytes (Lytle et al. 1998; Pulford et al. 2001; Deng et al. 2004).

Bioconcentration factor (BCF) and translocation factor (TF) of Cu and Pb in different tissues of *E. fluctuans*

All BCF values of Cu and Pb in different tissues of *E. fluctuans* at exposure periods of 3, 24, 48 and 96 h were > 1.0, with higher values recorded in root followed by those in leaf and the lowest in stem. The BCF values, which were higher for Cu than for Pb, mostly recorded an increase with time of exposure, though this pattern was less evident in the case of Pb (Supplementary Tables S1 and S3). On the other hand, the BCF values for Cu in all the three tissues at all hours of exposure mostly declined with increasing Cu concentrations, though the decline was less pronounced in the case of Pb. Thus, *E. fluctuans* was more efficient as a Cu accumulator at lower concentrations of the metal, though the BCF values remained as high as > 90 at 10.2 mg Cu L⁻¹. The BCF values ranged from 8.03 in stem – 421.82 ± 1.68 in root for Cu; and 1.40 ± 0.01 in stem – 40.59 ± 0.07 in root for Pb. Bioconcentration Factor is the ability of the plant to absorb metals into their body parts from an aqueous medium (Maldonado-Magaña et al. 2011). Higher values of BCF imply the ability of the plant to accumulate metals at higher levels (Bedabati Chanu and Gupta 2016). The extremely high BCF values for Cu (> 100) in *E. fluctuans* are similar to that found in many hyperaccumulators (Sun et al. 2009), though the low TF values rule out its inclusion in the category of hyperaccumulators. TF values for both Cu and Pb were all < 1 (Supplementary Tables S2 and S4), indicating that both these metals taken up by the plant remained confined mostly to the roots (Juang et al. 2011).

Heavy metal removal by *Enhydra fluctuans* from water

The removal of Cu from aqueous medium by *E. fluctuans* (Table 3) revealed that the final reduction after 96 h exposure at various concentrations ranged from 98.8–99.7 %, with a mean reduction of 99.2 %. The removal of Pb ranged from 97.1–99.1 %, with a mean reduction of 98.2 % (Table 4). These findings suggest that though not a hyperaccumulator, *E. fluctuans* shows high potential for removal of Cu and Pb from aqueous medium which may be aided by their higher biomass, rapid growth, and capability of metal absorption from aqueous medium as was also observed in several other species of macrophytes (Mant et al. 2007; Mishra and Tripathi 2008). Putra et al. (2015) reported that water lettuce (*Pistia stratiotes* Linn.) could remove ~ 99% of Pb from an aqueous medium.

Table 3
Cu concentrations in water samples at different hours of treatment with *E. fluctuans*. Percent reductions of Cu are given in parentheses

Cu concentrations mg L ⁻¹ (% reduction)				
0 hour	3 hour	24 hour	48 hour	96 hour
0.55 (0)	0.42 (23.6)	0.02 (96.4)	0.009 (98.4)	0.005 (99.1)
2.9 (0)	2.2 (24.1)	0.233 (92.0)	0.011 (99.6)	0.009 (99.7)
5.7 (0)	5.1 (10.5)	0.823 (85.6)	0.096 (98.3)	0.055 (99.0)
10.2 (0)	9.6 (5.9)	1.52 (85.1)	0.201 (98.0)	0.121 (98.8)

Table 4
Pb concentrations in water samples at different hours of treatment with *E. fluctuans*. Percent reductions of Pb are given in parentheses

Pb concentrations mg L ⁻¹ (% reduction)				
0 hour	3 hour	24 hour	48 hour	96 hour
11.5 (0)	9.2 (20.0)	5.8 (49.6)	0.55 (95.2)	0.33 (97.1)
20.1 (0)	18.7 (7.0)	11.6 (42.3)	0.565 (97.2)	0.392 (98.1)
35.7 (0)	29.9 (16.3)	15.7 (56.0)	0.852 (97.6)	0.572 (98.4)
50.2 (0)	47.5 (5.4)	27.9 (44.4)	0.92 (98.2)	0.46 (99.1)

Conclusions

Due to its widespread occurrence in several parts of the world, the plant *E. fluctuans* is a suitable candidate for its possible utility in phytoremediation of metal-contaminated water as well as in the assessment and biomonitoring of metals and metalloids in freshwater habitats. The present investigation revealed that the plant could survive in fairly high Cu and Pb (up to 10.2 Cu and 50.2 Pb mg L⁻¹) contaminated medium. It could also strongly accumulate these metals, especially in the roots, with BCF values for Cu reaching > 400 and those for Pb > 40. Further, the plant could remove Cu and Pb from aqueous medium at an average rate of 99.2 and 98.2 percent, respectively, after a relatively short exposure time of 96 h. These characteristic features, coupled with its ability to asexually propagate by fragmentation, makes it a plant worth exploring for its potential for phytoremediation of water contaminated with toxic metals and metalloids.

Declarations

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Authors' contributions Sultana Parven carried out the experimental work, statistical analysis, prepared the first draft and revisions of draft. Abhik Gupta and Aparajita De designed the experiment, helped in the analysis of statistics, developed the first draft and finalized.

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Data availability The datasets used and/or analyzed during the current study are available from the corresponding author on reasonable request.

Compliance with ethical standards

Ethics approval and consent to participate Not applicable

Consent for publication Not applicable

Competing interests The authors declare that they have no competing interests.

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