

**ORIGINAL RESEARCH ARTICLE**

## Submerged hydrophytes as a tool for the removal of heavy metals

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**Abstract:** Aquatic macrophytes serve as a remarkable biological filter. By absorbing the dissolved metals and pollutants through their tissues, they can efficiently detoxify water streams. The majority of human activities result in the discharge of toxic substances, including heavy metals, as byproducts into water, sediments, and the environment. This study assesses the efficacy of the usage of submerged aquatic macrophytes, such as *Ceratophyllum demersum* L. (Family: Ceratophyllaceae) and *Potamogeton pectinatus* L. (Family: Potamogetonaceae), gathered from El-Burullus Lake, which is located northeast to deltaic vicinity of Egypt, to purify waste-polluted water from heavy metals: cadmium, lead, zinc and manganese. Concentrations of these metals in the wastewater samples were measured by means of atomic absorption spectroscopy. Results showed that *C. demersum* has an elimination performance of 63% for heavy metals studied, which was lower than that of *P. pectinatus* measured at 75%. Therefore, aquatic macrophytes studied are promising candidates for remediating wastewater, comparing extent of eco-toxicity, and preventing pollution occurring in the aquatic environments.

**Keywords:** Wastewater treatment; *Potamogeton pectinatus* L.; *Ceratophyllum demersum* L.; Bioremoval of pollutants

## 1. Introduction

Pesticides, herbicides, organic and inorganic fertilizers, and other harmful substances find their way into the aquatic environment via anthropogenic, agricultural, industrial, and human activities taking place in the biosphere.<sup>1</sup> Ninety-five substances, including heavy metals introduced into the aquatic environment, have been classified as harmful.<sup>2</sup>

It is known that many plant species can flourish in metal-rich settings, with some capable of accumulating extremely high concentrations of poisonous metals in their tissues—concentrations that are critical to their development and growth. Among these common metals are lead (Pb), zinc (Zn), copper (Cu), molybdenum (Mo), nickel (Ni), chromium (Cr), iron (Fe), magnesium (Mg), cobalt (Co), and manganese (Mn).<sup>3-5</sup> On the other hand, some plants may become hazardous when these heavy metals accumulate excessively.<sup>6</sup> Biodegradation is one of the many techniques that is both ecologically and economically favorable.<sup>7-9</sup> To cast off metals, pesticides, petroleum compounds, explosives, polyaromatic hydrocarbons, and specific herbal pollutants, phytoremediation has been carried out extensively. The method has been found to successfully remove the above elements and is known to be maintenance-friendly.<sup>10,11</sup> To cast off metals, pesticides, petroleum compounds, explosives, polyaromatic hydrocarbons, and specific herbal pollutants, phytoremediation has been carried out extensively. It may be applied for fee successfully and is pretty easy to maintain.<sup>10,11</sup> Phytoremediation is possibly supported through manner of way of the public, and monitoring the plant life to make certain they will be doing their intended duties is an easy task. Physico-chemical or specific natural techniques are not as green, passive, solar energy-driven, sustainable, or luxurious as phytoremediation has proven to be in terms of environmental cleaning.<sup>12,13</sup> *Potamogeton pectinatus* (syn. *Stuckenia pectinata*) is a monocotyledon plant of the Potamogetonaceae family, commonly called sago pondweed or fennel pondweed, also known as ribbon weed, is a cosmopolitan water plant species that grows in fresh and brackish water on all continents except Antarctica. It is an aquatic plant that is completely submerged, with no leaves floating or emerging.<sup>14</sup>

The long, narrow, linear leaves of *P. pectinatus* are made up of two thin, parallel tubes and are <2 mm broad. The primary distinction from other pondweeds with thin leaves is that the stipule is attached to the leaf base; when the leaf is plucked, the sheath and stipule separate, like the sheath and ligule of grass. The seeds

float and the blooms are pollinated by the wind. On the rhizomes, starch-rich tubers are generated. Vegetative reproduction uses tubers and plant pieces, while sexual reproduction uses seeds. Fruit lengths range from 3 to 5 mm. Various kinds of water birds would consume the plant in its entirety.<sup>15,16</sup> Additionally, the *P. pectinatus* is excellent at bioremoving toxins and exogenous compounds from contaminated wastewater. It also shows potential for the removal of heavy metals from polluted wastewater, such as Fe, Mn, cadmium (Cd), selenium (Se), Co, Ni, Cr, Pb, and Cu, by capturing sizable quantities of these elements.<sup>2,17</sup>

A monocotyledon submerged perennial, rootless, free-floating aquatic macrophyte, *Ceratophyllum demersum* L. is a member of the Ceratophyllaceae family. Four species (and a few subspecies) of aquatic plants that are found worldwide in freshwater settings make up the genus *Ceratophyllum*. The base of *C. demersum*'s stem is often buried in silty or sandy soils as it develops. Usually, it is observed floating in slow-moving, stagnant water. It is a delicate, free-floating, rootless, yet heavily leafy, annual or perennial freshwater fragrant plant that reproduces vegetatively and by seed. Its leaves are so closely spaced at the tips that they resemble a bushy animal tail. This plant has feathery leaflets that are separated into many small segments and are arranged in whorls on the stem, resembling the tail of a raccoon. Additionally, the midribs of the leaves have several microscopic teeth that, when dragged through the fingers, give the plant a rough texture. The little flowers are situated near the leaf base. The blossoms appear every year, although rarely seen. *C. demersum* L. lives in fresh and brackish shallow water that is rich in nutrients. Under these circumstances, it may quickly absorb and deposit heavy metals in its tissues. This plant has been utilized for phytoremediation because it may be used to monitor heavy metal contamination since it collects residues of Pb and Cd. Because it floats freely, it needs to absorb a lot of nutrients from the water, capturing high levels of nitrogen from its natural habitat. Additionally, it affects cyanobacteria and phytoplankton allelopathically.<sup>12,18</sup>

The traditional remedy techniques for extracting biologically polluted heavy metals or effluents of wastewater include ion exchange, discount of contaminants, and chemical precipitation, membrane filtration, nanotechnological remedy, electrochemical elimination, and superior oxidation.<sup>19</sup> Additionally, there are numerous organic approaches that use useless or alive, unfastened or immobilized cells of algae or plant tissues, as cells have carbohydrates and polypeptides of

their partitions with aldehydes, hydroxyl, amines ketones, carboxyl organizations and phosphates accountable for metallic cation adsorption and chelation.<sup>20</sup> Unfortunately, the usage of these traditional approaches is limited because of disadvantages such as low selectivity, incomplete elimination, excessive electricity consumption, high cost, or generation of excessive poisonous waste. Therefore, this calls for safe, low-cost, and potent techniques for extracting heavy metals from polluted water.

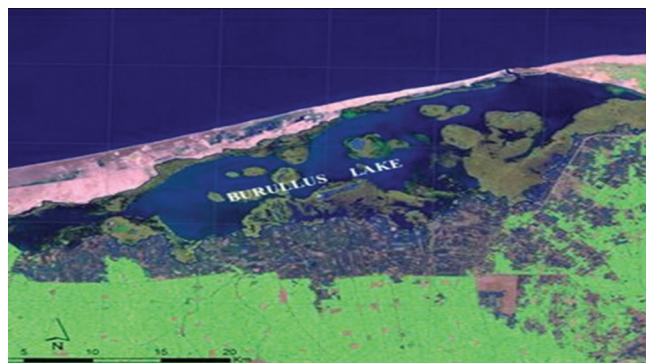
Biosorption is the use of dried organisms as adsorbents.<sup>20</sup> There are numerous methods for metallic uptake that use microbial cells, with the underlying principle consistent with extracellular accumulation, through which several heavy metals from wastewater are removed with the aid of covalent or ionic bonding.<sup>21</sup> Metal biosorption from aqueous options is a potential wastewater remedy strategy. This method is a special technique of capitalizing on organic material's potential to absorb heavy metals ions from wastewater through metabolically mediated or physicochemical uptake pathways.<sup>21</sup> Biosorption is considered an effective approach for extracting heavy metals from wastewater because of its attractive benefits, such as low cost and superior performance. In addition, they may compensate for the drawbacks of commercial resins, which lessen the performance of adsorption in wastewaters to the decrease in steel concentrations; this motivates its manufacturing in massive portions and in cost-effective ways.<sup>22</sup> Also, algae are ideally fitted for comparing water quality and had been proven to be powerful bioindicators due to their ubiquity, rapid reproduction rates, and high sensitivity to chemical changes, eutrophication and pollution.<sup>22,23</sup> Recently, Alhaithloul *et al.* (2025)<sup>24</sup> evaluated the water quality of the Bahr El-Baqar drains and investigated heavy metals in some vegetable plants.

Therefore, this study aimed to evaluate the effectiveness of *C. demerssum* and *P. pectinatus* through laboratory experiments as sustainable biomaterial candidates for removing heavy metals from contaminated wastewater in a larger-scale treatment.

## 2. Materials and methods

### 2.1. Study site

Samples of *P. pectinatus* L. and *C. demerssum* L. were obtained from their native habitat in the deltaic area of Egypt, which is located northeast of Lake El-Burullus (Figure 1). One of the delta lakes, Lake El-Burullus is situated between the Damietta and Rosetta branches, between latitudes 31° 21~ and 31° 35~ N and longitudes 30° 30~ and 31° 10~ E. The lake is elongated in shape,



**Figure 1.** The location where samples were collection from Lake El-Burullus situated at the Northern Deltaic region of Egypt. Source: Nature Conservation Egypt (<https://www.natureegypt.org>).

connected to the sea via the Boughaz El-Burullus, which has an aperture of 50 m in width.

### 2.2. Field water quality

The water quality of the original aquatic environment was measured to determine the best conditions for growing the plants under investigation. Using the Misuraline model ML 1010 (Misura Line, Romania), the water quality of the original water habitat was determined, along with pH levels and temperatures (°C). A Secchi disc was used to measure the field water's depth (m) and clarity. Salinity was determined using an electronic TDS meter (HANNA, model HI99300, Romania) to measure conductivity (µS/m). Using the HANNA model HI 9146, Hanna Product SKU, USA), dissolved oxygen and biological oxygen demand (mg/L and saturation percentage) were assessed as soon as the samples were collected. Subsequently, water samples were deep-frozen to allow for further examination of dissolved cations (Ca, Mg, Na, and K) and anions (HCO<sub>3</sub><sup>-</sup>, NH<sub>4</sub><sup>+</sup>, and P) as markers of the nutrient levels in the lake from which the plant samples were taken. The molybdate blue technique was used to estimate the soluble reactive phosphate.<sup>25</sup> Nessler's technique, which uses an alkaline solution of mercury chloride as a reagent for calorimetric measurement, was utilized to detect ammonia. A flame photometer (CORNING M410, CORNING, USA) was employed to determine K, Na, Ca, and Mg. Pye Unicam Sp 1900 Recording Flame Atomic Absorption Spectrophotometry (Tianjin Gangdong Scientific Technology Co., Ltd., China) was used to evaluate the levels of Pb, Zn, Cd, and Mn.<sup>26</sup>

### 2.3. Plants sampling

Plants of a similar size and growth stage were selected for the experiment, and the samples were cleaned with

new, distilled water to get rid of any remaining particles and parasites. The associated species' life form and longevity were recorded for the plants studied under investigation (Figures 2, and 3). Furthermore, the taxa under study were identified accordingly.<sup>27</sup>

#### 2.4. Experimental analysis

Heavy metal solutions for experiments are commonly prepared by dissolving metal salts in distilled water. By placing equal amounts of clean, fresh *P. pectinatus* and *C. demerssum* plants (1 kg for each glass tank in 50 cm × 80 cm × 50 cm field with 200 L of waste water) with varying concentrations of Cd, Pb, Zn, and Mn alternating (0.1, 0.25, 0.50, 1 g/L) throughout observation, the concentrations of heavy metals in the aqueous solution were monitored and measured.

The conduct time was determined by comparing the concentration of 1 g/L to the results of the subsequent experiment, which varied the exposure period for each element (6, 12, 18, 24, and 30 h).

After being cleaned with diluted nitric acid, the samples were placed in Teflon beakers. They were then allowed to evaporate at 80 – 90°C, and finally, 65%

nitric acid was added and filtered through filter paper. Atomic absorption spectroscopy was used to estimate the final concentrations of Cd, Pb, Zn, and Mn in the solutions, according to a protocol reported elsewhere.<sup>25</sup>

#### 2.5. Statistical analysis

Both experiments were carried out in duplicate. Using the SPSS software, the data was statistically analyzed using one-way analysis of variance to evaluate the variations in the plant treatment variables.<sup>28</sup> When there were substantial differences, a Duncan test was used.

### 3. Results

Table 1 reveals the physicochemical parameters of water of Lake El-Burullus where the tested plants were collected: the depth of water ranged 100 – 110 cm; the site temperature ranged 27 – 28°C; the transparency of water reached 54 cm; the pH value was 6.9; and the salinity value was 5,200 mmhos/cm. The dissolved oxygen was 4.5 mg/L and the biological oxygen demand was 18.0 mg/L. The nutrient composition data are as follows: K = 123 mg/L, Mg = 960 mg/L, Ca = 37 mg/L, Na = 175 mg/L mg/L, PO<sub>4</sub> = 9 mg/L, HCO<sub>3</sub> = 122 mg/L, and NH<sub>4</sub> = 12 mg/L.

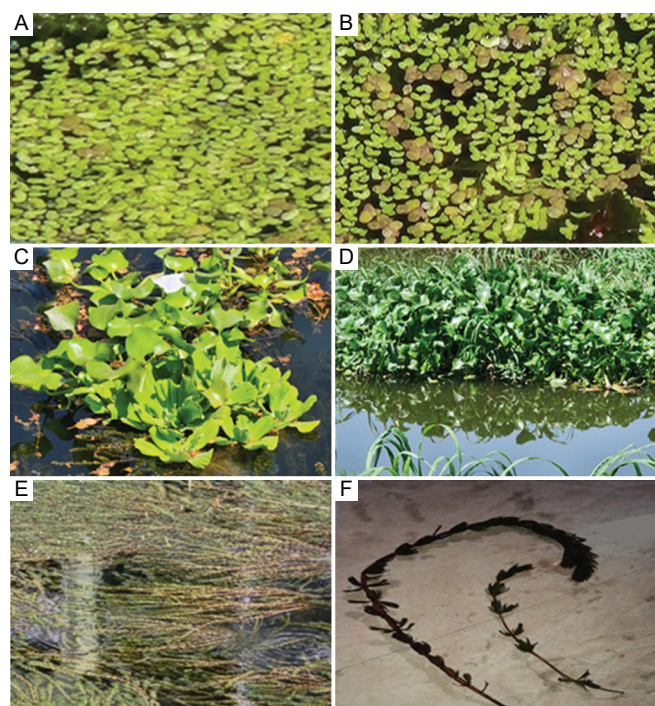


Figure 2. Photograph showing the aquatic macrophytes (floating and submerged) in Lake El-Burullus with the most dominant species: (A) *Lemna gibba*, (B) *Azolla filiculoides*, (C) *Pistia stratiotes*, (D) *Eichhornia crassipes*, (E) *Potamogeton perfoliatus*, and (F) *Ceratophyllum demersum*

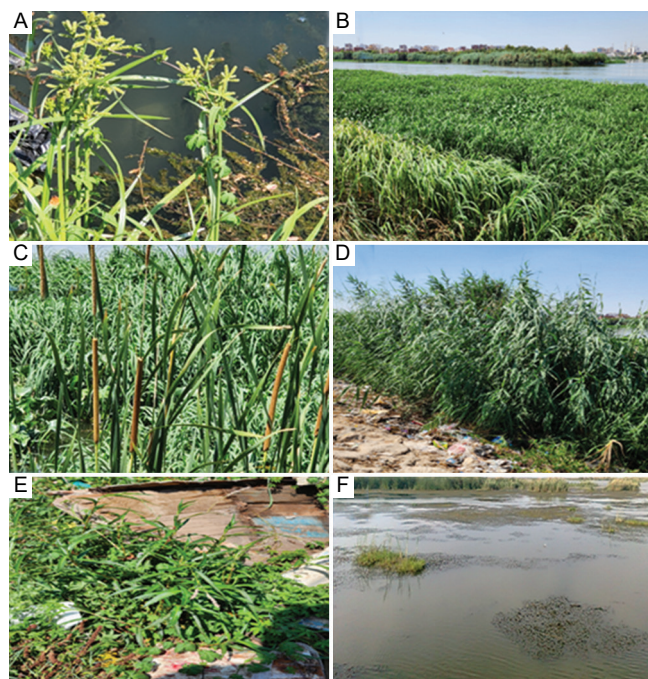


Figure 3. Photograph showing the amphibious macrophytes, (A) *Cyperus articulatus*, (B) *Paspalum paspalidum*, (C) *Typha domingensis*, (D) *Phragmites australis*, and (E) *Persicaria salicifolia*, and (F) Top view of aquatic vegetation in Lake El-Burullus

A floristic list of associated species for *C. demerssum* L. and *P. pectinatus* L. is provided (Figure 2). There are 11 species listed, representing 9 families and 11 genera (Table 2). The weeds in the research region may be divided into two categories based on their lifespan:

Annuals (also known as ephemerals) and perennials. Regarding the life span: all associated species belong to helophytes (including free-floating, submerged, and amphibious fresh hydrophytes) except *Persicaria salicifolia* (Brouss. ex Willd.) Assenov which belongs to Hemicryptophytes.

**Table 1. Data of various water quality parameters for sites where studied plants were collected**

Water quality parameter	Value
Depth (cm)	100.0±5.77 <sup>a</sup>
Temperature (°C)	27.0±1.56 <sup>a</sup>
Transparency (cm)	54.0±3.12 <sup>a</sup>
pH	6.9±0.40 <sup>a</sup>
Salinity (mmhos/cm)	5200.0±300.22 <sup>c</sup>
DO (mg/L)	4.5±0.26 <sup>a</sup>
BOD (mg/L)	18.0±1.04 <sup>a</sup>
K (mg/L)	123.0±7.10 <sup>a</sup>
Mg (mg/L)	960.0±55.43 <sup>b</sup>
Ca (mg/L)	37.0±2.14 <sup>a</sup>
Na (mg/L)	175.0±10.10 <sup>a</sup>
HCO <sub>3</sub> (mg/L)	122.0±7.04 <sup>a</sup>
PO <sub>4</sub> (mg/L)	9.0±0.52 <sup>a</sup>
NH <sub>4</sub> (mg/L)	12.0±0.69 <sup>a</sup>

Notes: <sup>a,b,c</sup>Letters are used to indicate statistically significant differences between group means. Groups sharing the same letter are not significantly different from each other, while groups with different letters are significantly different.

Abbreviations: BOD: Biological oxygen demand; DO: Dissolved oxygen.

The results of metal removal efficiency of Cd, Pb, Zn and Mn at different concentrations of 0.1, 0.25, 0.50, 1 g/L for both studied plants, in the preliminary experiment to compare metal concentrations and capability of these plants in metal accumulation. *C. demerssum* accumulates the highest concentration of Zn at (1 g/L of Zn) (Figure 4A), while *P. pectinatus* accumulate the highest concentrations of Cd (Figure 4B), Pb (Figure 4C), and Mn (Figure 4D) at the same concentration (1 g/L).

In the next experiment, 1 g/L for different elements (as a better concentration for Cd, Pb, Zn and Mn) with different exposure times was used to calculate the percentage of their absorptions as an indicator of these elements removal by hydrophytes. Tables 3 and 4 correspond to the metal removal results for *C. demerssum* and *P. pectinatus*, respectively. The removal efficiency of *P. pectinatus* was higher than that of *C. demerssum*.

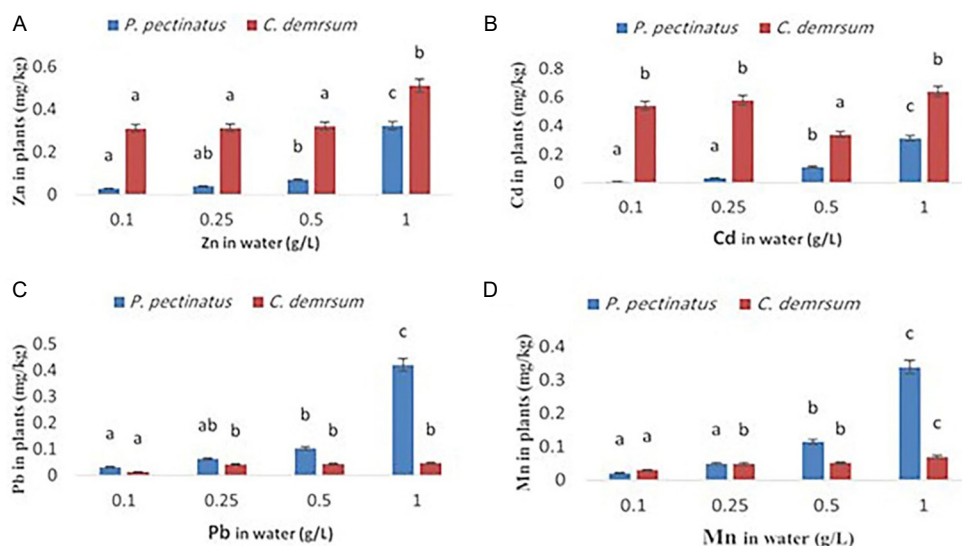
The heavy metal removal efficiency of *P. pectinatus* and *C. demerssum* is shown in Figure 5. Figure 5A shows the Cd removal efficiency of *P. pectinatus* and *C. demerssum* at different concentrations and different exposure time, with *P. pectinatus* and *C. demerssum* recording high rate of absorbed Cd at 85% and 72%,

**Table 2. Floristic list for the associated species presents at the studied site in Lake El-Burullus located at the northern deltaic region of Egypt**

Family	Species	Life span	Life form	Habitat	Chorotype
Lemnaceae	<i>Lemna gibba</i> L.	Ephemeral	Hy	Free floating	COSM
Araceae	<i>Pistia stratiotes</i> L.	Perennial	Hy		PAN
Pontederiaceae	<i>Eichhornia crassipes</i> L.				
Azollaceae	<i>Azolla filiculoides</i> Lam.		F		COSM
Ceratophyllaceae	<i>Ceratophyllum demersum</i> L.			Submerged	
Potamogetonaceae	<i>Potamogeton pectinatus</i> L.	Annual			
Poaceae	<i>Phragmites australis</i> (Cav) Trin	Perennial	G, HE	Amphibious	COSM
	<i>Echinochloa stagnina</i> (Retz.) P. Beauv.		G, HE		PAL
	<i>Paspalum paspalidum</i> L.		HE		PAN
Typhaceae	<i>Typha domingensis</i> Pers.		Ch		SA+IN
Polygonaceae	<i>Persicaria salicifolia</i> (Brouss. ex Willd.) Assenov		G		PAL

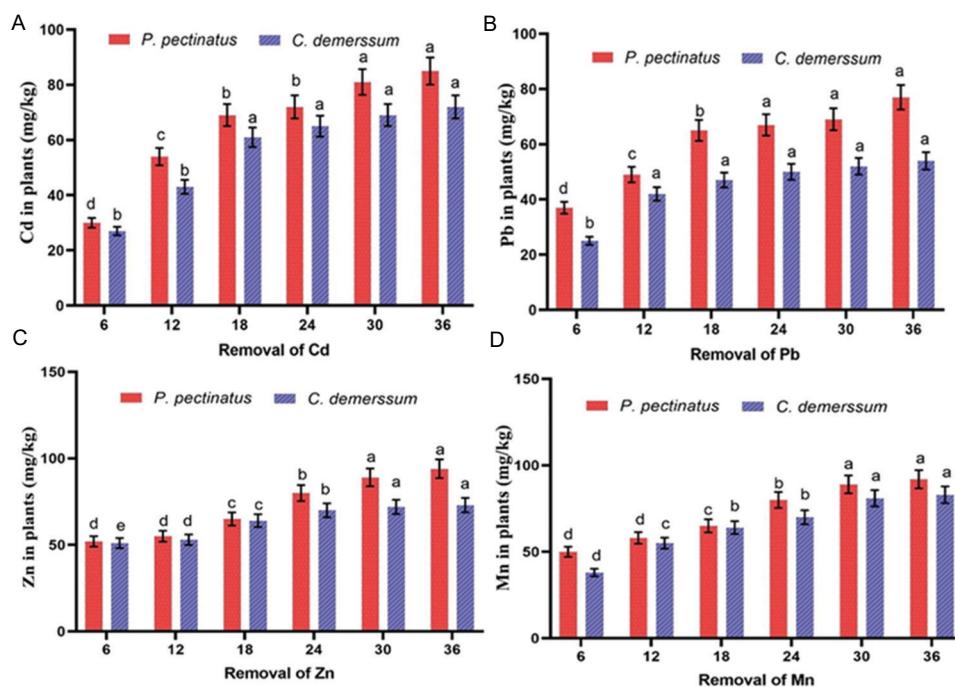
Abbreviations: Ch: Chamaephytes; COSM: Cosmopolitan; G: Geophytes; He: Helophytes; Hy: Hydrophytes; PAL: Palaeotropical; PAN: Panropical; SA+IN: Sudanic-Arabian+Indo-Pacific (These are geographical areas).

### Hydrophytes for removing heavy metals



**Figure 4. Removal efficiency of Zn (A), Cd (B), Pb (C), and Mn (D) by *Potamogeton pectinatus* (blue) and *Ceratophyllum demersum* (red)**

Notes: The mean difference is significant at the 0.05 level. <sup>a,b,c</sup>Letters are used to indicate statistically significant differences between group means. Groups sharing the same letter are not significantly different from each other, while groups with different letters are significantly different.



**Figure 5. Removal efficiency of Cd (A), Pb (B), Zn (C), and Mn (D) at different exposure times by *P. pectinatus* (red) and *C. demersum* (blue)**

Notes: The mean difference is significant at the 0.05 level. <sup>a,b,c</sup>Letters are used to indicate statistically significant differences between group means. Groups sharing the same letter are not significantly different from each other, while groups with different letters are significantly different.

respectively, after 36 h of growth in the water containing Cd, and low rate at 30% and 27%, respectively, after

6 h of exposure. The removal rate increased gradually with time until it became stable. [Figure 5B](#) reveals that

**Table 3. Metal removal efficiency by *Ceratophyllum demersum***

Metal concentration (g/L)	Zn	Cd	Pb	Mn
0.10	0.312±0.018 <sup>a</sup>	0.54±0.031 <sup>b</sup>	0.014±0.001 <sup>a</sup>	0.03±0.002 <sup>a</sup>
0.25	0.313±0.018 <sup>a</sup>	0.58±0.033 <sup>b</sup>	0.042±0.003 <sup>b</sup>	0.049±0.004 <sup>b</sup>
0.5	0.322±0.019 <sup>a</sup>	0.34±0.020 <sup>a</sup>	0.044±0.003 <sup>b</sup>	0.052±0.003 <sup>b</sup>
1.0	0.512±0.030 <sup>b</sup>	0.64±0.037 <sup>b</sup>	0.047±0.003 <sup>b</sup>	0.07±0.004 <sup>c</sup>
F-value	20.651*	17.586*	39.604*	23.999*

Notes: \*The mean difference is significant at the 0.05 level; Different superscript stars refer to significant differences at ( $p \leq 0.05$ ) level. \*:  $p < 0.05$ , significant \*\*:  $p < 0.01$ , high-significant. <sup>a,b,c</sup>Letters are used to indicate statistically significant differences between group means. Groups sharing the same letter are not significantly different from each other, while groups with different letters are significantly different.

**Table 4. Metal removal efficiency by *Potamogeton pectinatus***

Metal concentration (g/L)	Zn	Cd	Pb	Mn
0.10 g/L	0.03±0.002 <sup>a</sup>	0.01±0.001 <sup>a</sup>	0.032±0.002 <sup>a</sup>	0.021±0.001 <sup>a</sup>
0.25 g/L	0.04±0.002 <sup>ab</sup>	0.034±0.002 <sup>a</sup>	0.063±0.004 <sup>ab</sup>	0.05±0.003 <sup>a</sup>
0.5 g/L	0.071±0.004 <sup>b</sup>	0.112±0.006 <sup>b</sup>	0.104±0.006 <sup>b</sup>	0.115±0.007 <sup>b</sup>
1 g/L	0.324±0.019 <sup>c</sup>	0.313±0.018 <sup>c</sup>	0.421±0.024 <sup>c</sup>	0.339±0.020 <sup>c</sup>
F-value	207.829*	203.200*	200.881*	189.739*

Notes: \*The mean difference is significant at the 0.05 level; Different superscript stars refer to significant differences at ( $p \leq 0.05$ ) level. \*:  $p < 0.05$ , significant \*\*:  $p < 0.01$ , high-significant. <sup>a,b,c</sup>Letters are used to indicate statistically significant differences between group means. Groups sharing the same letter are not significantly different from each other, while groups with different letters are significantly different.

a high level of Pb removal from polluted water occurs after 36 h of exposure and growth of *P. pectinatus* and *C. demersum*, recording 77% and 54%, respectively. The removal efficiency decreased with time. The results presented in Figure 5C reveal that the Zn removal efficiency was high, reaching 94% with *P. pectinatus* and 73% with *C. demersum* after 36 h of growth, while the rate of absorption of Zn metal decreased gradually with time. The Mn removal efficiency, as demonstrated in Figure 5D, reached the maximum 92% and 83% with *P. pectinatus* and *C. demersum*, respectively, at 36 h of planting in the polluted water. Our findings also showed that the removal efficiency was the highest at 30 h of exposure time for both *C. demersum* (Table 5) and *P. pectinatus* (Table 6).

#### 4. Discussion

In artificial wetlands, macrophytes have a significant impact on the biological, chemical, and physical processes of treating contaminated water. They can remove suspended materials from the water, prevent erosion by lowering turbulence and water body flow velocities, and aid in the stabilization of sediments. In shallow seas, macrophytes are also known to inhibit the growth of wind waves. Lower wave heights cause a decrease in the resuspension of bottom sediments.<sup>29</sup>

Chen *et al.*<sup>30</sup> stated that heavy metals such as Pb, Cd, and others that are not required for plant development and metabolism accumulate in the food chain and cause environmental pollution that is hazardous to human health and the environment. Using plants to lessen environmental pollution is an approach known as phytoremediation.<sup>31</sup> This method has demonstrated potential in recent years for eliminating heavy metals from soil and water. Additionally, phytoremediation is comparatively clean, affordable, and effective.<sup>9</sup>

Our experiments on heavy metal removal from the polluted water suggest that *P. pectinatus* and *C. demersum* may treat and purify water after 36 h of planting. Additionally, they could quickly gather and absorb metals from the water. According to the metal saturation of plant tissues, the removal efficiency gradually dropped over time.<sup>8</sup>

Using *P. pectinatus*, wastewater may be treated to eliminate lead before it is utilized or flushed down the drain. Based on the calculation by Daud *et al.*,<sup>32</sup> the *P. pectinatus* species could greatly lower the amount of heavy metals in the leachate from landfills, with removal efficiency for Cd and Cu reaching 70% and 91%, respectively. *Ulva lactuca* is a macro alga that may be utilized in a sustainable manner to extract heavy metals from aqueous solutions.<sup>33</sup> Research has shown that *Ulva* is superior at cleaning wastewater and eliminating metals from it.

**Table 5. Heavy metal removal efficiency of *Ceratophyllum demersum* at different exposure times**

Exposure time	Cd	Pb	Zn	Mn
6	27.0±1.56 <sup>a</sup>	25.0±1.44 <sup>a</sup>	51.0±2.94 <sup>a</sup>	38.0±2.19 <sup>a</sup>
12	43.0±2.48 <sup>b</sup>	42.0±2.42 <sup>b</sup>	53.0±3.06 <sup>ab</sup>	55.0±3.18 <sup>b</sup>
18	61.0±3.52 <sup>c</sup>	47.0±2.71 <sup>bc</sup>	64.0±3.70 <sup>bc</sup>	64.0±3.70 <sup>bc</sup>
24	65.0±3.75 <sup>c</sup>	50.0±2.89 <sup>bc</sup>	70.0±4.04 <sup>c</sup>	70.0±4.04 <sup>cd</sup>
30	69.0±3.98 <sup>c</sup>	52.0±3.00 <sup>c</sup>	72.0±4.16 <sup>c</sup>	81.0±4.68 <sup>de</sup>
36	72.0±4.16 <sup>c</sup>	54.0±3.12 <sup>c</sup>	73.0±4.21 <sup>c</sup>	83.0±4.79 <sup>e</sup>
F-value	27.1	16.078	6.802	19.192

Notes: Data are expressed as an average of at least three separate experiments. The statistical significance of the results was assessed by Duncan's multiple range test ( $p=0.05$ ). The mean difference is significant at the 0.05 level. <sup>a,b,c,d,e</sup>Letters are used to indicate statistically significant differences between group means. Groups sharing the same letter are not significantly different from each other, while groups with different letters are significantly different.

**Table 6. Heavy metal removal efficiency of *Potamogeton pectinatus* at different exposure times**

Exposure time	Cd	Pb	Zn	Mn
6	30.0±1.7 <sup>a</sup>	37.0±2.14 <sup>a</sup>	52.0±3.00 <sup>a</sup>	50.0±2.89 <sup>a</sup>
12	54.0±3.12 <sup>b</sup>	49.0±2.83 <sup>b</sup>	55.0±3.18 <sup>a</sup>	58.0±3.35 <sup>ab</sup>
18	69.0±3.98 <sup>c</sup>	65.0±3.75 <sup>c</sup>	65.0±3.75 <sup>a</sup>	65.0±3.75 <sup>b</sup>
24	72.0±4.16 <sup>c</sup>	67.0±3.87 <sup>cd</sup>	80.0±4.62 <sup>b</sup>	80.0±4.62 <sup>c</sup>
30	81.0±4.68 <sup>cd</sup>	69.0±3.98 <sup>cd</sup>	89.0±5.14 <sup>bc</sup>	89.0±5.14 <sup>c</sup>
36	85.0±4.91 <sup>d</sup>	77.0±4.45 <sup>d</sup>	94.0±5.43 <sup>c</sup>	92.0±5.31 <sup>c</sup>
F-value	27.011	16.953	17.108	16.222

Notes: Data are expressed as an average of at least three separate experiments. The statistical significance of the results was assessed by Duncan's multiple range test ( $p=0.05$ ). The mean difference is significant at the 0.05 level. <sup>a,b,c,d</sup>Letters are used to indicate statistically significant differences between group means. Groups sharing the same letter are not significantly different from each other, while groups with different letters are significantly different.

Due to the *P. pectineus*'s ability to extract and absorb heavy metals from water, it may be utilized as a valuable bioremediation agent to rid the water of heavy metals and restore its suitability for irrigating woody tree forests and gardens. The proficiency of *P. pectinatus* to absorb and retain heavy metals in its tissues has been confirmed in previous studies.<sup>12,34</sup> Furthermore, it has the potential to extract and retain nutrients from wastewater.<sup>35,36</sup> *C. demersum* could extract heavy metals from water, which may subsequently be used as an irrigation tool for woody tree forests, gardens, and recently reclaimed land.<sup>17</sup>

The possibility of macrophytes as inexpensive agents for eliminating heavy metals from textile effluent has been examined previously.<sup>37,38</sup> They found that floating macrophytes may be used in textile effluent phytoremediation and were highly effective in removing heavy metals from contaminated water. Examples of these species are *Salvinia molesta*, *Eichhornia crassipes*, and *Pistia stratiotes*. Ugya *et al.*<sup>39</sup> also

showed the advantages of employing a few free-floating plants: *E. crassipes*, *Lemna minor*, and *P. stratiotes* for the filtration and remediation of industrial wastewater.

Rezania *et al.*,<sup>40</sup> pointed out that phytoremediation is an inexpensive, ecologically friendly approach that reduces heavy metal levels in wastewater more effectively than emerging and submerged plants. Bioremediation is defined as a treatment that uses naturally occurring organisms to break down hazardous substances into less toxic or nontoxic substances.<sup>41</sup> This treatment method covers bioaccumulation, biosorption, and phytoremediation. Phytoremediation, as defined by Sharma *et al.*,<sup>42</sup> is the direct and natural use of green plants to take up and absorb poisons through their roots and move them to the upper part of the plant. Studies by Bhatia and Goyal,<sup>43</sup> Baudhdh *et al.*,<sup>44</sup> and Nafea and Šera<sup>8</sup> showed that it is feasible to remove inorganic and/or organic contaminants (metals, pesticides, persistent organic pollutants) from contaminated soil, sludge, sediments, and water.

*P. stratiotes* showed an efficiency rate of 71% in removing lead from water, whereas *L. gibba* had an efficiency rate of 52%. When the concentration of heavy metals in water dropped to an extremely low level due to the *L. gibba* plant's capacity to remove and absorb them from the water at low concentrations, the water could once again be utilized to irrigate gardens and timber trees. The results of Amin *et al.*,<sup>34</sup> which demonstrated that the duck weed *P. pectinatus* is capable of absorbing and depositing heavy metals in its tissues, may support this. Additionally, it has been demonstrated that the duck weed *L. minor* has the capacity to remove and accumulate nutrients from wastewater.<sup>35,36</sup> *P. stratiotes* may extract and absorb heavy metals from water, lowering the metal concentration to an extremely low level, which is suitable for watering woody trees.<sup>17</sup>

In their evaluation of *C. demersum* as the accumulative bioindicator for trace metals, Sharma *et al.*<sup>42</sup> noted that the accumulation of aluminum (Al), Zn, and Cu was threefold at higher doses compared with the lower doses, but the bioconcentration factors were very low, indicating that this plant is not a hyper accumulator of these metals, consistent with the present results. According to Hak *et al.*,<sup>23</sup> *C. demersum*'s bioaccumulation and physiological reactions to Cu and Zn exposure exerted a greater deleterious impact than either the metal alone.

The potential of aquatic macrophytes as a low-cost bioreactor for removing heavy metals from textile effluent has been examined by Wickramasinghe and Jayawardana.<sup>38</sup> They found that the aquatic macrophytes *S. molesta*, *P. stratiotes*, and *E. crassepis* were highly potent in removing heavy metals from waste-polluted water when they were utilized in phytoremediation of textile effluent. In their review on aquatic plants' ability to remove heavy metals from wastewater, Rezanian *et al.*<sup>40</sup> concluded that phytoremediation is an inexpensive, environmentally friendly remediation technique and that free-floating plants are more efficient at removing heavy metals from wastewater than emerged or submerged plants. According to our results and observations, the submerged aquatic hydrophytes *P. pectinatus* and *C. demersum* can be used for the wider-scale removal of heavy metals from contaminated and polluted water with relatively low costs.

## 5. Conclusion

*P. pectinatus* and *C. demersum* demonstrate their potential as viable long-term options for treating different heavy metal-contaminated water and removing heavy metals (Zn, Cd, Pb, and Mn) from

the contaminated water. To prevent the plants from accumulating metals until the highly hazardous or fatal dose level, it is instrumental to regularly replace the plants being used at the sites of water remediation with the fresh ones and the used plants should be treated as hazardous items and discarded properly after wrapping. Alternatively, the used plants can be subject to metal retrieval for commercial purposes.

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## Conflict of interest

The authors declare there are no actual or potential competing interests.

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## Availability of data

Data is available from the corresponding author upon reasonable request.

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