Journal of Environmental Health and Sustainable Development

Phytoremediation Capacity of Egeria Densa for Heavy Metal Contaminated Wastewater and Its Applicability for Constructed Wetland

Md. Swadhin Hossain [1], Mahfuza Parveen [1]*, Mohammad Rajib [1,2], S.M Mahmudur Rahman [1]

[1] Department of Environmental Science and Disaster Management, Daffodil International University, Dhaka 1216, Bangladesh.
[2] Institute of Nuclear Minerals, Atomic Energy Research Establishment, Bangladesh Atomic Energy Commission, Dhaka 1349, Bangladesh.

ARTICLE INFO

ORIGINAL ARTICLE

Article History:
Received: 18 February 2025
Accepted: 20 April 2025

***Corresponding Author:**
Mahfuza Parveen

Email:
mahfuza.esdm@diu.edu.bd

Tel:
+88 017 98145583

Keywords:
Environmental Pollution,
Wastewater,
Water Purification,
Phytoremediation,
Wetlands, Plants.

ABSTRACT

Introduction: Phytoremediation is a process for restoring the environment that utilizes plant physiology and metabolism. The study was performed on a laboratory scale to assess the heavy metal removal efficiency of *Egeria densa*, its response to contaminated wastewater, and its potential for application in constructed wetlands.

Materials and Methods: Synthetic wastewater samples with low, moderate, and high concentrations of Cr, Cu, Fe, and Zn were prepared. *E. densa* was cultivated in these samples for 14 days under controlled conditions, and parameters such as heavy metal removal efficiency, growth rates, chlorophyll content, and water quality indices were analyzed.

Results: Results showed that in low heavy metal-contaminated wastewater, the removal efficiency of the *E. densa* for Cr, Cu, Fe, and Zn was 93.7%, 91.20%, 87.2%, and 100%, respectively. In highly heavy metal-contaminated wastewater, the heavy metal removal efficiency of the *E. densa* for Cr, Cu, Fe, and Zn was 36%, 54%, 35.2%, and 75%, respectively. Therefore, the study revealed that elevated heavy metal concentrations adversely affected the plant growth rate.

Conclusion: *E. densa* is a viable option for the constructed wetland system in scenarios where wastewater ranges from low to moderately contaminated respecting heavy metals. These findings can serve as a reference for future studies and provide valuable insights for researchers looking to implement *E. densa* as constructed wetland vegetation.

Citation: Hossain S, Parveen M, Rajib M, et al. ***Phytoremediation Capacity of Egeria Densa for Heavy Metal Contaminated Wastewater and Its Applicability for Constructed Wetland.*** J Environ Health Sustain Dev. 2025; 10(2): 2643-55.

Introduction

Water contamination and scarcity have posed significant environmental challenges. Rapid urbanization, industrialization, and agricultural activities over the past few decades have increased the discharge of polluted wastewater[1]. Wastewater carries an immense quantity of pollutants that are exceedingly noxious to aquatic ecosystems and human health. Moreover, certain water pollutants, such as heavy metals, are hazardous and harmful, affecting ecosystems and human health[2]. Many heavy metals, such as chromium (Cr), lead (Pb), mercury (Hg), and metalloids, can be hazardous even at low concentrations [3]. Wastewater reclamation is a critical approach to safeguarding the environment and human health.

This issue can be addressed using various methods. These techniques can be classified into

[DOI: 10.18502/jehsd.v10i2.19008]

various categories, such as physical, chemical, biological, and engineering procedures [4]. Engineering and physical treatment methods have successfully enhanced the quality of water and sediment to a certain degree. However, these methods also have negative consequences, including disruption of natural ecosystems and financial burden due to expensive initial investment and ongoing maintenance expenses [5]. Chemical techniques offer a rapid solution for purifying contaminated water. However, these solutions are only temporary and can result in the generation of additional waste, leading to potential environmental and health risks[6]. At low concentrations, most of these methods are expensive and inefficient. The drawbacks of these methods necessitate the development of alternative, low-cost water treatment systems. Consequently, phytoremediation is the preferred approach for removing heavy metal contaminants from the environment. This technology utilizes raw or genetically modified plant species to remediate contaminated water sources, effectively extracting or eliminating pollutants from wastewater [7]. It represents a cost-effective, energy-efficient, and environmentally friendly approach to extensive water treatment applications.

Modern phytoremediation methods are successfully utilized in constructed wetland technology for wastewater treatment. Several studies have used macrophyte plants in constructed wetland technology to remove pollutants and heavy metals from wastewater [8,9]. Choosing a suitable plant according to the pollutants and climatic conditions is essential in constructed wetland technology to achieve optimal pollutant and heavy metal removal efficiency [10]. The potential of multiple submerged aquatic macrophytes for wastewater remediation has been studied. For instance, studies have shown that the submerged aquatic plants *Typha angustifolia*, *Erianthus arundinaceus*, and *Phragmites australis* are effective in treating wastewater from the pulp and paper industry, which contains heavy metals [11]. The aquatic plant *Ceratophyllum demersum L.* accumulates As from contaminated wastewater [12]. A previous study demonstrated that *Canna indica L.* effectively eliminated organic pollutants from wastewater [13]. The positive outcomes of phytoremediation using aquatic plants have attracted significant interest from researchers and scientists.

Among various species, *E. densa*, a submerged aquatic macrophyte, holds a distinct position because of its high adoption capability and tolerance to a wide range of temperatures [14]. It is fast-growing, cost-effective, and capable of thriving in varied hydrological and climatic conditions, making it suitable for tropical countries such as Bangladesh. Extensive research has been conducted on various macrophytes to determine their pollutant and heavy metal removal capacities. Despite this growing body of research, one common and widely available macrophyte, *Egeria densa*, remains underexplored. Previous studies have mostly focused on other species, such as *Typha*, *Phragmites*, and *Ceratophyllum*, while there is a lack of detailed investigation on *E. densa* under varied heavy metal concentrations relevant to South Asian River systems. This gap in research not only limits our understanding of the *potential of E. densa* for heavy metal removal from wastewater but also prevents ecologists, engineers, and scientists from utilizing it in constructed wetland systems. This gap in knowledge suggests a potential opportunity to examine the heavy metal removal capacity of *E. densa* from wastewater and its potential utilization in constructed wetlands. This study represents the first assessment *of E. densa* applicability in constructed wetlands in Bangladesh. Its novelty lies in demonstrating the plant's effective heavy metal removal capabilities, while examining previously underexplored physiological and morphological responses. This study aimed to quantify the heavy metal (Cr, Cu, Fe, Zn) removal efficiency of *E. densa*, establish its tolerance thresholds for heavy metal exposure, and evaluate its suitability for treating contaminated wastewater in constructed wetlands.

Materials and Methods

Experimental setup

[DOI: 10.18502/jehsd.v10i2.19008]

The study used three distant synthetic wastewater samples, referred to as sample 1 (S1), sample 2 (S2), and sample 3 (S3), and each sample was represented by three replicate points to ensure the accuracy and reliability of the experiment, referred to as R1, R2, and R3. Furthermore, a control sample was used and designated as (C). The study used 1000 ml of synthetic wastewater per beaker, each containing three branchless *E. densa* plants. The evaporation rate of water from the beaker was 8.17 ml per day. The distilled water occupied the evaporation gap, providing 16.34 ml of water over 2 days. The experimental setup was subjected to 10 h of continuous natural light at an average temperature of 27 °C. *E. densa* was planted on March 28, 2024, followed by uprooting on April 12, 2024, for subsequent analysis of its height, weight, roots, and branches. The experiment was conducted over 14 days. Figure. Figure 1 a-b illustrates a pictorial representation of the media employed and the experimental setup of the study.

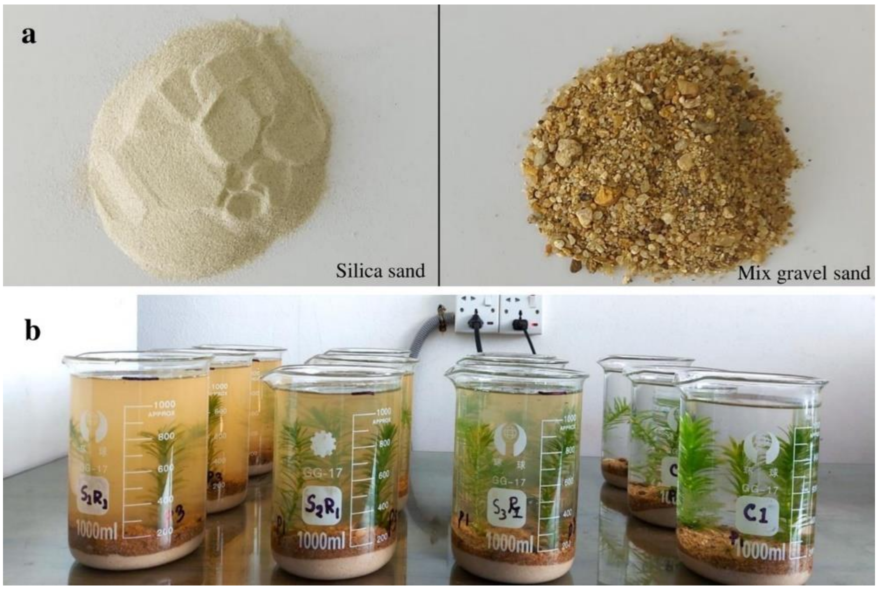

Figure 1: Pictorial plates of employed media (a), Experimental setup (b).

Wastewater preparation

The chemical composition of heavy metal-contaminated wastewater varies considerably across different sites. This study involved the preparation of synthetic wastewater reflecting the heavy metal concentrations observed in various rivers across Bangladesh [15,16,17]. This study employed three distinct concentrations of synthetic wastewater samples. Required amounts of $K_2Cr_2O_7$, $CuSO_4(H_2O)_5$, $ZnSO_4(H_2O)_7$, $FeSO_4.7H_2O$ powders, and H_2SO_4 solution were introduced into distilled water to prepare synthetic wastewater with the pollutant concentrations listed in Table 1. Sample 1 showed significant heavy metal contamination, sample 2 showed moderate contamination, and sample 3 showed minimal contamination.

Table 1: Heavy metals concentration of synthetic wastewater samples

Parameter (mg/l)	Sample 1	Sample 2	Sample 3
Cr	0.5	0.25	0.125
Cu	0.5	0.25	0.125
Fe	5.0	2.5	1.25
Zn	0.20	0.10	0.05

Sediment preparation

The study utilized processed silica from river-born sand with a SiO_2 presence of approximately 94%, along with mixed gravel sand, which is traditionally used for construction purposes. The sand was used in two layers: the bottom layer comprised silica sand, whereas the subsequent layer located above it was composed of mixed sand containing a mixture of gravel and sand. Both sands were used in equal amounts, weighing 250 g

[DOI: 10.18502/jehsd.v10i2.19008]

each; a total of 500 g of sand was used. Before use, the sand was cleaned and made dirt-free.

Macrophyte preparation

Submerged macrophytes *Egeria densa* were collected from a meticulously maintained aquatic nursery in Kataban, Dhaka, Bangladesh. The collected plants were cultured for 45 days in the Environmental Science and Disaster Management (ESDM) laboratory at the Daffodil International University (DIU). Before cultivation, the plants were rinsed with distilled water, and the necessary nutrients were added to the aquarium water. After 45 days, selected mature plants were collected from an aquarium and cut at 11 cm along the tip. The plants were thoroughly rinsed with distilled water before being placed in experimental beakers.

Experimental analysis

Laboratory analyses were conducted at the ESDM laboratory at DIU. Before *E. densa* plantation on the medium (day 1), the morphological parameters of *E. densa*, such as plant length, weight, branch length, and root length, were analyzed. Furthermore, the physicochemical parameters (pH, EC, TDS, and COD) and heavy metals (Cr, Cu, Fe, and Zn) of synthetic wastewater were analyzed. On the last day of the experiment (day 14), the plants were uprooted from the experimental medium and the same parameters measured on day 1 were measured. Water quality parameters were measured using standard methods [18]. pH, EC, TDS, and temperature were recorded using a Hanna HI9814 meter, and COD was analyzed using a Hanna HI-83399 multiparameter photometer. Chlorophyll concentration was measured using a HACH DR 6000 UV-VIS spectrophotometer. All data were statistically analyzed using SPSS and Microsoft Excel software. Equations 1 and 2 were used to determine the shoot growth rate (SGR) and root growth rate (RGR), and to assess the shoot weight growth rate (SWGR), respectively [19,20].

$$\text{Shoot Growth rate} = \frac{\text{Final(cm)} - \text{Initial (cm)}}{Day} \quad (1)$$

$$\text{Shoot weight Growth rate} = \frac{\text{Final}(weight) - \text{Initial (weight)}}{Day} \quad (2)$$

Determined metal index (MI)

This study adopted the Metal Index to determine the level of heavy metal contamination in wastewater [21]. The MI reflects different heavy metal concentrations in a particular sample and allocates one value representing the total pollution level. Equation 3 represents the MI Formula.

$$\text{MI} = \sum_{i=1}^{N} \frac{C_i}{(\text{MAC})_i} \quad (3)$$

Where C_i denotes the i^{th} heavy metal concentration in the sample analysis, and the maximum allowed concentration of the i^{th} heavy metal is referred to as MAC. The MI value was categorized into six distinct classes: "seriously affected (> 6), strongly affected (4-6), moderately affected (2-4), slightly affected (1-2), pure (0.3-1.0), and very pure (< 0.3)" [22]. For this study, the MAC value was obtained from the Bangladesh drinking water quality standard [23].

Determination of the chlorophyll content

The first step involved extracting chlorophyll from the stems and leaves of the plant to ascertain the chlorophyll content. The researchers then thoroughly washed and sterilized the test tubes and inserted the measured leaves and plant stems into the test tubes with appropriate labeling. The test tube was then filled with 10 ml of 80% acetone, and the test tube was enclosed by placing a layer of aluminum foil over it. For chlorophyll extraction, the test tube stand was placed in a dark place for twenty-four hours. The chlorophyll samples were extracted, and the amount of chlorophyll in each sample was determined. The chlorophyll content was evaluated using three specific wavelengths (470, 646, and 663 nm) according to Equations 4-6 [24].

$$\text{Chlorophyll a} = 12\ (Abs_{663}) - 3.11\ (Abs_{646}) \quad (4)$$

$$\text{Chlorophyll b} = 20.78\ (Abs_{646}) - 4.88\ (Abs_{663}) \quad (5)$$

$$\text{Chlorophyll (a + b)} = 17.67\ (Abs_{646}) + 7.12\ (Abs_{663}) \quad (6)$$

[DOI: 10.18502/jehsd.v10i2.19008]

The equations have been used in several studies related to estimating the plant's chlorophyll content [25,26].

Results

The phytoremediation capacity of Egeria densa

Table 2 presents the treatment efficacy for various physicochemical parameters of water samples, including the control, high heavy metal-contaminated wastewater in sample 1, moderate heavy metal-contaminated wastewater in sample 2, and low heavy metal-contaminated wastewater in sample 3.

Table 2: Treatment efficiency on physicochemical and heavy metal parameters across different wastewater samples

Parameter	Control		Sample 1		Sample 2		Sample 3	
	Initial	final	Initial	Final	Initial	Final	Initial	Final
pH	6.98	7.02	6.59	6.97	6.88	7.12	6.77	7.15
EC (mS/cm)	0.05	0.05	0.47	0.50	0.26	0.19	0.16	0.08
TDS (ppm)	40	43	360	387	200	147	120	60
COD (mg/l)	0	0	40	25	21	7.67	9	0.67
Cr (mg/l)	0	0	0.50	0.32	0.25	0.09	0.125	.0087
Cu (mg/l)	0	0	0.50	0.23	0.25	0.083	0.125	0.011
Fe (mg/l)	0	0	5	3.24	2.5	0.95	1.25	0.16
Zn (mg/l)	0	0	0.20	0.050	0.10	0.027	0.05	0.00

Changes in pH, EC, TDS, COD, and heavy metal concentrations were observed pre- and post-treatment, indicating a distinct impact on water quality. The initial pH levels of all samples were within the slightly acidic to neutral range, with minor increases after treatment. The control sample showed minimal change (6.98–7.02), whereas sample 1 increased from 6.59 to 6.97, sample 2 from 6.88 to 7.12, and sample 3 from 6.77 to 7.15. These findings suggest that the treatment process contributed to pH stabilization. Electrical Conductivity (EC) remained stable in the control (0.05 mS/cm) but varied in the treated samples. Sample 1 showed a minor increase (0.47 to 0.50 mS/cm), whereas samples 2 and 3 demonstrated decreases (0.26 to 0.19 mS/cm and 0.16 to 0.08 mS/cm, respectively), suggesting the effective removal of ionic elements. Total Dissolved Solids (TDS) increased slightly in the control (40–43 ppm) and sample 1 (360–387 ppm) groups, likely due to the decay of dead plants. However, it decreased significantly in sample 2 (200 to 147 ppm) and sample 3 (120 to 60 ppm), indicating the efficient removal of dissolved pollutants. Chemical Oxygen Demand (COD) was considerably reduced in all samples. Sample 1 COD declined from 40 to 25 mg/L, sample 2 from 21 to 7.67 mg/L, and sample 3 from 9 to 0.67 mg/L, respectively. These reductions suggest effective organic matter removal across the samples, with the most significant COD reduction observed in Sample 3.

Heavy metals, including chromium (Cr), copper (Cu), iron (Fe), and zinc (Zn), also exhibited substantial reductions in their concentrations. Cr decreased from 0.50 mg/L to 0.32 mg/L in sample 1, 0.25 mg/L to 0.09 mg/L in sample 2, and 0.125 mg/L to 0.0087 mg/L in sample 3. Cu was reduced from 0.50 to 0.23 mg/L in sample 1, 0.25 to 0.083 mg/L in sample 2, and 0.125 to 0.011 mg/L in sample 3, respectively. Fe levels fell from 5 to 3.24 mg/L in sample 1, 2.5 to 0.95 mg/L in sample 2, and 1.25 to 0.16 mg/L in sample 3, while Zn was near zero in sample 3 (0.05 to 0.00 mg/L).

Comparison of heavy metal removal efficiencies across three wastewater samples

The heavy metal removal efficiencies of *E. densa* were evaluated across three wastewater samples, as shown in Figure. 2. The variation in removal efficiency highlights the adaptability and effectiveness of *E. densa* at different contamination levels.

[DOI: 10.18502/jehsd.v10i2.19008]

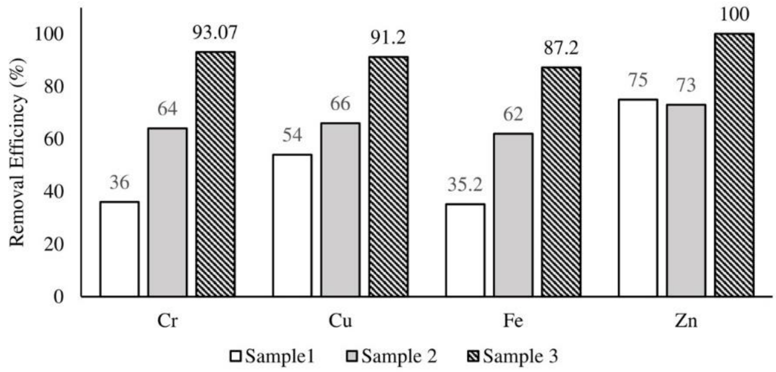

Figure 2: Heavy metal removal efficiencies of *Egeria densa* across wastewater samples with varying contamination levels

For Cr, the removal efficiency varied significantly, with the lowest efficiency observed in sample 1 at 36%, increasing to 64% in sample 2 and reaching 93.07% in sample 3. A similar trend was observed for Cu, where the efficiency improved from 54% in sample 1 to 66% in sample 2 and peaked at 91.2% in sample 3. The removal of Fe exhibited a progressive increase, with efficiencies of 35.2%, 62%, and 87.2% for samples 1, 2, and 3, respectively. Zn removal efficiency was notably high across all samples, with 75% for sample 1, 73% for sample 2, and a perfect 100% for sample 3.

Metal Index (MI)

MI measures the levels of various metals present in water and compares them to the maximum allowable concentration (MAC) set for each metal. Figure 3 shows the MI values in water at the initial and final phases of the study.

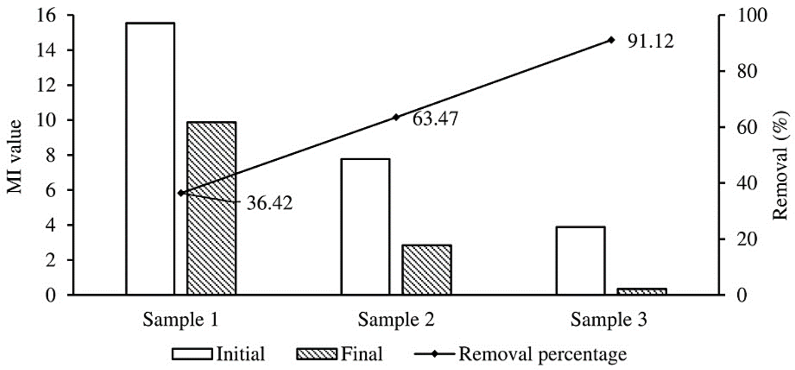

Figure 3: MI value in the water during the initial and final phases of the study with removal efficiency

The effectiveness of the applied treatment method was assessed by comparing the MI values before and after treatment across three water samples with varying levels of contamination. Sample 1, with the highest initial MI value of 15.54, was classified as being seriously affected. After treatment, the MI value decreased to 9.88, yet it remained within the seriously affected classification, reflecting a 36.42% removal efficiency. This suggests that although some pollutants were removed, the treatment was not fully effective in significantly improving water quality in samples with severe contamination. In contrast, Sample 2, which also began as seriously affected with an MI value of 7.77, showed a more substantial reduction after treatment, reaching an MI value of 2.83 and achieving reclassification to moderately affected. The removal efficiency for this sample was 63.47%, indicating that the treatment was notably more effective for samples with moderate contamination levels. Sample 3, with an initial MI value of 3.88, classified as moderately affected, demonstrated the most significant improvement post-treatment. Its MI value dropped to 0.34, achieving a 91.12% removal efficiency, and reclassifying it as pure. This result highlights the high effectiveness of the treatment for samples with lower initial contamination.

Heavy metal tolerance level of Egeria densa

The heavy metal tolerance of *E. densa* was assessed using synthetic wastewater samples containing varying concentrations of Cr, Cu, Fe, and Zn. The experiment demonstrated that *E. densa* has a defined tolerance range for each heavy metal, beyond which the survival of the plant was compromised. The results revealed that *E. densa* tolerated Cr concentrations of up to 0.25 mg/L. In sample 2 (Cr: 0.25 mg/L), the plants exhibited normal growth, whereas in sample 1 (Cr: 0.5 mg/L), signs of stress and mortality were observed, indicating that concentrations above 0.25 mg/L are lethal for *E. densa*. A similar trend was observed for Cu. The plant remained viable at Cu concentrations of up to 0.25 mg/L in sample 2 but failed to survive at 0.5 mg/L in sample 1, confirming a tolerance limit of 0–0.25 mg/L. For Fe, the plants demonstrated greater resilience, with a tolerance threshold of up to 2.5 mg/L. In sample 3 (Fe: 1.25 mg/L), *E. densa* maintained healthy growth, but its survival declined in sample 1, where Fe levels were elevated to 5.0 mg/L. A similar pattern was observed for Zn, where the plant thrived at concentrations of 0.05 mg/L in sample 3 and 0.10 mg/L in sample 2 but exhibited severe stress and mortality at 0.20 mg/L in sample 1. Based on these observations, the tolerance thresholds were established as follows: Cr (0–0.25 mg/L), Cu (0–0.25 mg/L), Fe (0–2.5 mg/L), and Zn (0–0.10 mg/L). Beyond these concentrations, *E. densa* exhibited complete mortality (Cr/Cu>0.25 mg/L; Zn>0.10 mg/L), indicating a narrow tolerance range for most of the tested metals.

Heavy metal contaminated wastewater impact on Egeria densa

Shoot growth rate (SGR)

Figure 4a shows the SGR of the plants across all samples. This study evaluated the mean plant growth across all water samples. The control sample had the maximum growth rate of 0.0640 cm/day, whereas sample 1 displayed the lowest growth rate of -0.3730 cm/day. The subsequent sample growth rates were as follows: sample 2: -0.0223 cm/day; sample 3:0.0573 cm/day. A decline in SGR was noted in samples 1 and 2. The growth rates of the control and sample 3 plant shoots were compaable.

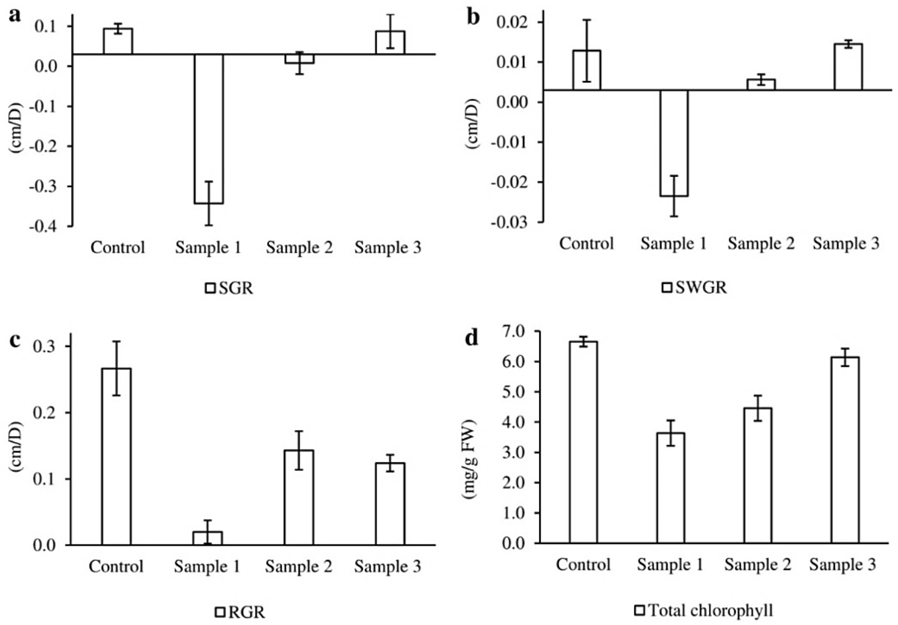

Figure 4: plant (a), SGR (b) SWGR, (c) RGR, and (d) Total chlorophyll across all the samples

[DOI: 10.18502/jehsd.v10i2.19008]

Shoot weight growth rate (SWGR)

Figure 4b illustrates the growth rate of the plant shoot weight for all samples. The control group showed a positive SWGR, indicating normal growth in the control group. In contrast, Sample 1 exhibited a significantly negative SWGR, suggesting that this treatment severely inhibited shoot growth; Sample 2 displayed a near-zero SWGR, indicating minimal or stagnant growth; and Sample 3 showed an increased SWGR compared to the control, implying that this treatment enhanced shoot biomass accumulation.

Root growth rate (RGR)

Figure 4c demonstrates that after 14 days of experimentation, the highest RGR was found in the control sample (0.26 cm/D), while the lowest plant RGR was identified in sample 1 (0.0198 cm/D). The growth rate of *E. densa* in sample 2 was 0.143 cm/D mm per day, while in sample 3, it was 0.123 cm/D. High heavy metal concentrations could be responsible for *the low RGR of E. densa* in sample 1.

Table 3: Growth rate percentage of *Egeria densa* across wastewater samples

Sample	Growth rate percentage (%)		
	SGR	SWGR	RGR
Control	8.15	19.13	373.33
Sample 1	-47.48	-53.70	27.78
Sample 2	-2.85	7.79	200
Sample 3	7.30	26.88	173.33

Table 3 presents the growth rate (%) of *E. densa* in the wastewater samples. The control group displayed the highest overall growth rates, with SGR, SWGR, and RGR values of 8.15%, 19.13%, and 373.33%, respectively, indicating optimal growth conditions. Sample 1 showed the most severe decline in growth, with negative values across all parameters (SGR: -47.48%, SWGR: -53.70%, and RGR: 27.78%), indicating a substantial inhibitory effect. Sample 2 exhibited mixed results; SGR slightly decreased (-2.85%), SWGR increased to 7.79%, and moderate improvements in RGR (200%) were observed. Sample 3 demonstrated improved growth compared to the control, with SGR (7.30%) and SWGR (26.88%), all showing positive increases, although RGR (173.33%) was lower than that of the control group.

State of chlorophyll content

After 14 days of experimentation, the total chlorophyll levels in the stems of *E. densa* are illustrated in Figure. 4d. The total chlorophyll concentration varied in all samples. In the control sample, the concentration of total chlorophyll was the highest at 6.65 mg/g FW, whereas in sample 1, the concentration was the lowest at 3.63 mg/g FW. The total chlorophyll concentrations in samples 1 and 3 were 4.45 mg/g FW and 6.13 mg/g FW, respectively. Based on the observed results, it can be concluded that in wastewater with low levels of heavy metal contamination, as in sample 3, the plants effectively balanced the total chlorophyll content. Sample 2 had a slightly balanced concentration of total chlorophyll. Compared to the control and sample 3 plants, the total chlorophyll concentration was relatively low in sample 2.

Correlation between heavy metal concentration and plant health

The correlations between heavy metal concentrations and plant health parameters were assessed to evaluate the impact of heavy metal stress. As shown in Figure. 5 a–d, MI inversely correlated with SGR, SWGR, RGR, and total chlorophyll content. The linear regression equations indicated similar slopes and intercepts for all parameters, with coefficient of determination values ranging from -0.8943 to -0.9995, highlighting consistent patterns of decline. Among the parameters, MI showed the strongest correlation coefficient with SWGR (−0.9995) and the weakest with total chlorophyll content (−0.8943).

[DOI: 10.18502/jehsd.v10i2.19008]

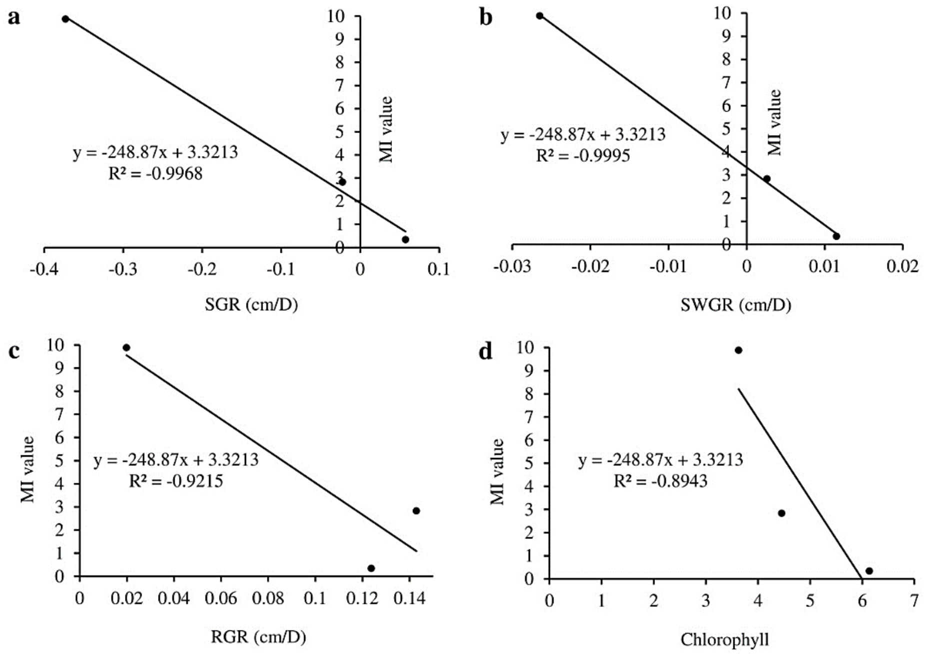

Figure 5: Correlation between (a) MI and SGR, (b) MI and SWGR, (c) MI and RGR, (d) MI and chlorophyll

Figure 5 illustrates the correlation between the metal index (MI) and shoot growth rate (SGR). A consistent and significant negative relationship was observed for all measured parameters.

Discussion

The findings of this study show that *Egeria densa* effectively reduces heavy metal concentrations in contaminated water through phytoremediation. The results implied that Sample 3 achieved the highest removal efficiency. This is likely because this sample contained a lower concentration of heavy metals, which enhanced the phytoremediation capacity of *E. Densa*. Furthermore, previous studies have reported similar findings, demonstrating that submerged aquatic macrophytes are effective in removing heavy metals from wastewater [27,28,29,30].

The trend in the removal efficiencies of Cr, Cu, Fe, and Zn demonstrated a clear relationship between the contamination levels and removal performance. Removal efficiencies improved as contamination levels decreased; *E. densa* removed up to 100% Zn and over 90% Cr and Cu in sample 3. This suggests that *E. densa* performs better in wastewater with lower contamination levels, possibly because of favorable uptake conditions in such environments. These consistently higher removal rates in samples 2 and 3 indicate *the potential of E. densa* as a sustainable tool for bioremediation, particularly in moderately to lightly contaminated systems. Similar studies have reported that other submerged aquatic macrophytes, such as *Pontederia cordata*, *Lemna minor*, and *Eichhornia crassipes*, also exhibit promising heavy metal removal capacities, but their tolerance thresholds are often higher or their adaptation is more limited in tropical regions than that of *E. densa* [31,32]. In contrast, the present study demonstrated that *E. densa* can achieve up to 100% removal of Zn and over 90% for Cr and Cu at lower contamination levels, surpassing the performance of *Typha angustifolia* under similar conditions [33]. This highlights *the high suitability of E. densa* for use in constructed wetlands, especially in South Asian contexts with variable contamination profiles and climatic conditions.

The effectiveness of the treatment method was further assessed using the Metal Index (MI). The findings clearly show an inverse relationship between the initial contamination level and treatment effectiveness. While the process reduced pollutants in all samples, it was most effective at moderate to low contamination levels. This pattern

[DOI: 10.18502/jehsd.v10i2.19008]

suggests that although *E. densa* is effective, it may not be sufficient alone for treating highly polluted wastewater. Additional or enhanced treatment strategies may be needed for highly polluted water, such as sample 1, as demonstrated by previous studies on phytoremediation limitations in heavily contaminated systems [34].

The results indicate that *E. densa* is highly sensitive to elevated concentrations of Cr, Cu, and Zn, whereas it tolerates moderate levels of Fe, likely due to its essential role in plant metabolism. The narrow tolerance range for Cr and Cu suggests a limited capacity for the detoxification or exclusion of these toxic metals. Although Zn is also a micronutrient, its threshold remains low, possibly due to its bioavailability, chemical form, or physiological sensitivity. Notably, these thresholds are substantially lower than those reported in a previous study, where *E. densa* tolerated a Zn concentration of 20 mg/L under 7-day exposure conditions [35]. Variations in exposure duration, growth conditions (e.g., light and nutrients), and genetic adaptations may contribute to differential tolerance. The observed Zn tolerance threshold of *E. densa* (0.10 mg/L) is significantly lower than the mean Zn concentration reported in groundwater from the Chittagong industrial area (0.49 mg/L) [36]. This disparity suggests that *E. densa* alone may be insufficient for effective remediation in highly contaminated environments without the support of complementary pretreatment systems. These findings highlight *the potential of E. densa* for phytoremediation within the specified concentration limits, while underscoring its vulnerability to excessive heavy metal contamination.

Regarding Wastewater with low levels of heavy metal contamination (sample 3), the plants effectively balanced the total chlorophyll content. Sample 2 slightly balanced the total chlorophyll concentration. Compared to the control and sample 3 plants, the total chlorophyll concentration was relatively low in sample 2. The findings also indicate that wastewater with high levels of heavy metal contamination (sample 1) significantly decreased the total chlorophyll concentration compared to the control sample. This reduction aligns with *the response of Ceratophyllum demersum* to multi-metal stress [37]. Several previous studies demonstrated that heavy metal concentration negatively impacts plant growth and chlorophyll concentration [38,39,40]. A consistent and significant negative relationship was observed across all measured parameters. The sharp decline in SGR, SWGR, and RGR reflects impaired growth processes, whereas the reduction in chlorophyll content highlights disruptions in photosynthesis under heavy metal exposure. These findings revealed that increasing heavy metal concentrations led to severe plant growth inhibition and physiological dysfunction. The strong MI-SWGR correlation ($r = -0.9995$) mirrors phytotoxicity models for submerged macrophytes [37].

This study suggests that *E. densa* is suitable for constructed wetland systems when wastewater is contaminated with heavy metals from low to moderate levels. The sustainable growth of *E. densa* requires moderate temperatures, sufficient light, nutrient-rich water, and a slightly acidic to neutral pH [41]. Furthermore, *E. densa* can survive across a wide range of temperatures and can grow even under unfavorable environmental conditions [42,43,44]. In Bangladesh, all conditions are conducive to the sustainable growth of *Egeria densa*. It could be utilized in several types of constructed wetlands in Bangladesh when the effluent contains low-to-moderate concentrations of heavy metals. It can be used in horizontal-flow constructed wetlands, vertical-flow constructed wetlands, free-water-surface constructed wetlands, or hybrid constructed wetlands. These findings may serve as a reference for future research and offer useful insights for scholars intending to utilize *E. densa* as vegetation in constructed wetlands.

Conclusion

Egeria densa effectively removed heavy metals (Cr, Cu, Fe, and Zn) from low heavy metal contaminated river water, with removal rates ranging from 87.2% to 100%. When treating moderately heavy metal-contaminated water, *the efficiency of E. densa* in removing these metals can

[DOI: 10.18502/jehsd.v10i2.19008]

range from 62% to 73%. Finally, *E. densa* is unsuitable for treating river water with high concentrations of heavy metals. The heavy metal tolerance levels of *E. densa* are as follows: Cr (0–0.25 mg/L), Cu (0–0.25 mg/L), Fe (0–2.5 mg/L), and Zn (0–0.10 mg/l). Water with low heavy metal contamination supports increased SGR and SWGR in *E. densa* while maintaining balanced chlorophyll concentrations. In water with moderate heavy metal concentrations. In water with moderate heavy metal contamination, plants can survive but exhibit reduced SGR, SWGR, and chlorophyll concentration. However, highly contaminated water is unsuitable for their survival. The study revealed a strong negative correlation between MI and SGR, demonstrating that higher heavy metal concentrations lead to a decline in the growth rate. These findings suggest that *E. densa* is well-suited for constructed wetland systems that treat effluents with low to moderate heavy metal levels.

Acknowledgements

The authors would like to thank the Department of Environmental Science and Disaster Management, Daffodil International University, for their kind support throughout this study.

Conflict of Interest

The authors declare no conflicts of interest.

Funding information

This research was funded by the International Atomic Energy Agency (IAEA)'s Coordinated Research Project (CRP)#F22076 through Research Contract#26702 entitled Determination of radionuclide sorption capacity of constructed wetland materials for hydrodynamics study.

Ethical Considerations

The present study was approved by the Ethics Committee of the Faculty of Health and Life Sciences, Daffodil International University, Bangladesh (ethical code: BD.DIU.REC.2024.128).

Code of Ethics

The authors adhered to the ethical guidelines and regulations throughout the study, ensuring the integrity and reliability of the research process. Data collection, analysis, and reporting were conducted transparently with due regard to scientific and ethical standards.

Data availability

Data are available from the corresponding author upon reasonable request.

Author contributions

Md. Swadhin Hossain was involved in the conceptualization, methodology, investigation, formal analysis, writing-original draft preparation, and visualization. Mahfuza Parveen was involved in supervision, methodology, validation, writing-review and editing. Mohammad Rajib ensured supervision, writing-review and editing, funding acquisition; and S.M Mahmudur Rahman carried out the investigation.

Reference

1. Sathya R, Arasu MV, Al-Dhabi NA, et al. Towards sustainable wastewater treatment by biological methods—a challenges and advantages of recent technologies. Urban Clim. 2023; 47:101378.
2. Mitra S, Chakraborty AJ, Tareq AM, et al. Impact of heavy metals on the environment and human health: novel therapeutic insights to counter the toxicity. J King Saud Univ Sci. 2022;34(3):101865.
3. Aransiola SA, Maddela NR, editors. Phytoremediation in Food Safety: Risks and Prospects. 2024.
4. Gao H, Xie Y, Hashim S, et al. Application of microbial technology used in bioremediation of urban polluted river: a case study of Chengnan River, China. Water. 2018;10(5):643.
5. Peilin G, Meng C, Lichao Z, et al. Study on water ecological restoration technology of river. InIOP Conference Series: Earth and Environmental Science. IOP Publishing; 2019. p. 032025.
6. Saleh IA, Zouari N, Al-Ghouti MA. Removal of pesticides from water and wastewater: chemical, physical and biological treatment approaches.

[DOI: 10.18502/jehsd.v10i2.19008]

Environ Technol Innov. 2020;19:101026.

7. Parnian A, Furze JN. Vertical phytoremediation of wastewater using Vetiveria zizanioides L. Environmental Science and Pollution Research. 2021;28(45):64150–5.
8. Yu G, Wang G, Chi T, et al. Enhanced removal of heavy metals and metalloids by constructed wetlands: a review of approaches and mechanisms. Sci Total Environ. 2022; 821:153516.
9. Saeed T, Alam MK, Miah MJ, et al. Removal of heavy metals in subsurface flow constructed wetlands: application of effluent recirculation. Environ Sustain Indic. 2021;12:100146.
10. Wu H, Wang R, Yan P, et al. Constructed wetlands for pollution control. Nat Rev Earth Environ. 2023;4(4):218–34.
11. Arivoli A, Mohanraj R, Seenivasan R. Application of vertical flow constructed wetland in treatment of heavy metals from pulp and paper industry wastewater. Environmental Science and Pollution Research. 2015;22:13336–43.
12. Xue P, Yan C, Sun G, et al. Arsenic accumulation and speciation in the submerged macrophyte Ceratophyllum demersum L. Environmental Science and Pollution Research. 2012;19:3969–76.
13. Patro A, Gupta S, Dwivedi S, et al. Spectrometric evidences to resolve the debate on direct role of macrophyte in organic pollutant removal and degradation in constructed wetlands. Chemical Engineering Journal. 2024;483:148740.
14. Thiébaut G, Gillard M, Deleu C. Growth, regeneration and colonisation of Egeria densa fragments: the effect of autumn temperature increases. Aquat Ecol. 2016;50:175–85.
15. Hossen MA, Mostafa MG. Assessment of heavy metal pollution in surface water of Bangladesh. Environmental Challenges. 2023;13:100783.
16. Islam MAS, Hossain ME, Majed N. Assessment of physicochemical properties and comparative pollution status of the Dhaleshwari River in Bangladesh. Earth. 2021;2(4):696–714.
17. Dey M, Akter A, Islam S, et al. Assessment of contamination level, pollution risk and source apportionment of heavy metals in the Halda River water, Bangladesh. Heliyon. 2021;7(12):e08625.
18. APHA. Standard methods for the examination of water and wastewater. 10th ed. Washington, DC: American Public Health Association; 2012.
19. Parveen M, Abdullah M, Rahman SMM, et al. Improvement of wastewater quality of Dhaleswari river, Bangladesh using submerged macrophyte Egeria densa. Journal of Sustainability Perspectives. 2022;2:449–58.
20. Rahman SMM, Parveen M, Shammi M. Responses of aquatic macrophyte Egeria densa to saline waters and biochar amended substrate. Journal of Ecological Engineering. 2025;26(1):234–47.
21. Tamasi G, Cini R. Heavy metals in drinking waters from Mount Amiata (Tuscany, Italy). Possible risks from arsenic for public health in the Province of Siena. Sci Total Environ. 2004;327(1–3):41–51.
22. Eldaw E, Huang T, Elubid B, et al. A novel approach for indexing heavy metals pollution to assess groundwater quality for drinking purposes. Int J Environ Res Public Health. 2020;17(4):1245.
23. BDWS. The environment conservation rules 1997. Dhaka: Government of the People's Republic of Bangladesh; 1997.
24. de Alkimin GD, Santos J, Soares AMVM, et al. Ecotoxicological effects of the azole antifungal agent clotrimazole on the macrophyte species Lemna minor and Lemna gibba. Comp Biochem Physiol C Toxicol Pharmacol. 2020;237:108835.
25. Dey AK, Sharma M, Meshram MR. An analysis of leaf chlorophyll measurement method using chlorophyll meter and image processing technique. Procedia Comput Sci. 2016;85:286–92.
26. Parry C, Blonquist JM, Bugbee B. In situ measurement of leaf chlorophyll concentration: analysis of the optical/absolute relationship. Plant Cell Environ. 2014;37(11):2508–20.
27. Ansari AA, Naeem M, Gill SS, et al. Phytoremediation of contaminated waters: an eco-friendly technology based on aquatic macrophytes application. Egypt J Aquat Res. 2020;46(4):371–6.
28. Xin J, Ma S, Li Y, et al. Pontederia cordata, an

[DOI: 10.18502/jehsd.v10i2.19008]

ornamental aquatic macrophyte with great potential in phytoremediation of heavy-metal-contaminated wetlands. Ecotoxicol Environ Saf. 2020;203:111024.

29. Abdelaal M, Mashaly IA, Srour DS, et al. Phytoremediation perspectives of seven aquatic macrophytes for removal of heavy metals from polluted drains in the Nile Delta of Egypt. Biology. 2021;10(6):560.
30. Demarco CF, Afonso TF, Pieniz S, et al. Potential phytoremediation of aquatic macrophyte species for heavy metals in urban environments in the southern area of Brazil. Sustainability. 2022;15(1):419.
31. Gupta A, Tyagi T. Phytoremediation and therapeutic potential of neglected plants: an invasive aquatic weeds and ornamental plant. InBiotechnological innovations for environmental bioremediation. Springer; 2022. p. 259–90.
32. Pang YL, Quek YY, Lim S, et al. Review on phytoremediation potential of floating aquatic plants for heavy metals: a promising approach. Sustainability. 2023;15(2):1290.
33. Woraharn S, Meeinkuirt W, Phusantisampan T, et al. Rhizofiltration of cadmium and zinc in hydroponic systems. Water Air Soil Pollut. 2021;232(5):204.
34. Goher ME, Hassan AM, Abdel-Moniem IA, et al. Evaluation of surface water quality and heavy metal indices of Ismailia Canal, Nile River, Egypt. Egypt J Aquat Res. 2014;40(3):225–33.
35. Harguinteguy CA, Pignata ML, Fernández-Cirelli A. Nickel, lead and zinc accumulation and performance in relation to their use in phytoremediation of macrophytes Myriophyllum aquaticum and Egeria densa. Ecol Eng. 2015;82:512–6.
36. Hossain MS, Rahman A, Asefa EM, et al. Assessing spatial distribution of heavy metal contamination in groundwater and associated human health risk in the Chittagong industrial area, Bangladesh. J Hazard Mater Adv. 2025;18:100728.
37. Lyu R, Gu B, Zhang T, et al. Simultaneous removal of Cd (II), Ni (II), and Pb (II) from water by a submerged macrophyte pondweed (Potamogeton malaianus). Water Environment Research. 2021;93(11):2637–47.
38. Xiao H, Peng S, Liu X, et al. Phytoremediation of nutrients and organic carbon from contaminated water by aquatic macrophytes and the physiological response. Environ Technol Innov. 2021;21:101295.
39. Nguyen TQ, Sesin V, Kisiala A, et al. Phytohormonal roles in plant responses to heavy metal stress: implications for using macrophytes in phytoremediation of aquatic ecosystems. Environ Toxicol Chem. 2021;40(1):7–22.
40. Singh S, Kaushik A, Bendi A, et al. Constructed wetlands as bioeconomic solutions: rhizofiltration with macrophytes for heavy metal removal. Emergent Mater. 2025;8(1):75-83.
41. Haramoto T, Ikusima I. Life cycle of Egeria densa Planch., an aquatic plant naturalized in Japan. Aquat Bot. 1988;30(4):389–403.
42. Dong J, Dai D, Yang Y, et al. Growth and morphological responses of Scenedesmus obliquus to submerged macrophyte Egeria densa. Aquat Ecol. 2023;57(1):127–38.
43. Siththisarn S, Theerawitaya C, Samphumphuang T, et al. Molecular and physiological responses of Egeria densa under salt stress. Flora. 2023;300:152226.

[DOI: 10.18502/jehsd.v10i2.19008]