



Seasonal Dynamics of Heavy Metal Bioaccumulation Potential in *Nymphaea Nouchali* and *Lemna Minor* from the Penna River, Nellore, Andhra Pradesh, India

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Abstract

Background: Anthropogenic heavy metal pollution in lotic ecosystems represents a severe global threat to biodiversity and public health due to elemental toxicity and food chain biomagnification.

Objective: This study investigates the spatiotemporal dynamics of toxic metal contamination and evaluates the comparative bioconcentration efficiencies of two native aquatic macrophytes, *Nymphaea nouchali* (rooted floating) and *Lemna minor* (free-floating), along the River Penna at Nellore, Andhra Pradesh, India.

Methods: Surface water and macrophyte samples were collected across three seasons (Pre-monsoon, Monsoon, and Post-monsoon) from two distinct locations: Sangam Barrage (agricultural influence) and Penna Bridge (urban sewage influence). Elements—Arsenic (As), Cadmium (Cd), Lead (Pb), and Mercury (Hg)—were quantified using optimized Flame Atomic Absorption Spectrometry (FAAS) following specialized acid and dry-ash digestions.

Results: The results revealed a distinct spatial gradient, with the urbanized Penna Bridge station consistently exhibiting elevated elemental loads. Surface water concentrations of Pb reached an absolute peak of 0.160 ± 0.015 mg/L during the monsoon at Penna Bridge, more than doubling the levels observed at Sangam Barrage (0.070 ± 0.017 mg/L). Temporally, dissolved metal levels peaked during the monsoon, showing that regional precipitation drives surface runoff kinetics and catchment scouring rather than simple volumetric dilution. Ecotoxicological tissue profiling demonstrated species-specific extraction efficiencies. The free-floating *Lemna minor* functioned as a superior sink for Pb and Hg, maintaining a Pb Bioconcentration Factor (BCF) up to 0.800 and an Hg BCF up to 1.667. This high capacity is facilitated by full-surface vegetative immersion which maximizes passive biosorption. Conversely, the foliar structures of *Nymphaea nouchali* exhibited a highly efficient pathway for Cadmium translocation, achieving a maximum BCF of 5.000 during both monsoon and pre-monsoon stress phases. Analysis of Variance (ANOVA) indicated that environmental shifts were statistically non-significant ($p > 0.05$), reflecting the highly dynamic, pulse-fed nature of open open-channel river ecosystems.

Conclusion: Ultimately, both macrophytes serve as sensitive continuous bio-indicators. Managed populations can be strategically deployed as low-cost, sustainable phytoremediation tools to protect the ecological equilibrium of the River Penna basin.

Keywords: Phytoremediation; Bioconcentration Factor (BCF); Penna River; *Lemna minor*; *Nymphaea nouchali*; Spatiotemporal dynamics.

Introduction

Anthropogenic heavy metal pollution represents a critical global threat to environmental stability and public health due to the persistent nature, systemic toxicity, and bioaccumulative tendencies of these contaminants within aquatic and terrestrial ecosystems (Masindi & Muedi, 2018). Heavy metals degrade atmospheric, soil, and aquatic matrices, compromising drinking water safety and accelerating the biomagnification of hazardous elemental fractions across trophic webs (Siddiquee et al., 2015). Primary drivers of this environmental degradation include accelerating industrialisation, mining, substandard waste management infrastructure, agricultural dependencies (such as synthetic fertilizers and pesticides), sewage sludge application, livestock manure, untreated wastewater discharges, fossil fuel combustion, atmospheric deposition, and petrochemical spillage (Wuana & Okieimen, 2011).

Characterised by high atomic densities, heavy metals trigger severe cytotoxic and systemic toxicity even at trace exposure thresholds (Koller & Saleh, 2018). These elements are broadly categorised into essential and non-essential cohorts. Essential metals—including cobalt (Co), copper (Cu), chromium (Cr), iron (Fe), nickel (Ni), manganese (Mn), and zinc (Zn)—serve as physiological micronutrients, yet precipitate profound toxicity when concentrations exceed homeostatic limits. Conversely, non-essential metals such as arsenic (As), cadmium (Cd), mercury (Hg), and lead (Pb) exhibit severe toxicity at trace levels, lacking any recognized biological function (Sandeep et al., 2019). Upon absorption, these non-biodegradable elements accumulate progressively within the

tissues of living organisms, driving severe ecological degradation and chronic health pathologies (Ojuederie & Babalola, 2017). Elemental and complexed configurations of chromium, cadmium, mercury, lead, nickel, and thorium are categorized as exceptionally hazardous (Kinuthia et al., 2020). Human toxicological vulnerability escalates across the following hierarchical sequence: $\text{Co} < \text{Al} < \text{Cr} < \text{Pb} < \text{Ni} < \text{Zn} < \text{Cu} < \text{Cd} < \text{Hg}$ (Mansourri & Madani, 2016). Notably, mercury, cadmium, arsenic, and lead command intense ecotoxicological scrutiny because they are readily assimilated by vegetation, directly entering human food supplies and inducing severe degenerative syndromes (Nedjimi, 2021).

Bioaccumulation designates the net accumulation of pollutants within an organism from both ambient environmental and dietary inputs, occurring when xenobiotic uptake rates eclipse metabolic clearance and elimination rates (Kanamarlapudi et al., 2018). This phenomenon operates as a two-stage mechanism: an initial, rapid physicochemical biosorption of metal ions onto outer cellular surfaces, followed by an active, metabolically driven intracellular transport process (Mrvčić et al., 2012). The kinetics of this uptake are strongly modulated by localized physicochemical variables, including ambient pH, moisture availability, and thermal conditions (Verma & Jaiswal, 2016). Within floral systems, the mechanisms of root-mediated contaminant uptake, subsequent vascular translocation, and terminal sequestration inside aerial tissues are collectively defined as phytoaccumulation, phytoabsorption, or phytosequestration (Abdel-Shafy et al., 2017). Exploiting these pathways, phytoremediation utilizes specialized botanical species to extract, degrade, immobilize, or transform environmental contaminants via coordinated root, shoot, leaf, stomatal, and cell-wall complexes (Sharma, 2012).

To date, more than 450 plant species across 45 angiosperm families have been classified as hyperaccumulators, with notable representation within the Fabaceae, Brassicaceae, Asteraceae, Euphorbiaceae, and Lamiaceae families (Suman et al., 2018). For instance, in the Kanjli Wetland of India, aquatic macrophytes including *Eichhornia crassipes*, *Panicum antidotale*, *Lemna minor*, and *Potamogeton crispus* demonstrated pronounced heavy metal extraction capacities. Specifically, *E. crassipes* functioned as an effective hyperaccumulator for Cd, Co, Fe, Pb, and Zn, whereas *P. antidotale* selectively sequestered Cu and Mn (Navdeep Singh et al., 2017). Parallel investigations in Taihu Lake, China, confirmed the systematic uptake of Ti, V, Cr, Mn, Fe, Co, Ni, Zn, and Pb by *Potamogeton malaianus*, *Nymphoides peltata*, *Eichhornia crassipes*, and *Hydrilla verticillata*, revealing highly species-specific and tissue-dependent partition coefficients (Li Bai et al.). A complementary meta-analysis identified *Limnium laevigatum*, *Salvinia auriculata*, *Lemna minor*, *Colocasia esculenta*, *Typha latifolia*, and *Typha palustris* as highly efficient bioaccumulators of Zn, Pb, Cr, Ni, Cu, Fe, Mn, and Cd within municipal and industrial wastewater frameworks (Muliyadi et al., 2023). Furthermore, evaluations of lotus root demonstrated differentiated extraction efficiencies scaling in the order of $\text{Cd} (5.03\text{--}46.67\%) > \text{Hg} (2.42\text{--}7.20\%) > \text{As} (1.93\text{--}6.68\%) > \text{Pb} (0.02\text{--}1.82\%)$, reinforcing the species-specific nature of macrophyte accumulation kinetics (Chunhui Xiong et al., 2013).

The present study evaluates the spatial and temporal dynamics of heavy metal contamination and bioaccumulation profiles within selected aquatic macrophytes native to the River Penna at Nellore, Andhra Pradesh, India. Environmental matrices and macrophyte samples were collected from two strategically distinct locations: Sangam Barrage and Penna Bridge, which are heavily influenced by agricultural runoff and urban municipal sewage discharges, respectively. Target analytes included Arsenic (As), Cadmium (Cd), Lead (Pb), and Mercury (Hg)—non-essential elements characteristic of agricultural, industrial, and domestic effluents that exert high bioaccumulation pressures despite low environmental baselines. The resulting datasets are intended to enhance local environmental monitoring frameworks, refine public health risk assessments, and provide an empirical foundation for governmental conservation strategies targeting the preservation of the River Penna's biodiversity and ecological equilibrium.

Materials And Methods

1. Collection and Physicochemical Preservation of Water Samples

One-litre surface water samples were collected at uniform depths of 3–4 feet from two monitoring stations—Sangam Barrage and Penna Bridge—across three distinct seasons (Pre-monsoon, Monsoon, and Post-monsoon). To isolate the dissolved metal fractions, samples were filtered immediately upon collection through 0.45 μm membrane filters. The filtrates were subsequently acidified to a $\text{pH} < 2$ using ultra-pure, trace-metal grade nitric acid (HNO_3) to prevent elemental precipitation and adsorption to container walls. Quantitative determination of target heavy metals—Arsenic (As), Cadmium (Cd), Lead (Pb), and Mercury (Hg)—was executed utilizing Flame Atomic Absorption Spectrometry (FAAS). Optimized hollow cathode lamps and specific wavelength profiles were utilized for each element. The instrument's limits of detection (LOD) and quantification (LOQ) were rigorously verified using multi-element standard working solutions to ensure accurate trace-level quantification within the linear dynamic range.

2. Collection and Pre-treatment of Macrophyte Samples

Specimens of two distinct aquatic macrophyte morphotypes were gathered from both monitoring stations across all three seasonal cycles: *Nymphaea nouchali* (a rooted plant with floating leaves, from which only foliar tissues were extracted) and *Lemna minor* (a free-floating, unanchored species evaluated as a whole-plant matrix). Collected plant biomass was thoroughly washed under running tap water to remove adhering sediment particles, debris, and epiphytic growth, followed by successive rinses with deionized water. The cleaned plant tissues were dried to a constant weight in a forced-air oven maintained at 105 °C. Dry biomass was mechanically

pulverized into a homogeneous, fine powder using a laboratory mill and preserved in airtight, labelled polyethylene bags within a desiccator until chemical digestion.

3. Chemical Digestion and Analytical Quantification of Plant Matrices

Homogenized plant tissue samples (2.0 g dry weight each) were weighed into clean porcelain crucibles and subjected to dry ashing in a muffle furnace at 450–500 °C for 12 hours. After cooling to ambient temperature within a desiccated environment, the residual ash was completely solubilized in 5 cm³ of 20% hydrochloric acid (HCl). The resulting solutions were quantitatively transferred into 100 cm³ volumetric flasks. Crucibles were triple-rinsed with distilled water, and the washings were transferred to the respective flasks before diluting to volume with distilled water. The final analytical solutions were thoroughly mixed by inversion. Element concentrations (As, Cd, Pb, and Hg) were determined via Flame Atomic Absorption Spectrometry (FAAS) in compliance with standard protocols established by the American Public Health Association (APHA, 1990). Analytical quality was maintained using reagent blanks and triplicate sample measurements.

4. Statistical Evaluation and Data Architecture

Descriptive statistics, including mean values and standard deviations, were computed for water (mg/L) and plant tissue (mg/Kg) concentrations across all experimental combinations. Spatial, temporal, and inter-species variances were evaluated using Analysis of Variance (ANOVA). To quantify the extraction and partitioning efficiency of each macrophyte from the surrounding aqueous medium, the Bioconcentration Factor (BCF)—alternatively termed the Bioaccumulation Factor (BAF)—was calculated using the standardized equation proposed by Suresh et al. (1999):

$$BCF = \frac{C_{\text{plant}}}{C_{\text{water}}}$$

where C_{plant} represents the dry-weight metal concentration in the macrophyte tissue (mg/kg) and C_{water} represents the corresponding concentration of the metal in the ambient water sample (mg/L). Where plant tissue concentrations fell below the instrument detection thresholds, the BCF value was strictly locked to 0.000 to maintain pure mathematical consistency.

Results And Discussion

1. Spatiotemporal Heavy Metal Profiles in Surface Water (Table 1)

The concentrations of Arsenic (As), Cadmium (Cd), Lead (Pb), and Mercury (Hg) in the surface water of the River Penna varied across monitoring stations and seasons (Table 1).

Table 1: Spatiotemporal Heavy Metal Concentrations (mg/L) in Penna River Surface Water

Heavy Metals	Seasons	Sangam Barrage (mg/L)	Penna Bridge (mg/L)
Arsenic (As)	Pre-monsoon	0.080 ± 0.015	0.120 ± 0.006
	Monsoon	0.100 ± 0.037	0.130 ± 0.015
	Post-monsoon	0.060 ± 0.012	0.090 ± 0.006
Cadmium (Cd)	Pre-monsoon	0.007 ± 0.002	0.003 ± 0.001
	Monsoon	0.001 ± 0.000	0.006 ± 0.001
	Post-monsoon	0.005 ± 0.001	0.007 ± 0.001
Lead (Pb)	Pre-monsoon	0.040 ± 0.005	0.100 ± 0.012
	Monsoon	0.070 ± 0.017	0.160 ± 0.015
	Post-monsoon	0.060 ± 0.013	0.110 ± 0.020
Mercury (Hg)	Pre-monsoon	0.003 ± 0.001	0.006 ± 0.003
	Monsoon	0.006 ± 0.003	0.009 ± 0.002
	Post-monsoon	0.004 ± 0.001	0.007 ± 0.002

Note: Values represent the mean ± standard deviation (S.D.) across three replicates.

1.1 Spatial Distribution Trends and Source Apportionment

A sharp spatial gradient was observed across all four analyzed heavy metals, with the **Penna Bridge station consistently exhibiting significantly higher toxic loads** than Sangam Barrage.

- **Lead (Pb):** Showed the most severe contamination at Penna Bridge, peaking at an elevated level of 0.160 ± 0.015 mg/L during the monsoon season. This is more than double the maximum value observed at Sangam Barrage (0.070 ± 0.017 mg/L).
- **Arsenic (As):** Concentrations at Penna Bridge hovered at high levels, shifting from a low-flow baseline of 0.090 ± 0.006 mg/L (post-monsoon) to a peak of 0.130 ± 0.015 mg/L (monsoon), while Sangam Barrage values remained systematically lower.

This clear spatial variation points directly to localized pollution profiles. Sangam Barrage primarily intercepts agricultural runoff and rural drainage from the surrounding irrigation networks of Nellore. Conversely, the Penna Bridge station crosses directly through a high-density urban corridor. It serves as a major repository for untreated domestic municipal sewage, urban stormwater scouring, automobile repair workshop drainage (rich in engine lead residues), and plastic waste degradation products. This direct input explains the elevated concentrations of industrial indicator metals at this location.

1.2 Seasonal Variations and Runoff Kinetics

The temporal shifts in water-column metal burdens challenge conventional dilution models. Instead of decreasing during high-flow periods due to simple volumetric dilution, **heavy metal concentrations generally peaked during the monsoon season.**

- Dissolved As at Sangam Barrage increased from 0.080 mg/L (pre-monsoon) to 0.100 mg/L during the monsoon.
- Dissolved Hg at Penna Bridge expanded to its absolute maximum value of 0.009 ± 0.002 mg/L during the monsoonal period.

This behavior shows that monsoonal rainfall across Andhra Pradesh drives intense **surface runoff kinetics and watershed flushing**. Heavy downpours scour agricultural fields upstream of Sangam Barrage, washing agrochemical residues, arsenical pesticides, and phosphate fertilizers into the river basin. At Penna Bridge, storm drain flows wash accumulated roadside dust, vehicular combustion deposits, and industrial debris directly into the main river channel. During the post-monsoon phase, settling dynamics dominate as water velocities drop, causing suspended particulates to sink into the riverbed sediment and lowering dissolved metal values.

2. Heavy Metal Bioaccumulation in Aquatic Macrophytes (Table 2)

The accumulation behavior within the tissues of *Nymphaea nouchali* (leaves) and *Lemna minor* (whole plant) revealed strong, species-specific extraction preferences (**Table 2**).

Table 2: Heavy Metal Bioaccumulation (mg/Kg Dry Weight) in Aquatic Macrophytes

Heavy Metals	Seasons	Sangam Barrage: <i>Nymphaea</i> (mg/Kg)	Sangam Barrage: <i>Lemna</i> (mg/Kg)	Penna Bridge: <i>Nymphaea</i> (mg/Kg)	Penna Bridge: <i>Lemna</i> (mg/Kg)
Arsenic (As)	Pre-monsoon	ND	ND	0.011 ± 0.0015	0.040 ± 0.0105
	Monsoon	0.01 ± 0.0006	0.003 ± 0.0015	0.110 ± 0.0200	0.120 ± 0.0251
	Post-monsoon	ND	ND	0.060 ± 0.0152	0.070 ± 0.0059
Cadmium (Cd)	Pre-monsoon	0.002 ± 0.0001	0.001 ± 0.0003	0.015 ± 0.0025	0.007 ± 0.0015
	Monsoon	0.005 ± 0.0015	0.003 ± 0.0015	0.004 ± 0.0006	0.001 ± 0.0005
	Post-monsoon	0.01 ± 0.0002	ND	0.002 ± 0.0015	0.001 ± 0.0006
Lead (Pb)	Pre-monsoon	ND	0.040 ± 0.0100	0.012 ± 0.0025	0.080 ± 0.0058
	Monsoon	0.010 ± 0.0005	0.063 ± 0.0152	0.020 ± 0.0010	0.100 ± 0.0026
	Post-monsoon	0.010 ± 0.0006	0.047 ± 0.0152	0.018 ± 0.0015	0.050 ± 0.0126

Mercury (Hg)	Pre-monsoon	ND	0.005 ± 0.0020	0.001 ± 0.0006	0.003 ± 0.0015
	Monsoon	0.002 ± 0.0015	0.005 ± 0.0010	0.002 ± 0.0015	0.006 ± 0.0006
	Post-monsoon	ND	0.006 ± 0.0015	0.002 ± 0.0010	0.005 ± 0.0015

Note: ND indicates Non-Detect (below instrument operational detection limits).

2.1 Arsenic (As) Sequestration Dynamic

In the rural agricultural setting of Sangam Barrage, As was completely **Non-Detect (ND)** in both macrophyte matrices during the pre-monsoon and post-monsoon phases. It only appeared at trace thresholds during the monsoon (0.001 ± 0.0006 mg/Kg in *Nymphaea*; 0.003 ± 0.0015 mg/Kg in *Lemna*), matching the seasonal runoff peak in the water column.

However, at the heavily impacted Penna Bridge station, both plants absorbed substantial concentrations of As. *Lemna minor* achieved its highest accumulation profile during the monsoon phase at 0.120 ± 0.0251 mg/Kg, closely mirrored by *Nymphaea nouchali* at 0.110 ± 0.0200 mg/Kg. This simultaneous accumulation confirms a high concentration of bio-available metalloids at this station.

2.2 Inter-Species Lead and Mercury Sequestration

Lemna minor consistently outperformed *Nymphaea nouchali* in immobilizing Pb and Hg across nearly all sampling configurations:

- **Lead (Pb):** At Penna Bridge, *Lemna minor* accumulated substantial Pb burdens across all seasons, peaking at 0.100 ± 0.0026 mg/Kg during the monsoon, compared to just 0.020 ± 0.0010 mg/Kg in *Nymphaea* leaves. At Sangam Barrage, pre-monsoon Pb was completely undetected (ND) in *Nymphaea* leaves, while *Lemna* had already sequestered 0.040 ± 0.0100 mg/Kg.
- **Mercury (Hg):** *Nymphaea* foliar tissue failed to capture Hg (ND) during both pre- and post-monsoon cycles at Sangam Barrage. Meanwhile, *Lemna minor* maintained continuous accumulation, peaking at 0.006 ± 0.0015 mg/Kg in the post-monsoon period.

3. Statistical Evaluation and Ecosystem Variance (Table 3)

The Analysis of Variance (ANOVA) configurations evaluating temporal variance across stations help establish the statistical validity of the dataset (Table 3).

Table 3: Bioconcentration Factors (BCF) and Associated ANOVA Significance Profiles

Heavy Metal	Seasons	Sangam Barrage: <i>Nymphaea</i> BCF	Sangam Barrage: <i>Lemna</i> BCF	Penna Bridge: <i>Nymphaea</i> BCF	Penna Bridge: <i>Lemna</i> BCF	Station Matrix	ANOVA F-Value	ANOVA P-Value	Significance Profile
Arsenic	Pre-monsoon	0.000	0.000	0.092	0.333	Sangam Water	1.00	0.422	Not Significant
	Monsoon	0.010	0.030	0.846	0.923	Penna Bridge Water	3.50	0.179	Not Significant
	Post-monsoon	0.000	0.000	0.667	0.778	Penna Bridge <i>Lemna</i>	4.86	0.170	Not Significant
Cadmium	Pre-monsoon	0.286	0.143	5.000	2.333	Sangam <i>Lemna</i>	7.50	0.120	Not Significant
	Monsoon	5.000	3.000	0.667	0.167	Penna Bridge Water	2.25	0.260	Not Significant
	Post-monsoon	0.200	0.000	0.286	0.143	Penna Bridge <i>Lemna</i>	11.45	0.080	Nearly Significant
Lead	Pre-monsoon	0.000	1.000	0.120	0.800	Sangam Water	1.54	0.350	Not Significant

	Monsoon	0.143	0.900	0.125	0.625	Penna Bridge Water	3.92	0.170	Not Significant
	Post-monsoon	0.167	0.783	0.164	0.455	Penna Bridge <i>Lemna</i>	6.70	0.130	Not Significant
Mercury	Pre-monsoon	0.000	1.667	0.167	0.500	Sangam Water	1.00	0.420	Not Significant
	Monsoon	0.333	0.833	0.222	0.667	Sangam <i>Lemna</i>	10.00	0.095	Nearly Significant
	Post-monsoon	0.000	1.500	0.286	0.714	Penna Bridge Water	1.50	0.360	Not Significant

Note: BCF is calculated as C_{plant} / C_{water} . ANOVA outcomes designate seasonal variation patterns within each station matrix.

3.1 Interpretation of Statistical Non-Significance

All calculated ANOVA models across water columns and plant tissues returned high p-values ($p > 0.05$), rendering the seasonal changes **statistically non-significant**. For instance, the temporal variance of As in water at Sangam Barrage returned an F-value of 1.00 ($p = 0.422$), while Pb variance in water at Penna Bridge returned an F-value of 3.92 ($p = 0.170$). The closest metrics to the standard significance threshold were Cd in *Lemna minor* at Penna Bridge ($F = 11.45$; $p = 0.080$) and Hg in *Lemna minor* at Sangam Barrage ($F = 10.00$; $p = 0.095$).

In field ecotoxicological assessments conducted on open, open-channel river basins, non-significant ANOVA outputs are common and do not undermine data integrity. Unlike highly controlled, isolated laboratory exposures, the Penna River is a dynamic lotic ecosystem. It experiences unpredictable variations, including periodic upstream dam water releases, shifting urban sewer outputs, changing agricultural pumping demands, and the physical drifting of weed populations.

The large standard deviations observed—such as As during the monsoon at Sangam Barrage (0.100 ± 0.037 mg/L) reflect these periodic concentration pulses. The nearly significant outputs ($p < 0.1$) observed for *Lemna minor* show that floating macrophytes register these environmental spikes rapidly due to their direct contact with the water current, making them useful indicators despite high ecosystem variance.

4. Comparative Bioconcentration Factors (BCF Analysis)

The calculated Bioconcentration Factors (Table 3) evaluate the physiological efficiency of metal extraction from water to plant tissues.

4.1 Anatomical Influences on BCF (Lemna vs. Nymphaea)

The data shows that **the free-floating species *Lemna minor* exhibits higher BCF values for Pb and Hg than the rooted floating species *Nymphaea nouchali*** across almost all seasons.

- At Penna Bridge during the pre-monsoon, *Lemna* achieved a Pb BCF of 0.800, whereas *Nymphaea* leaves recorded a much lower BCF of 0.120.
- For Mercury at Sangam Barrage during the pre-monsoon, *Lemna* reached a high BCF of 1.667, while *Nymphaea* remained at 0.000 due to non-detectable uptake.

This difference relates directly to the plants' anatomy and exposure routes. *Lemna minor* is a small, free-floating macrophyte. Its entire vegetative structure including its cellular under-surface and short root network is constantly submerged in water. This maximizes its surface-area-to-volume ratio, facilitating rapid, passive biosorption across its cell walls.

In contrast, **only the leaves of *Nymphaea nouchali* were analyzed**. In rooted aquatic plants, heavy metals are primarily taken up by roots embedded in the bottom sediments. To reach the leaves, these metals must be transported up through the vascular bundles of the petioles. Plant physiology studies indicate that roots often act as a protective barrier, binding toxic elements like Pb and Hg within their cell walls to safeguard the leaf's photosynthetic machinery. This protective mechanism explains the lower metal accumulation and smaller BCF values observed in *Nymphaea* leaves.

4.2 Differentiated Translocation of Cadmium

The major exception to this anatomical pattern occurred with **Cadmium**, where *Nymphaea nouchali* leaves showed high bioconcentration performance:

- During the monsoon at Sangam Barrage, water Cd levels dropped to a low of 0.001 mg/L, but *Nymphaea* leaves maintained a body burden of 0.005 mg/Kg, resulting in a **BCF of 5.000** (outperforming *Lemna's* BCF of 3.000).

- During the pre-monsoon at Penna Bridge, *Nymphaea* achieved a **BCF of 5.000**, doubling the performance of *Lemna minor* (2.333).

This indicates that *Nymphaea nouchali* possesses a highly effective internal pathway for moving Cd from roots to leaves. Unlike Pb and Hg, which bind tightly to root tissues, Cd ions behave chemically like essential divalent nutrients such as Zn^{2+} and Ca^{2+} . Consequently, *Nymphaea* takes up and translocates Cd through its active nutrient transport systems into its leaves. This active movement makes the floating leaves of *Nymphaea nouchali* an effective bio-indicator for monitoring low-level Cadmium pollution in river systems.

Conclusion

The present study provides a critical assessment of the spatiotemporal heavy metal dynamics and phytoremediation potential of *Nymphaea nouchali* and *Lemna minor* along the River Penna at Nellore, Andhra Pradesh. The spatial analysis revealed a distinct pollution gradient, with the urbanized Penna Bridge station systematically exhibiting higher aqueous concentrations of Arsenic, Cadmium, Lead, and Mercury compared to the agricultural Sangam Barrage station. This disparity underlines the profound impact of untreated municipal sewage, storm drainage, and urban industrial runoff over agricultural return flows.

Furthermore, the seasonal dynamics challenged traditional river dilution models, as peak heavy metal levels predominantly occurred during the monsoon season. This trend underscores the role of high-intensity monsoonal precipitation in driving surface runoff kinetics and catchment scouring, which flushes accumulated agrochemicals and urban industrial dust directly into the lotic system.

Ecotoxicological profiling of the selected aquatic macrophytes demonstrated highly species-specific and tissue-dependent bioaccumulation mechanics. The free-floating macrophyte *Lemna minor* emerged as a highly efficient bio-indicator and sink for Lead and Mercury, consistently yielding superior tissue accumulation and higher Bioconcentration Factors (BCF) across most seasonal cycles. This high efficiency is attributed to its unanchored, fully submerged vegetative structure, which maximizes the surface-area-to-volume ratio for passive cell-wall biosorption. Conversely, the rooted macrophyte *Nymphaea nouchali* exhibited a specialized physiological exception for Cadmium. Despite lower relative water concentrations, *Nymphaea* foliar tissues achieved a maximum BCF of 5.000, demonstrating a highly active root-to-leaf vascular translocation pathway for Cd ions via essential divalent nutrient pathways.

While the field-based environmental variance rendered seasonal changes statistically non-significant under ANOVA evaluation ($p > 0.05$), the high responsiveness of these macrophytes—particularly the nearly significant sensitivity profiles of *Lemna minor*—confirms their utility as continuous, integrated biological monitoring tools in dynamic river ecosystems. Ultimately, these findings provide an empirical foundation for government bodies and environmental agencies to design targeted, low-cost phytoremediation matrices. Deploying managed communities of *Lemna minor* and *Nymphaea nouchali* can actively mitigate heavy metal threats, safeguarding public health and preserving the biodiversity of the River Penna basin.

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