

Bioremediation Potential of Seaweeds *Enteromorpha intestinalis* and *Ulva lactuca* in Mitigating Nitrogen and Phosphorus Pollution in the Lower Gangetic Delta (LGD)

Debatri Banerjee^{1,2}, Prosenjit Pramanick¹, Arpita Saha¹, Sufia Zaman¹ and Abhijit Mitra¹

¹Department of Oceanography, Techno India University, West Bengal, EM 4/1 Salt Lake, Sector V, Kolkata 700091, India

²Faculty of Environmental Studies, St. Xavier's University, Action Area III B, New Town, Kolkata 700160, West Bengal, India

Abstract

Eutrophication has become a critical environmental issue worldwide, primarily driven by elevated nutrient levels in aquatic ecosystems. Excessive nitrogen (N) and phosphorus (P) from domestic waste, tourism, agriculture, and aquaculture intensify this process in coastal and estuarine environments. In the Lower Gangetic Delta (LGD), nutrient runoff peaks during the monsoon, posing a significant threat to ecosystem health. This study assesses the bioremediation potential of two seaweed species, *Enteromorpha intestinalis* and *Ulva lactuca*, by evaluating their nutrient absorption capacity at two LGD sites: Chotomollakhali and Harinbari, each with varying salinity levels. Data from 2014–2018 reveal significant site-wise differences in Nitrogen and Phosphorus Enrichment Factors (NEF and PEF) for both species, indicating that salinity and nutrient concentration modulate their efficacy. Findings suggest that seaweed-based bioremediation can serve as a sustainable solution for nutrient pollution, supporting coastal management and mitigating the adverse effects of eutrophication.

Keywords: Lower Gangetic Delta (LGD), Eutrophication, *Enteromorpha intestinalis* and *Ulva lactuca*, Nitrogen Enrichment Factor (NEF), Phosphorus Enrichment Factor (PEF)

1. Introduction

Eutrophication, triggered by excessive nutrient loading in aquatic ecosystems, has emerged as one of the most challenging environmental problems in the present *era*. This nutrient enrichment often results from the accumulation of nitrogen (N) and phosphorus (P) due to human activities, including agricultural runoff, industrial waste, aquacultural waste, tourism waste, and domestic sewage discharge. In estuarine and coastal ecosystems, such nutrient influx can disrupt ecological balance, leading to harmful algal blooms, oxygen depletion, and deterioration of aquatic environment (Carpenter et al., 1998; Anderson et al., 2002; Kato et al., 2009; Lintern et al., 2020; Peñuelas and Sardans 2022; Baldrich et al., 2024; Lan et al., 2024).

The Lower Gangetic Delta (LGD), a vital mangrove dominated estuarine ecosystem in the Indian sub-continent, is increasingly impacted by nutrient pollution due to rapid urbanization, tourism, and aquaculture (Mitra, 2013). The monsoon season intensifies nutrient loading through runoff from inland sources, exacerbating the eutrophication process in coastal waters. Coastal aquaculture, particularly shrimp farming, has contributed to nutrient loading in the LGD, with adverse impacts on the adjacent mangrove ecosystems (Biao et al., 2004; Ahmed et al., 2008; Buschmann et al., 2008; Hossain et al., 2013). This situation calls for effective, sustainable approaches to managing nutrient pollution.

Bioremediation using seaweeds offers a promising solution, given their natural capacity to absorb and assimilate nutrients. Seaweeds, particularly species in the genera *Enteromorpha* and *Ulva*, have demonstrated a significant ability to uptake N and P from surrounding waters, suggesting potential applications for nutrient removal in eutrophic ecosystems (Holdt and Kraan 2011; Yang et al., 2015; Pechsiri et al., 2016; Hasselströma et al., 2018). This study focuses on assessing the bioremediation potential of *Enteromorpha intestinalis* and *Ulva lactuca* in the LGD, evaluating their nutrient uptake across varying salinity conditions at two sites, Chotomollakhali and Harinbari.

2. Materials and Methods

2.1. Study Area

The study was conducted at two sites in the Lower Gangetic Delta, Chotomollakhali and Harinbari, chosen for their variation in salinity levels. Chotomollakhali (22°10'40.00"N; 88°54'26.71"E), characterized by higher salinity, and Harinbari (21°46'53.07"N; 88°04'22.88"E), with lower salinity, provide a basis for assessing the impact of salinity on nutrient absorption by seaweeds. These sites experience nutrient influx during the monsoon season, making them suitable for evaluating the bioremediation potential of seaweed species in nutrient-rich waters.

2.2. Sampling Period and Procedure

Sampling was conducted annually in September, coinciding with the monsoon season from 2014 to 2018. This period represents peak nutrient runoff, providing an ideal scenario for studying nutrient absorption in seaweeds. Water samples were collected from each site to measure dissolved nitrogen (NO₃-N) and phosphorus (PO₄-P) concentrations. Samples of *E. intestinalis* and *U. lactuca* were simultaneously collected to analyse nutrient content within the seaweed tissue.

2.3. Measurement of Nitrogen and Phosphorus Concentrations

2.3.1. Water Analysis

Dissolved NO₃-N and PO₄-P levels were measured in collected water samples using standard spectrophotometric methods (APHA, 2017; Habibah et al., 2018), recorded in milligrams per liter (mg/L).

2.3.2. Seaweed Analysis

Seaweed samples were rinsed with deionized water to remove any attached particulates and then dried at 60°C to a constant weight. The dried samples were ground, and nutrient analyses were conducted to determine N and P concentrations in milligrams per kilogram (mg/kg) as per the procedure outlined by Caisso et al. (1968).

2.3.3. Enrichment Factor Calculations

The bioremediation potential of each seaweed species was evaluated using Nitrogen Enrichment Factor (NEF) and Phosphorus Enrichment Factor (PEF), as indicators as per the expressions stated here:

$$\text{NEF} = \frac{\text{N concentration in seaweed (mg/kg)}}{\text{NO}_3\text{-N concentration in water (mg/L)}}$$

and

$$\text{PEF} = \frac{\text{P concentration in seaweed (mg/kg)}}{\text{PO}_4\text{-P concentration in water (mg/L)}}$$

These factors provide a quantitative measure of each seaweed's nutrient uptake efficiency, normalized for ambient nutrient concentrations in the ambient estuarine water.

2.4. Statistical Analysis

Statistical analyses were performed to assess differences in the values of all selected variables across sites and years for both the species. Analysis of variance (ANOVA) was applied to determine significant effects of salinity and nutrient levels in the seaweed species. Data analyses were conducted using R software, with significance levels set at $p < 0.05$.

3. Results and Discussion

3.1. Dissolved Nitrogen and Phosphorus

The dissolved nitrogen (NO₃-N) levels across the stations of Chotomollakhali and Harinbari show a gradual increase from 2014 to 2018. In Chotomollakhali, a high-saline zone, the NO₃-N levels rose from 0.021 mg/L in 2014 to 0.042 mg/L in 2018. Similarly, in Harinbari, which

is a relatively low-saline area, the $\text{NO}_3\text{-N}$ levels increased from 0.019 mg/L in 2014 to 0.039 mg/L in 2018.

Phosphorus ($\text{PO}_4\text{-P}$) levels also display distinct trends at each station. In Chotomollakhali, $\text{PO}_4\text{-P}$ remained between 0.002 mg/L and 0.005 mg/L across the years, while in Harinbari, $\text{PO}_4\text{-P}$ fluctuated, ending at 0.003 mg/L in 2018 after a peak of 0.004 mg/L in 2017.

This upward trend in dissolved nitrogen ($\text{NO}_3\text{-N}$) levels across Chotomollakhali and Harinbari, alongside the slight variability in phosphorus ($\text{PO}_4\text{-P}$), suggests increasing nutrient loading, likely due to rising human activities at both sites over time (Figs. 1 and 2). The significant site-wise variations of $\text{NO}_3\text{-N}$, as revealed through ANOVA (Table 1) may be attributed to site-wise variations in human activities.

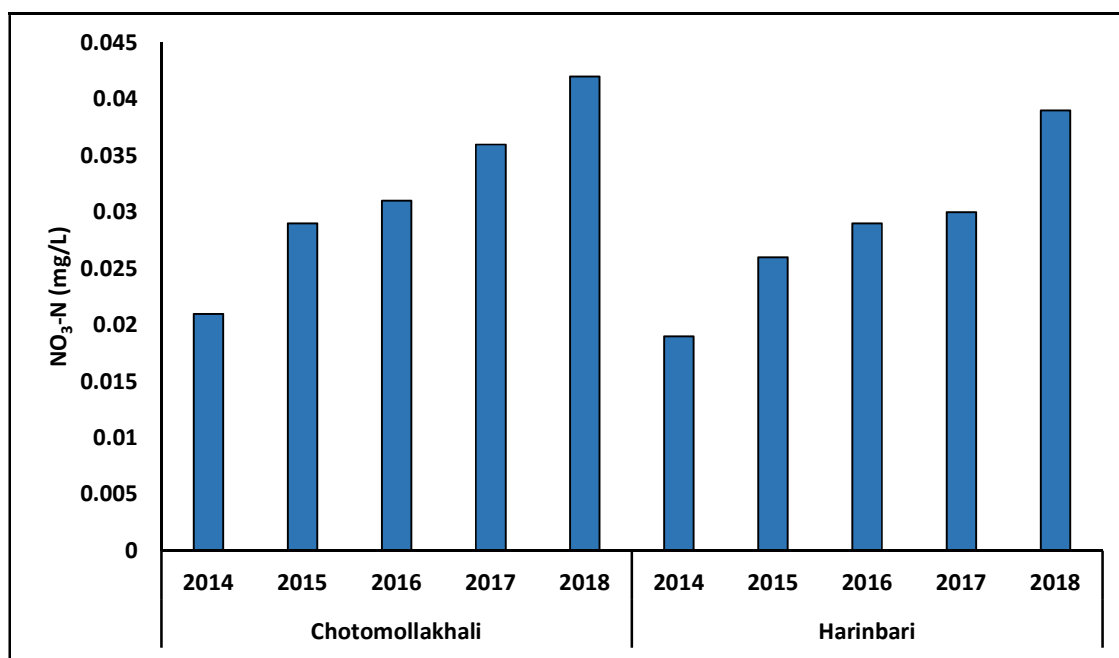


Fig. 1. Station-wise yearly variation of water nitrate-nitrogen concentration (in mg/L)

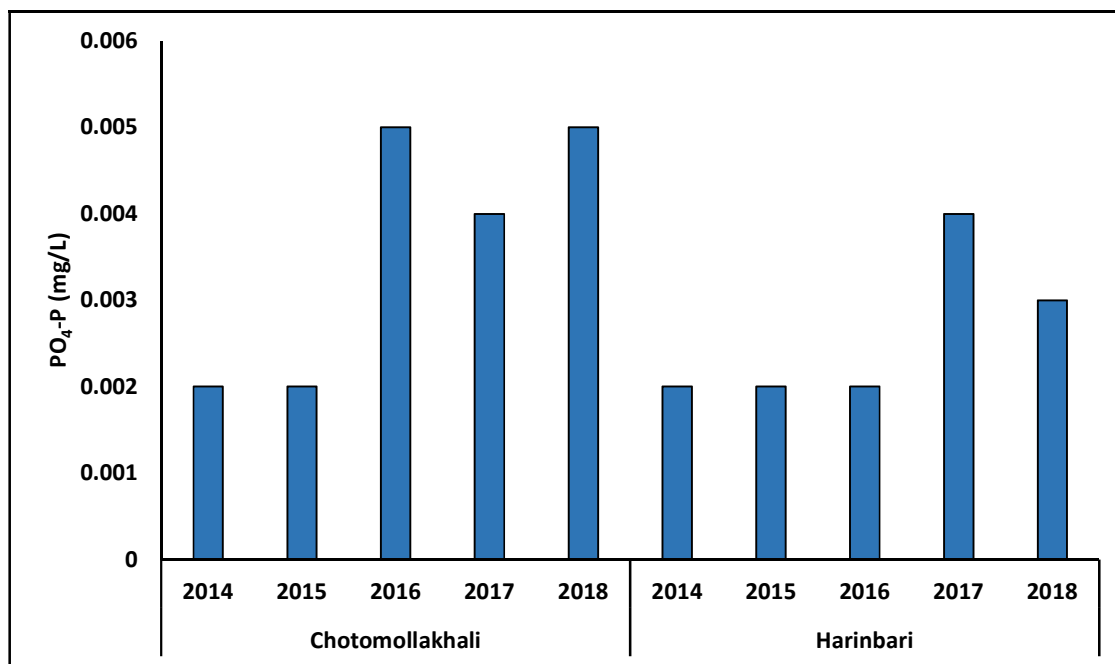


Fig. 2. Station-wise yearly variation of water phosphate-phosphorus concentration (in mg/L)

3.2. Nutrients in Seaweed tissue

The nitrogen (N) and phosphorus (P) levels in the seaweed species *Enteromorpha intestinalis* and *Ulva lactuca* reveal marked variations across both the high-saline Chotomollakhali and the lower-saline Harinbari sites from 2014 to 2018. In *E. intestinalis*, nitrogen levels in Chotomollakhali declined from 31,601 mg/kg in 2014 to 25,009 mg/kg in 2018, while Harinbari exhibited a more pronounced decrease, with nitrogen levels dropping from 30,931 mg/kg to 16,755 mg/kg over the same period. Similarly, *U. lactuca* in Chotomollakhali saw nitrogen levels decrease from 28,766 mg/kg to 20,446 mg/kg, with Harinbari's levels reducing from 26,002 mg/kg to 15,432 mg/kg.

For phosphorus, *E. intestinalis* in Chotomollakhali showed a slight decrease from 2,006 mg/kg in 2014 to 1,753 mg/kg in 2018, whereas Harinbari observed a decline from 1,853 mg/kg to 1,395 mg/kg. In *U. lactuca*, phosphorus levels in Chotomollakhali reduced from 1,938 mg/kg in 2014 to 1,280 mg/kg in 2018, while Harinbari saw a reduction from 1,805 mg/kg to 1,289 mg/kg. These patterns indicate a steady decline in nitrogen and phosphorus concentrations in both seaweed species (Figs. 3 and 4).

The significant differences in nutrient levels within the seaweeds between the sites ($p < 0.05$) can be attributed to biomass variations, with salinity acting as the primary driver (Table 1).

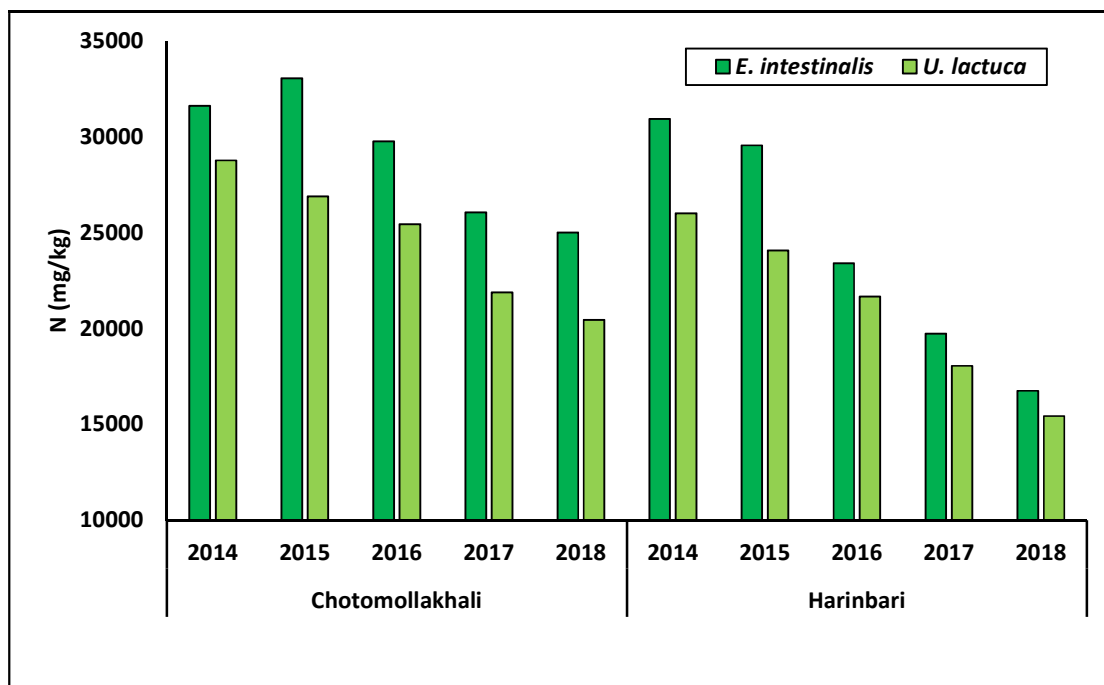


Fig. 3. Species-wise yearly variation of tissue nitrogen concentration (in mg/kg) at both the selected stations

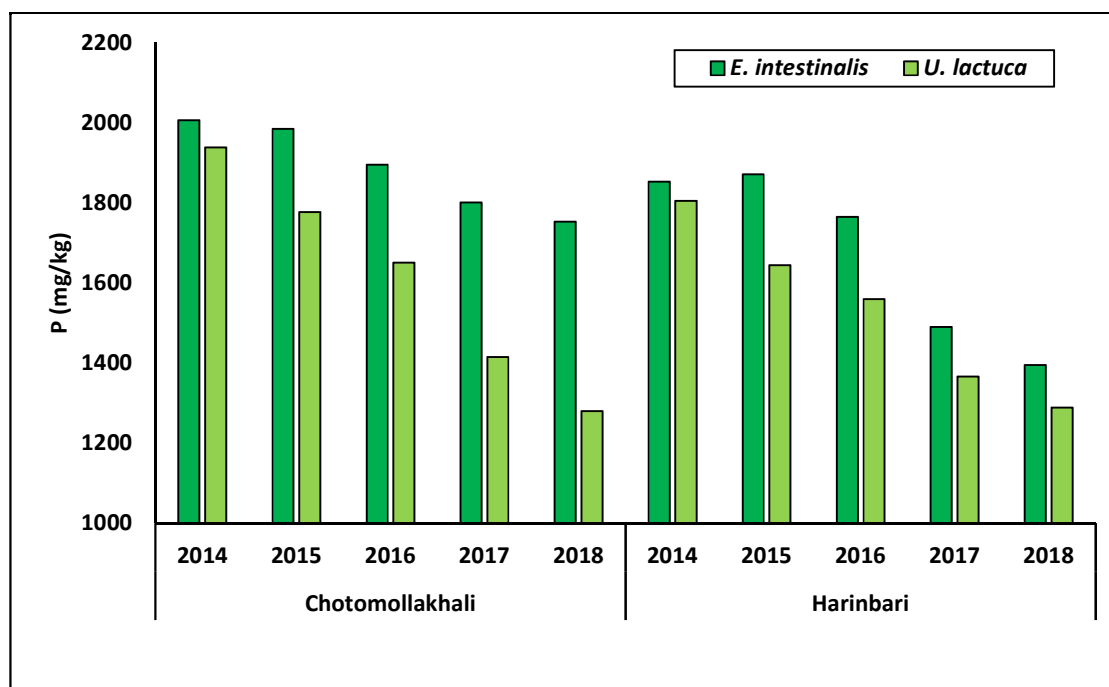


Fig. 4. Species-wise yearly variation of tissue phosphorus concentration (in mg/kg) at both the selected stations

3.3. Nitrogen Enrichment Factor (NEF)

The NEF values for both *Enteromorpha intestinalis* and *Ulva lactuca* revealed distinct patterns across the sites and years. Generally, *Enteromorpha* exhibited higher NEF values than *Ulva*,

indicating a superior nitrogen absorption capacity (Kamer and Fong, 2001) (Fig. 5). At Chotomollakhali, NEF values were consistently higher than at Harinbari, suggesting that elevated salinity in Chotomollakhali may positively influence nitrogen uptake. However, NEF values for both species declined over the years (2014–2018), potentially reflecting decreasing nutrient availability, saturation effects in seaweed tissues, or adaptive responses to changing environmental conditions.

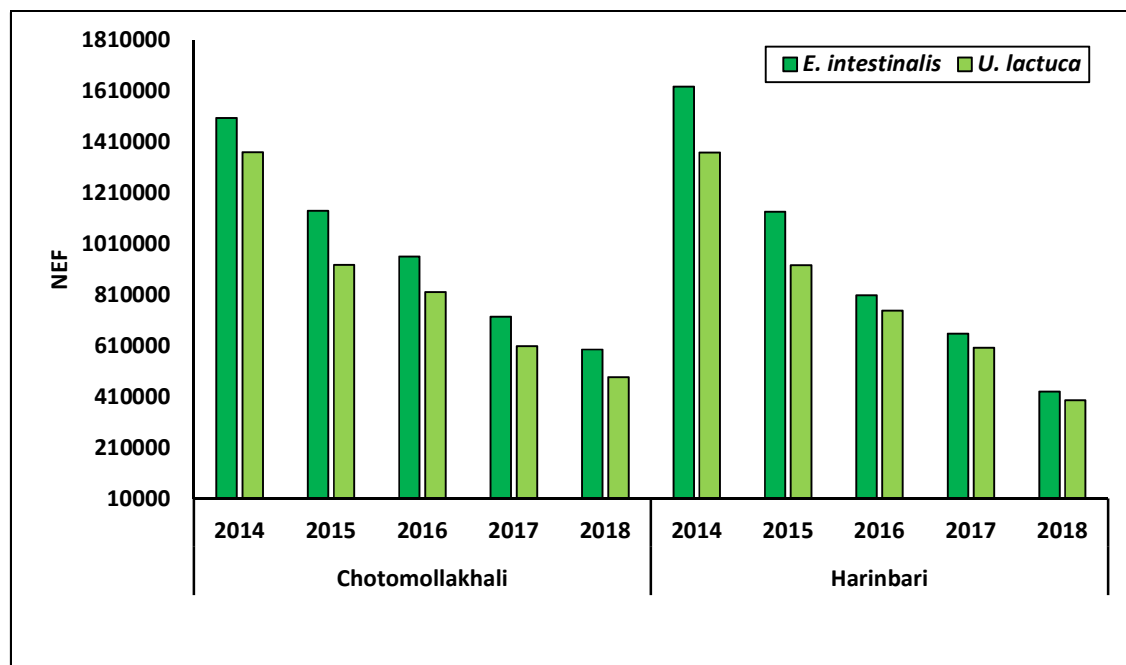


Fig. 5. Species-wise yearly variation of Nitrogen Enrichment Factor (NEF) at both the selected stations

3.4. Phosphorus Enrichment Factor (PEF)

In terms of phosphorus absorption, *Enteromorpha* again showed a stronger bioremediation potential, with higher PEF values compared to *Ulva* (Fig. 6). Chotomollakhali exhibited slightly higher PEF values than Harinbari, although data variability was notable. This variability could be due to factors such as fluctuations in ambient phosphorus levels, which might affect phosphorus uptake rates (Douglas et al., 2014). PEF values also displayed a decreasing trend over the study period, mirroring the trend observed for NEF values.

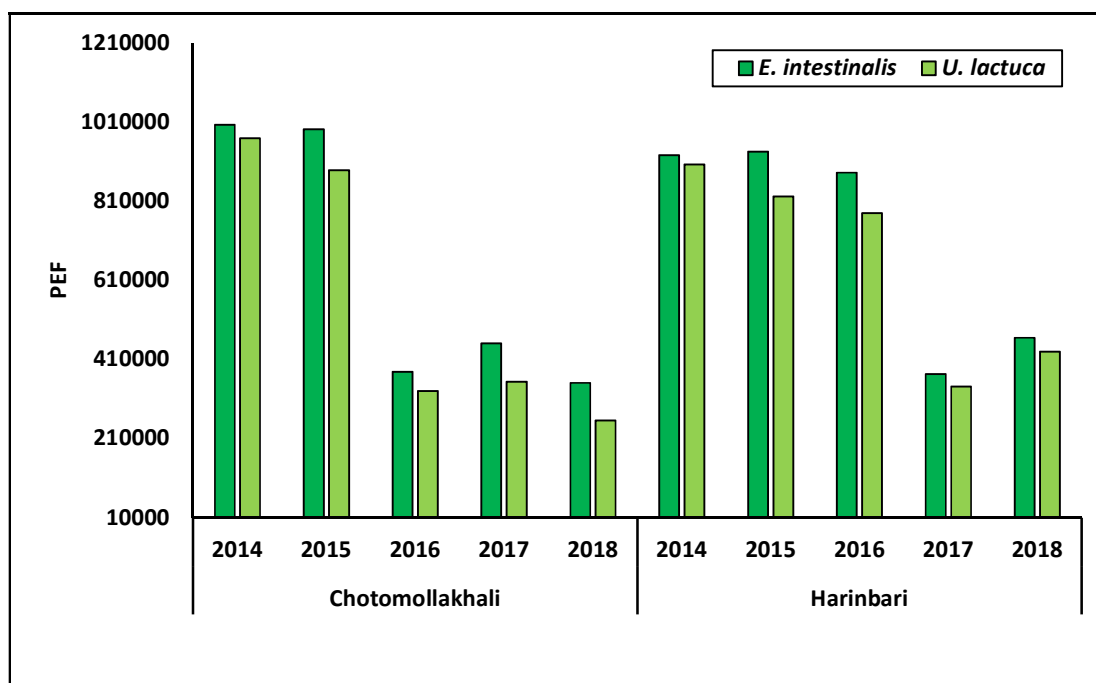


Fig. 6. Species-wise yearly variation of Phosphorus Enrichment Factor (PEF) at both the selected stations

3.5. Site-Specific Observations and Implications

The consistent observation of higher NEF and PEF values in Chotomollakhali suggests that salinity plays a critical role in modulating nutrient absorption in these seaweeds. *Enteromorpha intestinalis* demonstrated a robust capacity to absorb both nitrogen and phosphorus, making it a potentially effective species for bioremediation in higher salinity environments. This study's findings underscore the significance of site-specific factors such as salinity in shaping the efficacy of seaweed-based bioremediation.

The moderately high salinity in Chotomollakhali appears to provide a congenial environment for the optimal growth of *E. intestinalis* and *U. lactuca*, allowing for stable nutrient absorption and relatively consistent nutrient levels. This environment supports a balanced nutrient uptake, contributing to higher nutrient enrichment factors (NEF and PEF) in these seaweeds. In contrast, the extremely low salinity during the monsoon season in Harinbari often hampers the growth of these seaweed species, resulting in lower nutrient uptake and reduced tissue nutrient levels. This site-wise discrepancy in growth conditions is evident in the significant differences in nutrient concentrations and enrichment factors between the two sites, as confirmed by ANOVA analysis (Table 1). The lower salinity stress in Harinbari likely limits nutrient assimilation efficiency, leading to marked variability in nutrient accumulation between the two regions.

The observed decreasing trend in nitrogen (N) and phosphorus (P) levels in *E. intestinalis* and *U. lactuca*, alongside increasing dissolved nutrients in the water, raises a very interesting issue. We presume that this could be due to the facts that these seaweed species are experiencing either adaptive responses or supersaturation in nutrient uptake. As nutrient concentrations in water rise, seaweeds initially absorb more, but beyond a threshold, they may downregulate absorption as an adaptive mechanism to avoid toxicity. Additionally, if the seaweed populations have reached their carrying capacity for nutrient absorption (NEF and PEF), further nutrient accumulation in the water may no longer lead to proportional increases in their tissue nutrient levels. This supersaturation indicates a potential nutrient saturation point, where internal cellular regulation or metabolic limitations prevent further storage, despite external nutrient abundance.

This supersaturation suggests a nutrient threshold, where internal cellular regulation or metabolic constraints limit further nutrient storage in seaweeds, even with high external nutrient availability. To optimize nutrient uptake for bioremediation, further research is needed to determine the ideal timing for seaweed harvest. Harvesting at the right stage would allow the recycling of seaweed as biofertilizer (Panda et al., 2012), followed by the introduction of fresh thalli to the site. This cyclical approach could sustain effective bioremediation by maintaining seaweed populations actively engaged in nutrient absorption.

Table 1. ANOVA showing the yearly and site-wise variations of NO₃-N, PO₄-P, and seaweed nutrient level in Indian Sundarbans

Species	Parameters	Variables	Fcal	p-value	Fcrit
Water	NO ₃ -N	Between Sites	18.9630	0.0121	7.7086
		Between Years	83.4444	0.0004	6.3882
	PO ₄ -P	Between Stations	2.5000	0.1890	7.7086
		Between Years	2.1000	0.2450	6.3882
<i>E. intestinalis</i>	N	Between Sites	14.3390	0.0193	7.7086
		Between Years	10.2816	0.0222	6.3882
	P	Between Sites	17.7507	0.0136	7.7086
		Between Years	8.3825	0.0317	6.3882
<i>U. lactuca</i>	N	Between Sites	78.1488	0.0009	7.7086
		Between Years	71.3743	0.0006	6.3882
	P	Between Sites	8.6541	0.0423	7.7086
		Between Years	61.6175	0.0008	6.3882

4. Conclusion

This study reveals the promising bioremediation potential of *Enteromorpha intestinalis* and *Ulva lactuca* for mitigating nitrogen and phosphorus pollution in the Lower Gangetic Delta (Rahhou et al., 2023). Both seaweeds demonstrated significant nutrient accumulation potential,

with *Enteromorpha* showing superior nitrogen and phosphorus enrichment potential. The results suggest that seaweed-based bioremediation could be a viable strategy for controlling eutrophication, particularly in salinity-influenced estuarine areas like Chotomollakhali (Bews et al., 2021).

The observed decrease in nutrient uptake by seaweeds, despite rising water nutrient levels, likely stems from adaptive responses or supersaturation, where seaweeds downregulate absorption beyond a certain threshold to avoid toxicity. Once reaching their carrying capacity, further nutrient accumulation in water does not translate to proportional increases in tissue nutrient levels. This suggests a nutrient saturation point, where internal regulation limits further storage. To enhance bioremediation, research is needed to identify optimal harvest timing, enabling recycling as biofertilizer and introducing fresh thalli to maintain continuous nutrient uptake. In conclusion it can be advocated that incorporating seaweed bioremediation into coastal management plans could help reduce nutrient pollution, protect mangrove ecosystems, and sustain the health of the Lower Gangetic Delta's estuarine ecosystem.

References

1. Ahmed, N., Demaine, H. and Muir, J.F. 2008. Freshwater prawn farming in Bangladesh: history, present status and future prospects. *Aquaculture Research*, 39, 806-819.
2. Anderson, D.M., Glibert, P.M. and Burkholder, J.M. 2002. Harmful algal blooms and eutrophication: Nutrient sources, composition, and consequences. *Estuaries*, 25, 704-726.
3. APHA. 2017. Standard Methods for the Examination of Water and Wastewater (23rd ed). Method 4500 – NO₂-B.
4. Baldrich, Á.M., Díaz, P.A., Rosales, S.A., Rodríguez-Villegas, C., Álvarez, G., Pérez-Santos, I., Díaz, M., Schwerter, C., Araya, M. and Reguera, Ba. 2024. An Unprecedented Bloom of Oceanic Dinoflagellates (*Karenia* spp.) Inside a Fjord within a Highly Dynamic Multifrontal Ecosystem in Chilean Patagonia. *Toxins*, 16, 77.
5. Bews, E., Booher, L., Polizzi, T., Long, C., Kim, J.H. and Edwards, M.S. 2021. Effects of salinity and nutrients on metabolism and growth of *Ulva lactuca*: Implications for bioremediation of coastal watersheds. *Marine Pollution Bulletin*, 166, 112199.
6. Biao, X., Zhuhong, D. and Xiaorong, W. 2004. Impact of the intensive shrimp farming on the water quality of the adjacent coastal creeks from Eastern China. *Marine Pollution Bulletin*, 48, 543-553.
7. Buschmann, A.H., Varela, D.A., Hernández-González, M.C. and Huovinen, P. 2008. Opportunities and challenges for the development of an integrated seaweed-based

- aquaculture activity in Chile: determining the physiological capabilities of *Macrocystis* and *Gracilaria* as biofilters. *Journal of Applied Phycology*, 20, 571-577.
8. Caiozzi, M., Peirano, P., Rauch, E., and Zunino, H. 1968. Effect of Seaweed on the Levels of Available Phosphorus and Nitrogen in a Calcareous Soil. *Agronomy Journal*, 60(3), 324-326.
 9. Carpenter, S.R., Caraco, N.F., Correll, D.L., Howarth, R.W., Sharpley, A.N. and Smith, V.H. 1998. Nonpoint pollution of surface waters with phosphorus and nitrogen. *Ecological Applications*, 8, 559-568.
 10. Douglas, E.J., Haggitt, T.R. and Rees, T.A.V. 2014. Supply-and demand-driven phosphate uptake and tissue phosphorus in temperate seaweeds. *Aquatic Biology*, 23(1), 49-60.
 11. Habibah, N., Sri Dhyana Putri, I., Karta, I.W., Cok Dewi, W.H.S. and Choirul Hadi, M. 2018. A simple spectrophotometric method for the quantitative analysis of phosphate in the water samples. *Jurnal Sains dan Teknologi*, 7(2), 198-204.
 12. Hasselströma, L., Vischc, W., Gröndahla, F., Nylundc, G.M. and Pavia, H. 2018. The impact of seaweed cultivation on ecosystem services - a case study from the west coast of Sweden. *Marine Pollution Bulletin*, 133, 53–60.
 13. Holdt, S.L. and Kraan, S. 2011. Bioactive compounds in seaweed: functional food applications and legislation. *Journal of Applied Phycology*, 23(3), 543-597.
 14. Hossain, M.S., Uddin, M.J. and Fakhruddin, A.N.M. 2013. Impacts of shrimp farming on the coastal environment of Bangladesh and approach for management. *Reviews in Environmental Science and Biotechnology*, 12, 313–332.
 15. Kamer, K. and Fong, P. 2001. Nitrogen enrichment ameliorates the negative effects of reduced salinity on the green macroalga *Enteromorpha intestinalis*. *Marine Ecology Progress Series*, 218, 87-93.
 16. Kato, T., Kuroda, H. and Nakasone, H. 2009. Runoff characteristics of nutrients from an agricultural watershed with intensive livestock production. *Journal of Hydrology*, 368, 79-87.
 17. Lan, J., Liu, P., Hu, X. and Zhu, S. 2024. Harmful Algal Blooms in Eutrophic Marine Environments: Causes, Monitoring, and Treatment. *Water*, 16, 2525.
 18. Lintern, A., McPhillips, L., Winfrey, B., Duncan, J. and Grady, Cha. 2020. Best Management Practices for Diffuse Nutrient Pollution: Wicked Problems Across Urban and Agricultural Watersheds. *Environmental Science & Technology*, 54, 9159-9174.

19. Mitra, A. 2013. Sensitivity of mangrove ecosystem to changing climate, published by Springer, ISBN-10: 8132215087; ISBN-13: 978-8132215080. ISBN 978-81-322-1509-7 (eBook), XIX, pp. 323. DOI: <https://doi.org/10.1007/978-81-322-1509-7>.
20. Panda D, Pramanik K and Nayak B R. 2012. Use of Sea Weed Extracts as Plant Growth Regulators for Sustainable Agriculture. *Int. J. Bio-resource Stress Manag.*, 3 (3), 404–411.
21. Pechsiri, J.S., Thomas, J.B.E., Risén, E., Ribeiro, M.S., Malmström, M., Nylund, G., Jansson, A., Welander, U., Pavia, H. and Gröndahl, F. 2016. Energy performance and greenhouse gas emissions of kelp cultivation for biogas and fertilizer recovery in Sweden. *Science of the Total Environment*, 573, 347-355.
22. Peñuelas, J. and Sardans, J. 2022. The global nitrogen-phosphorus imbalance. *Science*, 375, 266–267.
23. Rahhou, A., Layachi, M., Akodad, M., El Ouamari, N., Rezzoum, N.E., Skalli, A., Oudra, B., El Bakali, M., Kolar, M., Imperl, J. et al. 2023. The Bioremediation Potential of *Ulva lactuca* (Chlorophyta) Causing Green Tide in Marchica Lagoon (NE Morocco, Mediterranean Sea): Biomass, Heavy Metals, and Health Risk Assessment. *Water*, 15, 1310.
24. Yang, Y., Chai, Z., Wang, Q., Chen, W., He, Z. and Jiang, S. 2015. Cultivation of seaweed *Gracilaria* in Chinese coastal waters and its contribution to environmental improvement. *Algal Research*, 9, 236-244.