

Evaluation of River Water Quality Using Phytoplankton Diversity Indices in a Riverine Ecosystem

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Abstract: Phytoplankton diversity indices reflects the health of the freshwater ecosystem; consequently these phytoplankton function as cost-effective bio indicators for determining the degree of pollution in our lifeline riverine ecosystem. In this study phytoplankton population density, their community composition and diversity indices were assessed as indicators of water quality along the Varanasi segment of the river Ganga in Uttar Pradesh. The Sedgwick-Rafter counting chamber method was used to quantify phytoplankton density while the key physicochemical parameters were analysed using standard methods. A total of 17 genera spanning four classes of Chlorophyceae, Bacillariophyceae, Euglenophyceae and Cyanophyceae were recorded consistently across all sites and seasons with Bacillariophyceae (diatoms) displaying dominance throughout. The Shannon-Wiener diversity index (H') varied from 2.612 to 2.739 Simpsons Index of Diversity (D) from 0.913 to 0.930 and Pielous index (J') from 0.922 to 0.967; the maximum diversity was consistently found at Manikarnika Ghat (Site IV) whereas the minimum occurred at Ramnagar (Site V) and during winter. Two-factor ANOVA with Replication showed a significant season $\times$ site interaction ($F = 4.74$ $p < 0.001$) for the physicochemical parameters while season and site alone were not statistically significant. Because these diversity indices fall within the moderately polluted category on established classification scales they suggest that the examined stretch is subject to moderate organic and anthropogenic stress even though it maintains a numerically stable and evenly structured phytoplankton community.

Keywords: Diversity Index, Physico-Chemical Parameter, Phytoplankton, River Ganga, Shannon-Wiener, Simpsons Index, Water Quality, Varanasi

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I. INTRODUCTION

➤ Ecological Role of Phytoplankton in Riverine Ecosystems

River ecosystems harbor diverse flora and fauna that exhibit ecosystem vitality and hydrochemical profile (Matta et al., 2018). Phytoplankton, tiny photosynthetic organisms, is particularly significant as they multiply rapidly and possess brief life spans, rendering them very sensitive to biotic and abiotic alteration. Moreover, they play a pivotal role in the aquatic trophic network and act as prime indicators of water quality and trophic conditions (Alam et al., 2015, 2020; Surya et al., 2018). Phytoplankton is crucial to riverine and other aquatic environments (Malik et al., 2021). In freshwater systems like rivers, lakes, and ponds, phytoplankton serves as the basal trophic level, transforming solar energy and dissolved nutrients into organic matter via photosynthesis. Negi and Rajput (2011) demonstrated a direct linkage between water quality and phytoplankton diversity. Nearly all aquatic ecosystems depend upon directly and indirectly on phytoplankton as their primary

food sources. Furthermore the other community organisms such as plankton, fin fish, and shellfish rely on these primary producers. The nutritional intake and yield of fish is highly affected by the population density of plankton in a given water body. The Ganga River is one of the most crucial river systems in South Asia, sustaining aquatic biodiversity and catering socioeconomic and nutritional foundation for millions (Singh et al., 2024).

➤ Phytoplankton as Bio Indicators of Water Quality

Phytoplankton remarkably changes their community composition based on slight alterations in physico-chemical determinants as compared to other indicators expressing these shifts. Phytoplankton populations respond quickly to environmental stress in terms of high reproduction and turnover rates, making them commonly utilized as bio-indicators of trophic conditions, pollution levels, and the well-being of aquatic ecosystems. Variations in phytoplankton communities directly imply the aquatic ecosystem health and its underlying environmental matrix (Gao et al., 2024).

Although the population density for each species indicates the structure and quantity of an indicator, it does not show how individuals are distributed, which is essential for evaluating community stability. Diversity indices consider both elements and are commonly acknowledged as the best metrics for assessing community diversity, as they integrate species richness and the relative abundance of individuals across species into a unified statistical measure of community health, stability, and trophic structure. The Shannon-Weaver index (Shannon & Weaver, 1949), based on information theory, quantifies community diversity by assessing the entropy of each species in relation to the elements of the system, whereas Simpson's index (Simpson, 1949) is a probability-based metric that evaluates the chance that two individuals selected at random are from the same species. These indices have become essential instruments for evaluating species diversity, community dynamics, and environmental quality, remaining in use as bio-indicators in many assessments of water quality throughout diverse river systems.

➤ *Phytoplankton Studies and Environmental Correlations in the Ganga River*

Kumar et al., (2023) have shown remarkable relationships between phytoplankton density and various physico-chemical determinants such as phosphate, dissolved solids, free carbon dioxide, bicarbonate, chloride, and silica in River Ganga. In another study on the Ganga River indicates that diatoms comprise a significant share of the predominant phytoplankton, whereas green algae and cyanobacteria typically thrive in eutrophic or in areas with minimal water movement (Kumar et al., 2022).

Shukla et al. (2015) studied the diversity of phytoplankton in the Ganga River at Allahabad (Prayagraj). They reported that the phytoplankton classes such as Bacillariophyceae, Chlorophyceae, and Cyanophyceae were found to be predominant. The highest and the lowest plankton density was observed at Chhatnag Ghat and Sangam, respectively which was further substantiated by Simpson's diversity index and a high Palmer pollution index implying that the entire river stretch (study area) was highly polluted. Pal et al. (2024) also documented that phytoplankton diversity was affected by human activities like mass bathing and idol immersion, and reported the dominance of Chlorophyta at Sangam and Rasulabad Ghat in the Ganga River, implying the increased organic pollution and human disruption. Sangam was identified to be relatively more contaminated than Rasulabad Ghat because of the huge population. Moreover the Palmer pollution index indicates high level of organic pollution at both sampling locations in which Sangam again pointing higher contamination than Rasulabad Ghat.

➤ *Anthropogenic Pressures and Rationale for the Present Study*

Due to several identified and unidentified discharge locations of the city and nearby areas, the significant amount of untreated or partially treated domestic wastewater is constantly aggravating river pollution. The various studies indicate that domestic sewage is a primary factor in the

notable decline of river water quality (Singh et al., 2015). In the context of Varanasi, Basak et al. (2015) revealed that the River Ghats of Varanasi has becoming a growing source of pollution from industrial discharges, household waste, and cremation debris. The on-going connection between the river, religious beliefs, culture, and the dense population inhabiting the Varanasi area has created a detrimental environmental stress zone, rendering it highly suitable for ecological assessment.

II. MATERIALS AND METHODS

➤ *Study Area and Sampling Sites:*

This study concentrated on the Varanasi section of the Ganga River (Uttar Pradesh), which faces significant stress from ritual, household, and industrial contributions owing to its roles in bathing, cremation, and idol immersion. Locations were chosen to represent various levels of human impact, from minimally affected upstream areas to heavily utilized Ghats (bathing, cremation, and sewage discharge sites), similar to the site-selection method used in earlier research on the water quality in this region (Basak et al., 2015). The five locations were chosen to represent the significant upstream-to-downstream progression of the Varanasi segment, spanning from the southern to the northern and eastern extremes of the Ghat series:

- Assi Ghat (upstream location): located at the southernmost end of the ghat series, at the junction with the Assi stream; relatively more undisturbed than the central ghats, and frequently mentioned as a reference point in regional water-quality research.
- Dashashwamedh Ghat (midstream location) — one of the most active ghats in the city, featuring the highest number of daily visitors participating in mass bathing and the daily Ganga Aarti ceremony; a central hub of pilgrim/tourist engagement.
- Rajghat (downstream location) — situated in the northern section of the city, near the historic Malviya (Dufferin) Bridge and close to where the river Varuna joins, combining its flow with the main current.
- Manikarnika Ghat (midstream, high-impact location) — a cremation area located by the river Ganga in central Varanasi on the western bank, thought to offer salvation to the soul post-death.
- Ramnagar (site on the right bank/opposite bank) — selected on the eastern bank of the Ganga River directly across from the city, close to Ramnagar Fort, to allow for a cross-river comparison.

These five sampling locations illustrate the primary upstream-midstream-downstream axis of the Varanasi section, along with a cross-river comparison site on the right bank (Ramnagar).

➤ *Identification and Enumeration of Phytoplankton:*

- Collection of Samples: Surface water samples (1 L, 0–30 cm depth) were gathered from the five sampling locations (Site I–V) along the Ganga River during the pre-monsoon, monsoon, post-monsoon, and winter

periods, utilizing a plankton net (mesh size 25–30 μm) or through direct grab sampling and filtration (APHA, 2017).

- **Preservation:** Samples were promptly stabilized in the field using a 4% formalin solution or Lugol's iodine solution (1 mL/100 mL sample) in suitably labeled bottles to prevent bacterial growth, thereby halting the revival of a living state, and to maintain the organisms' morphological characteristics for identification (APHA, 2017).
- **Sedimentation and Concentration:** In the lab, separate samples were allowed to rest for 24–48 hours to enable the suspended plankton to settle (Adoni, 1985). The supernatant was gently removed without disturbing the sediment, and the sample was concentrated to a specific volume — for instance, from 1000 mL to 25–50 mL. This factor of concentration was noted. The complete concentrated sample was uniformly mixed through gentle shaking/stirring prior to counting.
- **Enumeration via the Sedgwick-Rafter (S-R) Enumeration Chamber:** The density of the phytoplankton population was determined employing the Sedgwick-Rafter counting chamber technique (APHA, 2017; Welch, 1948). A 1 mL portion of the homogenized and concentrated sample was introduced into the S-R chamber (50 mm $\times$ 20 mm $\times$ 1 mm; total capacity 1000 mm³). The sample remained undisturbed for 15–20 minutes to enable organisms to settle on the chamber's bottom. Phytoplankton identification utilized existing literature (Bellinger and Sigee, 2010), with confirmed algal taxonomic names sourced from Algaebase (Guiry and Guiry, 2023). Organisms were categorized to the genus level (Prescott, 1962; Edmondson, 1959) and counted in ten randomly chosen horizontal strips beneath a compound microscope, employing a non-stratified random sampling approach. Counting was performed in duplicates (Replicate 1 and Replicate 2), with the average utilized for graph plotting.
- **Calculation of Density:** The density of organisms (N) was initially determined using the conventional Sedgwick-Rafter formula (APHA, 2017; Welch, 1948):

$$N (\text{organisms/mL}) = \frac{(C \times 1000)}{(A \times D \times F)}$$

Where N = the count of organisms per mL of the concentrated sample; C = the sum of all organisms counted across strips/fields; A = the area of a counted strip/field (mm²) for a complete 50 mm strip, A = 50 mm²; D = the depth of the Sedgwick-Rafter chamber (mm) = 1 mm (constant); F = the total number of strips (fields) counted; and 1000 = the conversion factor (mm³ chamber volume $\rightarrow$ μL , due to 1 mL = 1000 mm³). Given that every 1 mm-wide strip contains a volume of 50 mm³, the formula changes the total number of organisms counted per examined volume into organisms per mL.

The organism density (N) was subsequently transformed into population density per litre of the original river water utilizing the formula:

$$\text{Population Density (individuals/L)} = N \times \frac{V_c}{V_f}$$

Where N = organisms/mL, V_c = volume of the concentrated sample (mL), and V_f = volume of the original filtered water (L).

➤ *Physicochemical Characteristics*

Alongside biotic sampling of phytoplankton population density, essential physicochemical parameters were assessed at every site and season to evaluate their possible relationship with phytoplankton population density in this section of the river Ganga. Water temperature ($^{\circ}\text{C}$), pH, and dissolved oxygen (DO) were directly assessed using portable pH/DO meters, while biochemical oxygen demand (BOD) and nutrient levels (nitrate-N, mg/L and silicate, mg/L) were determined in the lab following standard protocols (APHA, 2017). These physicochemical factors are acknowledged as crucial elements influencing the community composition and population density of phytoplankton in river systems like the Ganga (Kumar et al., 2022).

➤ *Statistical Evaluation*

Data collected from the five sampling sites underwent statistical analysis using two-factor ANOVA (with replication) and various community diversity indices — the Shannon-Wiener Diversity Index (H') (Shannon & Weaver, 1949), Simpson's Diversity Index (D) (Simpson, 1949), Species Richness (S), and Pielou's Evenness Index (J') (Pielou, 1966) — to assess the spatial and temporal variations in the phytoplankton community.

- **Analysis of Variance (ANOVA):** A two-factor ANOVA with replication was performed in Microsoft Excel utilizing the Data Analysis ToolPak to assess the interaction effects of season and sampling site on the physicochemical parameters evaluated for the Ganga River. In this arrangement, “Season” (Factor 1: pre-monsoon, monsoon, post-monsoon, winter) served as the row variable, while “Sampling Site” (Factor 2: Site I–V) acted as the column variable. The null hypotheses examined were: (a) there is no notable difference across seasons; (b) there is no noteworthy difference among sampling sites; and (c) there is no significant interaction between season and sampling location. Every effect was assessed at a significance level of $\alpha = 0.05$, with significance marked when the computed F value surpassed the critical F value (F_{crit}) and/or when the P value fell below 0.05.
- **Examination of Diversity Index:** Phytoplankton population density (individuals/L) was calculated for each of the five sampling locations (Site I–V) throughout the four seasons to assess community diversity indices for each site–season pair. The relative frequency (p_i) of every genus was determined using:

$$p_i = \frac{n_i}{N}$$

Where n_i represents the population density of the i -th genus, while N denotes the overall phytoplankton population

density for the specific site–season sample. Species diversity was calculated using the following methods:

Species Richness (S) = count of genera at the specific site and season;

Simpson's Index of Diversity (D) = $1 - \sum (p_i^2)$;

Shannon-Wiener Diversity Index (H') = $-\sum (p_i \ln p_i)$; and

Pielou's Evenness Index (J') = $\frac{H'}{\ln(S)}$

III. RESULTS

To achieve the goal of the research named “Evaluation of River Water Quality Using Phytoplankton Diversity Indices in a Riverine Ecosystem” the current study was conducted throughout all four seasons — monsoon (August), pre-monsoon (April), post-monsoon (November), and winter (January) — at five sites covering around 12 km from the entry point to the downstream boundaries of Varanasi city. The study is divided into three sections: (i) physicochemical properties varying by site and season, (ii) phytoplankton density and composition analyzed by season and site, and (iii) diversity indices assessed in connection with water quality.

➤ Physicochemical Properties of River Water at Different Locations and Seasons:

Figure 1 summarizes the physicochemical parameters documented at various sampling locations and during different seasons. Seasonal examination showed a steady transition from pre-monsoon to monsoon, closely associated with the existing temperature pattern. Water temperature exhibited significant seasonal fluctuations, spanning from 18.2°C to 27.4°C, peaking in the pre-monsoon (26.2–27.4°C), followed by a gradual decrease during monsoon (24.1–25.2°C), reaching its lowest point in post-monsoon (18.2–19.7°C), and experiencing a slight increase in winter (21.4–22.8°C) — a pattern linked to variations in seasonal insolation and ambient air temperature, with post-monsoon cooling likely affected by greater cloud cover and remaining

monsoon moisture. Site IV showed consistently elevated temperatures in every season when compared to other sites, indicating a likely greater exposure to solar radiation or diminished water depth/flow, while Site V noted comparatively lower readings.

pH values varied between 7.1 and 8.5, consistently staying slightly alkaline, peaking in the pre-monsoon period (7.6–8.5) and experiencing a slight decrease during the monsoon (7.2–7.8), probably because of rainwater runoff dilution. Dissolved oxygen (DO) ranged from 6.1 to 8.7 mg/L, displaying an inverse correlation with temperature, as it was relatively lower in the warmer pre-monsoon and monsoon phases (due to decreased gas solubility at elevated temperatures and heightened microbial oxygen demand) and higher in the cooler post-monsoon and winter periods. BOD exhibited a comparable seasonal pattern, reaching its highest levels in the pre-monsoon period (3.3–4.8 mg/L) — with the maximum observed at Site IV — and decreasing in the monsoon phase (2.4–3.9 mg/L), likely due to temperature-influenced microbial action combined with the flushing and dilution of organic contaminants.

Nitrate-N levels peaked in the pre-monsoon phase (2.2–3.1 mg/L) and fell to their lowest during the monsoon (1.4–1.9 mg/L), indicating that agricultural runoff and human influence were less significant compared to dilution effects, while silicate showed the largest seasonal variation (6.9–10.8 mg/L), with consistently elevated pre-monsoon readings and minimal monsoon levels at all locations, highlighting a robust reliance on weathering, runoff inputs, and dilution processes. Site IV consistently exhibited the highest values for temperature, pH, BOD, nitrate, and silicate, indicating a greater anthropogenic and/or catchment effect at this location, while Site I typically displayed lower and more stable values throughout the seasons. These trends together suggest a distinct seasonal and temperature-driven effect, where increased pre-monsoon temperatures align with greater nutrient and organic inputs and lower DO, whereas cooling and dilution from the monsoon together moderate parameter levels at all locations.

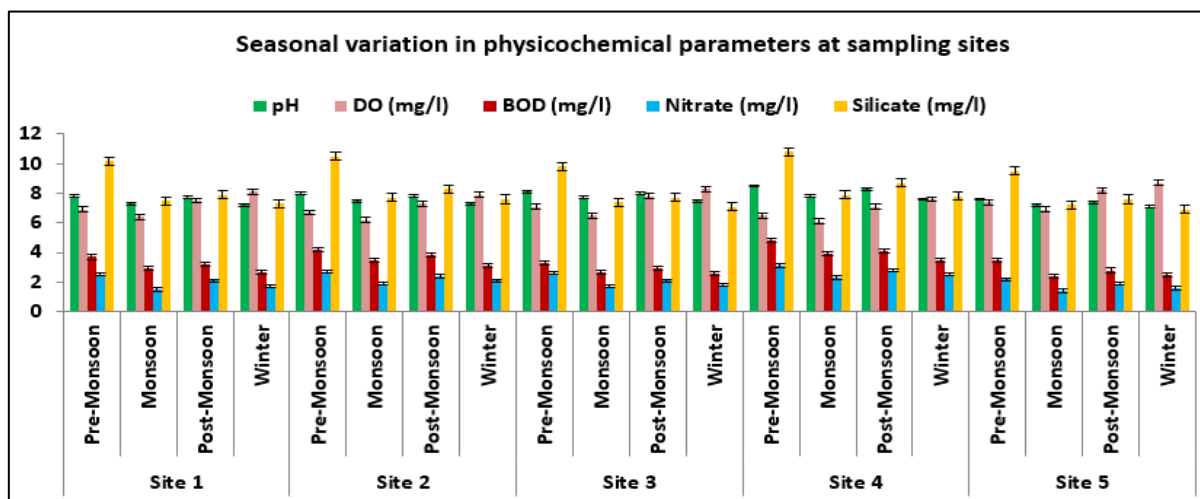


Fig 1 Physicochemical Parameters across All Sites and Seasons in River Ganga

A two-factor ANOVA with replication was used to evaluate differences in five physicochemical water-quality parameters (pH, DO, BOD, nitrate-N, and silicate) among the five sampling locations throughout the four seasons. The variation due to seasonality (Sample factor) was statistically non-significant ($F = 1.49$, $p = 0.208 > 0.05$), and the differences among sites/parameters (Columns factor) were likewise non-significant, yet they neared the threshold ($F = 1.64$, $p = 0.062 > 0.05$). The interaction between season and site (Season $\times$ Site) was found to be very significant ($F = 4.74$, $p < 0.001$), suggesting that the effect of seasons on physicochemical parameters differed across sites and varied based on the particular site–season pairing. This indicates that, although neither season nor location solely controlled water-quality variations, their interaction greatly influenced the detected physicochemical patterns, demonstrating location-specific reactions to seasonal water changes.

➤ *Phytoplankton Community Composition by Season and Site:*

Figure 2 illustrates the phytoplankton density (individuals/L) categorized by site and season. The radar chart illustrates the cumulative (season-aggregated) phytoplankton makeup across all five sampling locations (Site I–V), according to the total population density measured for each of the 17 genera. The most notable outward peaks at all five sites were attributed to Bacillariophyceae genera—especially *Navicula*, *Synedra*, *Fragilaria*, *Cymbella*, and *Nitzschia*—reinforcing the dominance of diatoms as the primary class in the river's phytoplankton community. In contrast, Chlorophyceae genera were found in a moderate mid-range band, while Cyanophyceae and Euglenophyceae genera were closely grouped near the center of the plot, indicating their persistently low contribution to overall density.

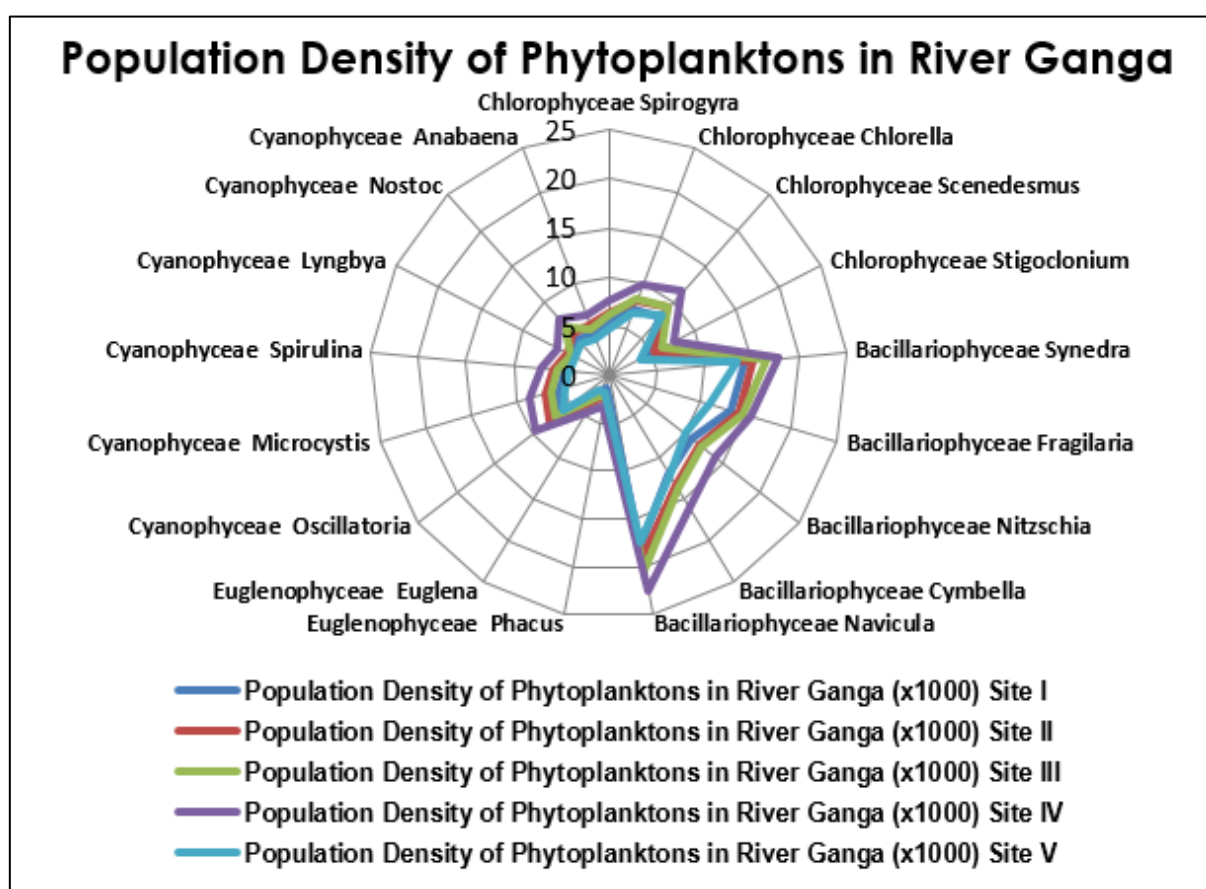


Fig 2 Phytoplanktons Community Composition Across Sampling Sites in River Ganga

Of the five sites, Site IV outlined the outermost polygon with the highest peaks across nearly every genus, particularly *Navicula* and *Synedra*, suggesting it hosted the densest overall phytoplankton abundance along the examined stretch. Site III and Site II displayed nearly overlapping, mid-sized polygons, signifying similar and moderately high densities among most genera, whereas Site I exhibited a noticeably smaller profile. Site V outlined the smallest, most compact polygon among all five sites, signifying that it had the lowest overall phytoplankton density in the research area. In general, the ranking of total phytoplankton abundance by site was Site IV > Site III ~

Site II > Site I > Site V, with a diatom-dominated radial skew maintained across all sites, indicating a largely uniform community composition but a distinct spatial gradient in overall productivity through the river.

➤ *Diversity Metrics of Phytoplankton Population Density:*

Figure 3 illustrates the diversity indices of phytoplankton population densities. The Shannon-Wiener diversity index (H') among the five sampling locations and four seasons of the Ganga River varied from 2.612 to 2.739, with the maximum values consistently observed at Site IV ($H' = 2.688$ –2.739) and the minimum at Sites I, III, and V in

winter ($H' = 2.612\text{--}2.626$), reflecting relatively rich and diverse phytoplankton communities across the study region, accompanied by a slight seasonal decrease in winter. Simpson's Index of Diversity (D) showed a comparable pattern, ranging from 0.913 to 0.930, reaching its highest

values at Site IV ($D = 0.923\text{--}0.930$) and lowest values in winter at Sites I, III, and V ($D = 0.913\text{--}0.915$), indicating a relatively balanced distribution of dominance among taxa within the community.

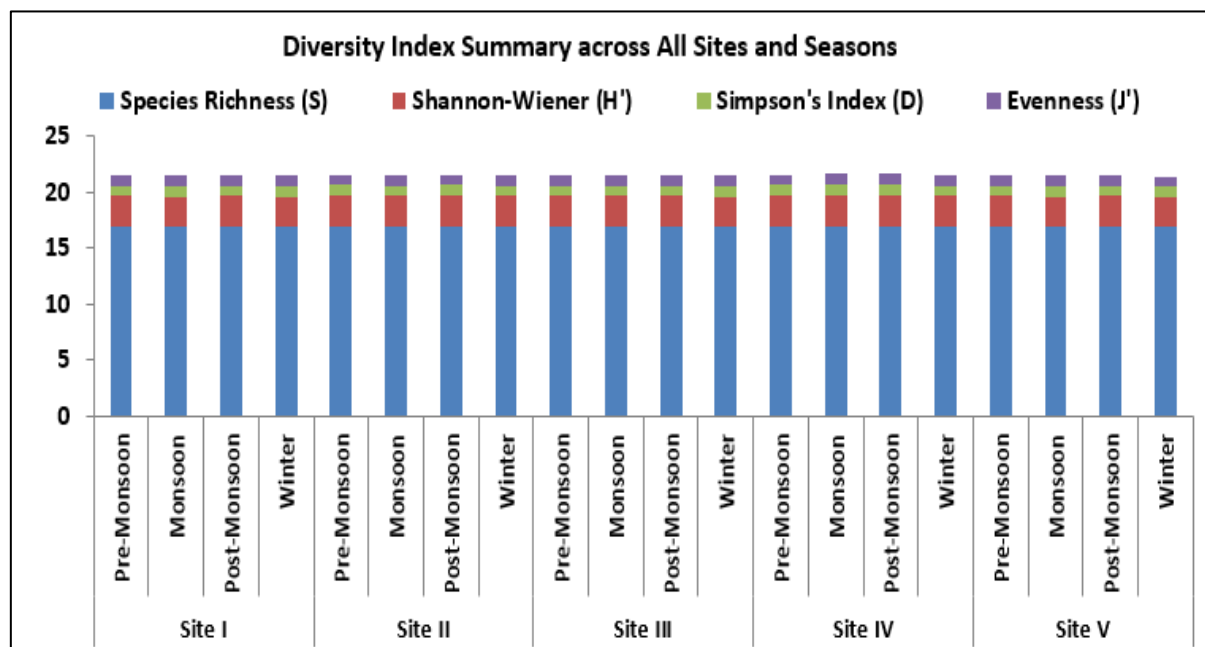


Fig 3 Diversity Indices of Phytoplankton Population Density in River Ganga

Species richness (S) was consistent at 17 genera throughout all locations and seasons, covering the classes Chlorophyceae, Bacillariophyceae, Euglenophyceae, and Cyanophyceae, highlighting that the variation in diversity indices was mainly influenced by changes in relative abundance (evenness) instead of the addition or removal of taxa. Similarly, Pielou's evenness index (J') stayed elevated throughout ($0.922\text{--}0.967$), with the highest evenness observed at Site IV during the monsoon ($J' = 0.967$) and the lowest at Site V in winter ($J' = 0.922$). In general, the consistently elevated H' , D , and J' values throughout all locations and seasons suggest a numerically stable and well-balanced structure of the phytoplankton community in the researched segment of the Ganga River, with Site IV standing out as the most diverse location, while winter is characterized by a period of relatively lower diversity.

Site IV consistently displays the greatest diversity across all three indices (reaching its peak during the Monsoon season), signifying the most varied and evenly balanced community.

Site V consistently exhibits the lowest levels of diversity in all three indices (hitting its lowest point in the winter season), signifying the least diverse and least equitable community.

The gaps between the maximum and minimum values are quite minor, indicating that diversity remains rather consistent across locations and times, with just slight fluctuations.

IV. DISCUSSIONS

The continual numerical supremacy of Bacillariophyceae (diatoms)—especially *Navicula*, *Synedra*, *Fragilaria*, *Cymbella*, and *Nitzschia*—at all five locations and throughout four seasons aligns with previous research on the river Ganga, where Shukla et al. (2015) also identified Bacillariophyceae, Chlorophyceae, and Cyanophyceae as the prevailing classes at Allahabad (Prayagraj). Apart from nutrient availability, the composition of phytoplankton can also be influenced by trace-metal contamination: experimental data indicate that metals like iron and arsenic can preferentially enhance specific algal groups while suppressing others (Ghosh et al., 2024), implying that the diatom prevalence noted here may signify not just nutrient enrichment but also a level of resistance to metal or organic pollutant accumulation in this region.

The prevalence of diatoms in lotic systems often correlates with moderate nutrient enrichment and organic loading, and their continued presence in the current Varanasi section aligns with the physicochemical findings of this study, which showed that nitrate-N and silicate levels—both recognized for promoting diatom growth—were at their peak during the pre-monsoon phase, coinciding with increased BOD, especially at Site IV. This reinforces the idea that the phytoplankton community in this area is primarily influenced by nutrient and organic-load changes instead of species turnover, as richness ($S = 17$) remained consistent across sites and seasons. Varsha and Singh (2025) similarly noted that seasonal hydrology significantly

influences phytoplankton productivity in the Ganga: during monsoonal flooding, nutrient concentrations are diluted and turbidity increases, which decreases phytoplankton density and chlorophyll-a, while the pre-monsoon phase is characterized by a risk of nutrient-driven eutrophication, with nitrogen being crucial for enhancing production; post-monsoon interactions, in turn, signify a natural "reset" where flood-driven nutrient dilution briefly alleviates loading but disrupts community structure. The seasonal variations observed in the current study — with diversity reaching its highest in pre- and post-monsoon and decreasing in winter — reflect the interaction between hydrology and nutrient cycling, emphasizing the importance of management strategies that address human-induced nutrient inputs instead of solely depending on natural seasonal flushing.

Evaluating the diversity metrics against recognized ecological thresholds offers additional understanding of the water-quality condition. According to the Shannon-Wiener index classification suggested by Wilhm (1975), H' values ranging from 1 to 3 denote a mesosaprobic (moderately polluted) state, values higher than 3 signify oligosaprobic (clean) water, while values lower than 1 suggest polysaprobic (heavily polluted) water. The H' values noted here (2.612–2.739) consistently falls within the mesosaprobic range across all sites and seasons, indicating that the Varanasi stretch experiences ongoing, moderate organic and anthropogenic stress, rather than being completely pristine or highly degraded. This corresponds with Shukla et al. (2015), who designated the river at Allahabad as contaminated based on findings from the Simpson and Palmer indices, and bolsters the depiction of the Varanasi ghats as a convergence of household waste, cremation-related, and ritual contributions (Basak et al., 2015).

An interesting, somewhat unexpected result is that Site IV (Manikarnika Ghat) — the cremation ghat facing the highest anthropogenic pressure — exhibited the greatest phytoplankton density, diversity (H'), and evenness (J') among the five sites, instead of the least. This is feasible in moderately (as opposed to highly) polluted environments, where ongoing organic and nutrient contributions from sewage, ritual offerings, and cremation-related substances can increase nutrient availability and promote phytoplankton biomass and evenness prior to the onset of toxic or hypoxic suppression. In contrast, Site V (Ramnagar), the reference site across the river with relatively less direct human influence, consistently showed the lowest density and diversity, probably indicating a decreased nutrient input and a differing flow/substrate pattern on the other bank.

This trend aligns well with the overall water-quality assessment for this section: Singh et al. (2024), evaluating nine locations (S1–S9) along the Ganga at Varanasi through multivariate statistics, the Water Quality Index (WQI), and the Nemerow Pollution Index (NPI), identified significant coliform levels, changed pH, and high BOD/COD across the basin, with sites S8, S9, and S2 — all downstream, heavily affected areas — being the most contaminated and unsuitable for bathing or drinking, whereas upstream

locations demonstrated relatively improved water quality. The WQI-focused fluctuation analysis during the pre-monsoon, monsoon, and post-monsoon phases highlighted untreated sewage discharge as the main pollution source and advocated for ongoing WAWQI monitoring that includes coliform count, BOD/DO, and COD for efficient management. Collectively, these results warn against directly linking increased phytoplankton diversity with improved water quality in nutrient-rich systems; instead, biological diversity indices should be assessed in conjunction with physicochemical and pollution-specific indices like the WQI and NPI for a thorough evaluation, as a location experiencing significant organic loading (Site IV / downstream Varanasi ghats) can display both high biological diversity and poor chemical water quality.

The findings from the two-factor ANOVA support this interpretation: although neither season nor site had a significant individual impact on the physicochemical parameters, their interaction was very significant ($F = 4.74$, $p < 0.001$). This suggests that the water quality in the Varanasi area is influenced by localized, specific reactions to seasonal variations instead of a single consistent factor, highlighting how phytoplankton communities are sensitive to various simultaneous environmental pressures rather than just to season or location separately.

Several constraints of the current study need to be recognized. Phytoplankton was categorized to the genus level instead of the species level, which might obscure more detailed diversity patterns and species-specific pollution tolerances that a more detailed taxonomic examination could uncover. The research was confined to four seasonal sampling occurrences; subsequent studies that include more frequent sampling, identification at the species level, prolonged monitoring, and direct statistical relationships between diversity indices, composite indices like WQI/NPI, and specific physicochemical factors (for instance, via regression or multivariate ordination) would enhance the diagnostic capacity of phytoplankton-based water-quality evaluation and aid in formulating integrated management approaches that reconcile ecological and socio-economic demands on the Ganga River (Varsha & Singh, 2025; Singh et al., 2024).

CONCLUDING REMARKS

This research demonstrates that the makeup of phytoplankton communities and diversity indices act as a sensitive and cost-effective means for assessing water quality in the Varanasi area of the River Ganga. Bacillariophyceae (diatoms) were the most numerically prevalent across all five locations and four seasons, with species richness consistently at 17 genera, showing that the variations in diversity observed spatially and seasonally were influenced by alterations in relative abundance instead of changes in taxonomic composition. The indices of Shannon-Wiener ($H' = 2.612$ – 2.739), Simpson's ($D = 0.913$ – 0.930), and Pielou's evenness ($J' = 0.922$ – 0.967) consistently categorized the examined stretch as mesosaprobic (moderately polluted) according to recognized ecological

classification scales (Wilhm, 1975), suggesting ongoing moderate anthropogenic and organic stress, rather than optimal or highly degraded states.

Manikarnika Ghat (Site IV), which experiences the most ritual and cremation-related activity, unexpectedly showed the highest levels of phytoplankton density and diversity, probably due to nutrient enrichment from ongoing organic contributions, whereas Ramnagar (Site V), the reference site across the river, registered the lowest values consistently. The extremely important season $\times$ site interaction effect ($F = 4.74$, $p < 0.001$) revealed by two-factor ANOVA reinforces that water-quality variations in this area are influenced by specific, combined seasonal-spatial factors instead of by season or site independently.

These results strengthen the importance of phytoplankton diversity indices as effective, sensitive bio-indicators for ongoing monitoring of the Ganga and other culturally important, human-impacted river systems. Considering the ongoing mesosaprobic condition observed at all locations and throughout different seasons, it is advised to implement specific management strategies — especially enhanced sewage collection and treatment at significant ghats like Manikarnika and Dashashwamedh. Future research that includes species-level taxonomic resolution, more frequent sampling, and direct multivariate correlations between phytoplankton diversity and physicochemical factors would enhance the diagnostic and predictive effectiveness of this biomonitoring method for the long-term ecological management of the Ganga River in Varanasi.

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