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Response of Diatoms to the Changing Water Quality in the *Myristica* Swamps of the Western Ghats, India

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Abstract: *Myristica* swamps are one of the rarest wetland ecosystems within the sub-tropical evergreen forests of the Western Ghats, India. As their name indicates, they harbor trees belonging to the ancient family *Myristicaceae*. Due to the waterlogged conditions and high humic decomposition, these swamps are acidic, harbor rare and endemic biotas, and provide ecosystem services to humans. Monitoring this rare ecosystem is crucial because the swamps that once formed a large hydrological network across the Western Ghats are now confined to isolated patches due to human disturbance such as agricultural interventions, roads, and dam construction. Due to the change in land use, there is also a drastic change in water chemistry and associated biodiversity. Biomonitoring is more precise than physical and chemical monitoring. So, the current study aimed to undertake a comprehensive analysis of the physical, chemical, and biological assessment of these swamps. The diatom assemblages are strongly affected by water chemistry and serve as a powerful indicator of environmental changes in the freshwater aquatic systems. However, there is no information on diatom assemblages in these swamps, and the present study aimed to determine the diatom assemblage structure in the *Myristica* swamps and their response to changing water quality. Diatom samples were taken at 17 different swamps across the central Western Ghats, and a set of environmental parameters was evaluated. Analysis revealed a total of 91 species of diatoms belonging to 27 genera across the 17 sites, from which 44 diatom species showed restricted distribution to this unique environment. Overall, the dominant diatom genera inside the swamps included, *Navicula* (19.8%), *Gomphonema* (16%), *Eunotia* (13.3%), *Ulnaria* (9.4%), *Achnanthidium* (8%), *Frustulia* (6.2%), *Planothidium* (5.2%), and *Brachysira* (2.8%). High diatom species richness was observed in the swamps having less anthropogenic disturbance, and diatom assemblage composition was primarily determined by dissolved oxygen, pH, and conductivity. The significant number of geographically restricted taxa in this study points towards our limited understanding of this tropical biome and calls for a dire need for more studies from here, not only to improve our knowledge concerning the diversity, ecology, and biogeography of these diatoms but to further encourage their use in applied (paleo) environmental sciences. Our results indicate that diatoms can prove useful environmental indicators even in harsh environments like swamps and can be a potential tool for assessing ecological and climatic change.

Keywords: wetlands; bacillariophyceae; water quality; community analysis; environment; indicators

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1. Introduction

Wetlands are territories between terrestrial and aquatic systems, characterized by waterlogged soils, aquatic vegetation, and typically high biodiversity [1]. India is home to various natural and human-made wetlands, ranging from marine mangrove swamps, tidal marshes, and coastal wetlands to freshwater marshes, swamps, and bogs. In India, notable freshwater swamps have been reported from the Siwalik and Doon valley, Brahmaputra valley [2], and the valleys of the Western Ghats [3,4]. The Western Ghats is one of the 36 global biodiversity hotspots, which lies in the western part of peninsular India as a series of mid (300–1200 m a.s.l) to high altitude hills (up to 2600 m a.s.l.) stretching over 1600 km from north to south and covering an area of about 160,000 km^2 [5]. The Western Ghats is home to four distinct freshwater swamp types: *Myristica*, *Elaeocarpus*, *hadlus*, and *Vayals* [6–8]. Among these, the *Myristica* swamps are one of the rarest ecosystems present in the tropical world. As their name indicates, they are water-saturated regions predominantly covered with trees belonging to the ancient family *Myristicaceae* [9,10]. *Myristica* swamps in India were first reported by Krishnamoorthy [3] from the Travancore region (present-day Kerala state) of the Southern part of the Western Ghats. Currently, there are three different patches of these swamps which are distributed across the Western Ghats: (1) Swamps located in Maharashtra and Goa in the Northern Western Ghats [11,12] (2) *Myristica* swamps situated in Karnataka in the Central Western Ghats [13] (3) *Myristica* swamps situated in the valleys of Shendurney, Kulathupuzha, and Anchal forest ranges in the southern Western Ghats [10].

Krishnamoorthy [3] reported the hydrological conditions of these swamps as most of the swamps remain entirely inundated during a greater part (8 months during monsoon, i.e., June-January) of the year. Wooded vegetations in swamps possess characteristic features, such as the presence of pneumatophores or breathing roots and stilt roots, which are necessary for the survival of trees in waterlogged conditions. Typically, water inside these swamps has acidic to neutral pH [10,14]. Since these swamps are located immediately after headwater, they are nutrients deficient (oligotrophic) with low electrical conductivity (EC). Due to the lack of mineral input, high humic decomposition, and leaching of organic compounds, the waters are acidic [10,15]. Thus, these swamps act as environmentally delineated from the surrounding environment by their acidic nature. At first sight, these extreme environments seem inhospitable places for life forms to thrive. Still, surprisingly, they are home to a diverse endemic biota, including *Semecarpus kathalekanensis* Dasappa & Swam., *Gymnacranthera canarica* (Bedd. ex King) Warb., or *Myristica magnifica* Bedd. [14]. The macroscopic flora studied by Krishnamoorthy [3]; Chandran et al. [16]; Bhat and Kaveriappa [14]; Roby et al. [17] indicate that these swamps typically display high floristic endemism, with studies recording 23 tree species endemics to these swamps. Many endemic biotas have been discovered inside these swamps in the last two decades, including many new species of frogs [18], mushrooms [19], butterflies [20], and diatoms [21]. Refer Ranganathan et al. [4] for a detailed review of the biodiversity of *Myristica* swamps. Despite their little-known biota, these swamps are virtually live museums of ancient life and of great interest to biologists, as most of the novel taxa described here, such as *Myristica magnifica*, *Gymnacranthera canarica*, *Syzygium stocksii* (Duthie) Gamble, *Madhuca bourdillonii* (Gamble) Raizada, and *Semecarpus kathalekanensis* represent ancient lineages [9].

Studies suggest that these swamps once covered the entire Western Ghats; however, the extensive network is now confined to only a few fragmented patches due to increased human pressure [9,22]. Recent studies highlight that many of these swamps are facing extinction at an alarming rate due to conversion into agricultural fields and anthropogenic interferences such as shifting cultivation, irrigation, and various developmental projects in the vicinity [14,16]. Additionally, Kumaran et al. [23] and Srivastava et al. [24] proposed that the weakening of monsoon stressors through the Holocene is a natural reason for the disappearance of these swamps from the Northern region of the Western Ghats, as the swamp trees community requires a rainfall of at least 3000 mm or above [10,13]. Thus, the increased need for water because of the increasing human population and changes in water availability due to climate change are the main stressors inside this ecosystem.

To monitor this extremely sensitive ecosystem, the first step is to measure the physical and chemical variables of soil and water over time to understand natural and human-made temporal variation. However, it has been shown that monitoring aquatic environments using biological elements can be effectively used in this case [25]. As aquatic organisms are regularly exposed to that environment, they can provide time-integrated information about the environmental conditions and the ecological health of the ecosystem [26]. In shallow water bodies like swamps or wetlands, because of their varied inundation time and depths, it is problematic to use fish and invertebrates as biological indicators [27], especially in monsoon-driven high energy systems like the Western Ghats. In such a scenario, diatoms can act as excellent bioindicators of this Ecobiome. Diatoms (Class: Bacillariophyceae) are siliceous, unicellular algae present in all aquatic and sub-aerial environments, forming the base of the food web, therefore responding rapidly to any environmental and anthropogenic changes [28,29]. Due to their sensitive nature and excellent preservation in lake sediments [30], diatoms can provide a continuous record of environmental history. Scientists use diatoms as potential bioindicators to monitor present and past environmental conditions across continents [29,31,32]. Additionally, diatoms also prove to be an excellent indicator of acidic systems, where the occurrence of other bioindicators is limited [33,34].

In spite of the fact that diatom can prove an excellent bioindicator to monitor this vanishing ecosystem, there is no comprehensive study on diatoms from this region, except a discovery of a new species *Germainiella chandranii* [21]. The present research focuses on documenting the diatom assemblage structure inside *Myristica* swamps to determine the environmental optima and tolerances of species to the limnological variables, especially across disturbance gradients. It aims to act as a reference study to characterize further the past and present environmental conditions of *Myristica* swamps in the central Western Ghats of India. The main objectives of this study were: (1) to describe the species composition and structure of diatom assemblages across the environmental gradients inside the swamps (2) to explore the relationships between diatom taxa and environmental variables. We hypothesized that the unique habitat would act as the primary driver of species inside this ecosystem.

2. Materials and Methods

2.1. Study Area

This study was carried out in *Myristica* swamps across the Uttara Kannada district (13.86° to 15.50° N and 74.08° to 75.08° E) in the central Western Ghats within the state of Karnataka (Figure 1). The district is hilly and thickly wooded with a tropical climate, documented by a well-defined rainy season of about six months between June and November, when the southwest monsoon brings most of the rainfall. The annual average rainfall here ranges from 2500 mm to 5000 mm [9].

From the *Myristica* swamps, diatom and water samples were collected from 17 different sites across the central Western Ghats between the latitudinal range of 13.71° to 14.66° N and longitudinal range of 74.56° to 74.99° E with an elevational range of 395 to 623 m a.s.l. Some swamps were individual and isolated (single site), while others were big (covering a few hectares of area). In the case of swamps covering a large area, we sampled 3–4 sites within the same latitude-longitude range. Hereafter, this kind of swamp will be called a swamp complex. The geo-coordinates of the sampling points in the different swamps are presented in Table 1. The distribution of the swamps in the study area and two images depicting the *Myristica* swamp ecosystem are illustrated in Figures 1 and 2, respectively.

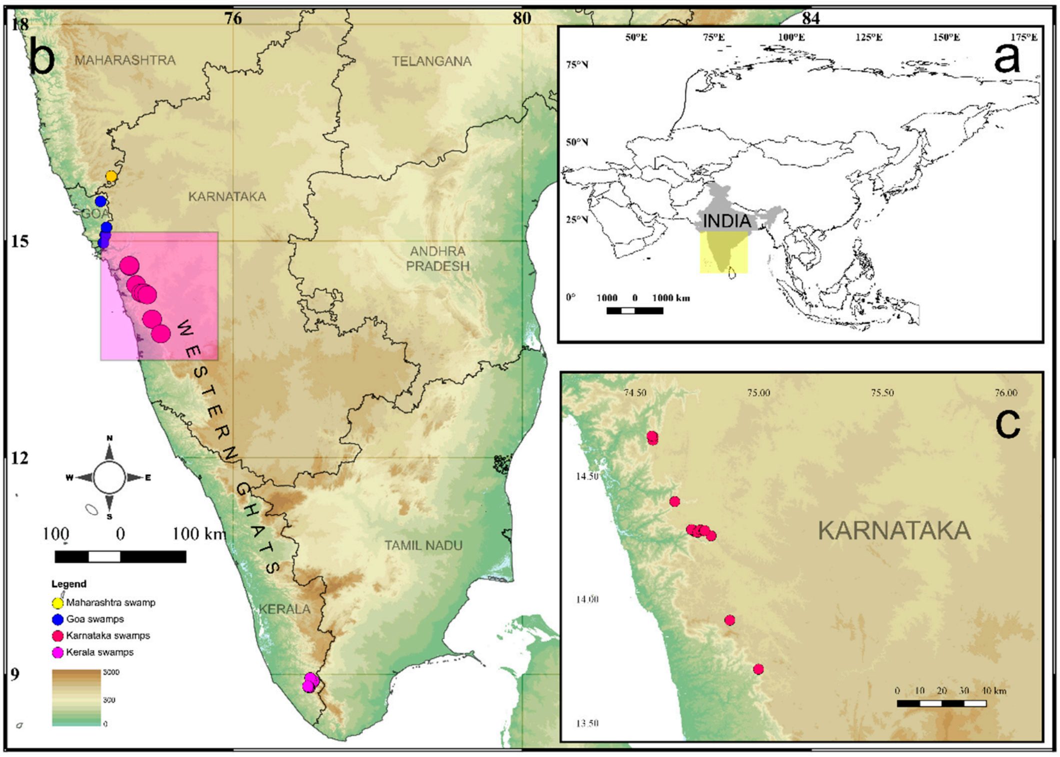

Figure 1. (**a**). Map of Asia with administrative boundaries with study area highlighted. (**b**). Map showing the distribution of *Myristica* swamps across the Western Ghats with topography and administrative boundaries. Inset (**c**). Map showing the location of sampling sites across the central Western Ghats, Karnataka. Magenta-pink color circles mark the study sites.

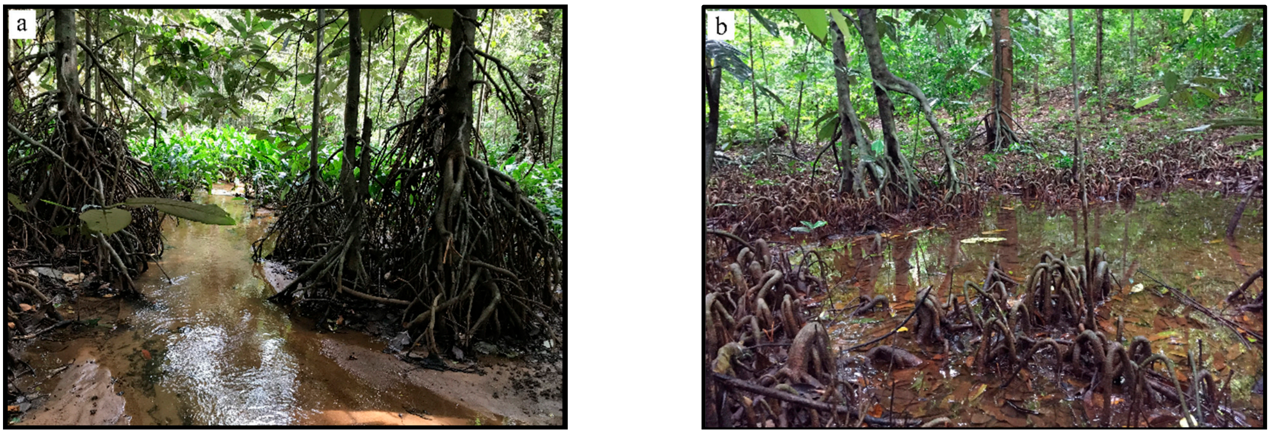

Figure 2. (**a**) Image showing the water inundation with the presence of stilt roots inside the *Myristica* swamp ecosystem; (**b**) Areas with water inundation along with the breathing roots (pneumatophores) inside the swamps.

Table 1. Location of sampling sites in the *Myristica* swamps of the central Western Ghats, Karnataka.

No	Swamp	Latitude (°N)	Longitude (° E)	Altitude (m a.s.l.)
1	Devikan swamp	14.65034	74.57084	451
2	Assoli swamp—1	14.65902	74.57178	419
3	Assoli swamp—2	14.66012	74.57059	446
4	Balajetty swamp	14.66122	74.56786	395
5	Hudidevarakudlu swamp	14.3969	74.6595	396
6	Torme swamp—1	14.2773	74.76942	617
7	Torme swamp—2	14.27856	74.76717	588
8	Mundigethagu swamp	14.27709	74.78032	608
9	Nagodi swamp	13.91614	74.88213	557
10	Hulikal swamp	13.71799	74.99848	571
11	Kathlekan swamp—1	14.27424	74.74807	556
12	Kathlekan swamp—2	14.27284	74.74468	540
13	Kathlekan swamp—3	14.27404	74.74564	562
14	Malemane swamp	14.28221	74.72691	530
15	New Kathlekan swamp	14.27064	74.7514	543
16	Mavingundi swamp—1	14.25621	74.80658	623
17	Mavingundi swamp—2	14.25646	74.80669	599

2.2. Environmental Variables Sampling and Analysis

Geographic coordinates were recorded at each sampling point using a Garmin® (Olathe, KS, USA) eTrex® 30x GPS. Physicochemical parameters of the water like pH, electrical conductivity (EC) (µS/cm), dissolved oxygen (DO) (mg/L), and water temperature (T) (°C) were measured in situ during the forenoon using a HACH HQ40D portable meter (Loveland, CO, USA). The water samples were collected in 100 mL pre-cleaned polyethylene bottles from the surface. One water sample was collected from each site following a standard procedure recommended by APHA [35]. Nitrate (NO_3^-) and phosphate (PO_4^{3-}) were measured on the same day with a method prescribed by HACH [36,37] using a HACH 1900 portable colorimeter and standard reagents (NitraVer® 3, NitraVer® 6, Amino acid, and Molybdate reagent). The detection limit for nitrate using nitrate, Low Range method was 0 to 0.50 mg/L NO_3^-, and for phosphate, it was 0 to 30.0 mg/L PO_4^{3-} using the Amino acid method.

2.3. Diatom Sampling, Preparation, and Analysis

A total of 20 diatom samples from different microhabitats were used for this study. In each habitat, samples were collected from the four different microhabitats, i.e., epipsammic (pipette collection of sand), epilithic (toothbrush scraping of stone), epiphytic (squeezing of plant material), and epibryophytic (squeezing of moss) across 17 different swamps. Sample collection was done during the post-monsoon season between 11–14 November 2017. After collection, the samples were stored in a Whirl-pak® sampling bag. Water samples were stored for 3–4 days on ice until they were transported to the lab for further analysis. In the laboratory, the diatom samples were oxidized by boiling them with concentrated nitric acid to remove the organic matter. The acid oxidized samples were then washed several times with distilled water by centrifuging them at 3500 rpm for 10 min until they attained a neutral pH. The cleaned samples were then air-dried onto glass coverslips and subsequently mounted onto the microscope slide using Naphrax® as the mounting medium [38]. Six hundred diatom valves were counted and identified per slide at 1000× magnification using an Olympus BX53 microscope equipped with an Olympus DP74 digital camera using differential interference contrast (DIC) microscope optics and images were captured with Olympus cellSens standard 1.16 imaging software. Diatom images were edited and compiled using GNU Image Manipulation Program (GIMP) 2.10.2 (available from: http://www.gimp.org/, The GIMP team, 1997–2021, retrieved on 20 November 2021) [39], and Inkscape Version 0.92.3 [40], respectively. Taxonomic identifications were mainly based on Krammer and Lange-Bertalot [41–44], Krammer [45], Lange-Bertalot [46], Lange-Bertalot et al. [47]. During the analysis, due to the higher number of geographically restricted flora, it was a limitation to use European literature for the identification. To address this problem, extensive searching of the species using regional flora from the tropical literature across southeast Asia such as Karthick et al. [48–50], Jüttner et al. [51], Taylor et al. [52,53], Chudaev et al. [54] were used, and subsequently, a voucher flora was developed based on the light microscopic images as described in Bishop et al. [55]. The nomenclature of diatoms was updated, according to DiatomBase [56].

2.4. Statistical Analysis

Histogram analysis was used to compare the range of all water quality variables across the sampling sites and to assess the general water chemistry status of these swamps. To document the diatom diversity and the richness patterns across the swamps, diversity indices, including Shannon–Wiener diversity (H), Pielou's evenness (J), and Simpson diversity ($1 - \lambda$), and Species richness (S) were calculated using the diatom abundance data. All these analyses were performed using PAST software Version 4.01 [57].

To determine the response of the diatoms to environmental variables and to assess the diatom-environment relationship, multivariate analysis was used. All environmental variables (except pH) were log-transformed [log10 (x + 1)] prior to analysis to normalize their distribution. Overall, the hydrochemical variables analyzed were pH, nitrate (NO_3^-), phosphate (PO_4^{3-}), electrical conductivity (EC), and dissolved oxygen (DO). Principle Component Analysis (PCA) and Canonical Correspondence Analysis (CCA) were carried out using CANOCO Version 5 [58]. PCA is a variable reduction technique that reduces the numerous variables to a smaller number of composite variables called "principal components" [59]. PCA was conducted on environmental variables to explore the main environmental gradients among the sampling sites. We first used PCA to examine which swamp is driven by which environmental variable and how these swamps are clustered under various environmental parameters. Then we used CCA to determine the relationships between the diatom assemblages and environmental variables. Prior to CCA analyses, the abundances of all taxa were expressed as relative counts, and taxa only with relative abundances of 1% or greater were taken into consideration. Name of each taxon with abbreviation code and % RA of diatom along with presence-absence data for all diatom taxa across the sampling sites are provided in Supplementary Data Table S1 and S2, respectively. Diatom assemblages data were log-transformed before analysis. Monte Carlo permutation tests (999 unrestricted permutations, $p \leq 0.05$) were used to test the significance of the first two CCA axis.

3. Results

3.1. Hydrochemistry

The summary of water chemistry variables is listed in Table 2. From the study, pH values indicated that most of the swamps were in acidic to circumneutral conditions ranging between pH 6.1 to 7.4 (Figure 3a). The electrical conductivity of the swamps was between 25–75 µS/cm, except for 3–4 swamps (Balajetty swamp, Hulikal swamp, and Malemane swamp) with higher ionic compositions between 115–200 µS/cm (Figure 3b). The nitrate and phosphate concentration inside the swamps was between 0.01–0.02 mg/L (Figure 3c) and 0.5–2 mg/L (Figure 3d)., except for two swamps with phosphate concentrations ranging between 3–4 mg/L. Dissolved oxygen content was between 6.5–8.5 mg/L and indicated well-oxygenated conditions of the swamps. The temperature of the study sites varied between 21–30 °C.

Table 2. Value of different water quality variables across the various sampling sites. Where EC = electrical conductivity, DO = dissolved oxygen, T = measured temperature of the water inside the swamp (T was measured during the forenoon), NO_3^- = nitrate, PO_4^{3-} = phosphate, and TDS = total dissolved solids. Site number (1, 2, 3, 4) represents sampling areas.

No.	Swamp	pH	EC (μS/cm)	DO (mg/L)	T (°C)	NO_3^- (mg/L)	PO_4^{3-} (mg/L)	TDS (mg/L)
1	Devikan swamp Site—1	7.45	35.50	6.65	25.7	0.02	3.95	16.61
2	Devikan swamp site—2	6.12	42.10	6.65	25.7	0.02	3.95	19.7
3	Devikan swamp site—3	7.48	52.50	6.65	25.7	0.02	3.95	19.7
4	Devikan swamp site—4	7.2	46.30	6.65	25.7	0.02	3.95	21.7
5	Devikan swamp site—5	6.40	45.30	5.98	24.4	0.02	2.95	21.04
6	Assoli swamp—1	7.28	79.80	6.9	24.8	0.02	2.32	37.6
7	Assoli swamp—2	7.21	77.50	7.06	25.2	0.01	3.41	
8	Balajetty swamp	7.01	168.80	6.24	25.8			80.1
9	Hudidevarakudlu swamp	7.24	74.30	7.63	24.9	0.01	2.08	34.9
10	Torme swamp—1	7.15	31.50	7.28	22.9	0.02	2.14	14.71
11	Torme swamp—2	6.86	30.00	6.99	22.4	0.01	2.29	14.01
12	Mundigethagu swamp	6.99	23.60	7.35	23	0.02	2.43	11
13	Nagodi swamp	7.36	40.70	7.24	28.4	0.01	0.85	19.08
14	Hulikal swamp	6.87	115.90	6.89	25.6	0.01	1.87	54.8
15	Kathlekan swamp—1	6.79	96.20	6.98	21.2	0.01	0.8	45.3
16	Kathlekan swamp—2	6.18	53.30	6.88	21.6	0.01	1.4	25
17	Kathlekan swamp—3	6.71	42.00	7.05	23.8	0.02	1.6	19.68
18	Malemane swamp	6.83	201.00	8.74	24.2	0.01	0.3	95.6
19	New Kathlekan swamp	6.82	40.70	6.95	24.1	0.01	2.6	19.07
20	Mavingundi swamp—1	6.67	68.80	7.04	23.7	0.01	3.33	32.4
21	Mavingundi swamp—2	6.98	44.40	6.82	23.2	0.01	2.98	20.8

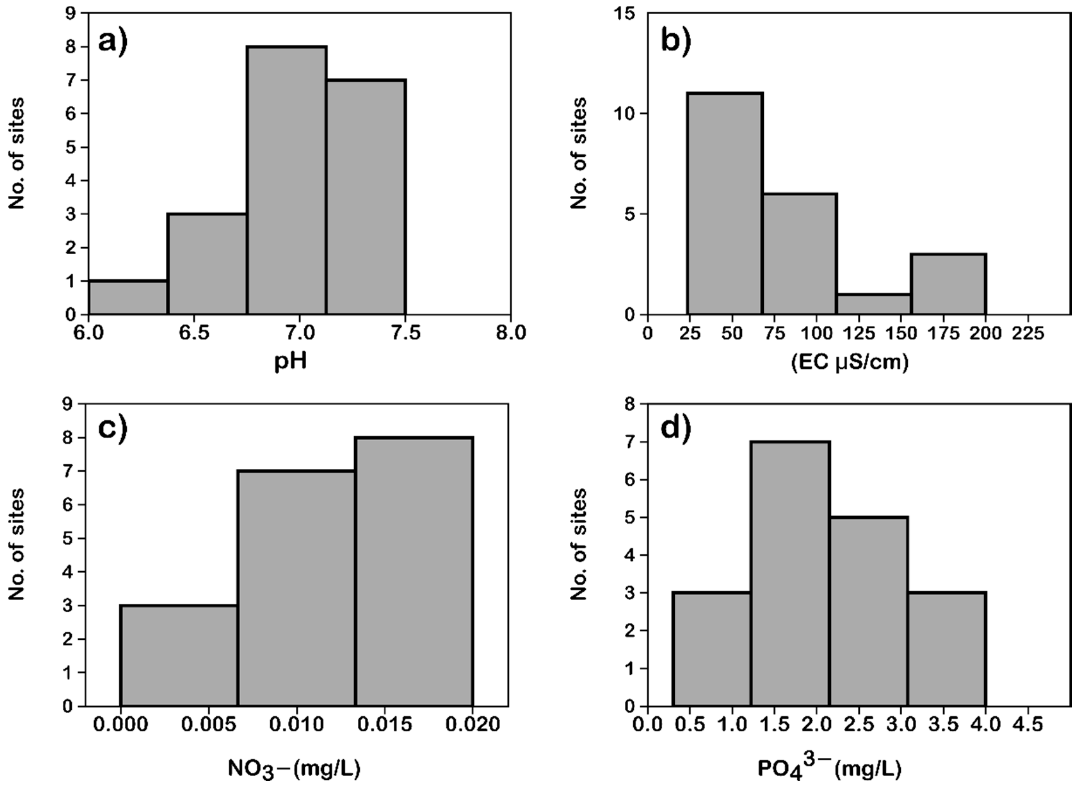

Figure 3. Histogram graphs illustrating the range of various water quality variables (**a**) pH, (**b**) electrical conductivity (EC), (**c**) nitrate (NO_3^-), and (**d**) phosphate (PO_4^{3-}) across the various *Myristica* swamps sites sampled.

3.2. Species Composition and Diversity

In the 20 samples, we found 91 species belonging to 27 genera, from which 44 diatom species are putative novel species and potentially endemic to *Myristica* swamps, as our ongoing survey across the Western Ghats did not yield any of these taxa. The most common diatom genera observed were *Navicula* Bory (19.8%), *Gomphonema* Ehrenberg (16%), *Eunotia* Ehrenberg (13.3%), *Ulnaria* (Kutzing) Compère (9.4%), *Achnanthidium* Kützing (8%), *Frustulia* Rabenhorst (6.2%), *Planothidium* Round & L.Bukhtiyarova (5.2%), *Brachysira* Kützing (2.8%), and *Neidium* Pfitzer (0.7%). The dominant diatom species identified were *Eunotia rhomboidea* Hustedt, *Eunotia* c.f. *incisa* W.Smith ex W.Gregory (Figure 4), *Eunotia bilunaris* (Ehrenberg) Schaarschmidt (Figure 4), *Frustulia jogensis* Gandhi, *Frustulia crassinervia* (Brébisson) Lange-Bertalot & Krammer (Figure 4), *Brachysira neoexilis* Lange-Bertalot (Figure 4), *Navicula obtecta* I.Jüttner & E.J.Cox (Figure 4), *Navicula nielsfogedii* J.C.Taylor & Cocquyt, *Navicula globulifera* var. *robusta* Hustedt, *Achnanthidium minutissimum* (Kützing) Czarnecki, *Gomphonema gandhii* B. Karthick & J.P.Kociolek (Figure 4), *Gomphonema parvulum* (Kützing) Kützing, *Ulnaria ulna* (Nitzsch) Compère, *Planothidium frequentissimum* (Lange-Bertalot) Lange-Bertalot (Figure 4), and *Neidium* sp. An overview of the commonly occurring diatom taxa in the study area has been shown in Figure 4.

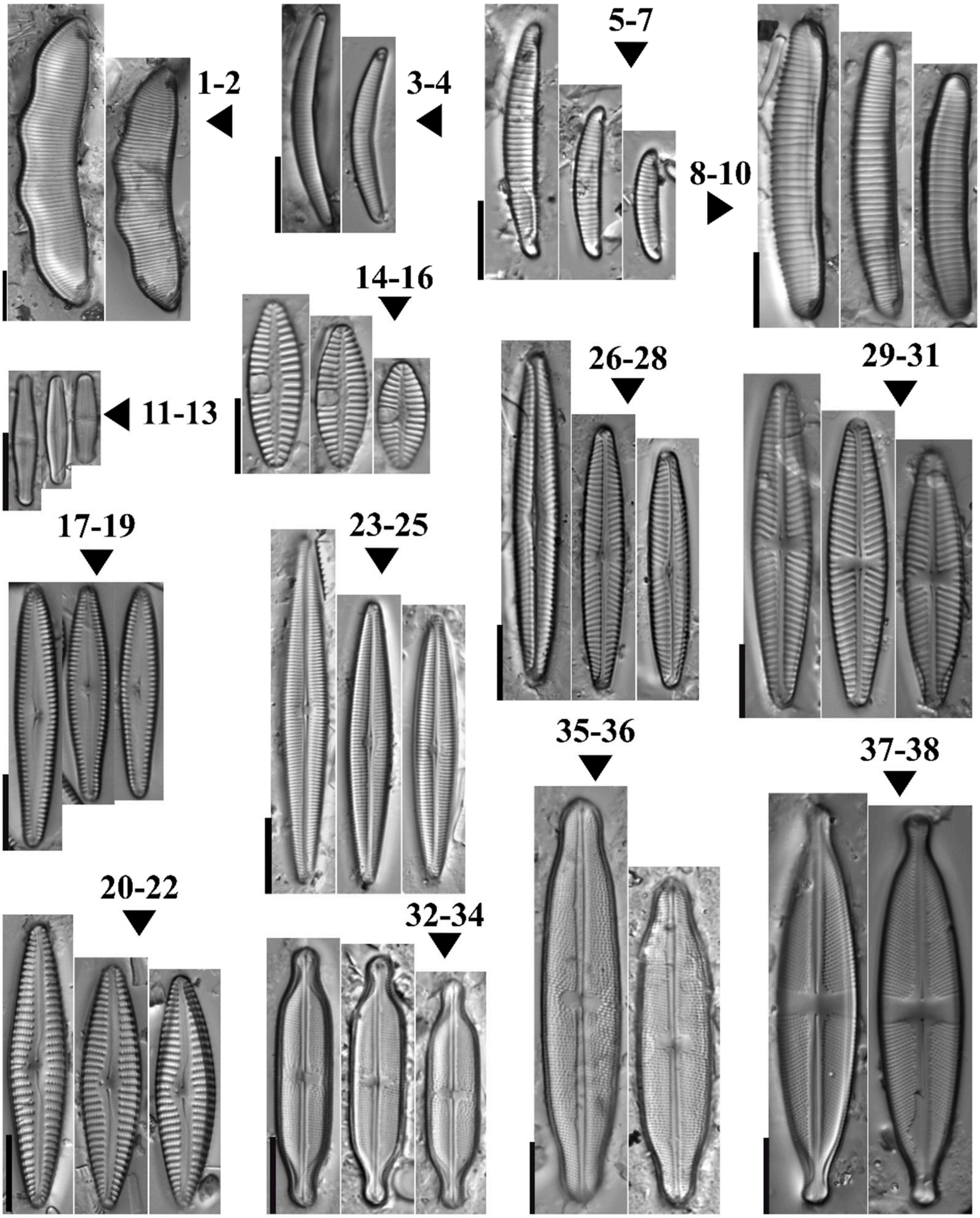

Figure 4. *Cont*.

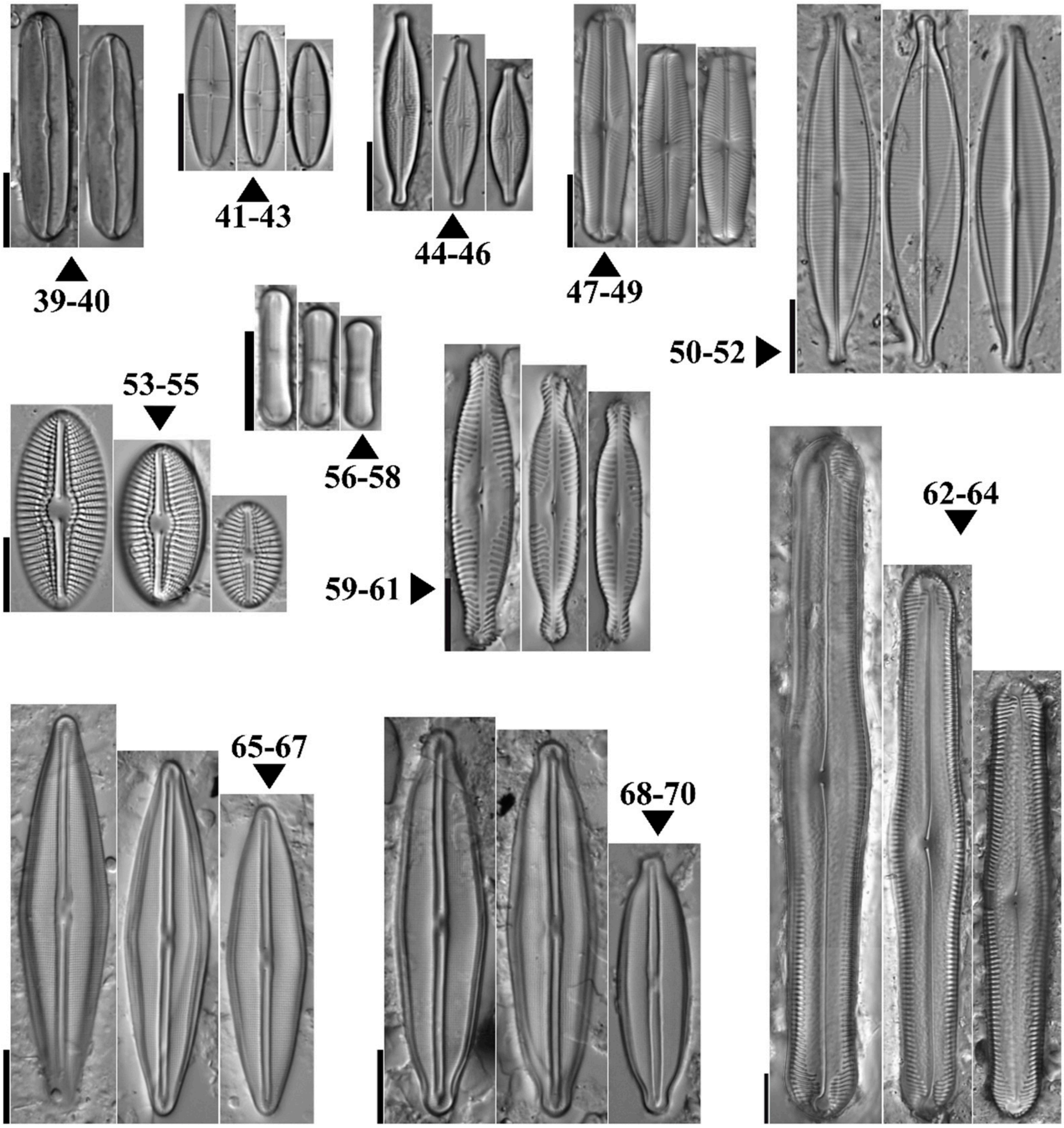

Figure 4. Plate showing diatom diversity inside the swamps. 1–2: *Eunotia* c.f. *zygodon*; 3–4: *Eunotia bilunaris* (Ehrenberg) Schaarschmidt; 5–7: *Eunotia* c.f. *incisa* W. Smith ex W. Gregory; 8–10: *Eunotia* sp.; 11–13: *Achnanthidium* sp.; 14–16: *Planothidium frequentissimum* (Lange-Bertalot) Lange-Bertalot; 17–19: *Gomphonema gandhii* B. Karthick & Kociolek; 20–22: *Oricymba japonica* (Reichelt) Jüttner, E.J. Cox, Krammer & Tuji; 23–25: *Navicula obtecta* I.Jüttner & E.J. Cox; 26–28: *Navicula angusta* Grunow; 29–31: *Navicula* sp.; 32–34: *Neidium* sp.; 35–36: *Neidium* sp.; 37–38: *Stauroneis amphicephala* Kützing; 39–40: *Germainiella* sp.;41–43: *Caloneis* sp.; 44–46: *Brachysira neoexilis* Lange-Bertalot; 47–49: *Sellaphora* sp.; 50–52: *Craticula riparia* (Hustedt) Lange-Bertalot; 53–55: *Diploneis* sp.; 56–58: *Humidophila contenta* (Grunow) Lowe, Kociolek, J.R. Johansen, Van de Vijver, Lange-Bertalot & Kopalová; 59–61: *Pinnularia* sp.; 62–64: *Pinnularia acrosphaeria* W. Smith; 65–67: *Frustulia* sp.; 68–70: *Frustulia crassinervia* (Brébisson ex W. Smith) Lange-Bertalot & Krammer.

Species richness (S) was higher at the New Kathlekan swamp site—1 (23 taxa) than other sites in this study, and it was lowest at Assoli swamp—1 (8 taxa). Shannon–Wiener diversity values (H) ranged from 1.10 (Devikan swamp site—2) to 2.80 (New Kathlekan swamp site—1). Pielou's evenness (J) ranged from 0.25 (Devikan swamp site—2) to 0.75 (Hudidevarakudlu swamp). Simpson diversity ($1 - \lambda$) was highest at the New Kathlekan swamp (0.91), followed by Kathlekan swamp—3 (0.90), and it was lowest at Devikan swamp site—2 (0.55) (Table 3).

Table 3. Diversity indices of the diatom assemblages were observed across the swamps. S = species richness, J = Pielou's evenness, H = Shannon index, $1 - \lambda$ = Simpson diversity index. Figures in bold represent the highest and lowest values of a particular index.

Swamp	Microhabitat	Species Richness (s)	Pielou's Evenness (J)	Shannon Index (H)	Simpson Diversity Index ($1 - \lambda$)
Devikan swamp site—1	Epibryophytic	17	0.54	2.22	0.85
Devikan swamp site—2	Epipsammic	12	0.25	1.10	0.55
Devikan swamp site—3	Epipsammic	14	0.48	1.91	0.78
Devikan swamp site—	Epilithic	18	0.54	2.28	0.86
Devikan swamp site—4	Epipsammic	17	0.41	1.96	0.80
Assoli swamp—1	Epiphytic	8	0.49	1.37	0.67
Hudidevarakudlu swamp	Epipsammic	14	0.75	2.36	0.89
Torme swamp—1	Epilithic	10	0.44	1.49	0.63
Mundigethagu swamp	Epilithic	13	0.49	1.86	0.76
Hulikal swamp site—1	Epilithic	13	0.48	1.84	0.80
Hulikal swamp site—2	Epilithic	16	0.66	2.37	0.88
Hulikal swamp site—3	Epiphytic	18	0.48	2.15	0.84
Kathlekan swamp—1	Epiphytic	12	0.73	2.17	0.86
Kathlekan swamp—2	Epidendric	14	0.73	2.32	0.88
Kathlekan swamp—2	Epiphytic	15	0.58	2.16	0.84
Kathlekan swamp—3	Epiphytic	18	0.73	2.58	0.90
Malemane swamp	Epipsammic	16	0.61	2.29	0.86
New Kathlekan swamp site—1	Epibryophytic	23	0.72	2.80	0.91
New Kathlekan swamp site—2	Epiphytic	21	0.72	2.72	0.91
Mavingundi swamp—1	Epilithic	16	0.55	2.18	0.86

3.3. *Relationship of Diatom Assemblages to Environmental Variables*

The first two axis of PCA (eigenvalues $\lambda_1 = 0.9637$, $\lambda_2 = 0.0333$) explained in total 99.7% of the variance in the data (axis 1: 96.37% and axis 2: 3.33%) (Figure 5). The ordination of data distinguished swamps into three groups. Sites 2 and 3 from the Kathlekan swamp complex were positively correlated with dissolved oxygen concentration and negatively correlated with elevated conductivity and nutrients. All Devikan–Assoli swamp complex sites showed a strong linear relationship with high phosphate (PO_4^{3-}) levels and moderate correlation with pH values. All Hulikal swamps, Hulidevarakudlu swamp, and Assoli swamps correlated with increased conductivity.

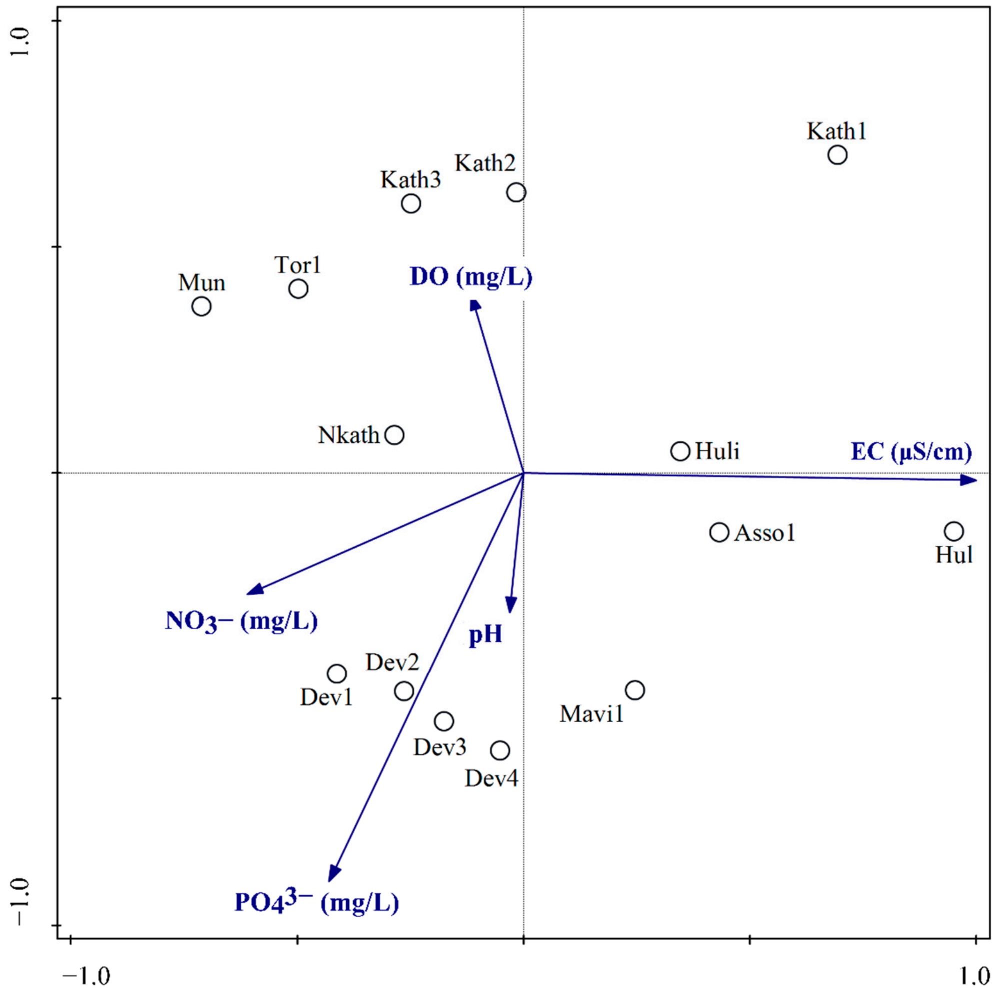

Figure 5. Principal component analysis (PCA) biplot illustrating the relationship between the environmental variables and water chemistry of the various sampling sites. (Dev = Devikan swamps; Asso = Assoli swamp; Huli = Hulidevarakudlu swamp; Tor = Torme swamp; Mun = Mundigethagu swamp; Hul = Hulikal swamps; Kath = Kathlekan swamp sites 1–3; Nkath = New Kathlekan swamp; Mavi = Mavingundi swamp).

According to the results CCA ordination diagram (Figure 6), the first two CCA axis (eigenvalues λ_1 = 0.5662, λ_2 = 0.4265) explained 54% of the total variation amongst the species composition data (axis 1: 31.3% and axis 2: 22.7%). The CCA ordination was plotted using 75 species (RA > 1%) and five physicochemical variables. The gradient length and analysis of the CCA plot illustrate that pH concentration was the most significant factor contributing to the first axis. In contrast, EC was the notable factor contributing to the second axis. DO, PO_4^{3-}, and NO_3^- were positioned between axis 1 and 2 with a negative relationship between nutrients and oxygen.

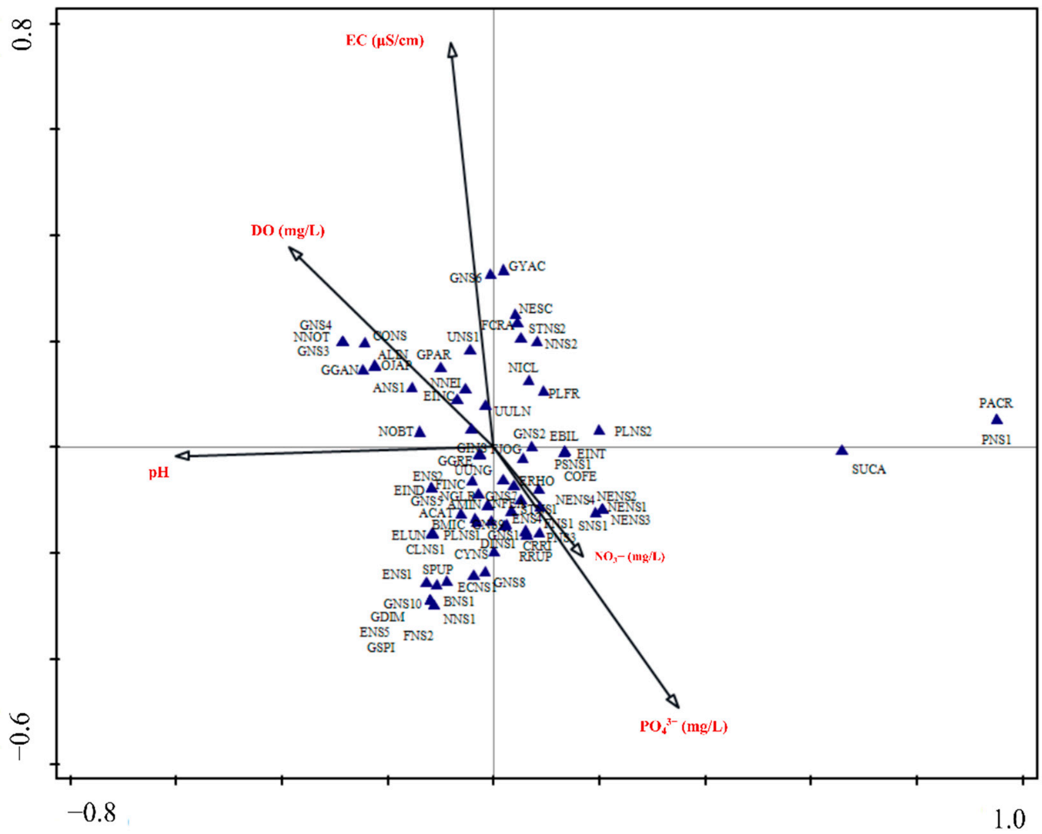

Figure 6. Canonical correspondence analysis (CCA) ordination illustrating the influence of environmental variables on the diatom assemblages in *Myristica* swamps. Only species contributing more than 1% of the variation are illustrated. The species codes are provided in the Supplementary Data.

The majority of the species on the left side of the graph are distributed under the increasing influence of elevated pH, conductivity, and dissolved oxygen. To the left side of the diagram, an abundance of the species like *Gomphonema gregarium* Kociolek, J.B.Woodward & C.Graeff, *Gomphonema diminutum* B.Karthick & J.P.Kociolek, *Gomphonema* sp. 5 and 7, *Achnanthidium minutisimum* (Figure 4), *Achnanthidium catenatum* (Bily & Marvan) Lange-Bertalot, *Sellaphora pupula* (Kützing) Mereschkovsky, *Planothidium* sp. 1, *Encyonema* sp., *Caloneis* sp. 1, and *Navicula globulifera* var. *robusta* prefers slightly alkaline condition with elevated pH level, whereas the abundance of species like *Navicula obtecta*, *Achnanthidium linannulum* B. Karthick, J.C.Taylor & P.B.Hamilton, *Gomphonema gandhii*, *Oricymba japonica* (Figure 4), *Navicula notha* Wallace, *Achnanthidium* sp. 1, *Cocconeis* sp., etc. are correlated with dissolved oxygen concentration and lower nutrients. Further, species like *Ulnaria* sp. 1, *Nitzschia clausii* Hantzsch, *Planothidium freequentissimum*, *Gyrosigma acuminatum* (Kützing) Rabenhorst, *Navicula* sp. 2, and *Navicula escambia* (Patrick) D.Metzeltin & Lange-Bertalot are influenced by higher conductivity (EC). On the right side of the plot, species like *Eunotia intermedia* (Krasske) Nörpel & Lange-Bertalot, *Eunotia rhomboidea*, *Neidium* sp. 1, 2, 3, and 4, and *Stauroneis* sp. 1 prefers slightly acidic condition with low pH level. The abundance of species like *Rhopalodia rupestris* (W. Smith) Krammer, *Cocconeis feuerbornii* Hustedt, *Craticula riperia* (Hustedt) Lange-Bertalot (Figure 4), *Sellaphora* sp., *Diploneis* sp. (Figure 4), and *Gomphonema* sp. 8 are influenced by high levels of nutrients (N and P) concentration and lower DO, corresponding to anthropogenic disturbances.

4. Discussion

This study demonstrated that studies on the diversity and composition of microbial flora inside the *Myristica* swamp forests are limited. Microbial communities have been recognized as key elements in ecosystem structure and functioning [60]. In this light, studying diatoms because of their unique characteristics such as (1) taxa with the capability to tolerate different environmental conditions [61], (2) with a restricted geographic distribution pattern, because each species has its own specific environmental preference [29] (3) a highly diverse group of microbial eukaryotes [62], and (4) high abundance across a wide range of aquatic ecosystems, can significantly help to understand various environmental drivers along with the history of that ecosystem. The other important fact is that natural acidic, poorly buffered (low alkaline) wetland environments are scarce worldwide. In addition, most of the information available about acidophilic communities in aquatic environments is focused on bacterial communities. However, microbial eukaryotes are also present and can play a critical role in these habitats [63].

Despite their importance, the *Myristica* swamp ecosystem in India is highly fragmented and being lost at an alarming rate due to human disturbance and climate change [9]. Thus, today's dire need is to pay considerable attention to developing tools for assessing the status and environmental changes occurring in these swamps. However, diatom-based assessment studies in swamps remain limited. Although other studies have been conducted on *Myristica swamps*, to our knowledge, this is the first report in which the water physicochemical parameters of an acidic swamp have been analyzed regarding the diatom assemblages.

4.1. Water Quality

Water chemistry is generally considered to be a primary factor controlling diatom distribution. Diatoms are sensitive to changing water chemistry, including pH, temperature, oxygen, and nutrient concentrations [64,65]. pH strongly affects diatom growth by causing physiological stress or indirectly by influencing other physicochemical variables [66]. The measured pH value in the current study indicated that most of the swamps were mildly acidic to circumneutral, with a pH value between 6.6 and 7.4. This confirms with the studies done on the soils of Uttara Kannada district, in the central Western Ghats, which indicate that humic decomposition and derivation of soil from the metamorphic parent rock led to laterite soil formation with a slightly acidic pH [67]. Conductivity is considered an enrichment indicator parameter and is an important determinant factor of diatom distribution in peatlands and wetlands [68,69]. Our study observed that swamp water was very low in mineral content and characterized by low conductivity (25–75 µS/cm), except for 3–4 swamps with higher ionic composition (201 µS/cm, Table 2). In agreement with Raghu et al. [70], who also reported that highly anaerobic conditions and permanently flooded soils led to a reduction in pH and electrical conductivity.

Furthermore, the significant effects of nutrients like (phosphorus and nitrogen) on diatoms have been frequently reported in lakes [71], but the importance of nutrients for diatoms in peatlands varies [72]. Wetlands are particularly susceptible to nutrient enrichment as these ecosystems act as sinks [73]. In our study, nitrate values measured amongst the majority swamps were low and generally reflected an oligotrophic state of these swamps. However, the high amount of phosphate values noted inside the swamps could be attributed to the high amount of biologically non-available P, which is not readily available to biological organisms in terms of their productivity [74]. Dissolved oxygen is crucial for the survival of organisms inside the water body [27]. In the current study, DO values across the swamps varied between 6.5–8.5 mg/L, indicating biological stable wetlands.

4.2. Diatom Community Composition, Diversity, and Distribution

In the 20 samples, we identified 91 diatom species, from which nearly 51.64% of the taxa were cosmopolitan, and 48.35% of taxa showed restricted geographical distribution. In agreement with the hypothesis, the unique habitat structure and its associated moisture level drove the high number of geographically restricted diatom taxa. To identify the geographically restricted taxa, the main constrain was the literature availability for their identification and confirmation. Most of the current literature available focuses on European flora; literature from the tropics is limited. Hence a regional (at least southeast Asia) level diatom database is essential for the identification as well as to further understand their environment optima. The voucher flora was built first, using all possible literature originating from tropical Asia, and the same will be published in the near future in a separate monograph due to the high number of new taxa.

The dominant diatom species identified in the present study were *Eunotia rhomboidea*, *Eunotia* c.f. *incisa*, *Eunotia bilunaris*, *Frustulia jogensis*, *Frustulia crassinervia*, *Brachysira neoexilis*, *Navicula obtecta*, *Navicula globulifera* var. *robusta*, *Gomphonema gandhii*, *Gomphonema parvuvlum*, *Ulnaria ulna*, and *Planothidium frequentissimum*. Each of these taxa possesses its own specific ecological preferences. *Eunotia bilunaris* extends from circumneutral to slightly acidophilous sites such as swamps and bogs. It is commonly observed in freshwater with low mineral content [75]. Diatoms like *Eunotia rhomboidea*, *Eunotia* c.f. *incisa* and *Frustulia crassinervia* are common in low pH and low mineral content environments like peat bogs, fen, etc., indicating pristine water with oligotrophic conditions [47,76]. *Brachysira neoexilis* is commonly found in circumneutral to slightly alkaline pH, in oligo to the mesotrophic environment [77]. *Navicula obtecta* prefers an electrolyte and nutrient-poor, pristine oligotrophic environment [51]. *Gomphonema gandhii* is found at circumneutral pH in electrolyte and nutrient-poor environments [48]. *Gomphonema parvulum* and *Ulnaria ulna* are common in oligo to mesotrophic freshwater environments, with circumneutral to slightly alkaline conditions [78,79].

The analysis of diatom taxa in the swamps revealed that species from the genus *Eunotia*, *Frustulia*, *Brachysira,* and *Neidium* are prominent in the swamps, indicating acidic to circumneutral oligotrophic conditions. The high abundance of the member of *Eunotia* like *Eunotia rhomboidea*, *Eunotia* c.f. *incisa*, *Eunotia bilunaris* can be correlated with the indication of circumneutral to acidic, oligotrophic water in agreement with the other studies which state that *Eunotia* is widely accepted as a freshwater genus frequently associated with oligotrophic, acidic, and mostly low conductivity waters [76,78]. On the other hand, an abundance of *A. minutissimum* can be considered a low nutrient indicator species [80]. In our study, we found taxa from the genera *Eunotia* and *Achnanthidium* that are able to withstand harsher environmental conditions. For e.g., *Eunotia* is tolerant of desiccation [78] and often dominant in *Sphagnum* bogs, where is prone to drying out [81], whereas, *Achnanthidium* is known to live in ephemeral conditions and are considered to be more or less tolerant to desiccation [78]. The presence of pollution tolerant species of the genera *Gomphonema* and *Navicula* indicated anthropogenic impacts inside the swamps [52,78]. Thus, diatom flora inside the swamps reflected that species such as *Eunotia bilunaris*, *Frustulia crassinervia, Navicula obtecta, Gomphonema gandhii* are known to be indicators of oligotrophic, circumneutral to acidic, and mostly low conductivity waters [78,82], on the contrary, *Gomphonema parvulum, and Ulnaria ulna* are indicators of meso-eutrophic conditions [78]. Thus, in the current study, we mainly found the dominant diatom taxa that can stand low pH, low concentrations of available nutrients (particularly nitrogen), low conductivity, and ephemeral conditions.

Species diversity is frequently used in ecological status assessment for explaining spatial and temporal patterns of biotic communities [83]. Some studies have found that species diversity may increase or decrease with the increase in pollution, and it mainly depends on the degree and the type of pollution [29]. The relation between species diversity and disturbance has been discussed much in recent times. In accordance with the intermediate disturbance hypothesis [84], species diversity is maximal when disturbances are at moderate frequency/intensity enabling the coexistence of species with different life strategies. Whereas, a study done by Chen et al. [83] showed that the species diversity of urban downstream sites was lower than in the upstream sites, owing to the high intensity of pollution in the downstream sites. We observed high species richness and diversity during this study when disturbances were low. We observed high species richness and diversity at New Kathlekan swamp sites compared to other sites, which is also supported by Chandran et al. [85], who reported high species diversity along with the discovery of many endemic higher plant taxa, and this could be attributed to less agricultural impact and anthropogenic influence in these swamps which may be due to establishment of sacred groves as a traditional conservation practice. Species diversity was low in the Assoli swamps. This could be due to comparatively high disturbances in these swamps, primarily due to conversion of swamps to areca nut plantations and paddy fields and diversion of swamp water for agriculture and plantation [86]. Thus, species richness and diversity can be used to evaluate pollution status across the sites. Low diatom richness may indicate harsh growing conditions due to, e.g., chemical stress and physical disturbances, which can cause further decline of freshwater biodiversity.

4.3. Diatom Distribution in Relation to Environmental Variables

Among the environmental variables measured and analyzed in this study, PCA analysis (Figure 5) revealed that conductivity, nutrient concentration, and dissolved oxygen were the environmental variables that distinguished the swamps into three different groups. In PCA analysis, all Kathlekan swamps, Torme swamps, and Mundigethagu swamp are distributed under the influence of fewer nutrients and comparatively less disturbed conditions. The study supported this, which indicated that waterlogged conditions in the swamps could lead to acidification and high leaching loss of nitrogen, creating pockets of less nitrogen in swamps [87]. Whereas, Devikan–Assoli swamps, Hulikal swamps, and Hulidevarakudulu swamps indicate degraded conditions by anthropogenic disturbances and agricultural runoff conditions.

Canonical correspondence analysis (CCA) identified pH, dissolved oxygen, and EC as the most important measured environmental variables that were associated with the distribution of diatom assemblages. From the CCA ordination plot (Figure 6), it is evident that the majority of the commonly occurring species have a trending affinity to pH, dissolved oxygen, and ion concentration. This suggests that these variables influence the diatom assemblage structure inside the swamps. The first group of species present on the right side of the plot, such as *Eunotia intermedia*, *Eunotia rhomboidea*, *Neidium* sp. 1, 2, 3, and 4, and *Stauroneis* sp. 1, were distributed under the influence of low pH and low EC, these are the group of taxa which usually present in ecosystems that are slightly acidic to circumneutral and oligotrophic. The same has also been confirmed by the other studies, which showed that species such as *Eunotia* c.f. *incisa*, *Eunotia blunaris*, *Eunotia minor*, and *Frustulia* sp. are known to be indicators of acidic, oligotrophic, and mostly low conductivity waters [78,82]. Whereas the second group of species on the first axis like *Navicula obtecta*, *Achnanthidium linannulum*, *Gomphonema gandhii*, *Oricymba japonica, Navicula notha, Achnanthidium* sp. 1, *Cocconeis* sp., are affected by high dissolved oxygen concentration. The significant contribution of dissolved oxygen in structuring diatom assemblages has also been observed in the studies done by Triest et al. [88], Shibabaw et al. [66].

Moreover, the third group of species, on the second axis of the ordination plot, like *Ulnaria* sp. 1, *Nitzschia clausii, Planothidium freequentissimum, Gyrosigma acuminatum, Navicula* sp. 2, and *Navicula escambia*, are distributed under the effect of increased conductivity (EC). Whereas the other species on the second axis, such as *Rhopalodia rupestris*, *Cocconeis feuerbornii, Craticula riperia, Sellaphora* sp., *Diploneis* sp., and *Gomphonema* sp. 8, are influenced by elevated levels of nutrients (N and P). The studies done by Kilham et al. [89], van Dam et al. [78], and Watanabe et al. [79] have confirmed similar results, which showed that species like *Nitzschia clausii*, *Ulnaria,* and *Gyrosigma* sp. indicate increased productivity and eutrophic water condition. Our results are comparable with the study done by Dalu et al. [31], who reported DO and turbidity as the principal predictors of diatom species richness in the Austral temperate rivers in South Africa. Thus, in CCA analysis, all over species distribution co-relates with the fact that the swamps situated over the left side of the diagram are relatively cleaner and oligotrophic in status with well-oxygenated water conditions (Torme swamp, Kathlekan swamps, and Mundigethagu swamps). Moreover, swamps scattered on the right side of the axis are relatively high in their nutrient load and under some anthropogenic or agricultural disturbance. The same can be reconfirmed from the studies in Uttara Kannada district in the central Western Ghats, revealing that 17 out of 51 observed swamps face extinction due to agricultural repurposing [14].

Overall, these swamps are biologically and ecologically rich, unique, valuable, and are home to rare and endemic biota. Despite this fact, a study on swamps across the Uttara Kannada district showed that this unique ecosystem is declining unceasingly and on the verge of extinction due to anthropogenic activities. Alteration of the ecological conditions inside these swamps will lead to further decline of freshwater biodiversity associated with swamps. To prevent this, the presumed widespread loss of these swamps needs detailed research on ecosystem services and the function they play in watershed dynamics. Moreover, a detailed review by Ranganathan et al. [4] on these swamps calls for a dire need for conservation initiatives to save the last few swamps. According to them, further research is needed to understand the abiotic factors governing the swamps. Further, to better protect these swampy forests, the study of Ranganathan et al. [4] recommended building a clear policy with the involvement of local communities. In this line, our study will provide more clarity about how diatoms and hydrochemistry data can be used to better understand the changing environment in the swamp and eventually which will impact the livelihood of local communities by altered water quantity and quality.

5. Conclusions

The study of *Myristica* swamps revealed a high number of diatom taxa unknown to science that will need formal description in the future, indicating that the diatom flora of the swamp is not well known. A better knowledge of the freshwater diatom flora inside the swamps will help improve our fundamental understanding of the diversity, ecology, and community associations of diatoms, which can be further used in future environmental research, including palaeoecological studies in this area. Our results showed that swamp diatom assemblages responded to environmental gradients at multiple scales. The important environmental gradients that affected the swamp diatom assemblages were pH, Dissolved Oxygen, and conductivity, which in turn, were influenced by land-use patterns inside the swamps. Repurposing of swamps for agriculture and water-flow alterations are major stressors inside the swamps, threatening the quality and availability of water and biodiversity. Thus, diatoms clearly reflected the intensity of human activities. Overall, the results of this study highlight the diatom assemblage structure in these swamps and further showed that diatom biodiversity could be used as a bioindicator to understand the possible impact of the most important water physicochemical parameters inside the swamps.

Supplementary Materials: The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/d14030202/s1, Table S1: Species name with abbreviation code and % RA of diatom across the sampling sites. Table S2: Presence absence data for diatom taxa across the sampling sites.

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References

1. Ramsar, C. *The List of Wetlands of International Importance*; RAMSAR Secretariat: Gland, Switzerland, 2013.
2. Rao, R.R. *Biodiversity in India: Floristic Aspects*; Bishen Singh Mahendra Pal Singh: Delhi, India, 1994.
3. Krishnamoorthy, K. *Myristica* Swamps in the evergreen forests of Travancore. *Indian For.* **1960**, *86*, 314–315.
4. Ranganathan, P.; Ravikanth, G.; Aravind, N.A. A review of research and conservation of *Myristica* swamps, a threatened freshwater swamp of the Western Ghats, India. *Wetl. Ecol. Manag.* **2021**, *30*, 171–189. [CrossRef]
5. Myers, N.; Mittermeier, R.A.; Mittermeier, C.G.; da Fonesca, G.A.B.; Kent, J. Biodiversity hotspots for conservation priorities. *Nature* **2000**, *403*, 853–858. [CrossRef] [PubMed]
6. Ganesan, R. Evergreen Forest swamps and their plant species diversity in Kalakad-Mundanthurai Tiger Reserve, South Western Ghats, India. *Indian For.* **2002**, *128*, 1351–1359.
7. Pushpakaran, B.; Gopalan, R. Study on seasonally waterlogged grasslands of Mudumalai Tiger Reserve. *Int. J. Sci. Res. Publ.* **2013**, *3*, 1–6.
8. Bassi, N.; Kumar, M.D.; Sharma, A.; Pardha-Saradhi, P. Status of wetlands in India: A review of extent, ecosystem benefits, threats and management strategies. *J. Hydrol. Reg.* **2014**, *2*, 1–19. [CrossRef]
9. Chandran, M.D.S.; Mesta, D.K. On the conservation of the *Myristica* swamps of the Western Ghats. In *Forest Genetic Resources: Status and Conservation Strategies*; Uma Shaanker, R., Ganeshaiah, K.N., Bawa, K.S., Eds.; Oxford & IBH Publications Co: New Delhi, India, 2001; pp. 1–19.
10. Nair, P.V.; Ramachandran, K.K.; Swarupanandan, K.; Thomas, T.P. *Mapping Biodiversity of the Myristica Swamps in Southern Kerala*; Research Report No. 326; Kerala Forest Research Institute: Thrissur, India, 2007; p. 255.
11. Santhakumaran, L.N.; Singh, A.; Thomas, V.T. Description of a sacred grove in Goa (India), with notes on the unusual aerial roots produced by its vegetation. *Wood* **1995**, *Oct–Dec*, 24–28.
12. Sreedharan, G.; Indulkar, M. New distributional record of the northernmost *Myristica* swamp from the Western Ghats of Maharashtra. *Curr. Sci.* **2018**, *115*, 1434.
13. Chandran, M.D.S.; Mesta, D.K.; Naik, M.B. *Inventorying and Conservation of the Myristica Swamps of Uttara Kannad*; Report; Research and Training Institute: Bangalore, India, 1999.
14. Bhat, P.R.; Kaveriappa, K.M. Ecological studies on *Myristica* swamp forests of Uttara Kannada, Karnataka, India. *Trop. Ecol.* **2009**, *50*, 329–337.
15. Varghese, A.V.; Kumar, B.M. Ecological observations in the freshwater swamp forests of southern Kerala, India. *J. Trop. For. Sci.* **1997**, *9*, 299–314.
16. Chandran, S.; Mesta, D.K.; Ali, S.; Gururaja, K.V.; Rao, G.R.; Ramachandra, T.V. Discovery of two critically endangered tree species and issues related to relic forests of the Western Ghats. *Open Conserv. Biol. J.* **2008**, *2*, 1–8. [CrossRef]
17. Roby, T.J.; Jose, J.; Nair, P.V. Checklist of Flora of *Myristica* swamps—A critically endangered freshwater ecosystem of Southern Western Ghats of Kerala India. *Ind. For.* **2014**, *140*, 608–616.
18. Gururaja, K.V.; Aravind, N.A.; Ali, S.; Ramachandra, T.V.; Velavan, T.P.; Krishnakumar, V.; Aggarwal, R. A new frog species from the central Western Ghats of India, and its phylogenetic position. *Zool. Sci.* **2007**, *24*, 525–534. [CrossRef]
19. Latha, K.D.; Raj, K.A.; Manimohan, P. *Laccaria violaceotincta*: A new species from tropical India based on morphology and molecular phylogeny. *Phytotaxa* **2019**, *392*, 140–146. [CrossRef]
20. Sujitha, P.C.; Prasad, G.; Sadasivan, K. Butterflies of the *Myristica* swamp forests of Shendurney Wildlife Sanctuary in the southern Western Ghats, Kerala, India. *J. Threat. Taxa* **2019**, *11*, 13320–13333. [CrossRef]
21. Thacker, M.; Roy, S.; Kociolek, J.P.; Lowe, R.L.; Karthick, B. A new species of *Germainiella* (Bacillariophyta) from the *Myristica* swamps of the Western Ghats, India. *Diatom Res.* **2019**, *34*, 181–191. [CrossRef]
22. Tambat, B.; Chaithra, G.N.; Reddy, K.V.V.; Vasudeva, R.; Ganeshaiah, K.N.; Shaanker, R.U. Threat assessment of *Myristica* swamps in central Western Ghats. *Int. J. For. Usufructs Manag.* **2012**, *13*, 3–12.
23. Kumaran, K.P.N.; Limaye, R.B.; Punekar, S.A.; Rajaguru, S.N.; Joshi, S.V.; Karlekar, S.N. Vegetation response to South Asian Monsoon variations in Konkan, western India during the Late Quaternary: Evidence from fluvio-lacustrine archives. *Quat. Int.* **2013**, *286*, 3–18. [CrossRef]
24. Srivastava, G.; Trivedi, A.; Mehrotra, R.C.; Paudayal, K.N.; Limaye, R.B.; Kumaran, K.P.N.; Yadav, S.K. Monsoon variability over Peninsular India during Late Pleistocene: Signatures of vegetation shift recorded in terrestrial archive from the corridors of Western Ghats. *Palaeogeogr. Palaeoclimatol. Palaeoecol.* **2016**, *443*, 57–65. [CrossRef]
25. Li, L.; Zheng, B.; Liu, L. Biomonitoring and bioindicators used for river ecosystems: Definitions, approaches and trends. *Procedia Environ. Sci.* **2010**, *2*, 1510–1524. [CrossRef]
26. Dalu, T.; Froneman, P.W. Diatom-based water quality monitoring in southern Africa: Challenges and future prospects. *Water SA* **2016**, *42*, 551–559. [CrossRef]
27. Kock, A.; Taylor, J.C.; Malherbe, W. Diatom community structure and relationship with water quality in Lake Sibaya, KwaZulu-Natal, South Africa. *S. Afr. J. Bot.* **2019**, *123*, 161–169. [CrossRef]

28. Battarbee, R.W.; Jones, V.J.; Flower, R.J.; Cameron, N.G.; Bennion, H.; Carvalho, L.; Juggins, S. Diatoms. In *Tracking Environmental Change Using Lake Sediments: Developments in Paleoenvironmental Research*; Smol, J.P., Birks, H.J.B., Last, W.M., Bradley, R.S., Alverson, K., Eds.; Springer: Dordrecht, The Netherlands, 2002; Volume 3, pp. 155–202. [CrossRef]
29. Stevenson, R.J.; Pan, Y.; van Dam, H. Assessing environmental conditions in rivers and streams with diatoms. In *The Diatoms: Applications for the Environmental and Earth Sciences*; Smol, J.P., Stoermer, E.F., Eds.; Cambridge University Press: Cambridge, UK, 2010; pp. 57–85.
30. Dixit, S.S.; Smol, J.P.; Kingston, J.C.; Charles, D.F. Diatoms: Powerful indicators of environmental change. *Environ. Sci. Technol.* **1992**, *26*, 22–33. [CrossRef]
31. Dalu, T.; Wasserman, R.J.; Magoro, M.L.; Mwedzi, T.; Froneman, P.W.; Weyl, O.L. Variation partitioning of benthic diatom community matrices: Effects of multiple variables on benthic diatom communities in an Austral temperate river system. *Sci. Total Environ.* **2017**, *601*, 73–82. [CrossRef]
32. Hamilton, P.B.; Hutchinson, S.J.; Patterson, R.T.; Galloway, J.M.; Nasser, N.A.; Spence, C.; Palmer, M.J.; Falck, H. Late-Holocene diatom community response to climate driven chemical changes in a small, subarctic lake, Northwest Territories, Canada. *Holocene* **2021**, *31*, 1124–1137. [CrossRef]
33. Battarbee, R.W.; Donald, F.; Charles, C.B.; Cumming, B.F.; Renberg, I. Diatoms as indicators of surface-water acidity. In *The Diatoms: Applications for the Environmental and Earth Sciences*, 2nd ed.; Smol, J.P., Stoermer, E.F., Eds.; Cambridge University Press: Cambridge, UK, 2010; pp. 98–121.
34. Birks, H.J.B.; Simpson, G.L. 'Diatoms and pH reconstruction' (1990) revisited. *J. Paleolimnol.* **2013**, *49*, 363–371. [CrossRef]
35. Water Environment Federation; American Public Health Association (APHA). *Standard Methods for the Examination of Water and Wastewater*, 21st ed.; American Public Health Association (APHA): Washington, DC, USA, 2005.
36. Hach, C.C. *Phosphorus, Reactive, Amino Acid Method Manual*, 9th ed.; Hach Co.: Loveland, CO, USA, 2014; p. 6.
37. Hach, C.C. *Nitrate, Cadmium Reduction Method Manual*, 11th ed.; Hach Co.: Loveland, CO, USA, 2019; p. 7.
38. Karthick, B.; Taylor, J.C.; Mahesh, M.K.; Ramachandra, T.V. Protocols for collection, preservation and enumeration of diatoms from Aquatic habitats for water quality monitoring in India. *IUP J. Soil Water Sci.* **2010**, *3*, 25–60.
39. The GIMP Team. University of California, Berkeley, 1997–2021. Available online: https://www.gimp.org/ (accessed on 20 November 2021).
40. Harrington, B.; Gould, T.; Hurst, N.; Mentalgu, Y. Inkscape 2004–2005. Available online: https://www.inkscape.org/ (accessed on 28 April 2020).
41. Krammer, K.; Lange-Bertalot, H. Bacillariophyceae 1. Teil: Naviculaceae. In *Süßwasserflora von Mitteleuropa*; Ettl, H., Gerloff, J., Heynig, H., Mollenhauer, D., Eds.; Gustav Fischer Verlag: Stuttgart, Germany, 1986; p. 876.
42. Krammer, K.; Lange-Bertalot, H. Bacillariophyceae 2. Teil: Bacillariaceae, Epithemiaceae, Surirellaceae. In *Süßwasserflora von Mitteleuropa*; Ettl, H., Gerloff, J., Heynig, H., Mollenhauer, D., Eds.; Gustav Fischer Verlag: Stuttgart, Germany, 1988; p. 596.
43. Krammer, K.; Lange-Bertalot, H. Bacillariophyceae 3. Teil: Centrales Fragilariaceae, Eunotiaceae. In *Die Süßwasserflora von Mitteleuropa*; Ettl, H., Gerloff, J., Heynig, H., Mollenhauer, D., Eds.; Gustav Fischer Verlag: Stuttgart, Germany, 1991; p. 576.
44. Krammer, K.; Lange-Bertalot, H. Bacillariophyceae 4. Teil: Achnanthaceae. Kritische Erga¨nzungen zu Navicula (Lineolatae) und Gomphonema. Gesamtliteraturverzeichnis Teil 1–4. In *Süßwasserflora von Mitteleuropa*; Ettl, H., Gerloff, J., Heynig, H., Mollenhauer, D., Eds.; Gustav Fischer Verlag: Stuttgart, Germany, 1991; p. 437.
45. Krammer, K. *Diatoms of Europe: Diatoms of the European Inland Waters and Comparable Habitats, The Genus Pinnularia*; Ruggell Koeltz Scientific Books: Konigstein, Germany, 2000; Volume 1, p. 703.
46. Lange-Bertalot, H. *Diatoms of Europe: Diatoms of the European Inland Waters and Comparable Habitats, Navicula Sensu Stricto; 10 Genera Separated from Navicula Sensu Lato Frustulia*; Ruggell Koeltz Scientific Books: Konigstein, Germany, 2001; Volume 2, p. 526.
47. Lange-Bertalot, H.; Hofmann, G.; Werum, M.; Cantonati, M.; Kelly, M.G. *Freshwater Benthic Diatoms of Central Europe: Over 800 Common Species Used in Ecological Assessment*; English Edition with Updated Taxonomy and Added Species; Koeltz Botanical Books: Glashütten, Germany, 2017; pp. 1–942.
48. Karthick, B.; Kociolek, J.P.; Mahesh, M.K.; Ramachandra, T.V. The diatom genus *Gomphonema* Ehrenberg in India: Checklist and description of three new species. *Nova Hedwig.* **2011**, *93*, 211. [CrossRef]
49. Karthick, B.; Hamilton, P.B.; Kociolek, J.P. *An Illustrated Guide to Common Diatoms of Peninsular India*; Gubbi Labs: Gubbi, India, 2013; p. 206.
50. Karthick, B.; Taylor, J.C.; Hamilton, P.B. Two new species of *Achnanthidium* Kützing (Bacillariophyceae) from Kolli Hills, Eastern Ghats, India. *Fottea* **2017**, *17*, 65–77. [CrossRef]
51. Jüttner, I.; Cox, E.J.; Ormerod, S.J. New or poorly known diatoms from Himalayan streams. *Diatom Res.* **2000**, *15*, 237–262. [CrossRef]
52. Taylor, J.C.; Harding, W.R.; Archibald, C.G.M. *An Illustrated Guide to Some Common Diatom Species from South Africa*; WRC Report No. TT282/07; Water Research Commission: Pretoria, South Africa, 2007.
53. Taylor, J.C.; Cocquyt, C.; Mayama, S. *Navicula nielsfogedii* JC Taylor & Cocquyt sp. nov., a new diatom (Bacillariophyta) from tropical and sub-tropical Africa. *Fottea* **2016**, *16*, 201–208. [CrossRef]
54. Chudaev, D.A.; Glushchenko, A.M.; Blagoveshchenskaya, E.Y.; Karthick, B.; Kulikovskiy, M.S. Morphology and taxonomy of *Navicula escambia* and *N. simulata* (Bacillariophyceae). *Nova Hedwig.* **2018**, *147*, 43–57. [CrossRef] [PubMed]

55. Bishop, I.W.; Esposito, R.M.; Tyree, M.; Spaulding, S.A. A diatom voucher flora from selected southeast rivers (USA). *Phytotaxa* **2017**, *332*, 101–140. [CrossRef]
56. Kociolek, J.P.; Balasubramanian, K.; Blanco, S.; Coste, M.; Ector, L.; Liu, Y.; Kulikovskiy, M.; Lundholm, N.; Ludwig, T.; Potapova, M.; et al. DiatomBase. Available online: https://www.diatombase.org (accessed on 30 December 2020).
57. Hammer, Ø.; Harper, D.A.T.; Ryan, P.D. Past: Paleontological statistics software package for education and data analysis. *Palaeontol. Electron.* **2001**, *4*, 9.
58. Ter Braak, C.J.F.; Šmilauer, P. *Canoco Reference Manual and User's Guide: Software for Ordination*; Version 5.0; Microcomputer Power: Ithaca, NY, USA, 2012; p. 496.
59. Gauch, H.G. *Multivariate Analysis in Community Ecology*; Cambridge University Press: Cambridge, UK, 1982.
60. Benito, X.; Fritz, S.C.; Steinitz-Kannan, M.; Tapia, P.M.; Kelly, M.A.; Lowell, T.V. Geo-climatic factors drive diatom community distribution in tropical South American freshwaters. *J. Ecol.* **2018**, *106*, 1660–1672. [CrossRef]
61. Passy, S.I. Framework for community functioning: Synthesis of stress gradient and resource partitioning concepts. *PeerJ* **2017**, *5*, e3885. [CrossRef]
62. Mann, D.G. The species concept in diatoms. *Phycologia* **1999**, *38*, 437–495. [CrossRef]
63. Aguilera, A.; Manrubia, S.C.; Gómez, F.; Rodríguez, N.; Amils, R. Eukaryotic community distribution and its relationship to water physicochemical parameters in an extreme acidic environment, Rio Tinto (southwestern Spain). *Appl. Environ. Microbiol.* **2006**, *72*, 5325–5330. [CrossRef]
64. Potapova, M.; Charles, D.F. Distribution of benthic diatoms in U.S. rivers in relation to conductivity and ionic composition. *Freshw. Biol.* **2003**, *48*, 1311–1328. [CrossRef]
65. Smol, J.P.; Wolfe, A.P.; Birks, H.J.B.; Douglas, M.S.V.; Jones, V.J.; Korhola, A.; Pienitz, R.; Ruhland, K.; Sorvari, S.; Antoniades, D.; et al. Climate-driven regime shifts in the biological communities of arctic lakes. *Proc. Natl. Acad. Sci. USA* **2005**, *102*, 4397–4402. [CrossRef]
66. Shibabaw, T.; Beyene, A.; Awoke, A.; Tirfie, M.; Azage, M.; Triest, L. Diatom community structure in relation to environmental factors in human influenced rivers and streams in tropical Africa. *PLoS ONE* **2021**, *16*, e0246043. [CrossRef]
67. Vijayakumar, P.K.; Vasudeva, R. Characterization of soil properties from freshwater swamps and adjoining evergreen forest area. *Karnataka J. Agric. Sci.* **2011**, *24*, 601–602.
68. Johnson, P.; Gerbeaux, P. *Wetland Types in New Zealand*; Department of Conservation and Ministry for the Environment: Wellington, New Zaeland, 2004.
69. Hajkova, P.; Bojková, J.; Frankova, M.; Opravilová, V.; Hajek, M.; Kintrová, K.; Horsak, M. Disentangling the effects of water chemistry and substratum structure on moss-dwelling unicellular and multicellular micro-organisms in spring-fens. *J. Limnol.* **2011**, *70*, 54. [CrossRef]
70. Raghu, H.B.; Vasudeva, R.; Nagesha, G.K.; Krishna, H.C.; Hombegowda, H.C. Dasappa and Boby, VU, 2006, Chemical properties of soils from freshwater swamps of Uttara Kannada. *Mysore J. Agric. Sci.* **2006**, *40*, 367–370.
71. Chen, X.; Bu, Z.; Stevenson, M.A.; Cao, Y.; Zeng, L.; Qin, B. Variations in diatom communities at genus and species levels in peatlands (central China) linked to microhabitats and environmental factors. *Sci. Total Environ.* **2016**, *568*, 137–146. [CrossRef]
72. Poulíčková, A.; Hájková, P.; Kintrová, K.; Bat'ková, R.; Czudková, M.; Hájek, M. Tracing decadal environmental change in ombrotrophic bogs using diatoms from herbarium collections and transfer functions. *Environ. Pollut.* **2013**, *179*, 201–209. [CrossRef]
73. Humphries, M.S.; Benitez-Nelson, C.R. Recent trends in sediment and nutrient accumulation rates in coastal, freshwater Lake Sibaya, South Africa. *Mar. Freshw. Res.* **2013**, *64*, 1087–1099. [CrossRef]
74. Reddy, K.R.; Wetzel, R.G.; Kadlec, R.H. Biogeochemistry of phosphorus in wetlands. In *Phosphorus: Agriculture and the Environment*; Sims, J.T., Sharpley, A.N., Eds.; Soil Science Society of America: Madison, WI, USA, 2005; Volume 46, pp. 263–316. [CrossRef]
75. Kim, Y.; Suk Choi, J.; Sin Kim, H. Epiphytic diatom communities from two mountain bogs in South Korea. *Nova Hedwig.* **2007**, *84*, 363–379. [CrossRef]
76. Lange-Bertalot, H.; Bąk, M.; Witkowski, A. *Eunotia* and some related genera. In *Diatoms of Europe—Diatoms of the European Inland Water and Comparable Habitats*; Lange-Bertalot, H., Ed.; A.R.G. Gantner: Ruggell, Liechtenstein, 2011; Volume 6, pp. 1–747.
77. Van de Vijver, B.; Schuster, T.M.; Kusber, W.-H.; Hamilton, P.B.; Wetzel, C.E.; Ector, L. Revision of European *Brachysira* species (*Brachysiraceae*, Bacillariophyta): I. The *Brachysira microcephala—B. neoexilis enigma*. *Bot. Lett.* **2021**, *168*, 467–484. [CrossRef]
78. Van Dam, H.; Mertens, A.; Sinkeldam, J. A coded checklist and ecological indicator values of freshwater diatoms from The Netherlands. *Neth. J. Aquat. Ecol.* **1994**, *28*, 117–133. [CrossRef]
79. Watanabe, T.; Ohtsuka, T.; Tuji, A.; Houki, A. *Picture Book and Ecology of the Freshwater Diatoms*; Uchida Rokakuho Publishing Company: Tokyo, Japan, 2005; p. 666.
80. Potapova, M.; Charles, D.F. Diatom metrics for monitoring eutrophication in rivers of the United States. *Ecol. Indic.* **2007**, *7*, 48–70. [CrossRef]
81. Hargan, K.E.; Rühland, K.M.; Paterson, A.M.; Finkelstein, S.A.; Holmquist, J.R.; MacDonald, G.M.; Keller, W.; Smol, J.P. The influence of water-table depth and pH on the spatial distribution of diatom species in peatlands of the Boreal Shield and Hudson Plains, Canada. *Botany* **2015**, *93*, 57–74. [CrossRef]
82. Cantonati, M.; Lange-Bertalot, H. Diatom monitors of close-to-pristine, very-low alkalinity habitats: Three new *Eunotia* species from springs in Nature Parks of the southeastern Alps. *J. Limnol.* **2011**, *70*, 209–221. [CrossRef]

83. Chen, X.; Zhou, W.; Pickett, S.T.; Li, W.; Han, L.; Ren, Y. Diatoms are better indicators of urban stream conditions: A case study in Beijing, China. *Ecol. Indic.* **2016**, *60*, 265–274. [CrossRef]
84. Fox, J.F. Intermediate-disturbance hypothesis. *Science* **1979**, *204*, 1344–1345. [CrossRef]
85. Chandran, M.D.S.; Rao, G.R.; Gururaja, K.V.; Ramachandra, T.V. Ecology of the swampy relic forests of Kathalekan from central Western Ghats, India. *Bioremediat. Biodivers. Bioavailab.* **2010**, *4*, 54–68.
86. Ramachandra, T.V.; Setturu, B.; Chandran, M.D.S. Geospatial analysis of forest fragmentation in Uttara Kannada District, India. *For. Ecosyst.* **2016**, *3*, 10. [CrossRef]
87. Raghu, H.B. Reproductive Biology and Conservation of *Semecarpus kathalekanensis*: A Threatened Swamp Species of the Central Western Ghats. Master's Thesis, University of Agricultural Sciences, Dharwad, India, 2000.
88. Triest, L.; Lung'ayia, H.; Ndiritu, G.; Beyene, A. Epilithic diatoms as indicators in tropical African rivers (Lake Victoria catchment). *Hydrobiologia* **2012**, *695*, 343–360. [CrossRef]
89. Kilham, P.; Kilham, S.S.; Hecky, R.E. Hypothesized resource relationships among African planktonic diatoms. *Limnol. Oceanogr.* **1986**, *31*, 1169–1181. [CrossRef]