

Article

Environmental Drivers and Aquatic Ecosystem Assessment of Periphytic Algae at Inflow Rivers in Six Lakes over the Yangtze River Basin

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Abstract: Periphytic algae is frequently utilized as a health indicator for ecosystems. Many research studies have been conducted in China on the periphytic algae community, but none has compared the periphytic algae community structure at inflow rivers among different lakes in the Yangtze river basin. The periphytic algae were investigated at 94 sites in inflow rivers of Dianchi Lake, Danjiangkou Reservoir, Dongtinghu Lake, Poyanghu Lake, Chaohu Lake, and Taihu Lake. Based on microscopic research, eight phyla and 126 genera of periphytic algae were found in the inflow river of six lakes, with Cyanobacteria and Bacillariophyta dominating. The CCA (Canonical Correspondence Analysis) was used to analyze the association between the periphytic algae community and environmental factors in the inflow river of six lakes, and the LefSe (Linear discriminant analysis effect size) analysis was used to find enriched species in the inflow river of six lakes. We discovered that TN (total nitrogen) and TP (total phosphorus) were the driving environment variables at the basin scale based on the combined results of the CCA and the Mantel Test. The TITAN (Threshold Indicator Taxa Analysis) analysis also revealed the indicator species and their TN and TP concentration thresholds. Finally, we assessed the ecosystem health of the inflow river at six lakes; biotic and abiotic indices yielded conflicting results, but utilizing both indices to assess ecosystem health using the Random Forest algorithm will yield objective and comprehensive results.

Keywords: Yangtze river basin; ecosystem assessment; environmental drivers; inflow river; periphytic algae



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1. Introduction

Periphytic algae can be found in a wide range of water bodies, from small puddles to enormous rivers. They are a key contribution to aquatic ecosystem primary productivity, as they play an important role in nutrient cycles and energy metabolism via photosynthetic metabolism [1]. Periphytic algae are vulnerable to environmental changes and human activities, such as habitat destruction, eutrophication, and heavy metal contamination. Periphytic algae community composition and relative abundance fluctuate depending on environmental factors [2]. Meanwhile, periphytic algae recover and spread faster than other species after disturbance, making them a helpful indicator of aquatic ecological conditions [3].

With the rapid industrialization and urbanization of human society, hydrodynamic circumstances, water quality, and biotic community have changed dramatically, and ecological environment degradation has worsened, resulting in a diverse structure and distribution of periphytic algal communities [4–6]. The relationship between periphytic algae and environmental heterogeneity has been studied extensively. According to functional marker

gene research, the community composition of periphytic algae is closely related to nutrient levels [7]. Pollution-tolerant taxa, such as bacillariophytes *Navicula*, dominated the structure of periphytic algae in Dianchi Lake, and TN, $\text{NH}_4\text{-N}^+$, and TP were significant aquatic environmental factors impacting the community composition of periphytic algae [8]. Seasonal fluctuation had a substantial impact on environmental parameters at the Danjiangkou Reservoir and its related water diversion channel, water temperature and nutrient concentration were the key factors determining the structure of the periphytic algae community [9], and the environmental factors plays an important role in shaping the periphytic algal community assembly in the water diversion channel based on marker gene analysis [10]. Different water levels in Poyang Lake resulted in environmental variety, resulting in varied habitats for periphytic algae [11]. Periphytic algae in Lake Taihu had a distinct community composition, with spatial variation and nutrient concentrations [12].

Many research studies on periphytic algae communities and their relationships with environmental variables have been conducted in a single Chinese lake, but few have been conducted at the basin scale between the inflow river of different lakes. This study focuses on the abundance, community composition and indicator species in the inflow river of six lakes. These six lakes, located in the Yangtze river basin, are Dianchi Lake, Danjiangkou Reservoir, Dongtinghu Lake, Poyanghu Lake, Chaohu Lake and Taihu Lake. The aim was to investigate the relationships between periphytic algae communities and environmental variables, to objectively evaluate the ecosystem health of the six lakes inflow river in the Yangtze river basin. To provide theoretical support and a scientific foundation for the maintenance of water ecological health.

2. Materials and Methods

2.1. Study Area

This study was conducted in the inflow rivers of six Yangtze River basin lakes (Dianchi Lake, Danjiangkou Reservoir, Dongtinghu Lake, Poyanghu Lake, Chaohu Lake, and Taihu Lake, Figure 1). In China, Dianchi Lake, Chaohu Lake, and Taihu Lake are examples of eutrophication inland lakes [13]. Poyang Lake and Dongting Lake are China's largest and second largest freshwater lakes, respectively, and they maintain a natural connection with the middle Yangtze River [14]. Danjiangkou Reservoir is the water source of the South-to-North Water Diversion Projects, which represent the world's largest water diversion projects [15].

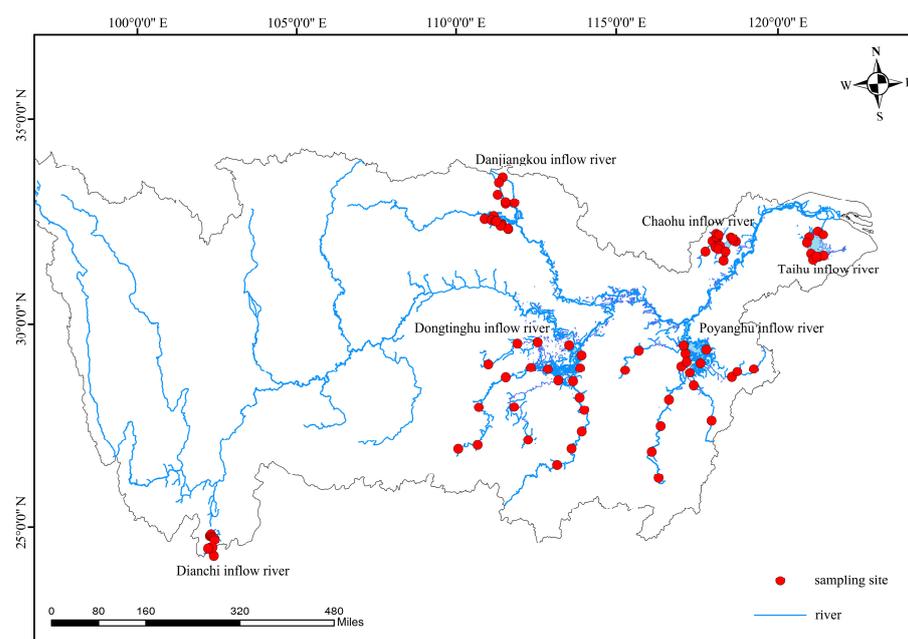


Figure 1. The location of the sampling sites in inflow rivers of six lakes.

2.2. Sampling and Analysis

Between June and August 2021, 94 sites were sampled for periphytic algae (Table 1). The periphytic algae sample approach is based on previous research [16]. At each site, sampling was done in the places where algae grew on the river's slope (10–30 cm from the water surface). A 100 cm² area was randomly picked, and the algae was scraped into the sample container with tweezers, a knife, and a brush. Sampling tools were then rinsed with distilled water, and the rinse water was then transferred to the sample bottle, and samples were fixed with a 1% Lugol iodine solution. After that, the samples were condensed and sedimented, with the volume of each sample set to 30 mL. After shaking and mixing well, 0.1 mL of the sample was placed into the counting frame; species identification and counting were carried out under a microscope (Olympus BX51, Olympus, Tokyo, Japan) at 400× or 1000× (for species in Bacillariophyta) magnification [17–19]. The concentration times were converted to the number of cells per unit area, and the number of cells was computed as the algae's abundance [20].

Table 1. Information of sampling sites.

Sample Sites	Longitude	Latitude	Area	Sample Sites	Longitude	Latitude	Area
1	109.4803	27.0753	Dongtinghu inflow river	51	114.29	28.73	Poyanghu inflow river
2	110.0222	27.1486	Dongtinghu inflow river	52	114.73	29.18	Poyanghu inflow river
3	110.11	28.09	Dongtinghu inflow river	53	114.8094	26.6581	Poyanghu inflow river
4	110.44	29.13	Dongtinghu inflow river	54	114.9242	25.9953	Poyanghu inflow river
5	110.9115	28.7746	Dongtinghu inflow river	55	115.1299	27.2832	Poyanghu inflow river
6	111.0989	28.0478	Dongtinghu inflow river	56	115.4378	27.9188	Poyanghu inflow river
7	111.3	29.59	Dongtinghu inflow river	57	115.88	28.69	Poyanghu inflow river
8	111.4266	27.2074	Dongtinghu inflow river	58	116.01	29.19	Poyanghu inflow river
9	111.64	28.98	Dongtinghu inflow river	59	116.02	29.19	Poyanghu inflow river
10	111.88	29.59	Dongtinghu inflow river	60	116.04	28.99	Poyanghu inflow river
11	112.1176	28.913	Dongtinghu inflow river	61	116.0439	28.7864	Poyanghu inflow river
12	112.18	26.54	Dongtinghu inflow river	62	116.0995	28.5081	Poyanghu inflow river
13	112.3867	28.6168	Dongtinghu inflow river	63	116.17	28.19	Poyanghu inflow river
14	112.61	26.91	Dongtinghu inflow river	64	116.4305	28.7169	Poyanghu inflow river
15	112.77	29.46	Dongtinghu inflow river	65	116.55	27.28	Poyanghu inflow river
16	112.8	28.57	Dongtinghu inflow river	66	116.64	29.03	Poyanghu inflow river
17	112.93	27.33	Dongtinghu inflow river	67	117.26	28.28	Poyanghu inflow river
18	112.95	28.17	Dongtinghu inflow river	68	117.43	28.4	Poyanghu inflow river
19	113.03	28.88	Dongtinghu inflow river	69	117.9	28.41	Poyanghu inflow river
20	113.05	27.85	Dongtinghu inflow river	70	110.57	32.7	Danjiangkou inflow river
21	113.1	29.19	Dongtinghu inflow river	71	110.78	32.66	Danjiangkou inflow river
22	102.77	24.49	Dianchi inflow river	72	110.84	32.75	Danjiangkou inflow river
23	102.61	24.66	Dianchi inflow river	73	110.89	32.63	Danjiangkou inflow river
24	102.69	24.69	Dianchi inflow river	74	110.91	32.63	Danjiangkou inflow river
25	102.73	24.69	Dianchi inflow river	75	111	33.27	Danjiangkou inflow river
26	102.78	24.88	Dianchi inflow river	76	111.03	32.52	Danjiangkou inflow river
27	102.78	24.92	Dianchi inflow river	77	111.06	33.56	Danjiangkou inflow river
28	102.74	24.95	Dianchi inflow river	78	111.18	33.68	Danjiangkou inflow river
29	102.71	24.97	Dianchi inflow river	79	111.22	33.03	Danjiangkou inflow river
30	102.65	24.99	Dianchi inflow river	80	111.22	33.08	Danjiangkou inflow river
31	102.67	25.01	Dianchi inflow river	81	111.24	32.44	Danjiangkou inflow river
32	102.68	25.03	Dianchi inflow river	82	111.25	32.43	Danjiangkou inflow river
33	116.95	31.43	Chaohu inflow river	83	111.48	33.04	Danjiangkou inflow river
34	117.19	31.66	Chaohu inflow river	84	111.07	32.54	Danjiangkou inflow river
35	117.26	31.5	Chaohu inflow river	85	111.07	32.55	Danjiangkou inflow river
36	117.27	31.66	Chaohu inflow river	86	120.02	30.88	Taihu inflow river
37	117.32	31.75	Chaohu inflow river	87	120.19	30.93	Taihu inflow river
38	117.33	31.45	Chaohu inflow river	88	120.13	30.94	Taihu inflow river
39	117.33	31.84	Chaohu inflow river	89	120.34	30.94	Taihu inflow river
40	117.35	31.54	Chaohu inflow river	90	119.99	31.03	Taihu inflow river

Table 1. Cont.

Sample Sites	Longitude	Latitude	Area	Sample Sites	Longitude	Latitude	Area
41	117.36	31.73	Chaohu inflow river	91	119.92	31.31	Taihu inflow river
42	117.39	31.52	Chaohu inflow river	92	120.01	31.45	Taihu inflow river
43	117.4	31.8	Chaohu inflow river	93	120.42	31.45	Taihu inflow river
44	117.44	31.16	Chaohu inflow river	94	120.28	31.55	Taihu inflow river
45	117.53	31.37	Chaohu inflow river				
46	117.7405	31.7096	Chaohu inflow river				
47	117.79	31.62	Chaohu inflow river				
48	117.79	31.68	Chaohu inflow river				
49	117.83	31.6	Chaohu inflow river				
50	117.87	31.58	Chaohu inflow river				

The multi-parameter water analyzer (YSI Professional Plus, Xylem, Yellow Springs, OH, USA) was used to determine the water temperature (Temp), pH, turbidity (TUB), electrical conductivity (Cond), and dissolved oxygen (DO). Total phosphorus (TP), total nitrogen (TN), ammonia nitrogen (AN), chemical oxygen demand by potassium permanganate (COD_{Mn}), and Silicon Dioxide (SiO₂) were measured by the Standard Methods for the Examination of Water and Wastewater in China [21].

2.3. Data Analysis

The Shannon–Wiener index, Margalef index, the Pielou index and dominant degree were calculated according to related studies [22,23]. Temp, pH, Cond, DO, TN, AN, TP and COD_{Mn} were used to calculate the water quality index (WQI) according to Pesce et al. [24].

The LEfSe method (<http://huttenhower.sph.harvard.edu/lefse/>, accessed on 27 April 2022) was used to detect the potential enriched species, and the Kruskal–Wallis test with value < 0.05 and LDA threshold > 5 was used to detect features with significantly different abundances. The mantel test analysis between environmental factors and periphytic algae used the ggcor package (version 0.9.5.2) in R, and the CCA and detrended correspondence analysis (DCA) were conducted based on the VEGAN package for R [25]. Indicator species were identified, and their threshold values were calculated using TITAN in the TITAN2 package in R [26]. This method compared the TN or TP mutation points of all species in the community, when numerous species exhibit similar responses at the same time within a short range of nutrient concentration. This range of nutrient concentration is considered as the community’s reaction threshold; for a more complete explanation of this method, please check the reference [27].

Based on the Random Forest Algorithm, the Shannon–Wiener index, Pielou index, Margalef index, and WQI were used to assess ecosystem health. These four indices were divided into five criteria based on related studies (Table 2) [22,24,28]. According to Table 1, 100 dataset groups are created at random for each assessment standard, for a total of 500 dataset groups for five standards. To develop a random forest model, 350 groups of datasets are randomly selected as training samples, and 150 groups of datasets are randomly selected as test samples. The results from this study are then substituted into the model to determine the water ecological health status of each region.

Table 2. Assessment standard of diversity indexes.

Assessment Standard	Shannon–Wiener Index	Pielou Index	Margalef Index	WQI
poor	0	0	0	(0, 25)
fair	(0, 1)	(0, 0.3)	(0, 0.6)	(25, 50)
average	(1, 2)	(0.3, 0.5)	(0.6, 1)	(50, 70)
good	(2, 3)	(0.5, 0.8)	(1, 1.6)	(70, 90)
excellent	(3, +∞)	(0.8, 1)	(1.6, 3)	(90, 100)

3. Results

3.1. Spatial Distribution of Periphytic Algae Community Structure

A total of 126 periphytic algae genera were discovered, distributing in eight phyla. Note that 56 genera were identified in Chlorophyta, 32 genera in Bacillariophyta, 26 genera in Cyanobacteria, and 10 genera in other phyla (Dinophyta, Cryptophyta, Chrysophyta, Euglenophyta and Rhodophyta).

Based on the cell density of each sampling site according to Table 1, the cell density and community composition of each area were computed. Figure 2a shows the periphytic algae cell density in each location. The median of cell density from highness to lowness in each area were: the Dianchi inflow river, the Chaohu inflow river, the Poyanghu inflow river, the Danjiangkou inflow river, the Dongtinghu inflow river and the Taihu inflow river. Based on Wilcoxon rank sum test, the cell density of the Dianchi inflow river is significantly higher than the Danjiangkou inflow river, the Dongtinghu inflow river and the Taihu inflow river; the cell density of the Chaohu inflow river is significantly higher than the Dongtinghu inflow river and the Taihu inflow river. Cyanobacteria was the dominant phylum in the community composition of periphytic algae in the Chaohu inflow river, the Poyanghu inflow river, and the Taihu inflow river. The dominant phyla in the Dianchi inflow river were Cyanobacteria and Bacillariophyta. The dominant phyla in the Danjiangkou inflow river and the Dongtinghu inflow river were Cyanobacteria, Bacillariophyta, and Chlorophyta.

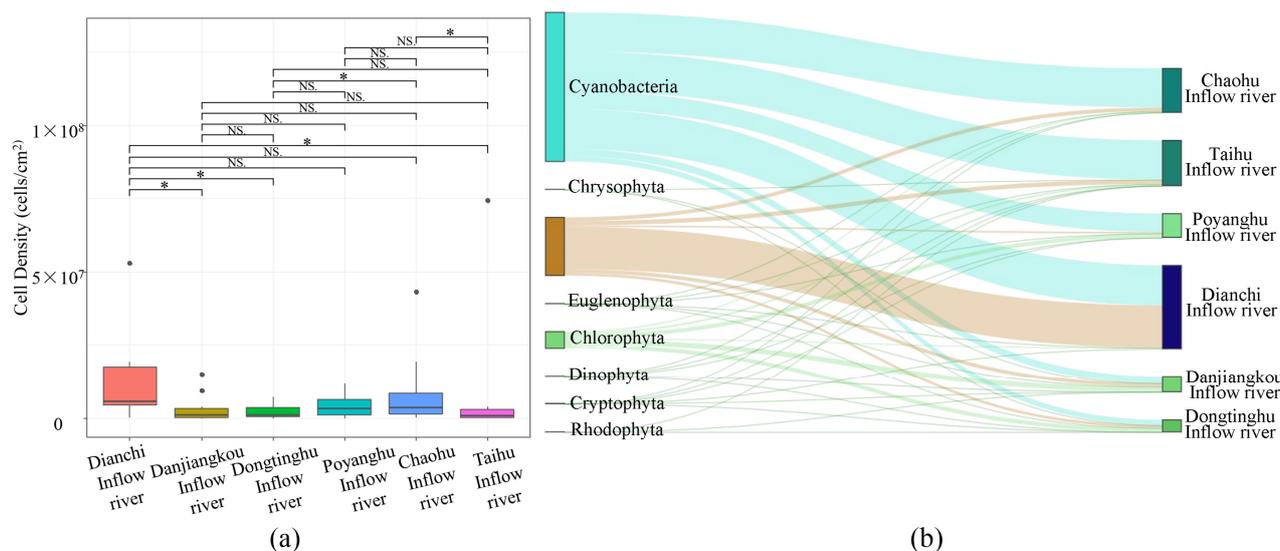


Figure 2. The cell density and community composition of periphytic algae in inflow rivers of six lakes. (a) Cell density; (b) Community composition. (* $p < 0.05$, and NS—not significant).

According to the LefSe analysis (Figure 3), when the threshold on the logarithmic LDA score for discriminative features was set to 5.0, the Poyanghu inflow river was abundant in Oscillatoriales, Oscillatoriaceae, and *Lyngbya*. Chlorophyta were abundant in the Dongtinghu inflow river, with *Gloeocystis* being the most common genus. Bacillariophyta were abundant in the Danjiangkou inflow river, with the genus level dominated by *Navicula* and *Achnantheidium*. Oscillatoriales, Pseudanabaenaceae, and *Pseudanabaena* were abundant in the Chaohu inflow river. There were no enriched species in other areas.

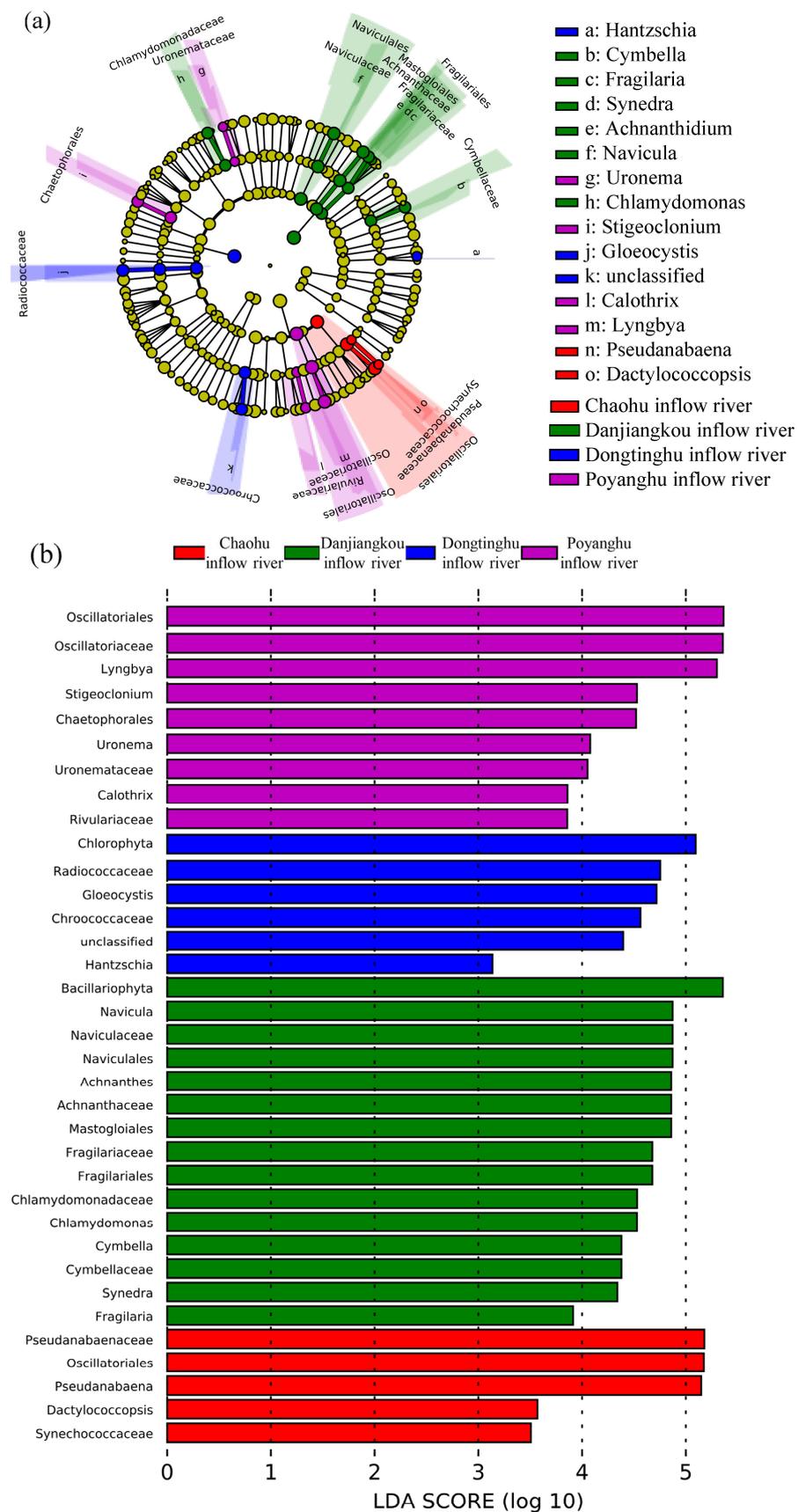


Figure 3. Comparison of periphytic algae variations based on LEfSe analysis. **(a)** Cladogram for taxonomic representation of significant differences among groups. **(b)** Histogram of the LDA scores for differentially abundant features among groups.

3.2. Relationship between Periphytic Algae Communities and Environment Variables

The length of the gradient was larger than 3.0 according to the DCA analysis, hence CCA was utilized to investigate the link between periphytic algae and environmental variables [29]. Prior to CCA, the Hellinger transformation was used to homogenize the variance of species abundance and down-weight rare species [30], and the multicollinearity of environmental factors was removed using the variance inflation factor [31], as shown in Figure 4.

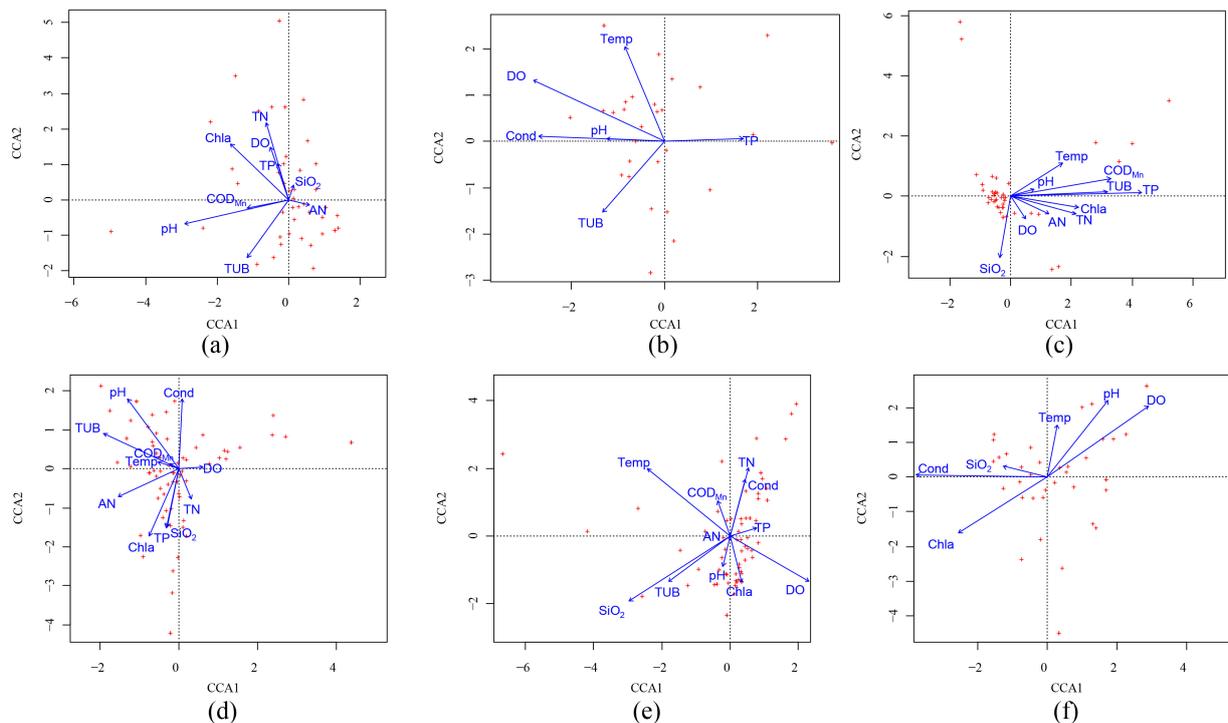


Figure 4. Canonical correspondence analysis (CCA) showed the relationship of the periphytic algae community and environment variable in each inflow river. (a) Chaohu inflow river; (b) Dianchi inflow river; (c) Danjiangkou inflow river; (d) Dongtinghu inflow river; (e) Poyanghu inflow river; (f) Taihu inflow river.

To find significant correlations between periphytic algae communities and environmental variables, the criterion of $p < 0.05$ and 1000 Monte Carlo permutations was used. The periphytic algal population in the Chaohu inflow river was significantly influenced by TN and pH. The periphytic algal population in the Danjiangkou inflow river was significantly influenced by TP, TUB, and COD_{Mn} . In the Dongtinghu inflow river, pH had a substantial impact on the periphytic algal community. The periphytic algae population in the Poyanghu inflow river was significantly affected by Temp and DO. The periphytic algae community in the Dianchi and Taihu inflow rivers had no environmental drivers.

3.3. Indicator Species and Ecological Community Thresholds

Figure 5 shows the CCA and Mantel test results based on periphytic algae community and environment variable data from all inflow rivers. Temp, Cond, TUB, TP, TN, COD_{Mn} , Chla, and SiO_2 all had a substantial impact on the periphytic algae population, according to CCA. Because Cyanobacteria and Bacillariophyta were the most prevalent phyla in all inflow rivers (Figure 2b), the Mantel test was used to investigate their relationships with environmental variables. Cyanobacteria were primarily influenced by AN, while Bacillariophyta were influenced by DO, TP, TN, and AN. TN and TP were the overlapped environment variables that significantly influenced the periphytic algae community composition in all inflow rivers, according to the results of the CCA and Mantel test.

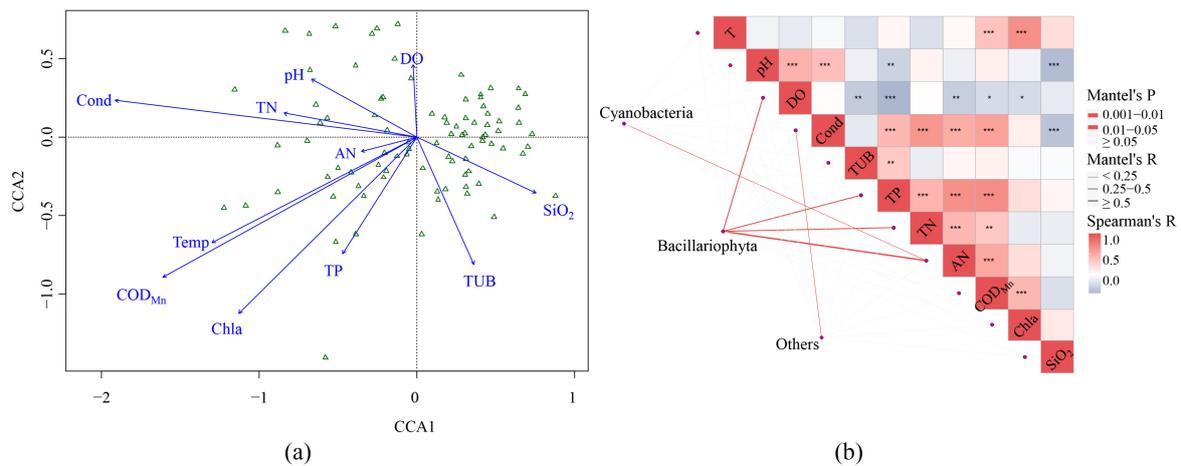


Figure 5. CCA and Mantel test showed the relationship of periphytic algae community and environment variable in all inflow river. (a) CCA; (b) Mantel test.

The indicator species and ecological community thresholds in all inflow rivers were further analyzed using TN and TP. Species with dominant degree higher than 0.0001 and occurrence higher than three times were used for analysis, TITAN identified positive (z+) and negative (z-) responding species for periphytic community change points in response to TN and TP (Figure 6).

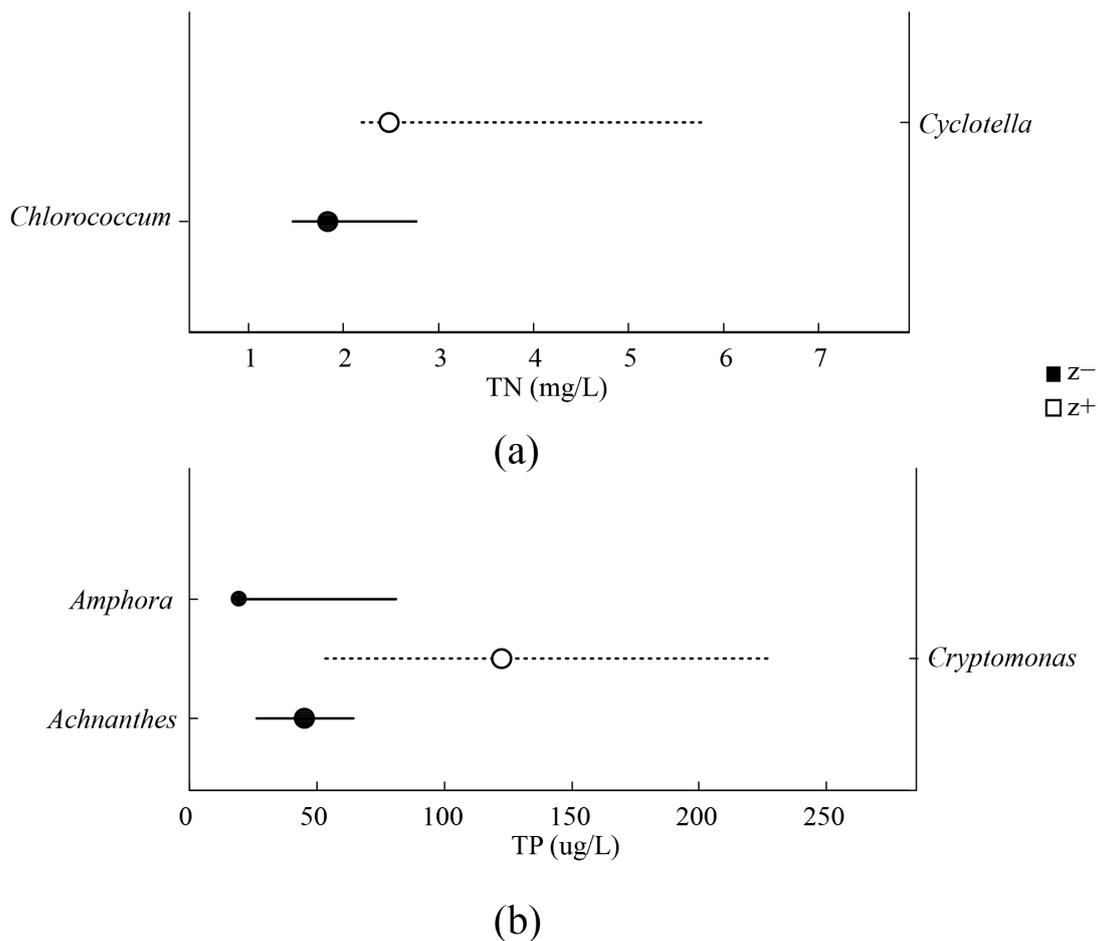


Figure 6. TITAN results showing significant indicator taxa. Circles showed taxa change points which were sized according to magnitudes of response. (a) Indicating species and thresholds of TN; (b) Indicating species and thresholds of TP.

Chlorococcum has been found as a negative reacting species to TN, with cell density increasing as TN decreases and a threshold of 1.835 mg/L. As a result, TN concentrations below 1.835 mg/L are insufficient for periphytic algae growth and reproduction. *Cyclotella* has been found as a positive responsive species, with cell density increasing as TN increases and *Cyclotella* thresholds of 2.355 mg/L. As a result, TN concentrations more than 2.355 mg/L are unsuitable for periphytic algae growth and reproduction. In all input rivers of the Yangtze river basin, the lowest acceptable TN concentration for periphytic algae is 1.835 mg/L.

The *Amphora* and *Achnanthydium* were shown to be negative reacting species for TP, with thresholds of 45 µg/L and 19.5 µg/L, respectively. As a result, TP concentrations less than 19.5 µg/L are insufficient for periphytic algae growth and reproduction. *Cryptomonas* has been found as a positive responsive species, with a 122.5 µg/L threshold. As a result, the lowest acceptable TP concentration of periphytic algae in the Yangtze inflow rivers is 19.5 µg/L.

3.4. Ecosystem Health Assessment in All Inflow River

The Shannon–Wiener index, Margalef index, and the Pielou index were used to assess the biotic health of each of the inflow rivers. The WQI is used to quantify water quality using multiple measurable parameters, allowing us to monitor changes in water quality and identify water trends [24], so the WQI tests were used to assess the abiotic health of each inflow river, and Wilcoxon rank sum test was used to compare different groups (Figure 7).

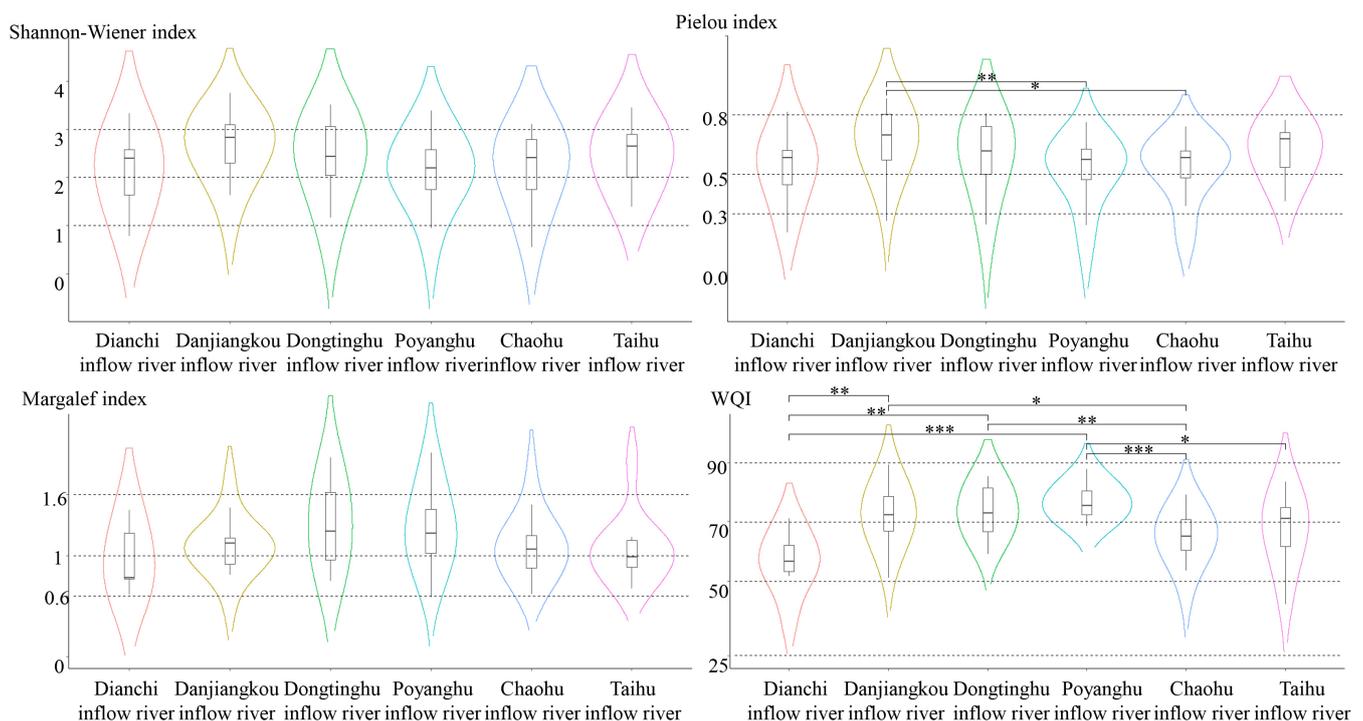


Figure 7. The Shannon–Wiener index, Pielou index, Margalef index and WQI of each inflow river. The dotted line indicates the assessment standard of each index. Wilcoxon rank sum test was used to compare different groups; *** indicates $p < 0.001$, ** indicates $p < 0.01$, * indicates $p < 0.05$.

The Shannon–Wiener index is a species diversity index emphasizing both richness and evenness [32], the Margalef’s index is a species diversity index emphasizing species richness [33], but there is no significant difference between any pairings of inflow rivers. The Pielou index is a species diversity index emphasizing evenness [34]; the periphytic algae community in the Danjiangkou inflow river had significantly higher evenness than the Poyanghu inflow river and the Chaohu inflow river.

The WQI for all inflow rivers ranges from 37.5 to 95, with an average WQI of 74.2 across all sites, indicating that the water quality of all inflow rivers was “good” (Table 2). The WQI in the Poyanghu inflow river is significantly greater than the WQI in the Di-anchi inflow river, the Chaohu inflow river, and the Taihu inflow river, according to the Wilcoxon rank sum test. The Dongtinghu and Danjiangkou inflow rivers have a higher WQI than the Dianchi and Chaohu inflow rivers. In conclusion, the water quality of the Dongtinghu inflow river and the Poyanghu inflow river is the best, followed by the Danjiangkou inflow river.

Figure 8 displays the random forest algorithm-based ecosystem health evaluation of each inflow river. In all inflow rivers except the Dianchi, more than 50% of sampling locations are in “good” condition. The ecosystem health of different inflow rivers was compared using the percentage of sampling sites with an assessment result better than “average” (i.e., “good” and “excellent”). The Dianchi inflow river, the Danjiangkou inflow river, the Dongtinghu inflow river, the Poyanghu inflow river, the Chaohu inflow river, and the Taihu inflow river all had excellent and good rates of 57%, 81%, 74%, 63%, 69% and 73%, respectively. So, the ecosystem health assessments from good to poor were: the Danjiangkou inflow river, the Dongtinghu inflow river, the Taihu inflow river, the Chaohu inflow river, the Poyanghu inflow river and the Dianchi inflow river.

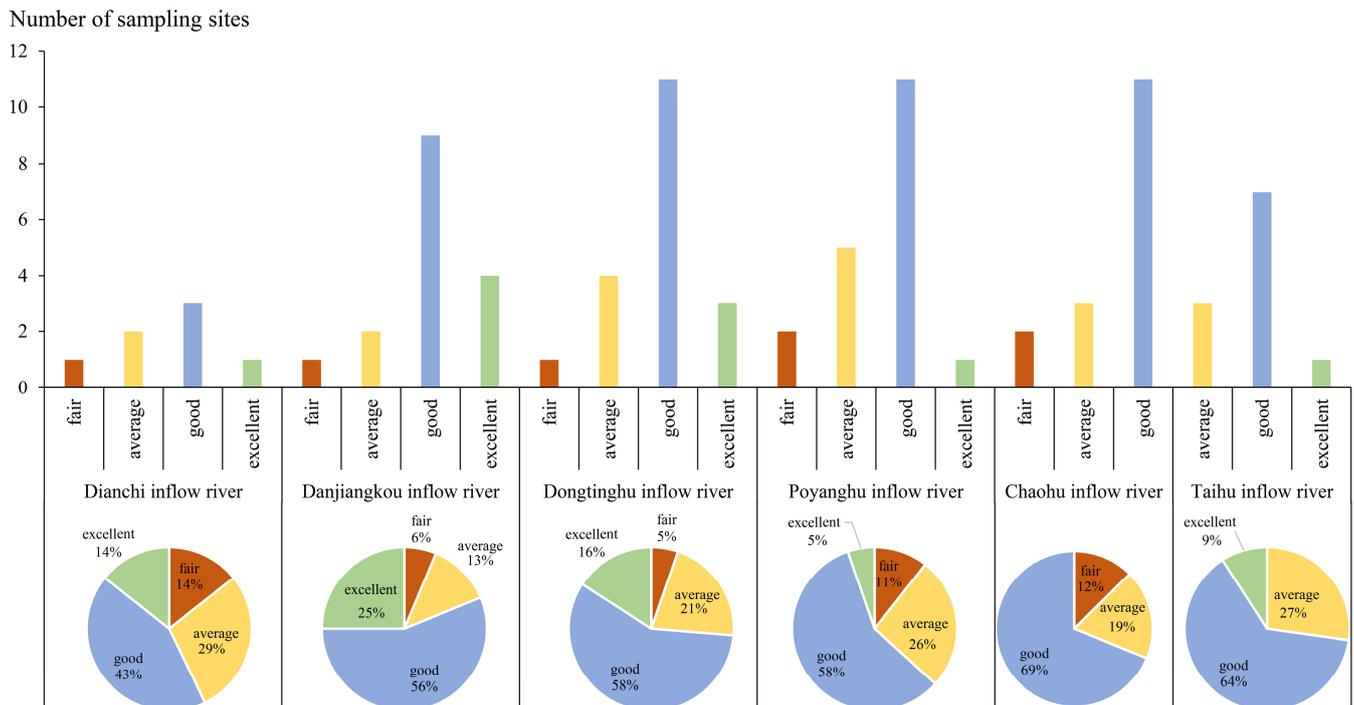


Figure 8. The ecosystem health assessment of each inflow rivers based on random forest algorithm.

4. Discussion

4.1. Spatial Distribution of Periphytic Algae Community Structure

In China, the structure of periphytic algal communities has been extensively researched. The periphytic algae in the Weihe river basin were examined by Liu et al., who discovered that Bacillariophyta is the dominant phylum [35]. Chlorophyta, Bacillariophyta, and Cyanobacteria were the most common periphytic algae in Poyanghu Lake [11]. Bacillariophyta was likewise found to be the most prevalent species in the Danjiangkou Reservoir and its associated water channel [10]. The dominant phyla of this study are Bacillariophyta and Cyanobacteria, which is consistent with prior findings. Bacillariophyta’s dominance is likely due to its adaptation to many environmental conditions and ecological tolerance, whereas Cyanobacteria’s dominance is likely due to the diverse methods used

by this group, such as storing phosphorus, fixing atmospheric nitrogen, absorbing CO₂, and so on [36].

When it refers to cell density, algae's growth and reproduction are inextricably linked to its surrounding nutrition environment. The driving environment variables for all inflow rivers, according to this study, are TN and TP. When we compared the TN concentration, TP concentration, and cell density of each area, we discovered that the Dianchi inflow river and Chaohu inflow river have higher cell density and TP concentration than the other areas, but a higher TN concentration in some areas may have lower cell density (the Danjiangkou inflow river has the second highest TN concentration, but the cell density is ranked fourth). Schindler et al. found that the nitrogen fixation of nitrogen-fixing cyanobacteria was adequate to allow cell density to continue to be generated in proportion to phosphorus, implying that TP contributes more to eutrophication than TN [37]. Here, we speculate that the increased cell density could be attributable to the high TP concentration in the Dianchi and Chaohu input rivers.

4.2. The Relationship between Environmental Variables and Periphytic Algae at Inflow Rivers in Six Lakes

The Dianchi inflow river has the largest density of periphytic algae, with Cyanobacteria and Bacillariophyta dominating the ecosystem. Cyanobacteria similarly dominated the periphytic algae in the Taihu inflow river, but we found no enriched species or environmental drivers in these locations.

The periphytic algae density in the Chaohu inflow river is the second largest; the community is dominated by Cyanobacteria, particularly *Pseudanabaena*, which is frequently reported as cyanobacterial bloom components [38]. Because the Chaohu Lake is frequently threatened by algae blooms [39], and algae distribution is influenced by water flow [40], the *Pseudanabaena* bloom in the Chaohu Lake should be monitored. The periphytic algae community in the Chaohu inflow river was significantly influenced by TN and pH; as TN is one of the most important nutrients for algae growth and reproduction, pH can directly influence the growth rate of algae [41].

Cyanobacteria, particularly *Lyngbya*, dominated the periphytic algae in the Poyanghu inflow river, which are found in a wide range of habitats and geographies [42]. Because water temperature is the most important physical driving element for Cyanobacteria species growth [43], and dissolved oxygen level is predominantly dependent on algal photosynthetic oxygen generation and respiration [44], the community structure is highly related to Temp and DO.

Periphytic algae in the Danjiangkou inflow river was dominated by Cyanobacteria, Bacillariophyta and Chlorophyta; the enriched species in the genus level are *Navicula* and *Achnanthydium*, which are both cosmopolitan in rivers [10]. TUB, TP and COD_{Mn} are the most driving environmental factors in the Danjiangkou inflow river. TUB is known to restrict algal growth by influencing light access [45]. Not only is TP an essential nutrient for algal growth and reproduction, but it also has a strong interaction with TUB [46]. COD_{Mn} is used to assess organic pollution in surface water; the organic matter was probably mineralized by microorganisms, providing nutrients to periphytic algae [9].

Cyanobacteria, Bacillariophyta, and Chlorophyta dominated periphytic algae in the Dongtinghu inflow river, and *Gloeocystis* was the enriched species, which commonly appears as the dominant species in oligotrophic water bodies during summer [19]. pH is a crucial environmental variable that influences the photosynthetic rate of periphytic algae. It affects protoplasmic ion balance and the solubility of numerous chemicals, including inorganic carbon [41].

4.3. The Periphytic Algae Indicator and the Threshold of TN and TP at Inflow Rivers in Six Lakes

This study found that the TN and TP significantly influenced the periphytic algal community composition in all inflow rivers (Figure 5). The community structure, biomass, and elemental composition of river periphytic algae can be affected by TN and TP con-

centrations [47]. The lowest optimum concentration of TN and TP for periphytic algae development were found to be 1.835 mg/L and 19.5 µg/L, respectively, in this study.

Low nutrition levels can result in low periphytic algae density when the TN concentration is less than 1.835 mg/L or the TP concentration is less than 19.5 µg/L. When TN and TP concentrations are above this threshold, higher TN and TP levels can result in increased periphytic algal density. The TN and TP thresholds of algae were 0.03 mg/L and 0.5 mg/L, respectively, according to Dodds et al. and Stevenson et al. [48,49]. Chambers et al. found that the TN and TP thresholds in streams were 0.21~1.20 mg/L and 0.01~0.03 mg/L, respectively. Different TN and TP thresholds in varied studies could be attributable to different geographical factors such as climate, land use types, and other physical and chemical elements [48,50].

4.4. The Ecosystem Health Assessment at Inflow Rivers in Six Lakes

This study assessed the biotic health of each inflow river in three aspects, the biodiversity (Shannon–Wiener index), evenness (Pielou index) and richness (Margalef index). Except for the periphytic algae community in the Danjiangkou inflow river, which showed significantly higher evenness than the Poyanghu inflow river and the Chaohu inflow river, no significant differences were found in other areas, indicating that most sites' biotic assessments range from "average" to "good." Abiotic examination revealed a more substantial difference (Figure 7). More significant difference was found in abiotic assessment. The water quality index (WQI) in the inflow rivers of Dongtinghu and Poyanghu is better than in other areas; these two lakes are naturally connected to the middle Yangtze river, so water and mass exchange between rivers and lakes are key processes that may keep the inflow rivers of Dongtinghu and Poyanghu healthy [14]. The water quality in the Danjiangkou inflow river is second only to the Dongtinghu inflow river and the Poyanghu inflow river, with most sites in these three areas rating "good" on the WQI. The Danjiangkou Reservoir, which delivers water to cities along the middle route of the South-to-North Water Transfer Project (the world's largest inter-regional water transfer project), is critical for maintaining the reservoir's water quality [51]. Dianchi Lake, Chaohu Lake, and Taihu Lake frequently experience harmful algal blooms, which are linked to elevated TP concentrations in these waterbodies [52]. The WQI showed low water quality in the inflow river of Dianchi Lake, Chaohu Lake, and Taihu Lake, and the evaluation result of most locations was "average," as in the previous study.

Our research uses the Random Forest method to measure ecosystem health using biotic and abiotic indexes. This technique may consider a variety of variables while avoiding overfitting, and it has been used to predict and evaluate unknown components that were difficult to forecast using earlier statistical methods [53]. The results showed that the majority of sites in inflow rivers were in "good" condition, with the Danjiangkou inflow river (which has poorer water quality than the Dongtinghu inflow river or the Poyanghu inflow river) having the highest excellent and good rate and the Poyanghu inflow river (which has the best water quality of all inflow rivers of the six lakes) having the second last excellent and good rate, which differs from the biotic or abiotic assessment result. The measurement of water quality and ecological health are both critical components of the water ecosystem health assessment. Aquatic organisms recycle substances and transfer energy through the water environment. Changes in the organization of aquatic organism communities are directly linked to changes in water quality indices. As a result, we can only undertake a fuller health evaluation of the aquatic environment if we combine the water quality and ecological assessments [54,55].

5. Conclusions

- (1) Periphytic algae from eight phyla and 126 taxa were discovered in the inflow rivers of six lakes in the Yangtze river basin, with Cyanobacteria and Bacillariophyta dominating. The driving environmental elements influencing the periphytic algae community structure are TN and TP. The lowest optimum TN concentration for periphytic algae

was found to be 1.835 mg/L, with *Chlorococcum* being the negative responding species and *Cyclotella* being the positive responding species. The lowest optimum TP concentration for periphytic algae is 19.5 µg/L; negative responding species include *Amphora* and *Achnanthydium*, while positive responding species include *Cryptomonas*.

- (2) For the periphytic algae density in the inflow river of six lakes, the median from high to low were the Dianchi inflow river, the Chaohu inflow river, the Poyanghu inflow river, the Danjiangkou inflow river, the Dongtinghu inflow river and the Taihu inflow river. In the Dianchi and Taihu inflow rivers, no driving environmental conditions or enriched species were discovered. *Pseudanabaena* was abundant in the Chaohu inflow river, and TN and pH were the primary environmental drivers. The Poyanghu inflow river was enriched in *Lyngbya*, and temperature and DO were the main driving environmental factors. The *Navicula* and *Achnanthydium* enriched the Danjiangkou inflow river, and the primary driving environmental elements were TP, TUB, and COD_{Mn}. The Dongtinghu inflow river was enriched in *Gloeocystis*, and the key driving environmental factor was pH.
- (3) In terms of biotic evaluation, the Danjiangkou inflow river showed significant higher evenness than the Poyanghu inflow river and the Chaohu inflow river; however, no significant differences were found in other areas. For abiotic assessment, the Dongtinghu inflow river and the Poyanghu inflow river have the best water quality, followed by the Danjiangkou inflow river, while the water quality in other places is relatively poor. Combining the biotic and abiotic assessments by the Random Forest algorithm, the ecosystem health from good to poor were: the Danjiangkou inflow river, the Dongtinghu inflow river, the Taihu inflow river, the Chaohu inflow river, the Poyanghu inflow river, the Dianchi inflow river.

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