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Seasonal variation of phytoplankton in the DaNing River and its relationships with environmental factors after impounding of the Three Gorges Reservoir: A four-year study

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Abstract

The aim of this study was to understand the seasonal succession of phytoplankton composition and the mechanism of algae bloom formation in the DaNing River after impounding of the Three Gorges Reservoir. A relatively long-term dataset of biotic and abiotic parameters of water quality in the river was subjected to correlation analysis and redundancy analysis (RDA). The results indicate that water temperature and TN/TP (total nitrogen divided by total phosphates) are key regulatory factors for phytoplankton abundance. The results of the correlation analysis and RDA suggest that temperature is a key regulatory factor for phytoplankton community composition and algae-bloom dominance in flood season; TN/TP and TN play governing roles in phytoplankton dynamics during the normal season; and transparency, TP and dissolved oxygen contributed significantly to phytoplankton community composition during the dry season.

Keywords: phytoplankton; algae bloom; redundancy analysis (RDA); DaNing River; Three Gorges Reservoir

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1. Introduction

The Three Gorges Reservoir (TGR) has been a focus of international interest because of its possible effects on the ecosystem. Since the filling of the TGR, algal blooms have been detected in the DaNing River [1, 2]. It is well known that an increase in the population of algae in an aquatic system is the prerequisite for algae blooming [3]. Therefore, it is necessary to obtain in-depth information on the seasonal succession of phytoplankton composition. This information is useful for exploring the formation and developmental mechanisms of algae blooming.

There is still no reasonable explanation of the mechanisms of algae bloom formation. Some studies have investigated the dynamics of the main nutrient input to Xiangxi Bay of the TGR [4] because there are empirical correlations between increasing bloom frequency and magnitude and the nutrient status of the water body [5, 6]. So far, little is known about the key regulatory factors of algae blooms because the available quantitative data on the dynamics of phytoplankton in river-style reservoirs are still inadequate. The aims of this paper are as follows: (1) to study annual and seasonal variations in environmental variables and the phytoplankton community; (2) to investigate relationships between phytoplankton and environmental variables; and (3) to discuss the seasonal key regulatory factors favoring outbreaks of algae blooms.

2. Materials and methods

2.1. Study sites and sampling

The DaNing River is a very important tributary to the TGR as well as a famous scenic spot. Its watershed covers an area of 4,426 km^2 in the central part of the Three Gorges Reservoir area [7]. Algae blooms have been observed in a branch backwater of the DaNing River (latitude 31º04′-31º44′N, longitude 108º44′-110º11′E) since June 2003 [1, 8]. The algae bloom usually takes place in May-October, with substantial variations between areas.

Water samples were collected monthly from 2004 to 2007 at three sites in the DaNing River (Fig.1): S1 (latitude 31^0 05′59.88″ N, longitude 109^0 53′40.08″ E), S2 (latitude 31^0 11′4.02 ″N, longitude 109^0 52′30.6″ E), and S3 (latitude 31^0 16′11.76″N, longitude 109^0 47′28.62″ E). Historically, most algae blooms have been observed at these three sites. All samples were taken from surface water (top 50 cm) according to standard methods [9]. At each sampling site, three parallel samples were collected and mixed thoroughly, and 500 ml of each mixed sample was stored in a sterile polypropylene bottle for transport to the lab as soon as possible. One-liter water samples for determination of phytoplankton were preserved with 1.5% Lugol's iodine and concentrated to 30 ml after sedimentation for 48 h.

2.2. Phytoplankton identification and counting

After mixing, 0.1-ml concentrated phytoplankton samples were counted directly with a standard light microscope (OLYMUS-CX31) at 400x magnification. The limitation of light microscopy for studying phytoplankton is that some cells are only identifiable to the genus level and assigned to a size category. Phytoplankton genera were identified according to Hu et al. [10]. Phytoplankton were grouped into taxonomic categories. The dominant genera were listed individually.

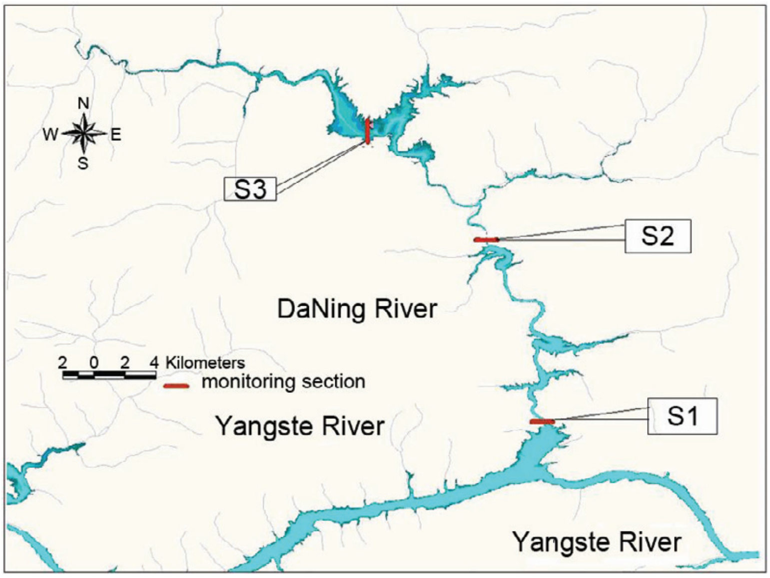

Fig. 1. Sampling sites in the DaNing River

2.3. Environmental Parameters

Environmental parameters included water temperature (Temp), pH, suspended substance (SS), transparency (Trans), dissolved oxygen (DO), pH, total nitrogen (TN), total phosphates (TP) and chlorophyll-a. Temp, Trans, DO, SS and pH were measured in the field, whereas potassium permanganate index (CODMn), TN, TP and chlorophyll-a were determined in the laboratory using State Environmental Protection Administration (SEPA, China) standard methods [9]. Transparency was measured with a Secchi disk. Water temperature, pH, DO and SS were measured using a YSI-6600 hand-held meter (YSI Inc.).

2.4. Data preparation and statistical analysis

Phytoplankton were represented by chlorophyll-a concentration. Chemical parameters included pH and COD_{Mn} as well as nutrients, represented by TP and TN. Physical parameters included Temp, Trans, SS and DO.

Utilizing the comprehensive nutritive index (TLI) method [11], the trophic states of the studied area were assessed. The comprehensive nutritive index (TLI) was calculated using the formula:

$$\mathrm{TLI}(\sum) = \sum_{j=1}^{m} W_i \bullet \mathrm{TLI}(j) \qquad (1)$$

Where TLI(Σ) represents the comprehensive nutritive index, TLI(j) is the nutritive index of variable j, and Wj is the relevant weight of the nutritive index of variable j.

Chlorophyll-a concentrations were used as a proxy for the abundance of phytoplankton [6]. To illuminate the relationships between chlorophyll-a and environmental variables, correlational analysis was performed using SPSS 17.0. Redundancy analysis (RDA) was used to investigate the relationships between phytoplankton genera and the environmental variables. RDA was performed using the software program CANOCO (version 4.15).

3. Results

3.1. Environmental characteristics

According to the hydrological conditions, the 40 months of the study period were divided into three groups, i.e., the flood season (June-October), the dry season (January-April and December), and the normal season (May and November). Seasonal average physicochemical parameters are displayed in Table 1. Fig. 2 shows the time series data of TN, TP, chlorophyll-a and TLI.

In this study, the integrated nutrition state index (TLI) values ranged from 26.24 to 45.74. Most of the studied months were beyond mesotrophic. The degree of eutrophication in flood season was higher than those in the normal and dry seasons, and the degree of eutrophication tended to increase as each season progressed. The results indicate that the degree of eutrophication showed more significant changes after the Three Gorges Reservoir‘s impoundment.

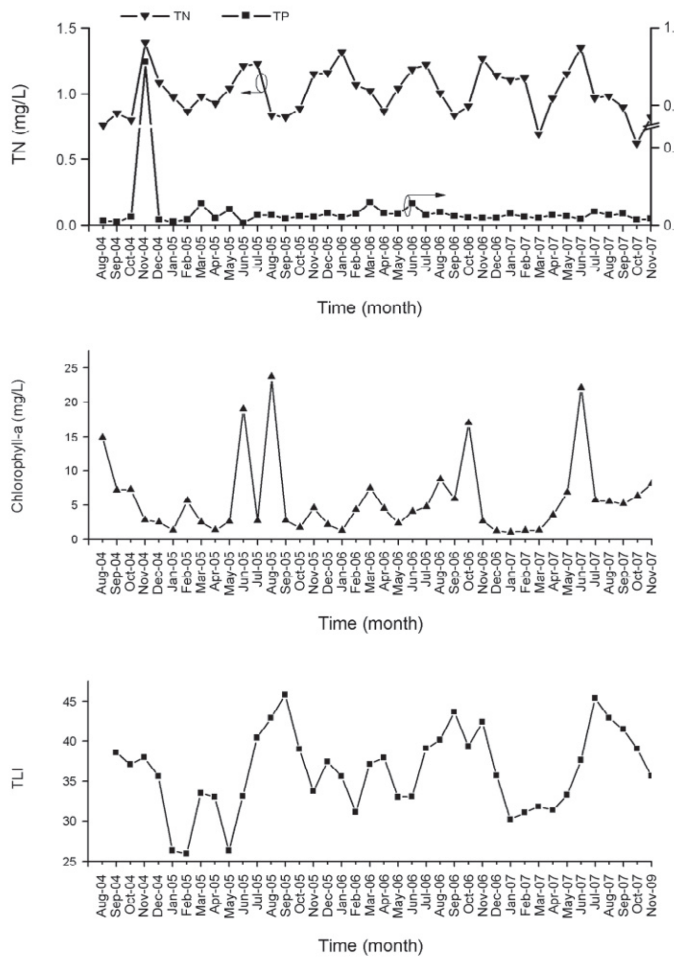

Fig. 2. Time series data of TN, TP, chlorophyll-a and TLI in backwater areas of the DaNing River, 2004-2007.

The values of Temp, COD_{Mn}, Trans and chlorophyll-a in the flood season were significantly different from those in the dry and normal seasons at a significance level of $P<0.05$ (ANOVA, Table 1). These findings indicate that DaNing River exhibits temporal heterogeneity of environment variables. Briefly, Temp and the concentrations of SS, COD_{Mn}, and chlorophyll-a in flood season were higher than those during the dry season, whereas Trans and the concentrations of TP, DO were comparatively lower during the flood season versus the dry season.

TN concentration was higher during the normal season than during the flood and dry seasons, whereas the concentration of TP was higher in the dry season than in the flood and normal seasons. In this study, the TN/TP ratios varied between 17.61 and 242.3. It should be noted that the TN/TP ratios of the water were mostly less than 29 but exceeded 29 to a marked extent between March and May.

The statistical results indicate the presence of an extremely significant positive correlation between Temp and chlorophyll-a concentration ($r=0.451$, $P<0.01$). This demonstrates that water temperature plays an important role in other environmental factors (suspended solids or transparency, nutrients, COD_{Mn}, and so on.). The correlation analysis results indicate that a complex relationship exists between chlorophyll-a and the main soluble nutrient concentrations, as captured by a significant positive correlation between TN/TP and chlorophyll-a concentrations ($r=0.346$, $P<0.05$), and there were no significant correlations among chlorophyll-a concentrations, TN and TP. Correlation analysis between the chlorophyll-a concentrations and environmental factors indicated that during flood season, the chlorophyll-a concentrations have a significant positive correlation with DO ($r=0.551$, $P<0.05$); in the normal season, chlorophyll-a concentrations have a significant positive correlation with pH ($r=0.782$, $P<0.05$).

3.2. Phytoplankton community composition

Damming not only changes the hydrological conditions of rivers but also causes variations in the phytoplankton composition and biomass at the same time [8]. The taxa observed in 2004-2007 included Bacillariophyta, Cyanophyta, Chlorophta, Pyrrophyta, Euglenophyta, Cryptophyta and Chrysophyta. The percentage composition of Bacillariophyta taxa showed a decreasing trend, and the percentage compositions of Cyanophyta and Chlorophta increased notably during 2004-2007 after impounding of the Three Gorges Reservoir (figure 3). Phytoplankton are often used to estimate the impact of large-scale dams on the aquatic ecosystem [12, 13]. Thus, the backwater regions of the DaNing River exemplified the ecological characteristics of the contact zone between riverine and lacustrine systems after impounding of the Three Gorges Reservoir.

Table 1. Seasonal variation in chlorophyll-a and related environmental variables in the DaNing River after impounding of the Three Gorges Reservoir, 2004-2007.

Time	Value	Temp (°C)	pH	SS ($mg•L^{-1}$)	DO ($mg•L^{-1}$)	COD_{Mn} ($mg•L^{-1}$)	TN ($mg•L^{-1}$)	TP ($mg•L^{-1}$)	TN/TP	Trans (m)	Chlorophyll-a ($mg•L^{-1}$)
Flood season	Maximum	35.33	8.7	78	11.75	2.68	1.35	0.06	242.3	2.73	23.74
	Minimum	19.83	7.72	2.47	5.54	1.43	0.62	0.005	21.33	0.8	1.69
	Mean	24.15	8.32	10.77	8.73	2.01	0.96	0.02	57.71	1.33	9.11
	Standard Deviation	3.45	0.2	1.71	1.34	0.32	0.2	0.01	50.71	0.58	6.96
Dry season	Maximum	16.67	8.63	7.23	12.23	1.89	1.32	0.06	114.53	5	7.41
	Minimum	9.83	8.07	1.7	7.47	0.86	0.69	0.01	17.61	2.15	1.03
	Mean	16.74	8.31	7.44	9.26	1.63	0.98	0.03	55.13	2.69	4.57
	Standard Deviation	1.97	0.15	1.46	1.31	0.32	0.15	0.01	25.49	0.75	2.01
Normal season	Maximum	22.5	8.49	4.35	10.69	2.02	1.26	0.04	68.3	4.03	9.3
	Minimum	17.67	8.16	1.25	7.51	1.2	0.82	0.02	25.81	1.55	2.3
	Mean	20.04	8.35	3.05	8.78	1.6	1.05	0.02	48.6	2.7	5.18
	Standard Deviation	1.89	0.14	1.17	1	0.26	0.15	0.01	13.62	0.95	2.89
	*P**	4.25E-13**	0.83	0.17	0.16	4.59E-06**	0.46	0.76	0.79	6.04E-08**	0.04595*

[*] Indicates statistical significance at $p < 0.05$ for all seasons combined.

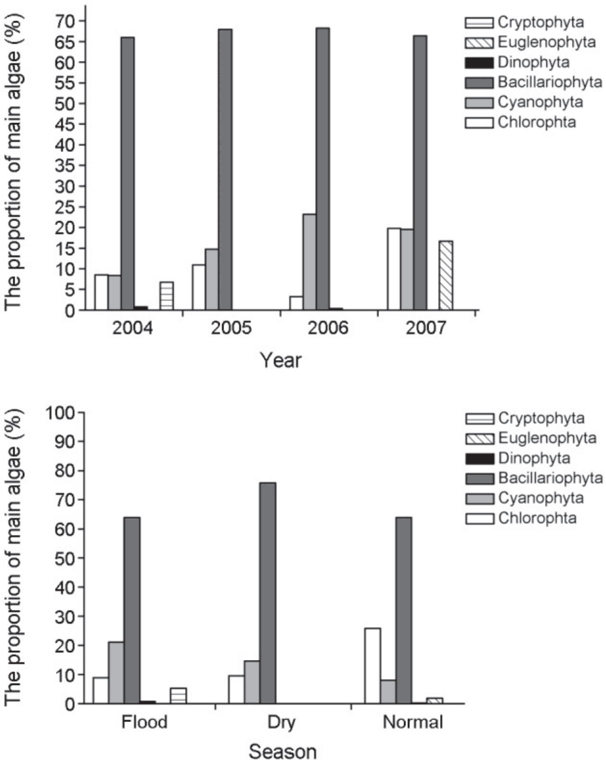

Fig. 3. Yearly and seasonal compositions (%) of algae in backwater areas of the DaNing River after impounding of the Three Gorges Reservoir, 2004-2007

In backwater areas of the DaNing River in the dry season, 30 genera belonging to 3 phyla were recorded. The observed taxa were Bacillariophyta, Cyanophyta, Chlorophta; Euglenophyta and Pyrrophyta were not recorded. Bacillariophyta were common in backwater areas of the DaNing River. In the flood, dry and normal seasons, Bacillariophyta accounted for 63.95%, 75.78% and 63.95% of the total taxa, respectively; thus, the maximum proportion of Bacillariophyta was observed in the dry season. The percentage of Cyanophyta taxa decreased in the dry season compared with the flood season.

In backwater areas of the DaNing River in the normal season, 24 genera belonging to 5 phyla were recorded. The observed taxa were Bacillariophyta, Cyanophyta, Chlorophta, Pyrrophyta and Euglenophyta.

In the flood, dry and normal seasons, Chlorophta accounted for 8.92%, 9.54% and 25.83% of the total taxa, respectively; that is to say, the maximum proportion of Chlorophta was observed in the normal season. In addition, the smallest percentage of Cyanophyta appeared in the normal season.

During the 2004-2007 period, an algal bloom (density > 10^6cells·L^{-1}) first occurred in August 2004 (flood season) at stations S1 and S2, and the bloom-forming algae were *Cyclotella* and *Anabaena*. The maximum density, appearing at station S1, reached 2.51×10^6 cells·L^{-1}. The blooms usually take place in May-October with substantial variations between different areas, except in July.

The algal blooms that occurred in backwater areas of the DaNing River from August 2004 to November 2007 are summarized in Table 2. The bloom-forming algae were *Pandorina*, *Peridinium* and *Chlorella* in May. *Microcystis* and *Peridinium* were the predominant algae genera in June. *Cyclotella* and *Anabaena* were the predominant algae genera in August. *Cyclotella* was the predominant algae genus in September. *Cryptomonas* and *Gymnodinium* were the predominant algae genera in October.

Table 2. Algal blooms occurring in backwater areas of the DaNing River from August 2004 to November 2007.

Time	Station	Total abundance of algae (cells·L^{-1})	Dominant genera
Aug-04	S1, S2	2.38×10^6	*Cyclotella* and *Anabaena*
Sep-04	S2	2.51×10^6	*Cyclotella*
Oct-04	S1	3.11×10^6	*Cryptomonas* and *Gymnodinium*
May-05	S1, S2	2.98×10^6	*Pandorina*, *Peridinium* and *Chlorella*
Jun-05	S2	2.39×10^6	*Microcystis* and *Peridinium*
May-06	S1, S2	1.33×10^6	*Chlorella*, *Peridinium* and *Cyclotella*
Jun-06	S1	2.01×10^6	*Microcystis* and *Peridinium*
Aug-06	S1, S2	1.99×10^6	*Cyclotella* and *Anabaena*
Sep-06	S2	2.33×10^6	*Cyclotella*
Oct-06	S2	2.19×10^6	*Cryptomonas* and *Gymnodinium*
May-07	S1, S2	6.35×10^6	*Cyclotella and Chlorella*
Jun-07	S2	3.49×10^6	*Microcystis* and *Peridinium*
Aug-07	S1, S2	4.39×10^6	*Cyclotella* and *Anabaena*

Oct-07	S2	2.19×10^6	*Cryptomonas* and *Gymnodinium*

3.3. Phytoplankton community composition in relation to environmental variables

Redundancy analysis (RDA) visualizes correlations of taxa with environmental factors and seasons (Fig 4). A total of 97.7% of the cumulative variance of the taxa-environment relationship was represented by the first two axes. Temperature (r=0.2457, P<0.05) and transparency (r=-0.3349, P<0.05) were positively and negatively correlated, respectively, with Cyanophyta in flood season. Thus, high temperature will increase the proportion of Cyanophyta as a fraction of total algal biomass. TN (r=0.4625, P<0.05) was positively correlated with Chlorophta in normal season, indicating that an increasing proportion of Chlorophta was associated with increasing TN concentration. Pyrrophyta was positively correlated with TN (r=0.1467, P<0.05) in the normal and dry seasons. Transparency (r=0.1389, P<0.05), TP (r=0.0917, P<0.05) and DO (r=0.0135, P<0.05) were positively correlated with Bacillariophyta in the dry season. In addition, Bacillariophyta was negatively correlated with Cyanophyta and Chlorophta. In other words, the percentage composition of Bacillariophyta generally decreased, whereas those of Chlorophta and Cyanophyta increased with the change in environmental conditions throughout the study period.

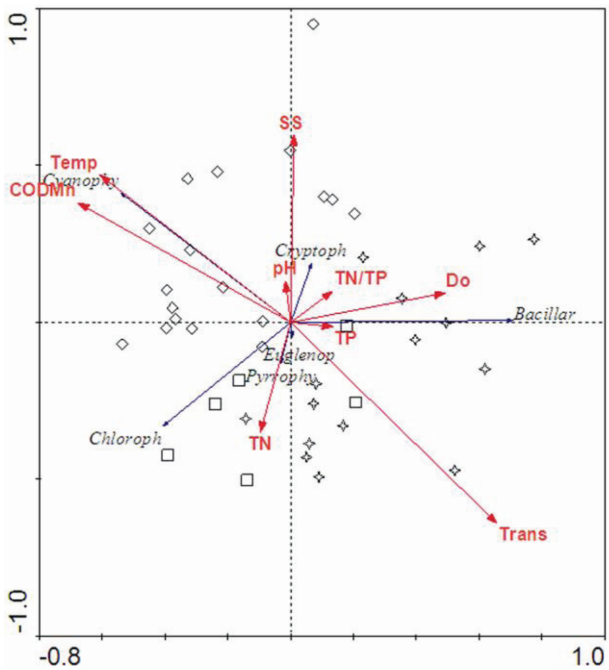

Fig. 4. The biplot of the first two axes of the RDA analysis for environmental factors associated with phytoplankton variation over the study period of 40 months. Stars denote dry season; squares, normal season; and diamonds, flood season.

4. Discussion

Previous researchers have reported that not only the abundance but also the composition of phytoplankton are influenced by temperature [14, 15]. In our experiment, the correlation analysis illustrates that increasing phytoplankton abundance was caused by increasing water temperature in the normal and flood seasons. The results of RDA suggest that the Cyanophyta may have been favored by the increasing temperature and/or decreasing transparency in flood season. Cyanophyta (e.g., *Microcystis*) were characterized by typical taxa that favor warmer temperatures [16, 17]. Transparency is mostly determined by the suspended solids in the water [18]. Phytoplankton blooms and its decomposed particles can cause increase of suspended solids levels, and thus reduce the transparency in flood season. Thus, it is most likely that the algae blooming in flood season (June and August) was strongly related to higher temperature. This explains the increasing Cyanophyta proportion in flood season.

The results of RDA indicate that increasing Chlorophta proportions must be caused by high TN concentration loading in the normal season from pumping of river water. This could partly explain the increasing Chlorophta proportion in the normal season. The bloom-forming algae were *Peridinium* and *Chlorella* in the normal season (May). It is well known that the dominance of Chlorophta is generally associated with higher TN concentrations [19, 20]. Furthermore, because of their high surface to volume ratio, *Chlorella* grow much more efficiently at high TN concentrations [10] In addition, Pyrrophyta were positively correlated with TN in the normal and dry seasons. Thus, the blooming of *Peridinium* and *Chlorella* was caused by high TN loading in May resulting from pumping of river water.

During our survey, TN concentration was higher in the normal season than during the flood and dry seasons, whereas the concentration of TP was higher in the dry season than during the flood and normal seasons. The reasons for this pattern are as follows: (1) nutrients that have accumulated in the surface soil are flushed into the river in the normal and flood seasons; (2) the high river discharge during these wet periods (normal and flood seasons) remobilizes TP nutrients stored in the sediment and causes the release of TP nutrients back into the water column in the dry season [4, 21]. The results of correlation analysis indicate the presence of a complex relationship between chlorophyll-a and the main soluble nutrient concentrations, specifically showing a significant positive correlation between TN/TP and chlorophyll-a concentrations but no significant correlations among chlorophyll-a concentrations, TN and TP. A likely reason for this finding may be that the backwater regions of the DaNing River are deep, and the water column is not well mixed. However, data to explain the contradiction are still scarce. Further efforts to interpret this contradiction will be summarized in a future paper.

In the present study, transparency and concentrations of TP and DO in the dry season were higher than during the flood and normal seasons. The results of RDA suggest that Bacillariophyta preferred higher transparency, TP and DO. This could partly explain the finding that the highest percentage composition of Bacillariophyta appeared in the dry season.

Generally, eutrophication processes not only were driven by environmental factors but also in turn modified the dynamics of physico-chemical variables and biological variables. In so doing, eutrophication acted on the relationships between the variables [22]. This study indicates that chlorophyll-a concentration has a significant positive correlation with DO in flood season. In contrast, some researchers have reported the opposite relationship. A likely reason may be that the oxygen production rate in the course of phytoplankton growth exceeded consumption due to decomposing in flood season. The results of the correlation analysis for the normal season indicate that chlorophyll-a concentrations have a significant negative correlation with pH. Bernard [23] gave an opposite report: phytoplankton growth enhanced the pH in a freshwater system. This can be ascribed to the production of acidic compounds in the course of algae decomposition because the algal blooms occurred in May. Theoretically, transparency is mostly determined by the presence and concentration of suspended solids in the water. However, suspended solids and transparency did not exhibit the expected negative relationship in this study. A likely reason for this finding may be that phytoplankton blooms and their decomposed particles predominated in reducing transparency in the flood and normal seasons.

5. Conclusions

It can be concluded that the increase in degree of eutrophication in the DaNing River was much more remarkable since the TGR impoundment. The DaNing River exhibits temporal heterogeneity of environment variables.

The results regarding succession of phytoplankton composition indicate that the DaNing River exhibits ecological characteristics typical of a contact zone between riverine and lacustrine systems as a result of the TGR's impoundment.

Since erection of the TGR, algal blooms have been observed in the normal and flood seasons. The DaNing River exhibits seasonal heterogeneity of phytoplankton community composition. Temperature and TN/TP played important roles in phytoplankton abundance during the study period. Correlation analysis and RDA revealed the key regulatory factors favoring the outbreak of algae blooms. Temperature is a key regulatory factor for phytoplankton community composition and the dominance of algae blooms in flood season (June and August). TN is a key regulatory factor for the community and the dominance of algae blooms in the normal season. Transparency, TP and DO contributed significantly to phytoplankton community composition in the dry season.

Acknowledgements

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