Environ Geochem Health (2014) 36:595–603
DOI 10.1007/s10653-013-9583-2

ORIGINAL PAPER

Changes in nutrient ratios and phytoplankton community structure caused by hydropower development in the Maotiao River, China

Fushun Wang · Baoli Wang · Cong-Qiang Liu · Xiaolong Liu · Yang Gao · Jing Zhang · Shuang Li

Received: 19 February 2013 / Accepted: 29 October 2013 / Published online: 10 November 2013

Abstract Reservoirs created for hydropower production have become an important feature impacting a river. Understanding the effects of river impoundment on the downstream environment is critical to decision-making for water resource protection. The changes caused by impoundment are changes in water quality and the resulting effect on the phytoplankton community structure. The impacts caused by a series of reservoirs along a river are still not well understood. In this study, we conducted an investigation of five reservoirs along the Maotiao River, China. We found that a series of impoundments plays a role in decreasing the phytoplankton biomass in downstream reservoirs. Within the studied area, nitrogen is not a limiting factor for phytoplankton growth. The ratio of silicon to phosphorus (Si:P) can become a major factor in the regulation of phytoplankton community structure. The Si:P ratio increased from upstream to downstream reservoirs, causing a concurrent increase in the percentage of Bacillariophyta, particularly during the winter. In addition, our results indicate that the creation of dams eliminates Si limitation downstream.

Keywords Nutrient stoichiometry · Phytoplankton · Dam construction · Maotiao river

F. Wang · Y. Gao · J. Zhang · S. Li
School of Environmental and Chemical Engineering, Shanghai University, Shanghai 200444, People's Republic of China
e-mail: fswang@shu.edu.cn

B. Wang (✉) · C.-Q. Liu
State Key Laboratory of Environmental Geochemistry, Institute of Geochemistry, Chinese Academy of Sciences, 46 Guanshui Road, Guiyang 550002, Guizhou Province, People's Republic of China
e-mail: baoliwang@163.com

X. Liu
Tianjin Key Laboratory of Water Resources and Environment, Tianjin Normal University, Tianjin 300387, People's Republic of China

Introduction

During the past several decades, eutrophication has been observed in coastal areas, particularly at the mouths of rivers, and has become an important environmental concern (Billen and Garnier 1997). Excessive anthropogenic input of nitrogen (N) and phosphorus (P) is considered to be the primary cause (Meybeck 1982; Rabalais et al. 1996; Muylaert et al. 2009). As a result of eutrophication, phytoplankton species succession occurs and the variations of N:P:Si (silicon) ratios have been suggested as being an important factor for this shift (Justic et al. 1995a; Sterner and Elser 2002; Lane et al. 2004). In this process, the growth of diatoms is restricted when Si:N and Si:P ratios are less than critical values and subsequently non-diatom plankton species flourish, which include noxious phytoplankton species (Justic et al. 1995b; Humborg et al. 1997, 2000).

As an important component of marine primary productivity, diatoms can be the basis of a healthy marine ecosystem. Nutrient sources from anthropogenic activities are primarily N and P. Silicon, however, seldom comes from anthropogenic sources. The recycling rates of N, P and Si vary greatly (Koszelnik and Tomaszek 2008; Howarth et al. 2011). Diatoms assimilate dissolved silicate in order to build frustules, which have a much lower re-mineralization rate than organic N and P during the decay of an organism (Muylaert et al. 2009). These factors together may intensify the limitation of silicon and induce a shift from diatoms to non-diatom phytoplankton (Conley et al. 2000). At the watershed scale, what happens at the mouth of a river is the result of different geochemical processes. Therefore, it is important to look upstream to identify the mechanism for silicon limitation downstream. The anthropogenic inputs of nutrients and the regulation of river flow within a watershed have been found to be important factors in the changing structure and fluxes of riverine nutrients (Friedl and Wüest 2002).

The construction of a dam has obvious impacts on a river. An artificial reservoir causes changes in hydrology, transparency and nutrient cycling, in comparison with the original river. As a result, reservoir ecosystems shift from river-type heterotrophy to lake-type autotrophy. In addition, an artificial reservoir, and a hydropower reservoir in particular, generally has the characteristics of a deepwater release. A water quality discontinuity will occur as a river crosses a dam, and changes in nutrient quantity and structure may be observed (Kelly 2001), with unknown impacts on the phytoplankton community structure downstream. These changes have not been well documented and are poorly understood. In this study, five reservoirs on the Maotiao River, in southwest China, were investigated with the major objective being to gain an understanding of how nutrient stoichiometry has been influenced by a series of hydropower dams and the effects of these changes on phytoplankton community structure.

Methods

Study area

The Maotiao River is a southern tributary of the Wujiang River, which is an important tributary to the Changjiang River (Fig. 1). This river is located in a karstic region, and the terrain is highly vegetated. The region annually experiences a subtropical rainy monsoon with an average annual rainfall of 1,200 mm and an annual mean temperature of 13.8 °C. The Maotiao River watershed is underlay variously by limestone, dolomitic limestone and shale.

Hongfeng (HF), Baihua (BH), Xiuwen (XW), Zhaixiangkou (ZXK) and Hongyan reservoirs (HY) are closely connected along the Maotiao River (Fig. 1). Currently, Hongfeng and Baihua reservoirs are in a state of eutrophication due to cage aquaculture. In this case, deep water readily becomes anoxic, especially during the warm seasons. The main hydrological features of these reservoirs are well understood (Wang et al. 2011).

Sampling

Water samples were collected monthly from July 2007 to June 2008 for the investigation of water chemistry. The samples were collected seasonally in July and October 2007 and in January and April 2008 for the investigation of phytoplankton community structure. The sampling stations are indicated in Fig. 1. For each reservoir, samples were taken at 0.5 m beneath the water surface from downstream of the dam, tributaries and the central part of the reservoir (0.4–0.5 km upstream the dam). Sites N and O could not be sampled in January due to a blocked access road.

Temperature (*T*), pH and dissolved oxygen (DO) were measured in situ with a portable pH and conductivity meter (YSI-6600v2). Water samples were filtered through a 0.45-μm polycarbonate membrane immediately after sampling and stored in 100-ml plastic bottles on ice in a cooler until analysis. The concentrations of dissolved silicon (DSi) were measured by inductively coupled plasma–optical emission spectrometry (ICP-OES).The concentrations of dissolved phosphorus (DP) and dissolved inorganic nitrogen (DIN), including NO_3^-, NO_2^- and NH_4^+, were measured using an automated flow-injection analyzer (SKALAR Sans Plus Systems). Analytical errors were less than 5 % for the NO_3^- and DSi determinations.

A surface water sample (1.5 l) was preserved with Lugol's solution for the quantitative analysis of phytoplankton. Phytoplankton for qualitative analysis was collected using a 64-μm nylon mesh and preserved with formaldehyde (2 % final concentration). The method of Zhang and Huang (1991) was used for

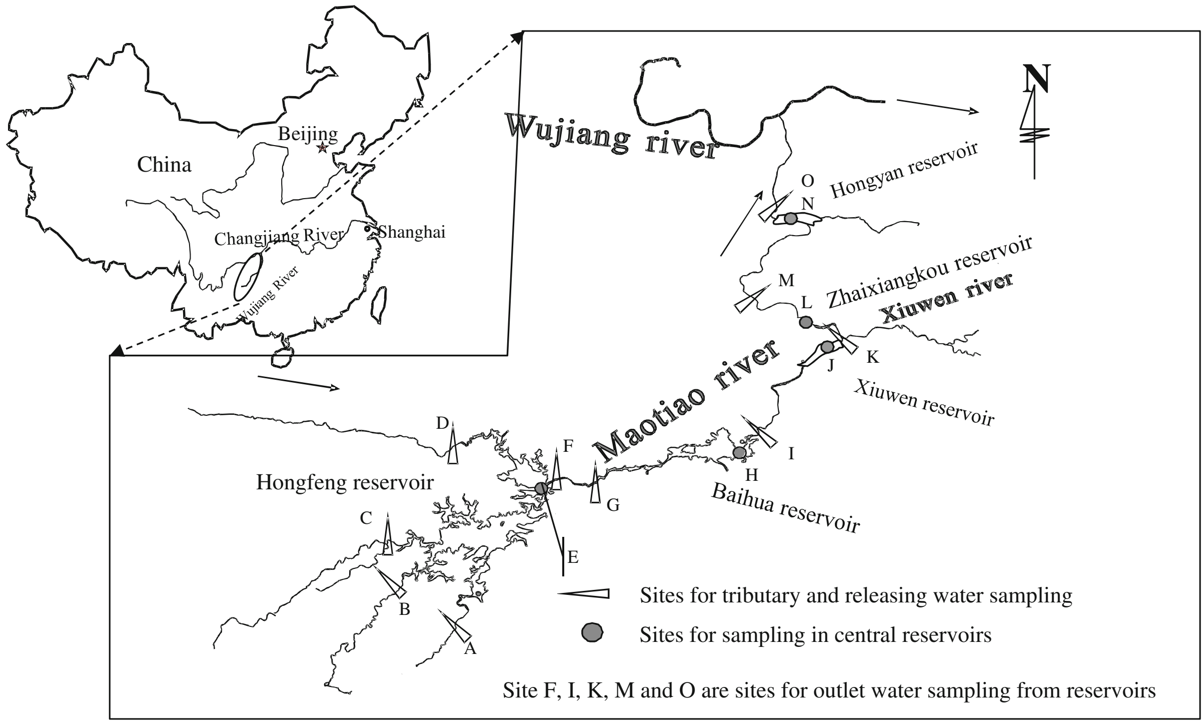

Fig. 1 Map showing sampling locations

taxonomic identification, cell counts and cell dimensions using a standard light microscope. The wet weight (mg/l) of phytoplankton biomass was calculated according to its biovolume and cell density (Zhang and Huang 1991).

Stoichiometirc values used for the assessment of nutrient limitation were Si:P > 22 and DIN:P > 22 for P limitation; DIN:P < 10 and Si:DIN > 1 for N limitation; Si:P < 10 and Si:DIN < 1 for Si limitation; and the concentrations of DSi $= 2$ μM, DIN $= 1$ μM and DP $= 0.1$ μM were used as threshold values (Justic et al. 1995b and the references therein). Pearson's correlation coefficient analyses were conducted using SPSS (version 11.5; SPSS Inc.).

Results

Nutrient limitation along the impounded Maotiao River

The basic parameters for water quality such as T, pH and DO are well understood (Wang et al. 2011). Here, we focused on the nutrient characteristics of the Maotiao River. The molar ratio of N:P varied from 0.7 to 3,200 and that of Si:N from 0.01 to 0.91. All the data from the four seasons fell outside the N-limitation area (Fig. 2a), suggesting that N is not a limiting factor for primary production in the Maotiao River, whereas the P-limitation demonstrated seasonal variation (Fig. 2b). Most samples during the winter and spring fell outside of the area of P limitation. In the winter, the range of the Si:P and N:P was from 0.06 to 172 and from 1 to 2,268, while in the spring season, these values were from 0.07 to 140 and from 0.7 to 3,203, respectively. Conversely, P deficiency in the surface water likely occurred in the summer and autumn. During these seasons, the Si:P and N:P varied from 0.28 to 474 and from 1 to 2,145, respectively. In comparison with P, more samples fell into the area of Si limitation in the winter with the values of the Si:N being from 0.01 to 0.13 and the Si:P being from 0.06 to 34. In fact, in the autumn, the Si availability was superior to that of the other seasons (Fig. 2c). In the spring and autumn, the Si:N and Si:P in the study area ranged from 0.01 to 0.91 and from 0.07 to 279.

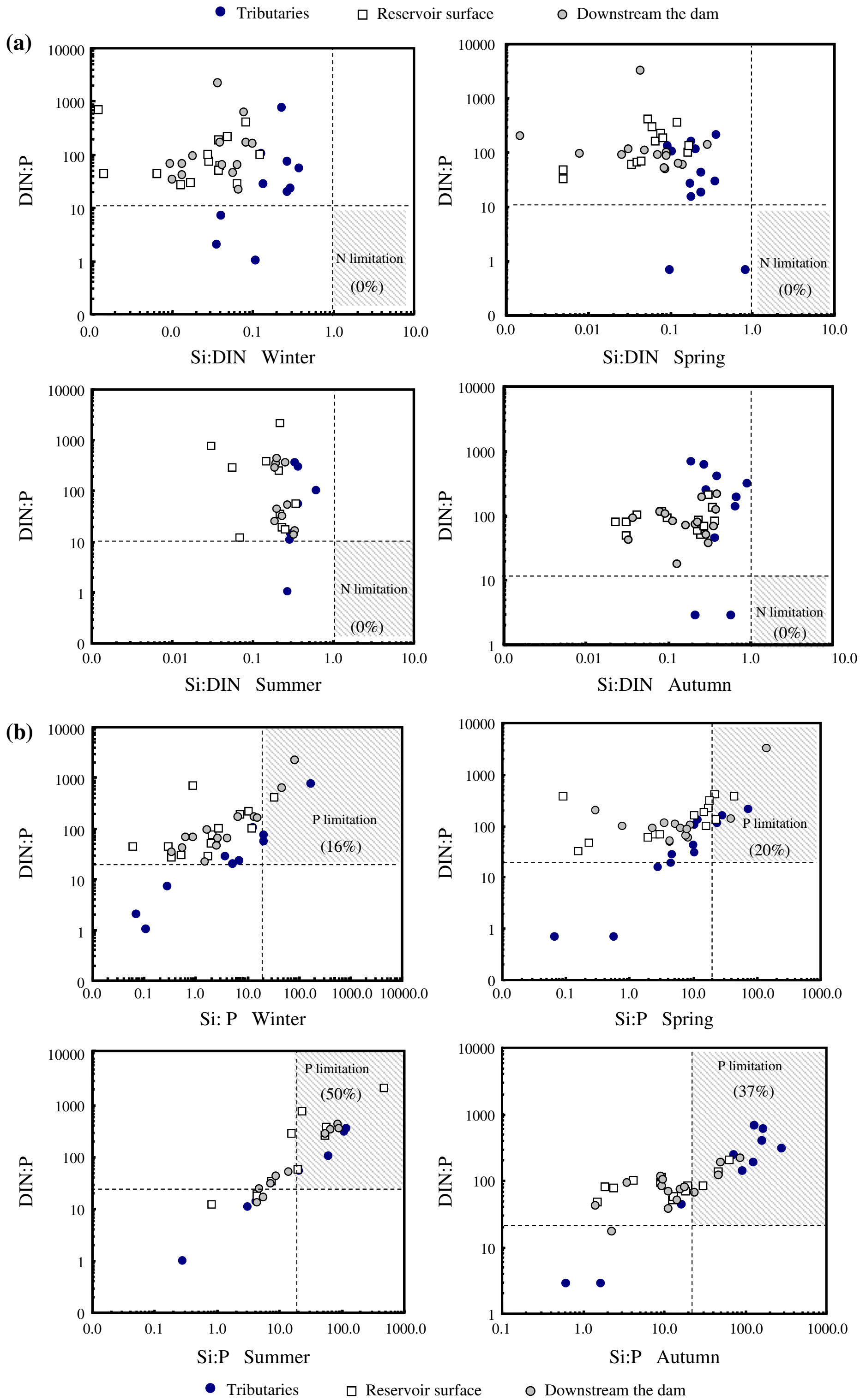

Fig. 2 Scatter diagrams of atomic nutrient ratios in surface waters of a series of reservoirs on the Maotiao River. The percentage of samples falling into the areas of N, P and Si limitation (%) is listed in parenthesis. Winter: from December to the following February; Spring: from March to May; Summer: from June to August and Autumn: from September to November

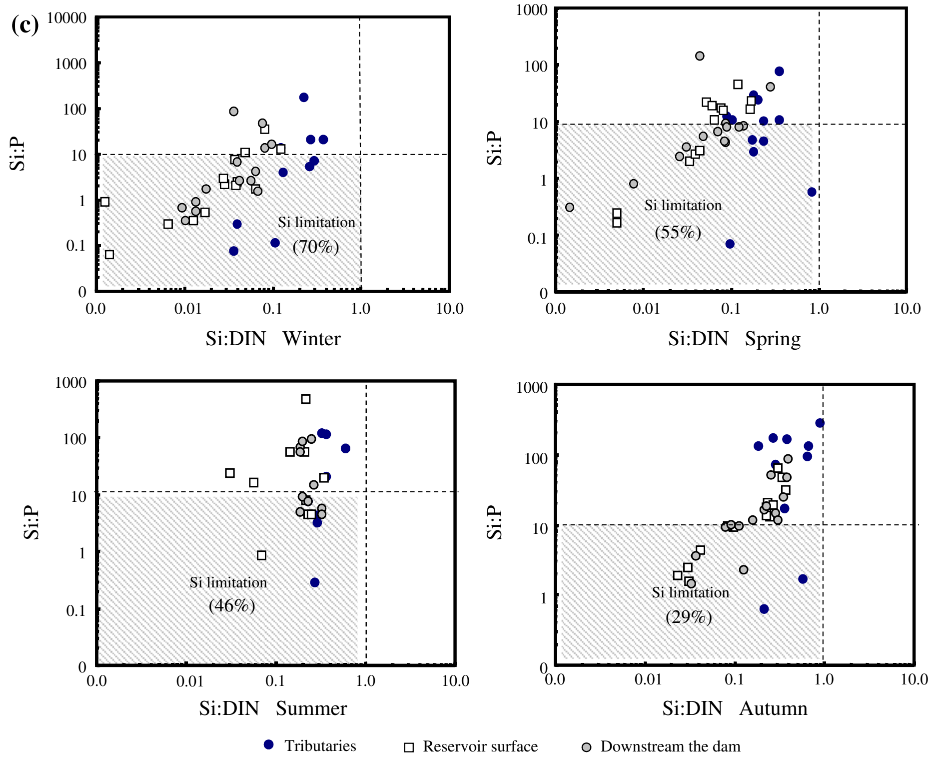

Fig. 2 continued

Longitudinal pattern of nutrient ratios

The changes in Si and P concentrations have had significant influences on the quantity and structure of phytoplankton as N is not a limiting factor to primary productivity in the Maotiao River. Therefore, we focused on the Si:P ratio in this study. In general, Si:P ratios in the major tributaries have shown large variations, ranging from 0.07 to 279 (Fig. 3), suggesting that nutrient concentrations have measurable effects in the inflow from the watershed. After entering the reservoirs, the "pulsating" characteristics of the Si:P ratios was minimized, and Si:P ratios showed a downward trend. For instance, the Si:P has a range of 0.09–4.28 in the upstream reservoir Hongfeng, while these values were between 1.98 and 47 in the downstream reservoir Hongyan in the winter and the spring; however, this trend was less distinct in the summer and the autumn (Fig. 3).

Phytoplankton biomass and community structure

In the Maotiao River, the upstream two reservoirs (Hongfeng and Baihua) received the majority of the nutrients from the watershed. As a result, these two reservoirs were higher in their trophic state. In addition, no important tributaries discharge into the reach between the Baihua and Hongyan reservoirs. The primary productivity in the five reservoirs showed the current situation of nutrient transport along the Maotiao River. As shown in Table 1, Hongfeng reservoir had the highest algal biomass among the studied reservoirs. For example, in April, this value reached 50.3 mg/l. Generally, algal biomass tended to decrease in the downstream reservoirs in April and July. However, this change was not obvious in October and January. In January, these reservoirs had an average biomass of 7. 5 mg/l, and in July, this value was 22.3 mg/l (Table 1).

In these reservoirs, four main algal groups were found. They were Chlorophyta, Bacillariophyta, Cyanophyta and Dinophyta. The phytoplankton community structure showed significant variations within seasons and across space (Table 1). In July, Cyanophyta dominated in the Hongfeng reservoir, accounting for 66 % of the total biomass, while Bacillariophyta only accounted for 2.2 %. However, in the downstream reservoirs, the proportion of Chlorophyta gradually increased,

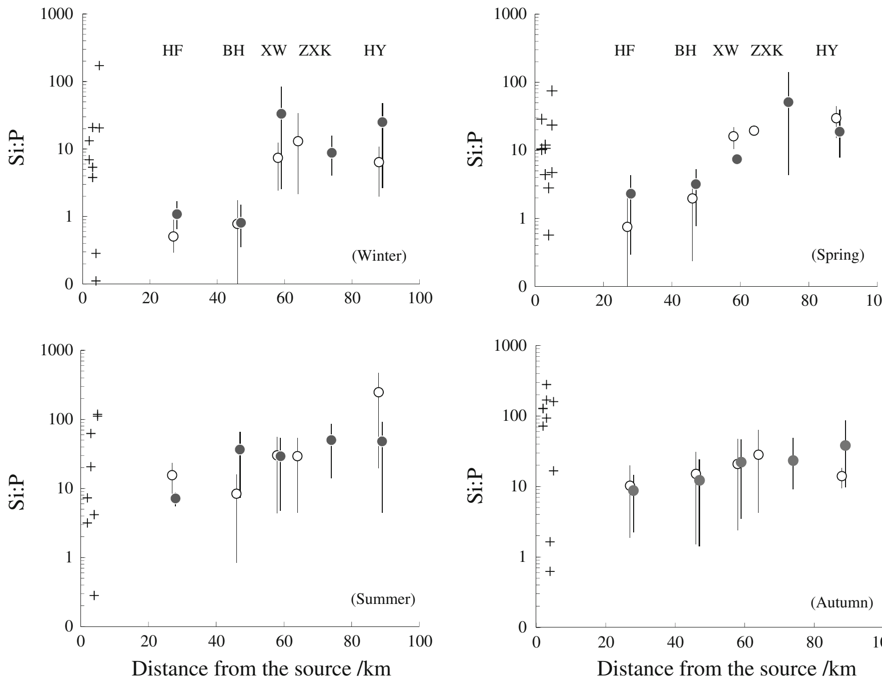

Fig. 3 Longitudinal variation of the ratio between dissolved Si and P. The + refers to the major tributaries; *open circles* represent reservoir samples and the *black circles* represent the downstream samples. The *error bar* represents the maximum and minimum values

corresponding to the decrease in Cyanophyta, and in the fourth reservoir (Zaixiangkou), it reached 94 %. Dinophyta was only abundant in July 2007, and its bloom dramatically increased the biomass in the fifth reservoir (Hongyan). The proportion of Bacillariophyta showed less variation in July. In October, despite total algal biomass having less longitudinal variation, the community structure showed a significant difference and the proportion of Bacillariophyta increased gradually from the upstream to the downstream reservoirs. Cyanophyta was mainly present in the Hongfeng and Baihua reservoirs, and in downstream reservoirs, it was replaced by Chlorophyta. In January, Cyanophyta had a very small contribution, while Bacillariophyta and Chlorophyta were the dominant phyla. Longitudinally, Bacillariophyta gradually became the dominant phylum. In April, the Hongfeng reservoir was dominated by Chlorophyta and showed the highest phytoplankton biomass. During this period, algal biomass decreased rapidly in the downstream reservoirs and the contribution of Bacillariophyta showed a clear increasing trend (Table 1).

Discussion

Generally, a terrestrial ecosystem has the feature of being N-limiting, while a freshwater ecosystem is often in a state of P limitation. In addition, among the fertilizers applied in agricultural production, nitrogenous fertilizer is applied in much larger amounts than phosphorus fertilizer (Yan et al. 1999). This further increases the load of N to a river, due to agricultural non-point discharges. Consequently, freshwater bodies are commonly found having a high N background. The excessive enrichment of nutrients results in the enhancement of primary productivity and leads to eutrophication (Muylaert et al. 2009). This process generally induces harmful algal blooms, causes oxygen deficiency in deep water and results in a breakdown of the aquatic food chain. In addition, the nutrient stoichiometry affects the phytoplankton community structure (Hein and Riemann 1995; Turner et al. 2003a, b). When the concentrations of nutrients reach a threshold value, algae, in principle, need a balanced ratio of nutrients, i.e., Si:N:P = 16:16:1 (Redfield 1958). When the ratio deviates from this value, certain algal groups may dominate (Justic et al. 1995b). Anthropogenic activities have caused the discharge of large amounts of nutrients such as N and P into the river, but riverine dissolved Si is generally derived from the chemical weathering of silicate rock. Consequently, dissolved Si limitation has gradually become a popular topic of study during the past few decades. As the studied area has a karstic geological

Table 1 Seasonal variation of phytoplankton community structure and biomass in a series of reservoirs on the Maotiao River

	HF	BH	XW	ZXK	HY
July 2007					
Cyan (%)	66	23	1	0	1
Chlo (%)	26	33	32	94	45
Baci (%)	2	4	5	4	3
Dino (%)	6	40	60	1	51
Othe (%)	0	0	2	1	0
Total (mg/l)	37.6	11.0	5.5	3.9	53.6
October 2007					
Cyan (%)	9	50	4	1	0
Chlo (%)	80	27	32	49	31
Baci (%)	8	22	62	48	57
Dino (%)	3	1	2	2	4
Othe (%)	0	0	0	0	8
Total (mg/l)	8.3	15.1	4.6	5.6	7.9
January 2008					
Cyan (%)	1	2	0	1	–
Chlo (%)	88	40	16	9	–
Baci (%)	10	44	82	89	–
Dino (%)	0	13	0	0	–
Othe (%)	1	1	2	1	–
Total (mg/l)	15.2	4.3	4.3	6.1	–
April 2008					
Cyan (%)	0	3	0	1	0
Chlo (%)	99	74	68	71	58
Baci (%)	1	20	30	26	38
Dino (%)	0	0	0	0	0
Othe (%)	0	3	2	2	4
Total (mg/l)	50.3	3.9	3.4	1.2	3.4

The total biomass (mg/l) and percentage of each algal group to the total biomass (%) were listed. Reservoirs: Hongfeng = (HF), Baihua = (BH), Xiuwen = (XW), Zhaixiangkou = (ZXK) and Hongyan = (HY)

Cyan Cyanophyta, *Chlo* Chlorophyta, *Baci* Bacillariophyta, *Dino* Dinophyta, *Othe* other phytoplankton and *Total* total phytoplankton biomass. "–" meant no sampling in January in HY

substrate, the dissolved Si concentration is lower than that found in other river draining silicate rock. In addition, among the study reservoirs, the first and second reservoirs (Hongfeng and Baihua) have been polluted by local agriculture, and they have become eutrophic. The low DSi and high P and N concentrations favor the growth of non-siliceous algae. This shift in phytoplankton composition affects the entire food web (Friedl et al. 2004).

Besides the stoichiometric disequilibrium caused by the variation of nutrients, the "reservoir process" can also become an important factor in changing the water chemistry. After impoundment, an autotrophic ecosystem will gradually be formed in a reservoir. That is, primary productivity will be enhanced. A portion of the newly produced organic matter will be recycled in a reservoir, and another portion will be buried in the sediment, depending upon water retention time. In general, a reservoir should act as a sink for riverine nutrients. Furthermore, the production and re-mineralization of algae may affect the ratios of nutrients within the water column. Based on these premises, a particular reservoir's inflows and outflows will have different nutrient ratios, as well as the nutrient concentrations when compared to other reservoirs. As the river passes through the reservoirs, this phenomenon may become more evident.

In the reservoirs on the Maotiao River, the longitudinal increase in the Si:P clearly demonstrates a reservoir's effect on nutrient ratios. Firstly, the reservoirs of this study employ deepwater introduction for power generation. In the first and second reservoirs (HF and BH), a seasonal thermal stratification develops due to the large volume and deep pelagic zone. During the period of stratification, chemical composition shows large vertical variability. In comparison with the epilimnion, the hypolimnion generally has low dissolved oxygen, lower temperatures and higher dissolved Si concentrations (Wang et al. 2010). It is postulated that deepwater discharge from the upstream reservoir will enhance the DSi level and mitigate the Si deficiency downstream. As a response, the abundance of Bacillariophyta of the total phytoplankton (CBTP) gradually increased with the increase in the Si:P ratio (Fig. 4). Secondly, in the first reservoir, blue and green algae were the dominant groups (Table 1). Nutrients were largely assimilated during algal production, and a large portion of the organisms produced settled into the sediments, leading to a net retention of nutrients. Consequently, after the multi-step reservoir processes along the river channel, the fluvial fluxes of nutrients should gradually be reduced. In addition, the utilization efficiency of P by algae is obviously higher than that of Si, which, to some extent, will cause an increase in the Si:P ratio downstream. Thus, the retention of P in a reservoir should be higher than that of Si.

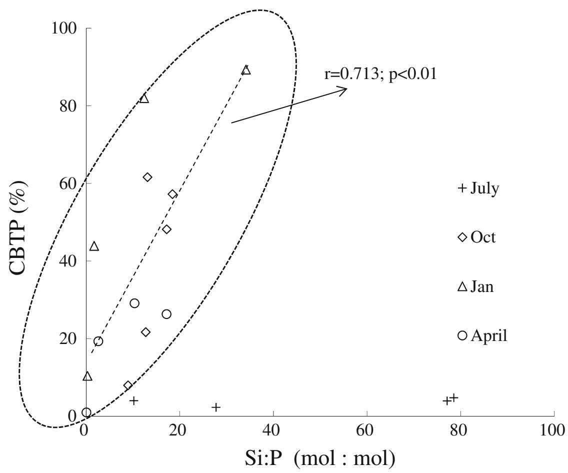

Fig. 4 A scatter diagram comparing the Si:P and the contribution of Bacillariophyta to total phytoplankton (CBTP, %). Pearson's correlation coefficient analyses were conducted for the data obtained on July and October 2007 and April 2008

In general, from upstream to downstream reservoirs, the Si:P ratio increased gradually. Based on the rough criteria for Si limitation (Si:P $\approx$ 10), Si deficiency was more serious in the upstream reservoirs, while in the downstream reservoirs, dissolved Si became less limiting with the increase in the Si:P ratio (Fig. 3). Correspondingly, the abundance of Bacillariophyta gradually increased, associated with a decrease in non-diatom phytoplankton (Table 1). The correlation analysis also showed that CBTP was positively correlated with Si:P ratios except in July (Fig. 4). This demonstrated that the phytoplankton community structure is controlled in large part by nutrient stoichiometry. Consequently, the alleviation of Si limitation favored the production of Bacillariophyta.

Conclusion

Due to the high N background in the river, the variation of P and Si concentrations become a major factor influencing the phytoplankton community structure. Based on the observations on the Maotiao River reservoirs, phytoplankton biomass was quite high in the upstream reservoirs (HF and BH), an indication of high nutrient retention. In these two reservoirs, blue and green algae became the dominant groups and diatoms were replaced, which was closely related to the Si limitation and the long hydrological retention time.

Our study showed that Si limitation generally occurred in upstream reservoirs which can significantly change the stoichiometry of nutrients in the impounded river. This is because that the production and re-mineralization of algae can have different assimilation and release rates, affecting the ratios of nutrients. On the other hand, deepwater discharge from a hydropower reservoir can also serve to mitigate a Si deficiency downstream. Along the series of reservoirs, the ratio of Si to P gradually increased and the CBTP showed an increasing trend. This demonstrated the control of nutrient stoichiometry on the changes in phytoplankton community structure.

Acknowledgments The authors thank Huihui Li, Zhiwei Han, Chipeng Zhang, Yan Yang and Ganrong Li for their careful assistances in field sampling. This research was financially supported by the National Natural Science Foundation of China (Nos. 41273128 and 41021062) and the Shanghai Municipal Education Commission (12YZ017).

References

Billen, G., & Garnier, J. (1997). The Phison River plume: Coastal eutrophication in response to changes in land use and water management in the watershed. *Aquatic Microbial Ecology, 13*, 3–17.

Conley, D. J., Stalnacke, P., Pitkanen, H., & Wilander, A. (2000). The transport and retention of dissolved silicate by rivers in Sweden and Finland. *Limnology and Oceanography, 45*, 1850–1853.

Friedl, G., & Wüest, A. (2002). Disrupting biogeochemical cycles: Consequences of damming. *Aquatic Sciences, 64*, 55–65.

Friedl, G., Teodoru, C., & Wehrli, B. (2004). Is the Iron Gate I reservoir on the Danube River a sink for dissolved silica? *Biogeochemistry, 68*, 21–32.

Hein, M., & Riemann, B. (1995). Nutrient limitation of phytoplankton biomass or growth rate: An experimental approach using marine enclosures. *Journal of Experimental Marine Biology and Ecology, 188*, 167–180.

Howarth, R. W., Billen, G., Chan, F., Conley, D., Doney, S. C., Garnier, J., et al. (2011). Coupled biogeochemical cycles: Eutrophication and hypoxia in coastal marine ecosystems. *Frontiers in Ecology and the Environment, 9*, 18–26.

Humborg, C., Ittekot, V., Cociosu, A., & Bdungen, B. V. (1997). Effect of Danube river dam on black sea biogeochemistry and ecosystem structure. *Nature, 386*, 385–388.

Humborg, C., Conley, D. J., Rahm, L., Wulff, F., Cociasu, A., & Ittekkot, V. (2000). Silicon retention in river basins: Far-reaching effects on biogeochemistry and aquatic food webs in coastal marine environments. *Ambio, 29*, 45–50.

Justic, D., Rabalais, N. N., & Turner, R. E. (1995a). Stoichiometric nutrient balance and origin of coastal eutrophication. *Marine Pollution Bulletin, 30*, 41–46.

Justic, D., Rabalais, N. N., Turner, R. E., & Dortch, Q. (1995b). Changes in nutrient structure of river-dominated coastal waters: Stoichiometric nutrient balance and its consequences. *Estuarine, Coastal and Shelf Science, 40*, 339–356.

Kelly, V. J. (2001). Influence of reservoirs on solute transport: A regional-scale approach. *Hydrological Processes, 15*, 1227–1249.

Koszelnik, P., & Tomaszek, J. A. (2008). Dissolved silica retention and its impact on eutrophication in a complex of mountain reservoirs. *Water, Air, and Soil pollution, 189*, 189–198.

Lane, R. R., Day, J. W., Justic, D., Reyes, E., Marx, B., Day, J. N., et al. (2004). Changes in stoichiometric Si, N and P ratios of Mississippi River water diverted through coastal wetlands to the Gulf of Mexico. *Estuarine, Coastal and Shelf Science, 60*, 1–10.

Meybeck, M. (1982). Carbon, nitrogen, and phosphorus transport by World rivers. *American Journal of Science, 282*, 401–450.

Muylaert, K., Sanchez-Perez, J. M., Teissier, S., Sauvage, S., Dauta, A., & Vervier, P. (2009). Eutrophication and its effect on dissolved Si concentrations in the Garonne River (France). *Journal of Limnology, 68*, 368–374.

Rabalais, N. N., Turner, R. E., Justic, D., Dortch, Q., Wiseman, W. J., & Sen Gupta, B. K. (1996). Nutrient changes in the Mississippi River and system responses on the adjacent continental shelf. *Estuaries, 19*, 386–407.

Redfield, A. C. (1958). The biological control of chemical factors in environment. *American Journal of Science, 46*, 205–221.

Sterner, R. W., & Elser, J. J. (2002). *Ecological stoichiometry: The biology of elements from molecules to the biosphere*. Princeton: Princeton University Press.

Turner, R. E., Rabalais, N. N., Justic, D., & Dortch, Q. (2003a). Global patterns of dissolved N, P and Si in Large Rivers. *Biogeochemistry, 64*, 297–317.

Turner, R. E., Rabalais, N. N., Justic, D., & Dortch, Q. (2003b). Future aquatic nutrient limitations. *Marine Pollution Bulletin, 46*, 1032–1034.

Wang, F., Yu, Y., Liu, C. Q., Wang, B., Wang, Y., Guan, J., et al. (2010). Dissolved silicate retention and transport in cascade reservoirs in Karst area, Southwest China. *Science of the Total Environment, 408*, 1667–1675.

Wang, F., Wang, B., Liu, C. Q., Wang, Y., Guan, J., Liu, X., et al. (2011). Carbon dioxide emission from surface water in cascade reservoirs–river system on the Maotiao River, southwest of China. *Atmospheric Environment, 45*, 3827–3834.

Yan, W., Yin, C., & Zhang, S. (1999). Nutrient budgets and biogeochemistry in an experimental agricultural watershed in Southeastern China. *Biogeochemistry, 45*, 1–19.

Zhang, Z. S., & Huang, X. F. (1991). *Methods in freshwater plankton study [in Chinese]*. Beijing: Science Press.