Bangladesh J. Bot. **46**(3): 1161-1170, 2017 (September) Supplementary

PHYTOPLANKTON ASSEMBLAGES IN TEN RESERVOIRS OF GUANGDONG: RELATIONSHIP BETWEEN MORPHOLOGICALLY BASED FUNCTIONAL GROUPS AND ENVIRONMENTAL FACTORS

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Keywords: Phytoplankton, Morphologically based functional groups, Environmental factors, Reservoir

Abstract

Prinicipal component analysis (PCA) and the canonical correspondence analysis of phytoplankton were carried out in 10 large reservoirs of Guangdong province during the wet period (July to August) of 2009 - 2011. There are 7 morphologically based functional groups (MBFG) in the Baipenzhu and Gongpin reservoirs, 5 MBFG in the Fengshuba reservoirs, and 6 MBFG in the other reservoirs. The results of the PCA show that physico-chemical factors of water accounted for 84.8% of MBFG. Results of canonical correspondence analysis (CCA) show that the Eigen values for axes 1 and 2 accounted for 40.1% of the cumulative variance of the species data, 74.0% of the variance of the MBFG-environment relationships. The results of CCA and Monte Carlo test seem to indicate that chemical oxygen demand, transparency depth, conductivity, ammoniac nitrogen and dissolved oxygen are the most important environmental variables in the modulation of the structure of MBFG. In addition, some reservoirs had a similar biovolume composition of MBFG which with similar or varied main environmental factors. It may indicate that the MBGF composition could be affected not only by main environmental factors but also the synergistic effect of multiple environmental factors.

Introduction

The classification of species based on morphology complemented with genetic studies has helped to understand the complexity of nature (Weithoff 2003). Ecologists have often been dissatisfied with the classification of taxonomic assemblages because they do not always reflect their ecological functions (Solbrig 1994). Phytoplankton ecologists have tried to group together species with similar morphological and physiological traits and similar ecological characteristics to define functional groups (Reynolds 1980, 1988). The phytoplankton was divided into 39 trait-differentiated functional groups (Reynolds *et al.* 2002, Padisák *et al.* 2009, Souza *et al.* 2008, Padisák *et al.* 2006, Borics *et al.* 2007, Callieri *et al.* 2006, Padisák *et al.* 2003). However, information of functional traits obtained following this approach is unavailable for the vast majority of the species (Kruk *et al.* 2010). It was still depending on the knowledge of morphological taxonomy to apply the method.

Recording of selected morphological traits can be used as a tool to better understand the ecology of species and the environmental features of their habitats (Naselli-Flores *et al.* 2007, Reynolds 2006, Weithoff 2003). Therefore, Salmaso *et al.* (2007) and Kruk *et al.* (2010) adopted the characteristics of morphological traits to discriminate phytoplankton into morpho-functional groups (MFG) (Salmaso and Padisák 2007) and morphologically based functional groups (MBFG) systems (Kruk *et al.* 2010), respectively. The MBFG system is based on easily to observe and measure morphological traits which with analyzed more than 700 freshwater species from more than 200 lakes.

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Guangdong province contains 31 large, 282 medium and over 6400 small reservoirs (Han *et al*. 2006). The reservoirs are very important source of water supply for the province. In the present study, present authors describe the phytoplankton communities based on the MBFG classification from 10 large reservoirs which located in tropical and subtropical areas of the region. Their objective is to clarify the role played by environmental variables in modulating the structure of MBFG in these large water reservoirs.

Materials and Methods

The Baipenzhu (BPZ), Dashuiqiao (DSQ), Feilaixia (FLX), Fengshuba (FSB), Gongping (GP), Hedi (HD), Heshui (HS), Nanshui (NS), Tangxi (TX) and Xinfengjiang (XFJ) were 10 large reservoirs (maximal capacity > ×10^8 m^3) in Guangdong province (Fig. 1). They are situated from 110°12′ E-20°21′ N and 116°51′ E-24°47′ N, and are subjected to both tropical and subtropical climatic conditions. The area experiences a heavy monsoon with seasonally contrasting patterns of precipitation, 70 - 85% of which occur in the wet period (from April to September) and 15 - 30% in the dry period (from October to March). The surface water temperature of the reservoirs ranged from 27 - 33.6 and 16.4 - 24.0°C in wet and dry period, respectively (Han *et al*. 2006).

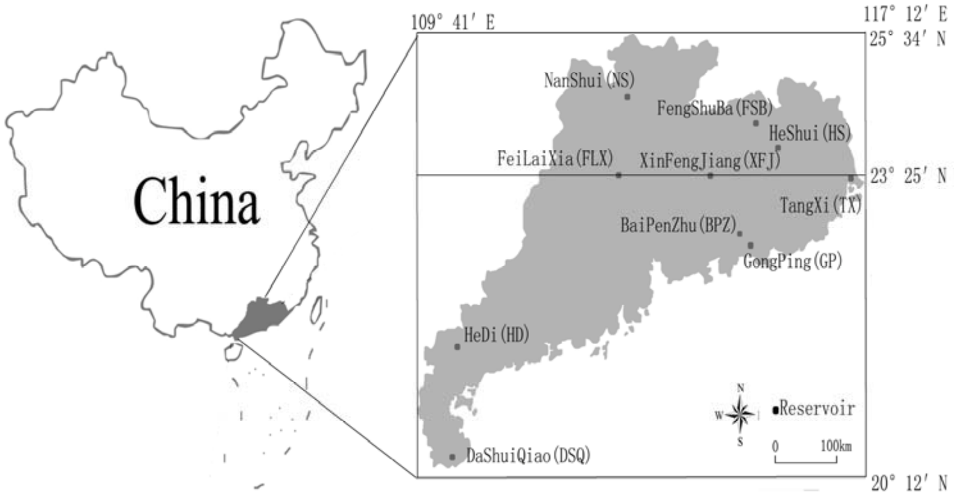

Fig. 1. Location of the ten reservoirs in Guangdong province, China.

Samples were collected with PTN-S200 Plankton net (Beijing Purity Instrument Co., Ltd. and WB-PM Plexiglass water harvesting device (Beijing Purity Instrument Co., Ltd.) at 0.5 m below the water surface at the center and dam of the 10 reservoirs during the wet period (July to August) of 2009 - 2011. Water temperature (Temp.), conductivity (Cond.) and dissolved oxygen (DO) were measured with an YSI multi-parameter meter (Professional Plus, YSI Inc., United States). Transparency depth (SD) was measured at the sites with a Secchi disk and pH with a Sanxin PHB-3 pH meter (Sanxin Instruments, Shanghai, China). Phytoplankton samples were preserved with Lugol's solution at a final concentration of 1% (v/v). The species in the water samples were identified, when possible, based on descriptions (Akiyama *et al.* 1981, Hu and Wei 2006, Wehr and Sheath 2003, John *et al.* 2002), and classified into different MBFG after Kruk *et al.* (2010). Phytoplankton enumeration was done following the methodology of Lund *et al.* (1958). Biovolume was calculated following Olenina *et al.* (2006).

Ammoniac nitrogen (NH_4-N), nitrate nitrogen (NO_3-N), orthophosphate (PO_4-P) and chemical oxygen demand (COD) were measured according to the standard methods of China (SEPAC 2002).

The mean value of each water quality parameter was transformed by log ($x_i + 1$) after Becker *et al.* (2009). PCA and CCA were calculated by CANOCO 4.5 software (Braak and Smilauer 2002) to detect the major environmental variables in the reservoirs and elucidate the relationships between MBFG and their environment (Naselli-Flores 2000). The significance with which environmental variables explain the variance of MBFG data was tested using Monte Carlo simulations with 499 unrestricted permutations by the (CANOCO 4.5 software) Braak and Smilauer 2002, Naselli-Flores 2000). Variables were considered to be significant at $p < 0.05$.

Results and Discussion

Environmental variables of 10 reservoirs were relatively stable during the study periods (Table 1). The results of principal component analysis (PCA) explained 84.8% of the variability in the first two axies (axis 1 = 51.5%; axis 2 = 33.3%). SD (r = –0.77), COD (r = 0.86) and Cond. (r = 0.79) were the most important variables for the axes 1 and 2 were pH (r = 0.80) and NO_3-N (r = 0.75). Based on the results, the reservoirs were divided into 6 types (Fig. 2).

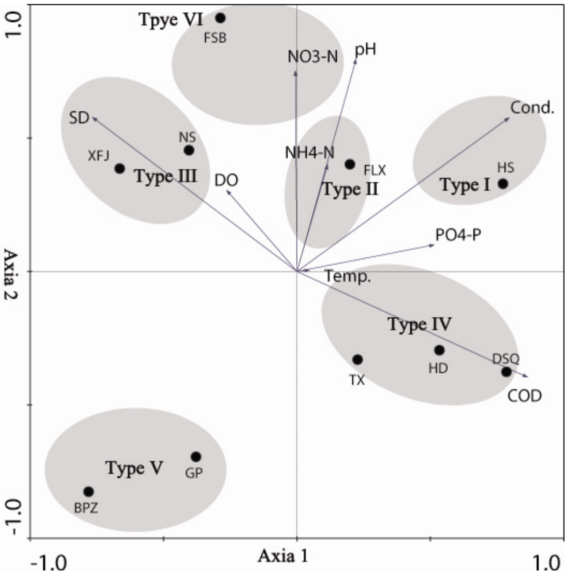

Fig. 2. PCA ordination biplot of samples and environmental variables in reservoirs.

The community of phytoplankton comprised of a total of 121 species belonging to 6 major taxonomic categories. The species of Chlorophyta, Bacillariophyta and Cyanophyta were principal composition which occupied 38.89 - 51.06, 15.94 - 23.40 and 11.76 - 29.63%, respectively. The number of species present in TX , HS, DSQ, BPZ, GP, FSB, HD, NS, XFJ and FLX reservoirs were 83, 82, 81, 75, 75, 71, 68, 64, 58 and 57, respectively. While the phytoplankton biovolume of the reservoirs as mentioned before was 0.78, 1.20, 2.91, 3.85, 5.00, 5.50, 8.34, 8.63, 11.82 and 13.19 mm^3/l, respectively.

Table 1. Main environmental variables of the 10 studied reservoirs.

Reser-voirs	Temp. (°C)	SD (m)	pH	Cond. (µs/cm)	DO (mg/l)	COD (mg/l)	NH_4-N (mg/l)	NO_3-N (mg/l)	PO_4-P (mg/l)
BPZ	30.80 ± 0.36	1.79 ± 1.07	6.43 ± 0.37	34.16 ± 5.70	6.33 ± 1.60	1.56 ± 0.30	0.08 ± 0.06	0.20 ± 0.06	0.005 ± 0.002
DSQ	30.72 ± 1.35	0.75 ± 0.12	7.04 ± 1.43	137.54 ± 10.79	5.89 ± 1.43	4.59 ± 1.47	0.24 ± 0.11	0.10 ± 0.07	0.009 ± 0.004
FLX	29.40 ± 1.91	1.67 ± 0.81	7.54 ± 0.38	123.04 ± 25.70	7.62 ± 1.32	2.20 ± 0.78	0.14 ± 0.04	1.04 ± 0.11	0.007 ± 0.003
FSB	31.90 ± 0.79	3.34 ± 1.46	7.80 ± 0.91	101.53 ± 18.05	7.08 ± 0.51	1.60 ± 0.20	0.58 ± 0.84	2.15 ± 0.69	0.007 ± 0.006
GP	30.53 ± 1.47	1.83 ± 1.28	6.33 ± 0.78	48.87 ± 3.99	5.70 ± 1.31	2.65 ± 0.56	0.17 ± 0.15	0.15 ± 0.09	0.004 ± 0.002
HD	30.77 ± 1.77	0.81 ± 0.17	6.36 ± 2.04	99.98 ± 11.37	6.73 ± 2.12	4.46 ± 1.99	0.32 ± 0.23	0.75 ± 0.56	0.005 ± 0.004
HS	31.02 ± 0.38	1.25 ± 0.22	7.99 ± 0.53	201.28 ± 24.14	5.07 ± 1.00	3.21 ± 1.20	0.10 ± 0.04	0.45 ± 0.10	0.006 ± 0.001
NS	29.45 ± 1.01	4.35 ± 0.79	7.02 ± 0.88	91.29 ± 5.62	6.67 ± 1.40	2.02 ± 0.59	0.22 ± 0.11	0.54 ± 0.03	0.006 ± 0.003
TX	31.25 ± 2.06	1.22 ± 0.55	7.34 ± 1.24	83.58 ± 8.43	7.10 ± 2.42	3.69 ± 0.34	0.09 ± 0.03	0.53 ± 0.19	0.009 ± 0.003
XFJ	30.91 ± 0.28	4.49 ± 0.22	7.60 ± 0.46	79.04 ± 10.06	6.55 ± 1.39	1.22 ± 0.08	0.04±0.01	0.37±0.06	0.004 ± 0.002

The dominant (relative biovolume > 30%) or co-dominant (relative biovolume 10 - 30%) species were *Ankistrodesmus aciculars* (A. Braun) Korschikoff (11.82% in HD), *Ceratium hirundinella* (Müll.) Schr. (37.18 and 33.81% in BPZ and FSB), *Chlorella* spp. (25.52% in NS), *Closterium venus* Kützing (20.90% in GP), *Cosmarium* spp. (12.36, 18.48, 26.16 and 19.98% in DSQ, TX, GP and XFJ, respectively), *Cryptomonas* spp. (14.63% in FLX), *Cyclotella meneghiniana* Kützing (14.78, 23.26 and 30.32% in FLX, NS and XFJ, respectively), *Glenodinium pulvisculus* (Ehr.) Stein (34.93 and 10.67% in HS and FLX), *Planktosphaeria gelatinosa* G. M. Smith (17.82% in BPZ), *Jaaginema angustissimum* (W. *et* G. S. West) Anag. *et* Kom (14.26, 38.25 and 25.36% in HS, HD and DSQ, respectively), *Melosira granulata* (Ehr.) Ralfs (10.98, 17.04 and 10.69% in FLX, NS and GP, respectively), *Raphidiopsis curvata* Fritsch *et* Rich (19.31% in HD), *Staurastrum* spp. (14.05, 17.63 and 23.10% in DSQ, TX and GP, respectively), and *Synedra acus* Kützing (11.88% in XFJ).

The phytoplankton could be divided into 7 MBFG in the BPZ and GP reservoirs, 5 MBFG in FSB (absence of Groups II and III), and 6 MBFG in the others (absence of Group II). However, the biovolume of Group III was very lower in GP, FLX, XFJ, HS and NS reservoirs (Fig. 3). Some reservoirs had similar dominant and/or co-dominant MBFG constituents, such as NS and XFJ (Groups IV and VI), GP and TX (Group IV), HS, BPZ and FSB (Group V), DSQ and HD (Group I).

The results of CCA (canonical correspondence analysis) showed that the Eigenvalues for axes 1 and 2 was 0.181 and 0.155, respectively. The cumulative variance of the MBFG was 40.1%. The MBFG-environment correlations of axes 1 and 2 were 0.845 and 0.854, respectively. The cumulative variance of MBFG-environment relationships was 74.0%.

The analysis confirms that the importance of COD, SD, Cond. (r with axis 1 was 0.69, –0.52, 0.33, respectively), NH_4-N and DO (r with axis 2 was 0.53 and –0.60) explained the variance on the MBFG data. The Monte Carlo test shows that COD, NH_4-N and Cond. were significant ($p < 0.01$) with axes 1 or 2 (Fig. 4). The analysis indicates that these variables provided a finer representation of the major controlling environmental gradients of the MBFG in the reservoirs. The Group I exhibited a negative relationship with DO and SD, and a positive relationship with Cond., PO_4-P and NH_4-N. Group II exhibited a negative relationship with COD and DO, and a positive relationship with SD. Group III exhibited a positive relationship with COD and DO. Group IV exhibited a negative relationship with NH_4-N and Cond., and a positive relationship with SD. Group V exhibited a negative relationship with SD. Groups VI and VII exhibited a negative relationship with NH_4-N, Cond. and COD, and a positive relationship with SD (Fig. 4).

Group I was composed of small organisms with high surface/volume (S/V) ratio which lacks siliceous exoskeleton, aerotopes or the presence of mucilage (Kruk *et al*. 2010). These organisms were well adapted to rapid resource acquisition and present a higher rate of population growth (Reynolds 2006). Therefore, Group I was present in all reservoirs. The dominant species of Group I was *J. angustissimum* in DSQ and HD reservoirs which with lowest SD. The results of CCA showed that biovolume and SD had a negative relationship (Fig. 4). It was indicated that some species of *Jaaginema* were adapted to low light and to survive in turbid mixed environments as in the lakes (Padisák *et al*. 2009, Reynolds *et al*. 2002). The results also showed that Group I exhibited a positive relationship with NH_4-N, PO_4-P and Cond. (Fig. 4). The observation agreed with a previous report indicating that organisms represented in Group I were more suitable to survive in water with higher concentration of dissolved nutrients (Kurk *et al*. 2010).

Group II was composed of small flagellated organisms with siliceous exoskeletal structures, such as the members of the class Chrysophyceae (Kruk *et al*. 2010). The Group II had a very low biovolume as in the natural lakes (Pacheco *et al*. 2010, Kruk *et al*. 2010). It was only present in the

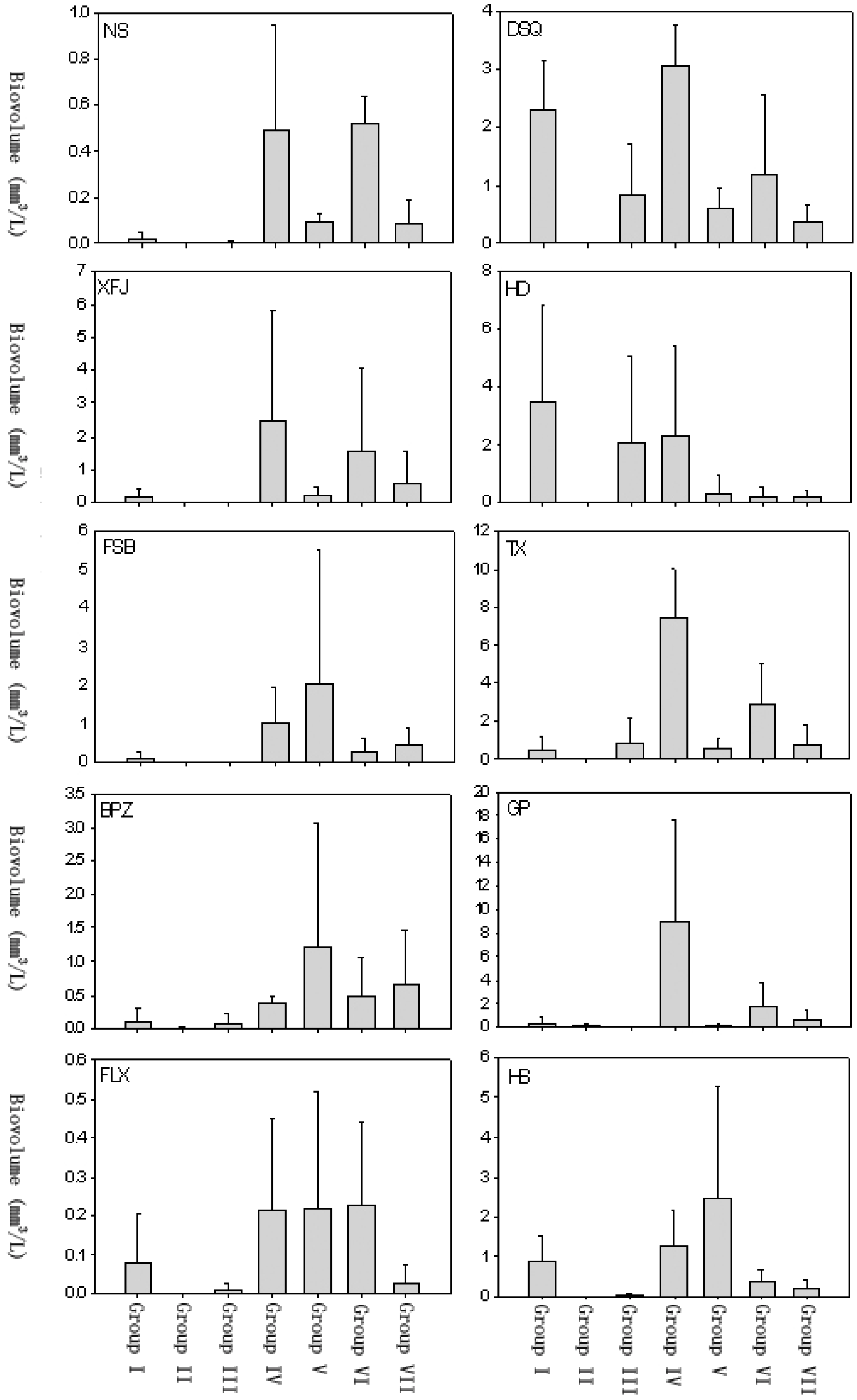

Fig. 3. The biovolume composition of MBFG in 10 reservoirs.

GP and BPZ reservoirs (Fig. 3) which with lower nutrient levels (Table 1). The results of CCA showed that the biovolume of Group II had little or unclear relationship with the NH_4-N and PO_4-P content (Fig. 4). It was indicated that these organisms were highly tolerant to low nutrient levels, because they were facultative mixotrophs (Kruk *et al.* 2010, Pacheco *et al.* 2010) and could produce resistance propagates (Sandgren 1988). However, these organisms was absent in the XFJ reservoir which had a lower concentration of nutrients (Table 1). It may be due to the different of main environmental variables in these reservoirs (Fig. 2). The results also showed that the Group II had a negative relationship with DO (Fig. 4). It may indicate that the higher DO could inhibit the growth of these organisms.

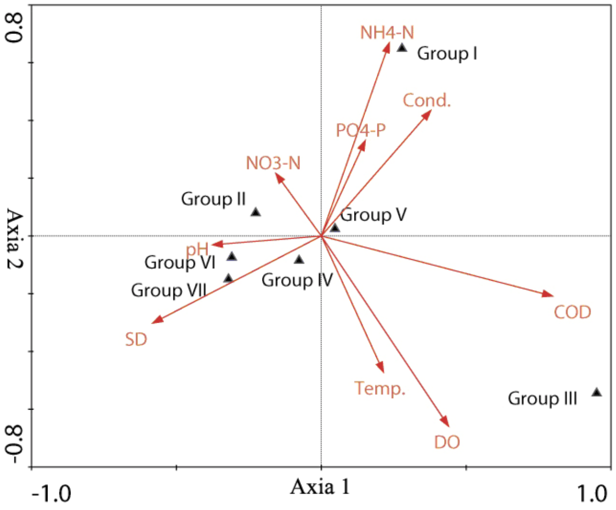

Fig. 4. CCA ordination biplot for biovolume of MBFG and environmental variables.

Group III was composed of organisms with large filaments with aerotopes (Kruk *et al.* 2010). These organisms had a high S/V ratio, presenting a greater tolerance to light deficient conditions (Naselli-Flores and Barone 2007). Interestingly, some members of the group had the capacity to fix nitrogen, showing a potential tolerance to nitrogen deficiency (Kruk *et al.* 2010). These organisms appeared in most of the reservoirs (Fig. 3). It was observed that *R. curvata* was the dominant species in the HD reservoir. This species was adapted to survive in turbid warm mixed environments with tolerance to low light and nitrogen (Kruk *et al.* 2010). The results of CCA exhibited Group III had a positive relationship with temp., DO and COD (Fig. 4), indicating the importance of oxygen for these organisms.

Group IV was composed of medium size organisms which lacked specialized traits such as flagella, siliceous structures or mucilage (Kruk *et al.* 2010). These organisms had potentially moderate tolerances to limiting resources and low to moderate sinking rates (Kruk *et al.* 2010). It has been observed that these organisms were particularly abundant in the DSQ, GP, TX, NS and XFJ reservoirs (Fig. 3). The dominant species in these reservoirs were *A. aciculars*, *Cl. venus*, *Cosmarium* spp. and *Staurastrum* spp. Those species were adapted to survive in oligo-hypertrophic environments, with tolerance to nutrient deficiency (e.g. *Cosmarium*, *Staurastrum*), mild light and the deficiency of a carbon source (e.g. *Staurastrum*, *Ankistrodesmus*) (Reynolds *et al.* 2002, Padisák *et al.* 2009, Soares *et al.* 2007). However, some of these species were sensitive to pH rise of (e.g. *Closterium*, *Staurastrum*) and nutrient deficiency (e.g. *Ankistrodesmus*)

(Reynolds *et al*. 2002, Padisák *et al*. 2009, Soares *et al*. 2007). Group III exhibited a negative relationship between biovolume and NH_4-N, PO_4-P and Cond., and a positive relationship with SD (Fig. 4). These results indicated that light was an important factor for this group and that these species might be adapted to environments which were poor in nutrients.

Group V was composed of unicellular flagellated organisms of medium to large size (Kruk *et al*. 2010). These organisms were present in all reservoirs (Fig. 3). These organisms were particularly abundant in the BPZ, FSB and HS reservoirs where *Cer. hirundinella*, *Glenodinium pulvisculus* were the dominant species. In the FLX reservoir, *Cryptomonas* spp. was the most abundant species. The CCA results showed the Group V had a negative relationship with SD, and unclear relationship with NH_4-N or PO_4-P (Fig. 4). It had been reported that those species were tolerant to low light (Padisák *et al*. 2009, Reynolds *et al*. 2002) and to conditions of reducing of nutrients (Graham *et al*. 2009).

Group VI was represented only by algae that belong to the class Bacillariophyceae, non-flagellated organisms with siliceous exoskeletal structures (Kruk *et al*. 2010). These organisms were present in all reservoirs (Fig. 3). These organisms require a source of silica for their exoskeletons (Reynolds 2006, Kruk *et al*. 2010). The dominant species in the reservoirs were *M. granulata* , *Cy. meneghiniana* and *Sy. acus*. These species were adapted to survive at higher trophicity (e.g. *Melosira*, *Cyclotella*), inorganically turbid (e.g. *Synedra*) and highly lotic (e.g. *Melosira*) environments (Reynolds *et al*. 2002, Padisák *et al*. 2006, Borics *et al*. 2007, Padisák *et al*. 2009). However, the results of the CCA indicate that the biovolume of Group VI had a positive relationship with SD and a negative relationship with NH_4-N, PO_4-P and Cond. (Fig. 4). Due to the high cell density and lack of motility, diatoms sink rapidly from the surface of reservoirs with sluggish flow, these results indicated that light was an important factor for this group and that these species may be adapted to environments with lower concentration of dissolved nutrients in the surface water column.

Group VII consists of large mucilaginous colonies which were sensitive to low supply of resources (Kruk *et al.* 2010). These organisms were present in all reservoirs in low abundance (Fig. 2). The representative species was *P. gelatinosa* which was adapted to survive in clear, deeply mixed meso-eutrophic environments (Padisák *et al*. 2009). The results of the CCA showed that the Group VII had a positive relationship with SD and negative relationship with NH_4-N, PO_4-P and Cond. These observations may indicate that light was an important factor for this group, and these were sensitive to lower availability of dissolved nutrients as in the lakes (Kruk *et al*. 2010).

It was unsurprising that similar environmental factors determine a similar biovolume composition of MBFG. For example Groups IV and VI were dominant in the XFJ and NS reservoirs (type III), such as Groups I and IV in the HD and DSQ reservoirs (type IV). However, it was also observed that different environmental factors could determine a similar composition of MBFG. For example, Group IV dominated in the GP (type V) and TX (type IV) reservoirs, and Group V dominated in the FSB (type VI), BPZ (type V) and HS (type I) reservoirs. These results may indicate that the synergistic effect of multiple factors could create a similar composition of MBFG of phytoplankton.

The phytoplankton composition observed in these reservoirs seems to be similar to the composition described for lakes in different parts of the world. Present results also indicated that the morphological based functional group (MBFG) classification for phytoplankton was suitable for studies in water reservoirs in Asia. Future research is needed to determine additional factors that may affect the composition and dynamics of the phytoplankton populations.

Acknowledgements

The authors are grateful to the members of the research team for their assistance in collecting the samples. This research was supported by “Major science and technology projects of water pollution control and governance of China (2008ZX07211-003)”.

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(Manuscript received on 7 April, 2017; revised on 7 August, 2017)