Asynchrony of spring phytoplankton response to temperature driver within a spatial heterogeneity bay of Three-Gorges Reservoir, China

Yaoyang Xu\textsuperscript{a,b}, Qinghua Cai\textsuperscript{a,*}, Lin Ye\textsuperscript{a}, Meiling Shao\textsuperscript{a}

\textsuperscript{a} State Key Laboratory of Freshwater Ecology and Biotechnology, Institute of Hydrobiology, Chinese Academy of Sciences, Wuhan 430072, PR China
\textsuperscript{b} Graduate University of Chinese Academy of Sciences, Beijing 100049, PR China

\textbf{Abstract}

The increased air temperature is expected to have important driver on spring phytoplankton dynamics. To test whether spatial heterogeneity modifies the synchronous responses of phytoplankton to regional temperature driver, we evaluate temporal coherences for physical factors (temperature, water stability and non-algal light extinction), nutrients (nitrogen, phosphorus and silicon), and biomass and density of phytoplankton by Pearson correlation analysis and synchrony for phytoplankton community dynamics by Mantel test and nonmetric multi-dimensional scaling (NMS), during spring bloom (February 23–April 28, 2005) within Xiangxi Bay, a high spatial gradient bay of Three-Gorges Reservoir (China). The high level of temporal coherences for surface water temperature ($r = 0.946$, $p < 0.01$) and relative water column stability ($r = 0.750$, $p < 0.01$) were found between pair sites (A and B), in which the increase trends occurred with increase in regional air temperature during the study period. However, the low synchrony for phytoplankton dynamics were indeed observed between Site A and B, especially for the density of common dominant taxa (Cyclotella spp.: $r = 0.155$, $p = 0.388$) and community structure (Mantel test: $r = 0.351$). Moreover, the local habitat characteristics such as nutrient (nitrogen and phosphorus) and non-algal light extinction showed low levels of temporal coherence. It indicated that local community of phytoplankton varies rather independently within the single lentic bay with high spatial heterogeneity and that dispersal of algal organisms among locations cannot overwhelm these local dynamics. Contrary to many studies, the present results argued that, in a small geographic area (i.e., a single reservoir bay of approximately 24 km length), spatial gradients also may influence spring phytoplankton response to regional temperature driver.

\textsuperscript{*}Corresponding author.

E-mail address: qhec@ihb.ac.cn (Q. Cai).

Introduction

Phytoplankton live at the interface between the abiotic and the biotic compartments of ecosystems, they play an important role in multiple nutrient cycles (Klausmeier et al., 2008). As one of the dominant features in the seasonal growth patterns of phytoplankton of oceans and lakes, spring phytoplankton bloom is particularly important with respect to food web dynamics of aquatic systems, so that the controlling mechanisms of spring phytoplankton succession has been received much attention (Henson et al., 2006; Ferris and Lehman, 2007; Peeters et al., 2007a; Nicklisch et al., 2008; Sommer and Lengfellner, 2008). Some studies have found that spring phytoplankton respond strongly to physical conditions such as irradiance, temperature and water stability, all of which are expected to be altered by climate change (Tirok and Gaedke, 2007). For example, Iriarte and Purdie (2004) commented on the relationship between North Atlantic Oscillation and spring phytoplankton bloom in the English Channel. Henson et al. (2006) studied the effect of meteorological conditions on interannual variability in timing and magnitude of spring bloom in the Irminger Basin, North Atlantic. Peeters et al. (2007b) analyzed to what extent the onset and early development of the spring phytoplankton bloom depends on seasonal changes in turbulent mixing, solar radiation and water temperature. Henson (2007) pointed out that the onset of water column stability is closely linked to the timing of the phytoplankton spring bloom.

During the last decades, aquatic systems were subject to change in climatic conditions and increase in nutrient loading, and it requires a better understanding of how phytoplankton response to multiple environmental stressors. Most previous research focused either on the effect of physical condition on phytoplankton dynamics (e.g. Iriarte and Purdie, 2004; Jasprica and Hafner, 2005; Henson et al., 2006; Miller and Harding, 2007; Peeters et al., 2007a) or on the effect of nutrient on phytoplankton biomass (e.g. Hunt and Matveev, 2005; Beamud et al., 2007; Çelik and Ongun, 2008; Kasprzak et al., 2008; Katano et al., 2008). Although...
Tirok and Gaedke (2007) investigated the individual and combined effects of three physical variables (irradiance, temperature and water stability) on algal dynamics using descriptive statistics, multiple regression models and a process-oriented dynamic simulation model, few studies has tried to reveal the relative importance of local (e.g. spatial heterogeneity) and regional (e.g. temperature increase) factors on the phytoplankton dynamics. As a simple and useful method, temporal coherence analyses were often used to explore whether the populations within a region are influenced more by intrinsic (local) or extrinsic (regional) factors, a key question in population ecology (Takahashi et al., 2008). Temporal coherence is defined as the phenomenon of synchronous fluctuations in one or more parameters among locations within a geographic region (Nakamoto et al., 1976; Magnuson et al., 1990). High level of synchrony indicated that the dynamics of population were determined by regional factors (e.g. temperature). In contrast, low level of synchrony suggested that local-scale regulators (e.g. nutrient) were most important. The study of this phenomenon is thus necessary to explain the underlying mechanisms that control spring phytoplankton bloom.

The reservoir-bay can provide a gold opportunity to gain a better understanding of the relative effect of regional climate driver and local spatial gradient on the spring phytoplankton development. A long reservoir-bay can be regarded as the canyon-shaped reservoir without dam which exhibited marked longitudinal gradients (Shao et al., 2010). For example, Ye et al. (2006, 2007) analyzed the spatial distribution of nutrient in Xiangxi Bay of Three-Gorge Reservoir (TGR, China), and found that phosphorus increase and nitrogen decrease along longitudinal gradients from the mouth to the headwater. Xu et al. (2009a) also illustrated the longitudinal gradients in inorganic suspended particles of Xiangxi Bay, one of non algal absorbing components which influence the underwater light condition for phytoplankton growth. Based on theoretical studies of temporal coherence, phytoplankton should exhibit coherent development patterns within a reservoir-bay with common climatic conditions if they are not influenced by spatial heterogeneity. Conversely, if asynchrony dynamics of phytoplankton is found, one may infer the predominance of local-scale regulators (e.g. nutrient and underwater light conditions). However, most previous studies of reservoir emphasized longitudinal gradients in biological, physical and chemical variables (e.g. Caputo et al., 2008), and few studies focused on temporal coherence of limnological variables within a single system (Lansac-Tôha et al., 2008).

Three-Gorges Reservoir (TGR), located in the mainstream of the Yangtze River (China), is one of the largest man-made lakes in the world, with capacity of 3.93 x 10^10 m^3, water level of 175 m and surface area of 1080 km^2 (Huang et al., 2006; Xu et al., 2009a). The reservoir includes 40 large reservoir-bays (watershed area > 100 km^2) and the surface area of these bays account for 1/3 of the total surface area of TGR (Cai and Hu, 2006; Huang et al., 2006). Since the reservoir was filled to an altitude of 135 m above sea level in June 2003, algal blooms often occur in these bays (Cai and Hu, 2006). At the times, scientists and policy makers were concerned that the bays of TGR was showing symptoms of eutrophication, because no algal blooms were reported before the construction of Three-Gorge Dam. The Xiangxi River is the largest tributary of TGR in Hubei Province. After the first impoundment, the lower ca. 24 km stretch of this river became Xiangxi Bay (a representative bay of TGR), and a series of limnological observations have been taken by Xiangxi Ecosystem Station, Chinese Academy of Sciences (CAS) and China Three Gorges Project Corporation (CTGPC) (Cai and Hu, 2006). In this study, we performed temporal coherence analyses to test whether spring phytoplankton response to regional temperature driver was altered by local spatial heterogeneity, based on bidaily monitoring of pair sites on abiotic factors and phytoplankton during spring bloom in Xiangxi Bay in 2005.

Materials and methods

Field sampling

Site A for daily monitoring and Site B for bidaily monitoring were set up in Xiangxi Bay in the present study (Fig. 1). Site A and B located ca. 5 km and 18 km upstream from the mouth of the bay, with the depth of ca. 23 m and 16 m, respectively. From February 23 to April 28, 2005, 300 mL surface water samples for nutrient measurements, including nitrate nitrogen (NO3- N), phosphate phosphorus (PO4-P), and silicate silicon (SiO2-Si), and 1.5 L surface water sample for phytoplankton quantitative investigations were collected between 10:00 and 14:00 using a 5 L Van Dorn sampler. The samples for dissolved substance analyses were stored in a pre-cleaned plastic bottle, acidified in situ with sulfuric acid to make pH ≤ 2 and held on ice until laboratory measurement (Huang et al., 2000; Cai, 2007). The quantitative samples for phytoplankton identification were preserved in situ with standard Lugol’s solution. A sedimentation method was used to concentrate 1.5 L of water samples to 30 mL of quantitative sub-samples for phytoplankton identification, and those sub-samples were fixed in 5% neutral formalin (Huang et al., 2000; Cai, 2007). An additional 300–600 mL of water was filtered through a microfilter (0.8 μm) for Chlorophyll a (Chl. a) determination, and the filter was immediately placed in a dark cooler and packed in ice until the laboratory analysis (Huang et al., 2000; Cai, 2007). In situ, transparency (SD) was measured with a 20 cm Secchi disc, and water temperatures (WT) of surface and bottom were recorded with a Horiba U-20 Water Quality Meters (HORIBA Advanced Techno, Japan). During the study period, the data of average daily air temperature was provided by Xingshan Weather Station, located about 18 km north from the Xiangxi Bay.

Fig. 1. The sampling sites of the study located in Xiangxi Bay, Three-Gorges Reservoir (China).
**Laboratory measurement**

From each sample, 0.1 mL of phytoplankton quantitative subsample was examined in a Fuchs-Rosenthal counting chamber under an Olympus microscope at 400× magnification in order to determine cell number of taxa, and all phytoplankton densities were expressed as the number of cells L⁻¹. Most phytoplankton taxa were identified to species or genus according to Hu et al. (1980), John et al. (2002) and Hu and Wei (2006). Small centric diatoms (about 10 μm diameter, disc-like species), mainly consisting of Cyclotella, were combined as one group (Cyclotella spp.) (Xu et al., 2009b). The concentrations of NO₃-N, PO₄-P and SiO₂-Si were analyzed following the standard methods of aquatic ecosystem (Huang et al., 2000; Cai, 2007). The pigments are extracted from the algal sample concentrated by filter in an aqueous solution of acetone. The Chl.  α concentration is determined on a spectrophotometer (Shimadzu UV-1601, Japan) by measuring the absorbance of the extract at various wavelengths (750, 663, 645, and 630 nm).

**Data analysis**

By comparing the density difference between bottom (D₅) and surface (D₁) water to the density difference between 4 °C (D₄) and 5 °C (D₃) of pure water, the dimensionless parameter (relative water column stability, RWCS) was calculated according to the following formula (Padišák et al., 2003):

\[
\text{RWCS} = \frac{D_b - D_3}{D_4 - D_5}
\]

Water density (D) was calculated as a function of the water temperature using the Krambeck equation (Rodríguez-Rodríguez et al., 2004):

\[
D = 0.999869 + 6.67413 \times 10^{-5}T - 8.85556 \times 10^{-6}T^2 \\
+ 8.23031 \times 10^{-8}T^3 - 5.51577 \times 10^{-10}T^4
\]

where \(T\) is the water temperature (°C).

Underwater light extinction (K) is the sum of extinction from phytoplankton themselves (Kₚ) as well as extinction from non-algal turbidity (Kₐ) caused by colored dissolved organic matter and inorganic solid. The non-algal turbidity extinction (Kₐ) was calculated according to the following formula (Caraco et al., 1997):

\[
K_{Na} = K - K_{p}
\]

Total light extinction (K) was calculated from SD as (Caraco et al., 1997):

\[
K = -1 \times \frac{\ln(\text{SDLight\%})}{\text{SD}}
\]

where SDLight% is the fraction of surface light penetration at the SD and is generally reported as 0.1 (e.g., 10%, Wetzel and Likens, 2000). The light extinction from phytoplankton themselves (Kₚ) was calculated as (Caraco et al., 1997):

\[
K_{p} = \text{EP} \times \text{Chl.} \alpha
\]

where EP is the biomass specific extinction (in m² mg Chl. α⁻¹) of phytoplankton and is here used as a value of 0.01 (McBride et al., 1993).

As estimate of temporal coherence requires the same time series of observation data, data analysis of the present study was made based on bidayal data which were simultaneity gained from Site A and B. We used the Pearson correlation coefficient to estimate temporal coherence for physical variables, nutrient, Chl. a and algal density. The difference of nutrients between Sites was evaluated with Pairied-Samples t-test. Both Pearson correlation analysis and Paired-Samples t-test were run with SPSS13.0. Among-samples separation was displayed in nonmetric multi-dimensional scaling (NMS). We choose autopilot mode in the NMS analysis and Sorenson coefficient in the distance measure. We tested differences in phytoplankton community type among NMS groups with multi-response permutation procedures (MRPP). Furthermore, we tested temporal coherence for phytoplankton community between Site A and B with Mantel test. Prior to multivariate analyses, abundance data of phytoplankton were transformed into relative abundance to better approximate normality. All multivariate analyses (NMS, MRPP and Mantel test) were run using PC-ORD 4.0 (McCune and Mefford, 1999).

**Results**

**Temporal coherence for physical factors**

Mean daily air temperature of Xiangxi Bay has increased substantially from February to April, ranging from 7.1 to 25.8 °C, with an average of 14.7 °C (Std. Deviation = 5.1 °C) over the study periods (Fig. 2). Mean water temperature of Site A and B were 14.5 °C and 14.8 °C, ranging from 9.8 to 21.9 °C and 10.1 to 22.9 °C, respec-
Fig. 3. Temporal coherence of nitrate nitrogen (NO₃-N), phosphate phosphorus (PO₄-P), and silicate silicon (SiO₂-Si) between Site A and B (see Fig. 1).

Fig. 4. Temporal coherence of nutrient ratio between Site A and B (see Fig. 1).

**Figuraty coherence for nutrient variables**

Concentrations of NO₃-N in Site A and B varied from 0.73 to 2.40 mg L⁻¹ and 0.06 to 0.83 mg L⁻¹, respectively, while that of PO₄-P in Site A and B ranged from 0.048 to 0.155 mg L⁻¹ and 0.079 to 0.416 mg L⁻¹, respectively. Low level of temporal coherence between Site A and B were detected for NO₃-N and PO₄-P (Fig. 3). Significant higher concentration of nitrogen was recorded in Site A throughout the study period (Paired test: \( r = 12.663, p < 0.01 \)), while obvious higher phosphorus were observed in Site B (Paired test: \( r = -9.819, p < 0.01 \)). Mean concentrations of SiO₂-Si were 1.63 mg L⁻¹ and 1.69 mg L⁻¹, for Site A and B, varying from 0.02 to 3.09 mg L⁻¹ and 0.32 to 2.67 mg L⁻¹, respectively. High level of temporal coherence was detected between Site A and B for silicon, which displayed obvious decline (Fig. 3).Silicate silicon showed no significant difference between Site A and B (Paired test: \( r = -0.549, p = 0.587 \)). Low level of temporal coherence between Site A and B were detected for nutrient ratio (Fig. 4). Site A had significantly higher NO₃-N:PO₄-P ratio (Paired test: \( r = 10.6, p < 0.001 \)) and SiO₂-Si:PO₄-P ratio (Paired test: \( r = 5.0, p < 0.001 \)) than Site B, which had significantly higher SiO₂-Si:NO₃-N ratio than Site A (Paired test: \( r = -6.357, p < 0.001 \)).

**Asynchronous dynamics of phytoplankton**

Spring algal blooms occurred in Site A and B, as showed by the dramatic variation of Chl. \( \alpha \) and algal density (Fig. 5). Mean concentrations of Chl. \( \alpha \) were 27.3 and 42.5 \( \mu \)g L⁻¹ for Site A and B, varying from 1.6 to 75.3 \( \mu \)g L⁻¹ and 1.6 to 156.1 \( \mu \)g L⁻¹, respectively. Algal density of Site A and B ranged from \( 0.78 \times 10^6 \) to \( 20.24 \times 10^6 \) cells L⁻¹ and \( 0.32 \times 10^6 \) to \( 17.75 \times 10^6 \) cells L⁻¹, respectively. For Chl. \( \alpha \) and total algal density, there were significant correlations between Site A and B, but temporal coherences were lower than that of water temperature and relative water column stability (Figs. 2 and 5).
The dominant taxa of phytoplankton spring bloom in Site A were *Asterionella formosa* and *Cyclotella* spp., accounting for 46.1% and 30.3% of the total abundance, respectively. Density of *A. formosa* and *Cyclotella* spp. ranged from $0.16 \times 10^6$ to $15.64 \times 10^6$ cells L$^{-1}$ and $0.01 \times 10^6$ to $13.67 \times 10^6$ cells L$^{-1}$, respectively. One density peak of *A. formosa* was observed at Site A from April 04 to April 28, while two density peak of *Cyclotella* spp. occurred from March 09 to March 21 and April 06 to April 16, respectively (Fig. 6). The dominant taxa of phytoplankton bloom in Site B were *Cyclotella* spp., *Peridiniopsis* spp., and *Chroomonas* spp., which accounted for 35.2%, 22.6% and 15.9% of the total abundance, respectively. Density of *Cyclotella* spp. ranged from $0.01 \times 10^6$ to $14.44 \times 10^6$ cells L$^{-1}$, and its peak occurred from April 06 to April 28. Three density peak of *Peridiniopsis* spp. occurred between February 23 and April 04, and one density peak of *Chroomonas* spp. were only observed on April 02. The phytoplankton spring bloom of Site A was dominated by the diatom (*A. formosa* and *Cyclotella* spp.) during the whole study period, as reported by Xu et al. (2009b), while the dinoflagellate (*Peridiniopsis* spp. and *Chroomonas* spp.) and diatom (*Cyclotella* spp.) dominated the phytoplankton spring bloom during February 23–April 04 and during April 06–April 28, respectively. Moreover, lowest coherence was also found for the common dominant taxa of phytoplankton spring bloom (*Cyclotella* spp.) between Site A and B.

The final two-dimensional NMS solution accounted for 87.5% of the variance in algal composition for Site A and B, based on the coefficient of determination between the distance in ordination space and distance in the original space for axis 1 ($r^2 = 0.574$) and axis two ($r^2 = 0.302$). In the NMS sample ordination space (Fig. 7), the 33 observation of Site A were well classified into the following five stages: stage A1 (February 23–March 07), stage A2 (March 09–March 21), stage A3 (March 23–April 04), stage A4 (April 06–April 16) and stage A5 (April 18–April 28), while that of Site B were well divided into the following two stages: stage B1 (February 23–April 02) and stage B2 (April 04–April 28). The five stages of diatom bloom at Site A were reported by Xu et al. (2009b). The comparisons of community types by MRPP further confirmed that significant shifts in phytoplankton composition occurred at Site A ($p < 0.001$) and Site B ($p < 0.001$). However, the low level of synchrony in community shifts of phytoplankton can be observed between Site A and B in the NMS space and was validated by Mantel test ($r = 0.351$).
Fig. 7. Non-metric multidimensional scaling (NMS) ordination of phytoplankton communities throughout the study in the Xiangxi Bay of the Three-Gorges Reservoir (China). Final stress for two-dimensional solution = 40.44. Different symbols indicate different community groups, and related sample scores are enclosed with ellipses.

Discussions

Lakes provide an excellent system to explore the influence of intrinsic vs extrinsic drivers of microbial communities, since the shoreline boundaries enable one to distinguish forces acting from within and from outside the system (Kent et al., 2007). Previous studies addressing temporal coherence were thus made across a series of distinct lakes at different temporal and spatial scales (Rusak et al., 1999; Baines et al., 2000; Arnott et al., 2003). For example, Rusak et al. (1999) investigated the temporal coherence (i.e., the correlation or synchrony between time series) of annual abundances among populations of freshwater zooplankton in eight lakes in Ontario, Canada, from 1980 to 1992. Baines et al. (2000) analyzed synchrony in the temperature, calcium, and chlorophyll dynamics of seven neighboring northern Wisconsin lakes to assess the effect of lake basin properties on their response to climate signals. Arnott et al. (2003) used temporal coherence to determine the response to climate change in boreal shield lakes. However, some recent studies emphasized that temporal coherence of limnological variables within a single ecosystem should also be extensively investigated, due to high heterogeneity of different sampling locations (Lansac-Tôha et al., 2008; Xu et al., 2010).

The present study applied temporal coherence of limnological variables to exploring the synchronous response of a single reservoir-bay ecosystem to regional temperature driver during the spring. Following the related concept (Lansac-Tôha et al., 2008), high temporal coherence was considered as a good indicator of the importance of regional climate drivers such as air temperature, while low temporal coherence suggests that local factors disturb the influence of daily temperature increase on the spring dynamics of aquatic ecosystem. Among three physical variables, water temperature and relative water column stability both had high coherences between Site A and B, indicating that regional air temperature plays a prevalence role on those two key physical variables for phytoplankton dynamics. The present result is shared with commonsense expectation which the increased air temperatures during the spring typically raise surface water temperature and thus water column stability (Wetzel, 2001). The above opinions were further proved by our regression analysis between air temperature, surface water temperature and relative water column stability.

Normally, the increased temperatures, water stability and surface irradiance start the phytoplankton succession in spring, building up a biomass maximum (Wetzel, 2001). Considering that our phytoplankton data were collected from a single reservoir-bay system with common surface irradiance, it would be expected that high level temporal coherences for phytoplankton were similar to those for surface water temperature and relative water column stability. However, the low synchrony for phytoplankton dynamics were indeed observed between Site A and B, especially for the common dominant taxa and community structure. The low levels of temporal coherence are probably accounted for by weak individual dispersal and high spatial heterogeneity that may asynchronize the dynamics of life organisms. During the study periods, Xiangxi Bay with the range of monthly residence times (RT) from 127 to 178 d (Xu et al., 2010), should be grouped into lentic systems (RT > 120 d, Søballe and Kimmel, 1987). The low potential of algal dispersing between Site A and B with a hydrological connection may thus be inferred considering weak transport of water flow in a lentic system. On the other hand, nutrient (nitrogen and phosphorus) and non-algal turbidity extinction showed low coherences between Site A and B, indicating that the single reservoir bay had high spatial heterogeneity of nutrient and underwater light conditions for phytoplankton growth. Hence, it is reasonable to believe that local community of phytoplankton varies rather independently within the single lentic bay with high spatial heterogeneity and that dispersal of algal organisms among locations cannot overwhelm out these local dynamics. Due to only two sites, it was a pity that the present study statistically failed to discriminate the relative importance of nutrient and underwater light conditions for regulating the asynchronous response of phytoplankton. In spite of that, we may deduce that non-algal turbidity extinction was more key local regulators in Xiangxi Bay, where the concentrations of nitrogen and phosphorus both already exceed the threshold values of the eutrophic state, and are probably sufficient high to achieve algal maximum growth rates.

Most previous studies often regarded the development of spring phytoplankton spring as a period cycle of seasonal events and emphasized the importance of large-scale climatic fluctuation on the development of spring bloom (Smetacek, 1990; Yin et al., 1997; Straile, 2000; Iriarte and Purdie, 2004; Miller and Harding, 2007). Some studies have confirmed the expected earlier onset of the spring bloom under warming conditions (Rees et al., 1999; Peeters et al., 2007a), which agree with the frequently reported earlier onset of spring events reported from terrestrial ecosystems (e.g. Walther et al., 2002), as well as with commonsense expectation. The traditional views were sometimes opposed by other researchers who reported retardations of spring bloom by climate warming have been reported (e.g. for the shallow German Bight of the North Sea, Wiltshire and Manly, 2004). The above controversy may be clarified by the present study that indeed confirmed asynchronous response of phytoplankton to spring temperature driver in a single reservoir bay with high spatial heterogeneity. In other words, local regulators may weaken the effects of temperature increase on the dynamics of spring phytoplankton. It is not thus surprising that the development of spring phytoplankton is retarded under climate warming.

Acknowledgements

This work was funded by National Natural Science Foundation of China (No. 40671197) and the Key Project of Knowledge Innovation Program of CAS (No. KZCX2-YW-427). We thank Shunchan Zhou, Xinqin Han, Xinghuan Jia, Fengqing Li, Xiaocheng Fu, Naicheng Wu.
Guangjie Zhou, Hongbo Tang and Jianlin Hu for their assistance in the field and the lab.

Special thanks are due to two anonymous reviewers for their useful comments for improving the manuscript.

References


