Full Length Research Paper

# Comparison of benthic recruitment with pelagic growth of bloom-forming cyanobacteria

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In eutrophic lakes, overwintering bloom-forming cyanobacteria such as *Microcystis* re-inoculate the water column in the spring, contributing to pelagic development as a cell source for initial pelagic growth and a direct inoculum for continued pelagic development as well. In the present study, recruitment and pelagic growth of cyanobacteria were monitored by using migration traps and phytoplankton chambers at a study site near Meiliang Bay (1.5 m deep) in Taihu Lake from March to June, 2008. Cell abundance from both sources was compared synchronously; in the meantime water temperature was recorded using an automatic sonde. Results showed that recruitment and pelagic growth of cyanobacteria already started at 11°C, and showed changing patterns over the time of the study. Benthic recruitment only accounted for a small portion of pelagic growth, which is similar to that found in deep lakes. Taken together, these findings suggest that studies on cyanobacteria blooms development should be mainly focused on the pelagic population development.

Key words: Colony, cyanobacteria, *Microcystis*, pelagic growth, recruitment.

# INTRODUCTION

In temperate lakes, bloom-forming cyanobacteria such as *Microcystis* overwinter as a life strategy to survive harsh environment in two niches: water column and surface sediments (Brunberg and Blomqvist, 2002; Reynolds et al., 1981; Tsujimura et al., 2000). The stock on the surface sediments can become quite large, sometimes larger than the pelagic population in some lakes (Takamura et al., 1984); in other lakes, pelagic overwintering abundance can be 10 times higher or more than benthic population (Brunberg and Blomqvist, 2002). When suitable environment returns, the benthic populations renew growth, start dividing, and form new colonies (Reynolds et al., 1981) or filaments (Karlsson-Elfgren, 2003). Immediately following, the colonies or filaments develop gas vacuoles and migrate into the water column. Benthic recruitment of cyanobacteria has been viewed as an advantage over other phytoplankton for dominance and ultimately bloom formation (Brunberg and Blomqvist, 2003; Head et al., 1999). A lack of benthic recruitment would reduce summer blooms by 50% (Verspagen et al., 2004). Although benthic recruitment of Microcystis measured in field studies only accounted for a small proportion of the overall pelagic population (Brunberg and Blomqvist, 2003; Karlsson-Elfgren et al., 2003), but they are the "seeds" for pelagic growth. Furthermore, it has been found that cyanobacteria originated from benthic stocks in deep lakes are unlikely to contribute much to pelagic development as they lack gas vesicles which enable them to "float" back to the water column (Tsujimura et al., 2000) and the recruitment is restricted to the hypolimnion (Hansson, 1996). On the other hand, the number of recruited colonies is larger in shallow regions of lakes than those in deep regions (Brunberg and Blomqvist, 2003; Hansson, 1996: Karlsson-Elfgren, 2003; Karlsson-Elfgren and Brunberg, 2004), which indicates that shallow areas are more important for pelagic development of phytoplankton. Additionally, the importance of shallow regions is highlighted by their high stock of biomass on the surface sediment. Following this line of findings, it is reasonable

to assume that recruitment could be more important in shallow lakes than in deep lakes.

Taihu Lake is a large and extremely shallow eutrophic lake located in the southeastern China (latitude 30°55'40" to 31°32'58" N; longitude 119°52'32" to 120°36'10" E). It has an area of 2,338 km<sup>2</sup>, a maximum depth of 2.6 m, an average depth of 1.9 m and a mean water residence time about 264 days (Hu et al., 2006). Taihu Lake receives inflows from nearby riverine networks, including over 200 streams, canals and rivers. Meiliang Bay, the northern part of Taihu is highly eutrophic. In the past few years, it suffers from annual Microcystis blooms. Especially, in the spring of 2007, an unprecedented heavy Microcystis blooms resulted in water supply crises. In the present study we measured the size of recruitment with migration traps and net growth rate of pelagic cyanobacteria with perspex phytoplankton chambers so as to compare their sizes throughout the water column on an area basis and find out the major origin of pelagic Microcystis after recruitment.

## MATERIALS AND METHODS

#### Study site description

The study site (31° 25′ 53.4″ N, 120° 10′ 42.5″ E) (Figurre 1) is between Meiliang Bay and offshore regions of Taihu Lake, where annual blooms appear. Cyanobacteria in the water column and sediments are dominated by *Microcystis* (>95% of the overall biomass), other genus of cyanobacteria only exist with very low amounts (Cao et al., 2005; Chen et al., 2003). The depth of the study site was 1.5 m and fluctuated within 10 cm during this study, which was taken into account in calculation.

## Sampling and analyses

Transparent perspex migration traps (volume = 20 L) designed by Cao et al. (2005) were used to estimate cyanobacterial recruitment from sediments. The detailed features and usage of the traps were illustrated elsewhere (Cao et al., 2005). In the current study, traps in triplicate sat flush on the sediment water within the traps was removed with a bilge pump powered by a 12 V accumulator. Ambient water entered the trap through the 3-um mesh nets (Sefar, Switzerland) covering the traps which were able to screen single cells of Microcystis. After trap installation, 25 L water was pumped from within the traps. The traps were deployed there for three days to allow benthic cyanobacteria to enter the traps; the water within the traps was collected using a bilge pump. For each sample, 1.0 L water was immediately fixed with acid Lugol's solution for phytoplankton examination: 250 ml water was transferred back into laboratory and immediately filtered with GF/C glass filters (Whatman, UK). The resulting filters were stored and determined for pigment concentration according to methods described by Downes and Hall (1998) and Yan et al. (2004). As the cyanobacterial population is nearly all composed of Microcystis (Cao et al., 2005) and phycocyanin (PC) content could mainly represent the biomass of Microcystis. The reason we used phycocyanin was that *Microcystis* cells showed wide heterogeneous colony size, with one colony consisting of from two to more than 1000 cells. Pelagic growth of cyanobacteria was measured by culturing the phytoplankton within transparent perspex chambers suspended in water column. The chambers (50 L) had an open bottom covered

with 3-µm mesh nets (Sefar, Switzerland) which could hold the passage of colonies and unicells through the nets.

The nets themselves were fastened to the chambers and also secured with wires. Water was added into and retrieved from the chambers after culture through an opening in the top of the chambers which was sealed with a perspex lid. In addition, water temperature was continuously monitored by an automatic sonde (6600, Yellow Spring Instruments, USA).

#### Data analyses

Recruitment of cyanobacteria was presented as the total of each 3day trap collection converted on 1 m<sup>2</sup> surface sediments. The *in situ* net growth rate was calculated from  $r = \ln (C/C_0)/T$ , where  $C_t$  is the PC concentration at the end of culture,  $C_o$  is the PC concentration at the beginning and T is the period of culture in days. As we look to assess the relative contributions of sedimental and pelagic sources, their absolute biomass contributed in three days were used instead of growth/recruitment rates during the same time.

# RESULTS

# Changes in water temperature

Environmental conditions were becoming suitable for algal growth in Taihu Lake during the study. Water temperature increased from 5.3°C in early March to 30.1°C in late June which showed recurrent fluctuations but steadily remained over 10°C after April 7 (Figure 1).

# Benthic recruitment and pelagic growth of cyanobacteria

The cyanobacteria in benthic and pelagic niches were both mainly composed of *Microcystis*, accounting for over 95% of total cyanobacterial biomass, while other genera such as *Anabaena* and *Aphanizomenon* were also present but only in rather less amounts. Concentration of benthic Chla (chlorophyll a) and Chlb (chlorophyll b) showed decreasing trends during the study, but that of PC approximately remained slight fluctuations (Figure 2). While these pigments showed two peaks in original water and one peak in traps (Figures 3 and 4). Moreover, concentrations of pigments in traps were far less than original water owing to filtration by the 3-µm mesh nets.

# Comparison of benthic recruitment with pelagic growth of cyanobacteria

Due to the difference in the units used to express the amounts of cyanobacteria from these two niches, it is difficult to directly compare their sizes. We calculated the amount of growth for each 3-day experiment, in the entire water column (from water surface to sediment surface) overlying on a unit area of sediments  $(1.0 \text{ m}^2)$  from that obtained for 1.0 L water. In other words, pigment content was transformed from microgram per liter water ( $\mu g L^{-1}$ )



Figure 1. Changes in water temperature during the study.



Figure 2. Concentration of benthic pigment.

into microgram per water column (microgram column<sup>-1</sup>). Benthic recruitment was already expressed as on a unit area (microgram m<sup>-2</sup>). This way, the pelagic growth and benthic recruitment could be compared directly. Accordingly, recruitment of cyanobacteria only accounted for a small proportion of pelagic increase in the water column (Table 1). The highest recruitment only accounted for about 2.3% of the pelagic growth in the chambers of the first test for each 3-d period. Then it declined persistently throughout the study.

#### DISCUSSION

Overwintering cyanobacteria on the surface sediments will first renew growth before inoculating the water

column (Reynolds et al., 1981), during which they develop gas vesicles and gain buoyancy to return to the column (Karlsson, 2003). Recruitment water of cyanobacteria, such as Microcystis (Brunberg and Blomqvist, 2003; Cao et al., 2005; Hansson et al., 1994), Gloeotrichia (Karlsson-Elfgren et al., 2003), Anabaena, Aphanizomenon, and Oscillatoria (Hansson et al., 1994; Head et al., 1999; Karlsson-Elfgren and Brunberg, 2003) contributes to pelagic phytoplankton in two ways: seed source for initial pelagic growth and direct inoculation for continued phytoplanktonic development. As the stock of overwintering cyanobacteria in the sediments is significantly larger than pelagic proportions (Brunberg and Blomqvist, 2003; Karlsson-Elfgren et al., 2003; Tsujimura et al., 2000), the recruitment of cyanobacteria has served as a 'seed bank' for the water column.



Figure 3. Concentration of pelagic pigment in original water.



Figure 4. Average concentration of pelagic pigment in traps.

 Table 1. Comparison of benthic recruitment with pelagic growth.

	23-Mar	3-April	19-April	11-May	26-May	18-Jun
Recruitment (µg m <sup>-2</sup> )	30.3	40.3	48.2	45.4	37.1	40.3
Pelagic growth (µg L⁻¹)	0.9	1.3	4.0	3.1	1.9	2.4
Pelagic growth (µg column <sup>-1</sup> )	1300.0	1900.0	6000.0	4700.0	2800.0	3500.0
Percentage of benthic recruitment to pelagic growth (%)	2.3	2.1	0.8	1.0	1.3	1.2

However, it is not clear which process starts first in nature and therefore it is difficult to assess their relative importance. Temperature is an important driving factor for the growth renewal of overwintering cyanobacteria in surface sediments. Tao et al. (2005) cultured sediment samples from Taihu Lake under a gradient of temperatures; they found no cyanobacterial recruitment below 14°C and observed the greatest amount of recruitment at 18 to 20°C. Similarly, it is confirmed by Li et al. (2004) who reported migration is initiated at 15°C and 30  $\mu$ E m<sup>-2</sup> s<sup>-1</sup> of light intensity, the optimum temperature for *Microcystis* growth being 20°C. In contrast to these laboratory findings, cyanobacterial recruitment already started in the present study at about 11°C, implying *in situ* migration might be facilitated by disturbance and initiated at the lower temperatures.

Previous studies showed that the metabolic activity of M. aeruginosa represented by esterase activity started to increase at about 9°C (estimated from the figure displayed by the authors) in surface sediments of Grangent Reservoir which suggests that it begin to regain activity at this temperature (Latour et al., 2004). As temperature continued to increase in our study, the size of recruitment also increased, confirming the driving effect of temperature. As for the initiation of pelagic growth, although there were no reports, presumably it starts at about the same or higher temperatures, owing to benthic recruitment is considered as a source of "seeds" for pelagic growth (Head et al., 1999; Reynolds et al., 1981). Although the contribution of benthic recruitment to pelagic development of cyanobacteria has been assessed in Esthwaite Water (Head et al., 1999) and Lake Erken (Karlsson-Elfgren et al., 2003), no such work has been undertaken in shallow lakes, especially in large and extremely shallow lakes such as Taihu Lake. In the present study, we assessed both the benthic recruitment and pelagic growth of cyanobacteria synchronously and pelagic compared their relative importance to populations. It was found that benthic recruitment only accounted for a small portion of pelagic growth in Taihu Lake. The inocula of cyanobacteria in Esthwaite Water (Head et al., 1999) and Lake Erken (Karlsson-Elfgren et al., 2003) were also found to be very small relative to the observed pelagic population development, indicating that it is not a significant source of cells. Our findings suggest that shallow lakes show similar patterns of benthic recruitment versus pelagic growth of cyanobacteria to those observed in deep lakes. However, it is noteworthy that this benthic contribution was direct inoculum for continued pelagic development. As recruitment already started and pelagic population grew in our test, it is unlikely to assess the contribution as seed source for initial pelagic growth. Moreover, the assessment may be worth further study as recruitment of *Microcystis* is lately found to be largely a passive process resulting from resuspension by wind-induced mixing or bioturbation (Verspagen et al., 2004). In most cases, resuspension is a frequent event in shallow lakes and more so in the case of Taihu Lake (Chen et al., 2003). Also, in our study, most of the tests were undertaken when the lake was calm. So the effects of resuspension were minimal and thus recruitment was underestimated.

Additionally, the traps themselves may also contribute to the underestimation of recruitment by diminishing resuspension (Cao et al., 2005). The pelagic cyanobacteria biomass did not increase steadily but showed a variable pattern in Meiliang Bay, which was subject to growth, death, influx from and outflux to neighboring offshore areas. Through a model calculation, benthic recruitment combined with either pelagic growth rate or residence time could account for the observed variations in Lake Erken (Karlsson-Elfgren et al., 2003). It was clear in our study that even the cyanobacteria kept growing in the chambers, the pelagic abundance were still quite variable, indicating other abiotic factors were in play, which are mainly depended on temperature, wind and turbidity (Chen et al., 2003). These findings suggest that we should mainly focus on the pelagic fraction of cyanobacteria in order to better understand cyanobacterial bloom development.

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