Interdisciplinary Studies on Environmental Chemistry—Biological Responses to Chemical Pollutants, Eds., Y. Murakami, K. Nakayama, S.-I. Kitamura, H. Iwata and S. Tanabe, pp. 367–372.

Succession of Harmful Algae *Microcystis* (Cyanophyceae) Species in a Eutrophic Pond

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(Received 16 May 2008; accepted 28 July 2008)

Abstract—In eutrophic freshwaters, succession of *M. aeruginosa* and *M. wesenbergii* was examined through field study during May and November 2006. The reciprocal succession between *M. aeruginosa* and *M. wesenbergii* was found during the study period. From the fact that the water temperatures during the dominance of *M. aeruginosa* were apparently higher (from 24.7 to 33.9°C) than those during *M. wesenbergii* dominance (from 19.6 to 28.6°C), we suggest that temperature is one important environmental factor controlling the succession of dominant *Microcystis* species.

Keywords: *Microcystis*, water temperature, succession, eutrophic pond

INTRODUCTION

Microcystis is one notorious genus forming waterblooms particularly in shallow eutrophic freshwater environments, and often cause serious problems in the management of water quality. Some *Microcystis* species produce toxin called "microcystin" (Park *et al*., 1998), which has harmful effects not only on domestic animals but also on human beings (Carmichael, 1992). Numerous studies have been conducted to reveal the mechanisms how *Microcystis* dominate the phytoplankton community and the related environmental factors affecting their prosperity in the habitat (Reynolds and Walsby, 1975; Fay, 1983; Takamura, 1988).

It has been found that *Microcystis* blooms often consist of multiple species. Their spatial and temporal dynamics in natural habitats have been studied by some authors (Watanabe *et al*., 1986; Amemiya *et al*., 1990; Tsujimura, 2003: Ozawa *et al*., 2005). In Japan, *Microcystis* blooms in lakes mainly consist of *Microcystis aeruginosa* and *Microcystis wesenbergii* (Watanabe *et al*., 1986). Generally, *M. aeruginosa* is toxic, while *M. wesenbergii* nontoxic (Watanabe *et*

al., 1988, 1991; Ozawa *et al*., 2005). Hence, to know the succession pattern of dominant *Microcystis* species and the mechanism controlling their succession is important for management of water quality as well as for understanding ecology of aquatic organisms. However, we still have insufficient information on the mechanism of the reciprocal succession between *M. aeruginosa* and *M. wesenbergii*.

In the present study, the mechanism inducing the reciprocal succession of the two *Microcystis* species was analysed though the field monitoring in the hyper eutrophic pond in Japan.

MATERIALS AND METHODS

Field study

The present study was carried out in Furuike Pond (33°49′ N, 132°48′ E, Matsuyama city, Ehime Pref., Japan) from May to November 2006. The pond is eutrophic due to anthrophogenic loading from the watershed. Its physical and chemical characterizations have been described in our previous studies (Nakano *et al*., 1998, 2003; Manage *et al*., 2001; Nishii *et al*., 2001). During *Microcystis* bloom period (May to November), weekly samplings were carried out to collect more detail data set. Water samples were taken with a column sampler which has a hydraulically operating flap at the bottom (60 cm long; 5 cm diameter). This sampler is designed for collecting water quantitatively from the surface to a certain depth. In the present study, the water samples were taken from the surface to near bottom. Surface water temperature and pH were determined using a thermistor (ABT-1, ALEC Electronics Co. Ltd.) and pH meter (B-212, HORIBA), respectively.

To determine the nutrient concentrations in the pond, 80 ml of water sample was filtered through a Whatman GF/F glass fiber filter (Whatman Inc., Clifton, USA). The filtrate was poured into an acid-washed plastic bottle and stored in a freezer (–20°C) before analyses. Dissolved inorganic nitrogen (DIN = NO_2–N + NO_3–N + NH_4–N) and soluble reactive phosphorus (SRP) concentrations were determined by colorimetric analysis with a continuous flow system (AutoAnalyzer 3, BRAN + LUEBBE).

To determine chlorophyll *a* concentration, a water sample was filtered through a 0.2 μm Nuclepore filter (CORNING Nuclepore) to retain seston. The retained seston was then transferred into a glass tube containing 8 ml of *N*,*N*-dimethylformamide to extract chlorophyll *a*. They were kept in a freezer at –20°C. The chlorophyll *a* concentration was determined using a fluorometer (Turner Designs, 10-AU) (Moran and Porath, 1980).

For the enumeration of phytoplankton, a 300 ml of a water sample was fixed with acid Lugol's solution at a final concentration of 1%. Phytoplankton were concentrated by natural sedimentation, and cell numbers of each species were counted with a haematocytometer under a microscope at ×200 magnification. *Microcystis* species was identified based on their morphology (Komárek, 1991). To obtain biovolume of *Microcystis*, colony sizes of each *Microcystis* species

Table 1. The results of field study on the environmental factors.

Month	Day	WT[1] (°C)	pH	DIN[2] (μg N l^{-1})	SRP[3] (μg P l^{-1})	Chl. *a*[4] (μg l^{-1})
May	16	19.0	9.5	4.0	3.4	189.0
	22	24.9	9.7	22.3	2.7	88.3
	30	24.7	9.8	2.5	1.6	481.8
June	5	26.6	9.3	8.2	2.9	191.3
	13	31.4	10.5	6.1	1.8	723.4
	19	26.3	10.1	0.5	1.3	677.8
	26	24.3	8.0	3.8	2.2	477.2
July	3	28.7	9.7	1.7	2.2	410.7
	10	28.6	9.7	13.7	3.3	398.2
	24	26.5	8.8	9.4	1.5	454.4
	31	32.5	9.3	17.0	2.2	179.9
August	7	33.7	9.5	8.8	1.4	514.9
	14	33.9	9.7	16.0	1.6	396.4
	21	33.4	9.8	20.0	1.7	583.3
	30	29.3	9.5	34.7	1.0	418.4
September	7	28.0	9.5	8.0	2.3	382.5
	12	26.3	9.2	35.7	1.0	444.7
	21	26.6	9.0	12.2	7.7	334.6
	27	25.7	9.4	13.6	1.7	270.1
Octorber	4	24.8	9.3	9.6	1.3	329.9
	11	24.8	9.5	7.8	1.1	296.4
	18	23.9	9.3	12.9	2.3	250.9
	24	19.6	8.9	17.8	3.3	272.4
November	9	18.1	8.6	8.0	2.1	447.1
	17	14.7	8.9	5.7	2.4	ND

[1]Water temperature.
[2]Dissolved inorganic nitrogen.
[3]Soluble reactive phosphorus.
[4]Chlorophyll *a* concentration.

were measured by Center for Microbial Ecology Image Analysis System (CMEIAS), and converted into carbon biomass using Strathmann's equation (1967).

We used Pearson Correlation Analysis to find significant correlations between biomass of dominant two *Microcystis* species and environmental variables. The statistical analysis was performed with software (Microsoft office Excel 2003, Statcel2).

RESULTS

Seasonal changes in environmental factors

Water temperature in Furuike Pond gradually increased from May, and then fluctuated between 24.3 and 32.5°C from June to July (Table 1). High water temperatures (29.3–33.9°C) were maintained during August, and water temperature

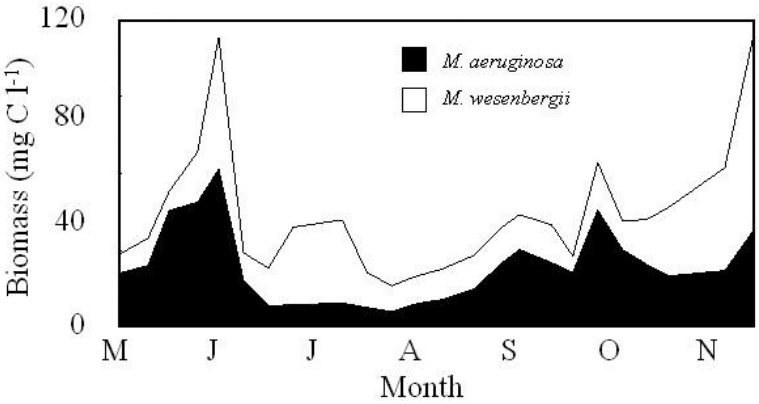

Fig. 1. Changes in abundance of *M. aeruginosa* (black) and *M. wesenbergii* (white) during the study period in the Furuike Pond.

continued to decrease from September to the end of the study period (Table 1). Seasonal changing pattern of pH was similar to that of water temperature, ranging between 8.0 and 10.5 (Table 1). The average DIN concentration in the pond was low (13 μg N l^{-1}), but high DIN concentrations were found from mid July with the highest concentration in September (35.7 μg N l^{-1}, Table 1). SRP concentrations ranged between 1.0 and 7.7 μg P l^{-1}, and the highest concentration was detected also in September (Table 1).

Seasonal changes in phytoplankton community and Microcystis biomass

Chlorophyll *a* concentration highly fluctuated between 88 and 723 μg l^{-1}, and a clear seasonal pattern was not found (Table 1). Phytoplankton community was dominated by cyanobacteria. *Microcystis* dominated phytoplankton community from May. *M. aeruginosa* predominated in June and from mid August to September, while *M. wesenbergii* dominated in July and from mid October onwards.

Remarkable biomass increase of *M. aeruginosa* was observed in June and August when the water temperature rapidly increased. However, such biomass increase was not observed for *M. wesenbergii* (Fig. 1). As a result, the water temperatures during the dominance of *M. aeruginosa* were relatively higher (from 24.7 to 33.9°C) than those during the dominance of *M. wesenbergii* (from 19.6 to 28.6°C).

DISCUSSION

M. aeruginosa often predominated in the early stage of bloom forming, followed by the dominance of *M. wesenbergii* (Takamura and Watanabe, 1987). Our results coincide with previous results. The succession of *Microcystis* species in the present study seems to be closely related to the changes of water temperature.

Previous studies demonstrated that optimal temperature of *M. aeruginosa* growth ranged between 30 and 35°C (Krüger and Eloff, 1978; Van der Westhuizen and Eloff, 1985; Watanabe and Oishi, 1985), and this temperature range overlaps well with our observation. Our results and previous studies indicate that favorable temperature for the growth of *M. aeruginosa* is higher than that of *M. wesenbergii*. Consequently, it can be thought that *M. aeruginosa* predominates during summer when water temperature is high while *M. wesenbergii* becomes predominant from autumn under decreased water temperature.

In the present study, we have demonstrated that temperature is one of the important environmental factors determining dominant *Microcystis* species. This result suggests that the dominance of *M. aeruginosa* occurs in freshwater ecosystems more frequently under global warming. Park *et al*. (1998) reported that higher amount of toxin was released from *Microcystis* cells when the water temperature was high, due to the dominance of toxic *M. aeruginosa*. Hence, toxic effects by *Microcystis* may become more serious in temperature freshwater ecosystems under global warming. Further physiological and toxicological studies on *Microcystis* species are quite necessary for water quality management and human health.

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