

# *In situ* study on the control of toxic *Microcystis* blooms using phytoplanktivorous fish in the subtropical Lake Taihu of China: A large fish pen experiment

Zhixin Ke, Ping Xie\*, Longgen Guo, Yaqin Liu, Hua Yang

*Donghu Experimental Station of Lake Ecosystems, State Key Laboratory for Freshwater Ecology and Biotechnology of China, Institute of Hydrobiology, the Chinese Academy of Sciences, Wuhan 430072, PR China*

Received 11 October 2006; received in revised form 29 January 2007; accepted 30 January 2007

## Abstract

Three large fish pens (0.36 km<sup>2</sup> of each) stocked with silver and bighead carp were set up in Meiliang Bay for controlling toxic *Microcystis* blooms. The responses of plankton communities and food consumption of silver and bighead carp were studied. Crustacean zooplankton were significantly suppressed in the fish pens. Total phytoplankton biomass, *Microcystis* biomass and microcystin concentration were lower in the fish pens than in the surrounding lake water, but the difference was not statistically significant. The present stocking density of silver plus bighead carp (about 40 g/m<sup>3</sup> in July) was likely too low to achieve an adequate control of *Microcystis*. Silver carp fed mainly on phytoplankton but bighead carp mainly on zooplankton: mean zooplankton contribution in the gut was 31.5% for silver carp and 64.7% for bighead carp. Compared with previous studies, both carp species preyed upon more zooplankton because of the abundant food resource. Daily rations of silver and bighead carp were estimated by Egger's model in the main growing season. Filtration rate was calculated from the daily ration and the density of plankton in the lake. During May–October, filtration rates of silver and bighead carp for phytoplankton were 0.22–1.53 L g<sup>-1</sup> h<sup>-1</sup> and 0.02–0.68 L g<sup>-1</sup> h<sup>-1</sup>, respectively, and filtration rates for zooplankton were 0.24–0.44 L g<sup>-1</sup> h<sup>-1</sup> and 0.08–1.41 L g<sup>-1</sup> h<sup>-1</sup>, respectively. Silver carp had a stronger ability of eliminating phytoplankton than bighead carp. To achieve a successful biomanipulation with a minimum effect of ichthyoeutrophication, the stocking proportion of bighead carp should be controlled in the future practice.

© 2007 Elsevier B.V. All rights reserved.

**Keywords:** Fish pens; Silver carp; Bighead carp; Biomanipulation; *Microcystis* blooms; Microcystins; Filtration rate

## 1. Introduction

Cyanobacterial blooms are causing severe problems in many warm-water lakes due to increasing eutrophication in recent years. One method of controlling cyanobacterial blooms is through biomanipulation.

The classical view of biomanipulation is the reduction of planktivorous fishes, which results in higher densities of herbivorous zooplankton and consequently in lowered densities of algae due to zooplankton grazing (Shapiro et al., 1975; McQueen, 1990). However, the ability of natural populations of zooplankton to control phytoplankton is still under debate, especially in the water dominated by cyanobacterial blooms (Bernardi and Giussani, 1990). In tropical or subtropical regions, for instance, zooplankton does not seem able to control

\* Corresponding author. Tel./fax: +86 27 68780622.  
E-mail address: [xieping@ihb.ac.cn](mailto:xieping@ihb.ac.cn) (P. Xie).

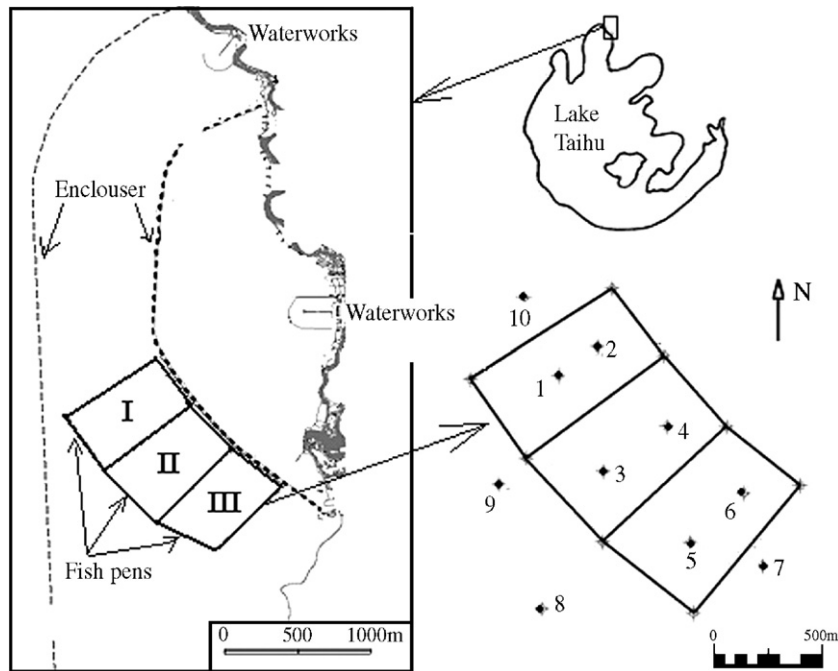


Fig. 1. The sketch of Lake Taihu and the location of fish pens in Meiliang Bay.

algal biomass, a role that is played instead mainly by omnivorous, filter-feeding fish (Xie and Liu, 2001).

Grazing pressure by planktivorous fish is a key factor in shaping the size of algae and zooplankton in lakes (Xie and Yang, 2000; Lu et al., 2002). Bio-control of algae through introduction of suitable herbivorous fishes has been one of the most environmentally sound management propositions recently. The large filter-feeding fish, silver and bighead carp and tilapia, are the attractive candidates for bio-control of plankton communities to eliminate odorous populations of cyanobacteria (Starling, 1993; Tucker, 2006). It is difficult to evaluate the effect of filter-feeding fish on phytoplankton communities, since fish suppresses phytoplankton as well as zooplankton grazers. However, despite many inconsistent effects of them on the control of water quality (Lieberman, 1996; Domaizon and Devaux, 1999), numerous studies have shown that opportunistic, size-selective grazing by filter-feeding fish can control cyanobacterial blooms, and thus improve water quality (Smith, 1985; Starling, 1993; Datta and Jana, 1998). This biomanipulation strategy is effective especially under eutrophic or hypertrophic conditions where the phytoplankton community is dominated by colonial species and the zooplankton community is dominated by microzooplankton (Fukushima et al., 1999; Xie and Liu, 2001).

The planktivorous filter-feeding silver carp (*Hypophthalmichthys molitrix*) and bighead carp (*Aristichthys nobilis*) are the most intensively cultured fish species in Asia and comprise much of the production of Chinese aquaculture (Liang et al., 1981). These two fish species have now become the dominant species in many lakes and reservoirs in southern China for two purposes: (1) to increase fish yield and (2) to control algal blooms. Silver and bighead carps are being used or tested in many Chinese lakes such as Lake Dianchi in Yunnan Province, Lake Chaohu in Anhui Province, and Lake Taihu in

Table 1

Annual mean ( $\pm$ SD) of physical and chemical parameters in the fish pens and the surrounding lake during the study period

Parameters	In fish pens	In the surrounding lake
Transparency (cm)	30.65 $\pm$ 4.88	30.59 $\pm$ 5.80
pH	8.1 $\pm$ 0.57	8.19 $\pm$ 0.44
Conductivity (mS/cm)	0.56 $\pm$ 0.01	0.56 $\pm$ 0.13
NH <sub>4</sub> -N (mg/L)	1.39 $\pm$ 1.28	1.42 $\pm$ 1.47
NO <sub>2</sub> -N (mg/L)	0.11 $\pm$ 0.09	0.10 $\pm$ 0.08
NO <sub>3</sub> -N (mg/L)	1.07 $\pm$ 0.52	1.06 $\pm$ 0.52
TN (mg/L)	2.52 $\pm$ 1.79	2.47 $\pm$ 1.83
TDN (mg/L)	2.24 $\pm$ 1.92	2.03 $\pm$ 1.68
PO <sub>4</sub> -P (mg/L)	0.014 $\pm$ 0.006	0.015 $\pm$ 0.008
TP (mg/L)	0.149 $\pm$ 0.055	0.152 $\pm$ 0.067
TDP (mg/L)	0.050 $\pm$ 0.014	0.054 $\pm$ 0.016

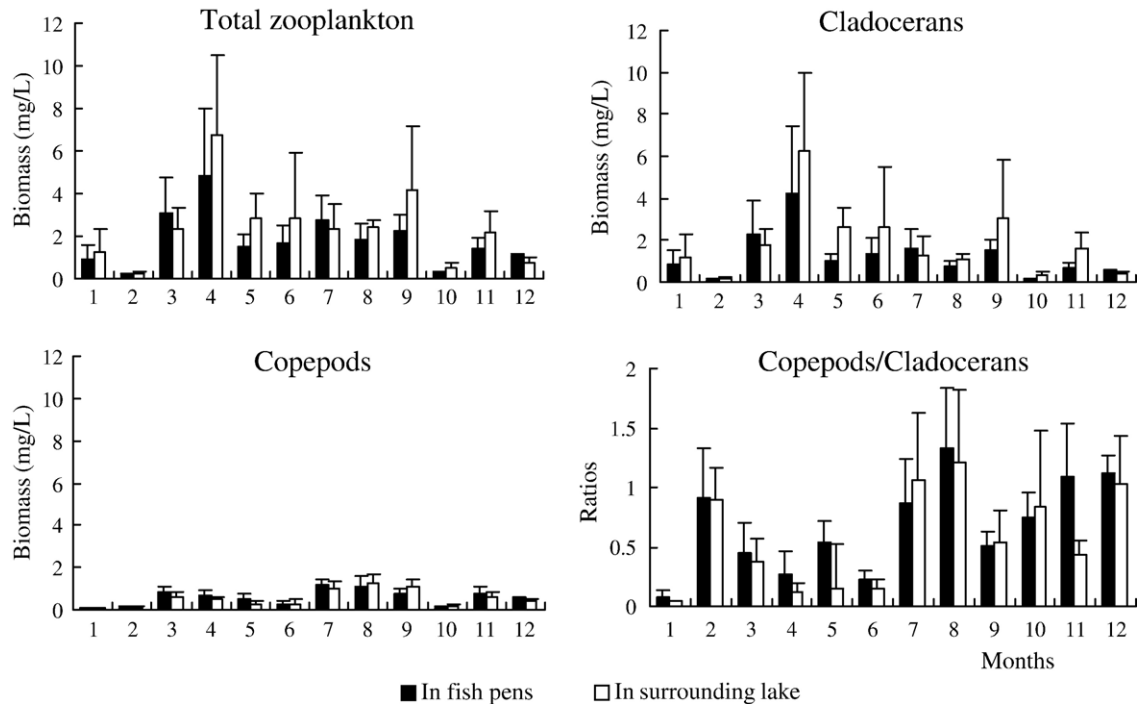


Fig. 2. Seasonal changes in the biomass of crustacean zooplankton in both the fish pens and the surrounding lake in 2005.

Jiangsu Province for the control of cyanobacterial blooms (Xie and Liu, 2001). Although silver and bighead carp have been tested for eliminating algal blooms in a variety of small-scale experiments, their ecological niches are variable, with food selection strongly dependent on food availability in the environment (Fukushima et al., 1999; Berthou and Amich, 2000). There is still little quantitative information on food consumption of silver and bighead carp in the practice of large-scaled biomanipulation.

The main purpose of this study was to examine the possibility of bio-filtering toxic *Microcystis* blooms in drinking water resources by utilizing phytoplanktivorous fishes stocked in large fish pens in Meiliang Bay of Lake Taihu.

**2. Methods**

Lake Taihu, the third largest freshwater lake in China, is located in the south of the Yangtze River delta. The total area of the lake is 2338 km<sup>2</sup>, with an average depth of 2 m and a total capacity of 47.6 × 10<sup>8</sup> m<sup>3</sup> (Jin and Hu, 2003). Because of a large population increase and rapid industrial and agricultural pollution in the lake’s drainage basin, Lake Taihu has undergone rapid eutrophication (William, 1996). The Meiliang Bay is located in the northern part of Lake Taihu with a surface area of 100 km<sup>2</sup> and a depth of 1.8–2.3 m. It acts not

only as principal water resource for Wuxi City, but also as an important tourist attraction. During the past few decades, heavy *Microcystis* blooms occurred regularly and may last 8 months each year. This region has been listed as one of the hypertrophic parts in Lake Taihu. Three large biomanipulation fish pens for stocking silver and bighead carps were built in Meiliang Bay for the control of cyanobacterial blooms, which was a part of the Lake Taihu restoration program (Fig. 1). The area of each fish pen was 0.36 km<sup>2</sup> and the mesh size of the net was 2 cm × 2 cm. From December 2004 to January 2005, 24,775 kg and 8005 kg silver and bighead carps fingerlings were averagely stocked into the three pens. The initial stocking density of silver and bighead carp was 11.47 g/m<sup>3</sup> and 3.71 g/m<sup>3</sup>, respectively. We chose this stocking density because it is close to the levels of

Table 2  
Biomass of crustaceans (means ± SD) and the difference of crustacean zooplankton between the fish pens and the surrounding lake in 2005 (\*\* significant difference)

	Pens	Surrounding lake	T-test (P value)
Total crustaceans (mg L <sup>-1</sup> )	1.83 ± 1.3	2.39 ± 1.77	0.049**
Cladocerans (mg L <sup>-1</sup> )	1.26 ± 1.12	1.86 ± 1.66	0.031**
Copepods (mg L <sup>-1</sup> )	0.57 ± 0.37	0.53 ± 0.4	0.513
Copepods/cladocerans	0.67 ± 0.4	0.57 ± 0.42	0.126

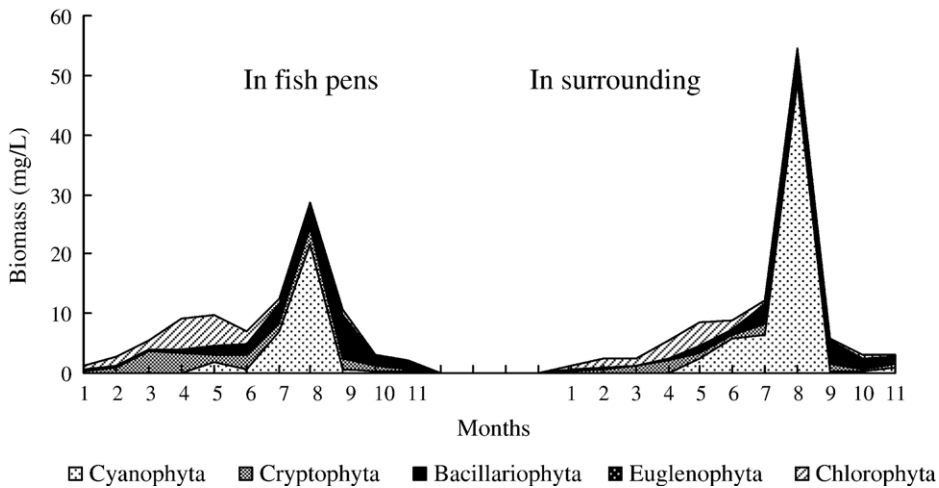


Fig. 3. Seasonal changes in the biomass of phytoplankton in both the fish pens and the surrounding lake in 2005.

Lake Donghu and many successful biomanipulation experiments (Xie and Liu, 2001). All fishes were harvested by seining in December 2005.

Fish samples were collected monthly in 2005, and about 30 silver carp and 30 bighead carp were randomly captured from the middle of each pen by seine or multi-mesh gillnets to examine individual growth (body length and body weight) on each sampling date. The standing stock of fish in each month was estimated from the total number of fish finally captured in the pens and the average body weight. Survival rate was estimated from the data of fish yield at the end of the study. To examine food items, five individuals of each carp species were killed and the guts were removed immediately by dissection in each month. Fore-gut contents were collected from the proximate end of the intestine to the middle of the first loop. Food in this part of the intestine was considered to be little digested since this part usually comprises less than 1/12 of the total intestine

length (Xie, 1999). Individual samples of gut contents were fixed in Lugol's iodine for a few minutes, and then preserved in 10% formaldehyde solution. In laboratory, the gut contents were homogenized in cool distilled water with an electronic stirrer for 3–5 min, and then examined under microscope.

In parallel with the fish sampling, zooplankton and phytoplankton in the water of the middle pen were sampled for comparison. Crustacean zooplankton were sampled with a plankton net (69 μm mesh) and preserved in 5% sucrose formalin. Phytoplankton and rotifer samples were preserved with 1% Lugol's iodine after sedimentation for 24 h. Wet weights of crustacean zooplankton were estimated according to the weight-body size regression of Huang et al. (1984). Phytoplankton cells and rotifers were counted and sized to derive volumes from appropriate geometric shapes. To count *Microcystis* cells, the samples were agitated by gentle

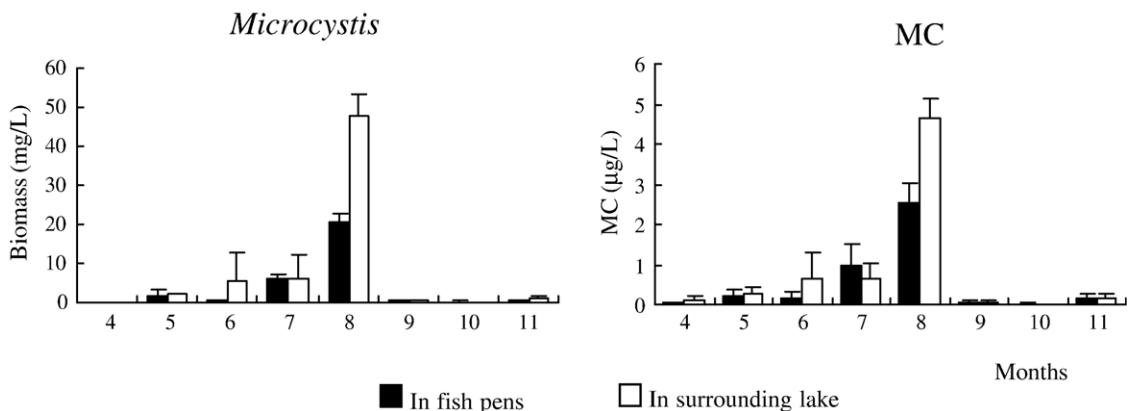


Fig. 4. Temporal changes of *Microcystis* biomass and MC concentration in both the fish pens and the surrounding lake from April to November.

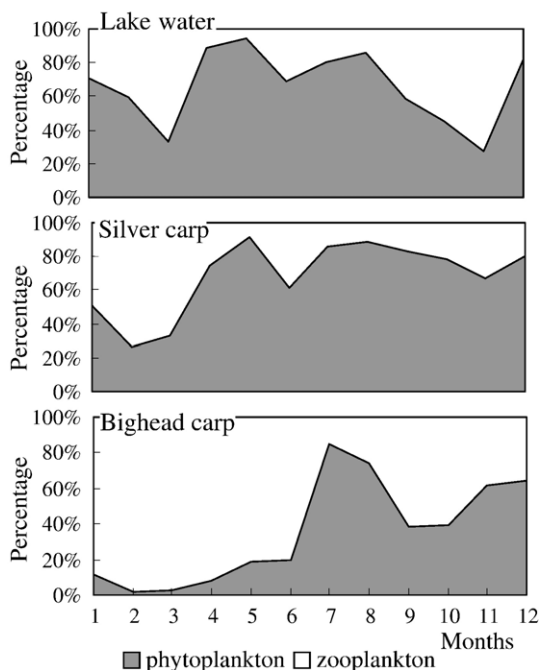


Fig. 5. Seasonal variation of phytoplankton and zooplankton components in the lake water and gut contents of silver and bighead carp in 2005.

ultrasonication to split the colonies into single cells. After mixing, 0.1 ml samples were counted directly under 400× magnification. Biomass (wet weight) of phytoplankton and rotifers were calculated assuming a wet weight density of 1 g cm<sup>-3</sup>.

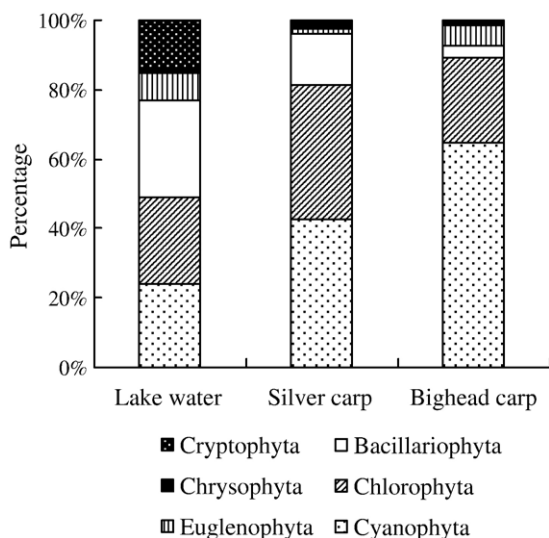


Fig. 6. Mean composition of phytoplankton in the lake water and gut contents of silver and bighead carp in 2005.

In the main growth season, daily rations of silver and bighead carp were estimated by the Eggers' model (1977):  $C=24SR$ , where  $S$  is the mean gut fullness during a 24-h trial and  $R$  ( $h^{-1}$ ) is the gut evacuation rate. To study the mean gut fullness and the feeding rhythm, five individuals for each carp were captured in the fish pens at a 4-h interval over a 24-h period in every month. The captured fish were dissected immediately and the total gut contents were weighted. The gut evacuation rate of silver and bighead carp was calculated according to the water temperature and exponential evacuation equations established by Chen (1990). We calculated the filtration rates of silver and bighead carp by the following formulation:  $F=1000C/24B$ , where  $F$  ( $L g^{-1} h^{-1}$ ) is the filtration rate for phytoplankton or zooplankton,  $C$  is the daily ration of fish for phytoplankton or zooplankton and  $B$  ( $mg L^{-1}$ ) is the biomass of phytoplankton or zooplankton in the lake water. To compare the changes between the fish pens and the surrounding lake, a total of 10 sampling sites were set (Fig. 1). Discrete water samples (0, 0.5, 1.0, 1.5 m) from each sampling site were respectively taken with a Patalas–Schindler trap on the same date of fishing sampling. Crustacean zooplankton and phytoplankton were also sampled. Surface temperature of the water was measured with a thermometer. The orthophosphate ( $PO_4-P$ ), total phosphorus (TP), total dissolved phosphorus (TDP), total nitrogen (TN), total dissolved nitrogen (TDN), nitrate ( $NO_3-N$ ), ammonium ( $NH_4-N$ ) and nitrite ( $NO_2-N$ ) were analyzed by the standard methods (APHA, 1992). Intracellular microcystins (MC-LR, MC-RR and MC-YR) were measured by a reverse-phase high-performance

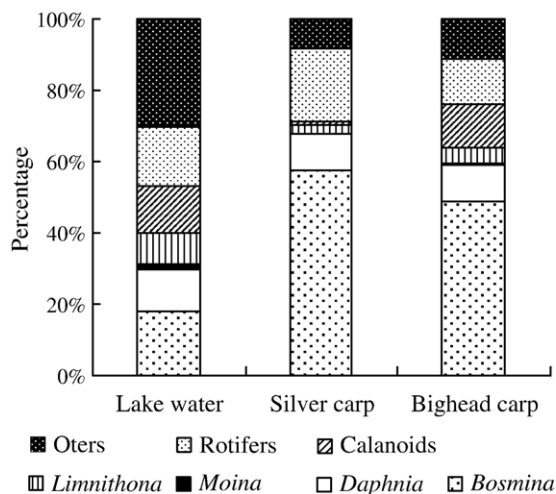


Fig. 7. Mean composition of zooplankton in the lake water and gut contents of silver and bighead carp in 2005.

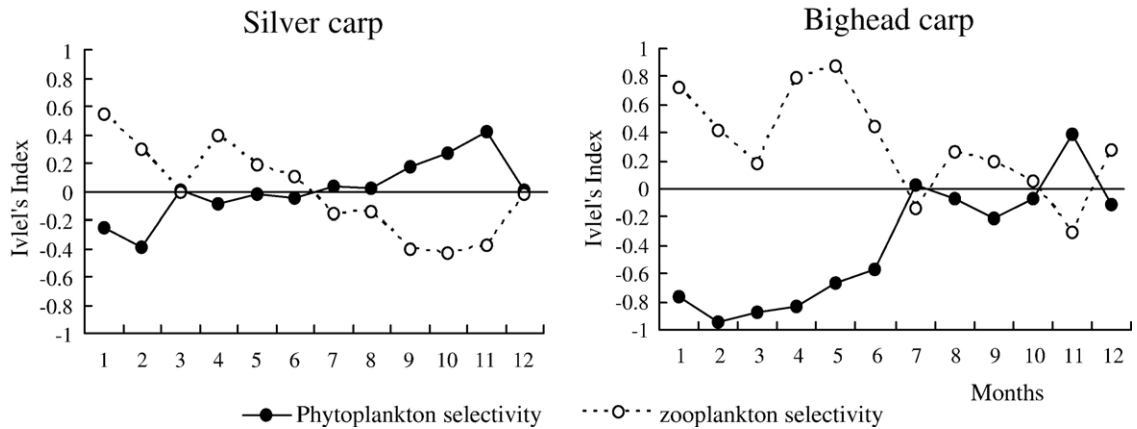


Fig. 8. Seasonal changes in food selectivity of silver and bighead carp in 2005.

liquid chromatography (HPLC) according to the methods of Park and Lwami (1998) and Zhang et al. (2006).

A *T*-test was performed to test for significant differences between the fish pens and the surrounding lake. Due to low replication and statistical power, we chose a probability level of  $\alpha < 0.10$  to reduce the chance of making the type II error of failing to reject a false null hypothesis.

### 3. Results

#### 3.1. Physicochemical parameters

The average surface temperature of the lake water was 17.2 °C, with the lowest 3 °C in January and the highest 31.2 °C in August. The mean values of the environmental variables were generally similar in both the fish pens and the surrounding lake (Table 1). According to OECD (1982), Meiliang Bay of Lake

Taihu is hypereutrophic. The annual mean TDN concentration was slightly higher in the pens than in the surrounding lake. During the study period, no physicochemical parameters were significantly different between the fish pens and the surrounding lake ( $P > 0.1$ ) except for pH ( $P = 0.08$ ). Annual mean chlorophyll a concentration was lower in the fish pens ( $43.73 \mu\text{g L}^{-1}$ ) than in the surrounding lake ( $48.06 \mu\text{g L}^{-1}$ ), but the difference was not statistically significant.

#### 3.2. Phytoplankton and crustacean zooplankton

The community structure of plankton was very simple and many species were only seasonally dominated in the water of Meiliang Bay. Crustacean zooplankton were mainly represented by the cladocerans *Daphnia*, *Ceriodaphnia*, *Bosmina* and *Moina*, and by the copepods *Cyclops*, *Mesocyclops*, *Limnoithona* and

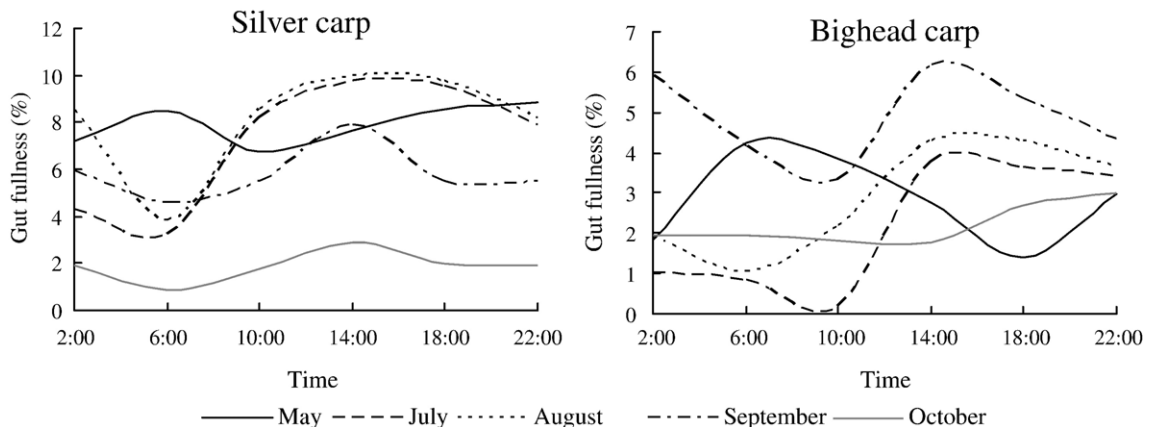


Fig. 9. Feeding rhythms of silver and bighead carp studied at a 4-h interval over a 24-h period in the main growing season.

Table 3  
Daily rations of silver and bighead carp estimated by Eggers' model

	Gut fullness (%)		Gut evacuation rate ( $\text{h}^{-1}$ )		Daily ration (%)	
	Silver carp	Bighead carp	Silver carp	Bighead carp	Silver carp	Bighead carp
May	7.91	2.85	0.078	0.081	14.75	5.51
July	7.13	2.13	0.097	0.101	16.61	5.18
August	8.12	2.90	0.102	0.107	19.93	7.41
September	5.80	4.87	0.095	0.099	13.28	11.61
October	1.86	2.16	0.079	0.082	3.53	4.25
Average	6.17	2.98	0.090	0.094	13.62	6.79

*Sinocalanus*. The dynamics of crustacean zooplankton in the fish pens were generally similar to those of the surrounding lake (Fig. 2). Annual mean biomass of crustacean zooplankton was significantly higher in the surrounding lake ( $2.39 \text{ mg L}^{-1}$ ) than in the pens ( $1.83 \text{ mg L}^{-1}$ ) ( $P < 0.05$ ). Biomass of cladocerans was also significantly higher in the surrounding lake than in the fish pens ( $P < 0.05$ ) (Table 2). The copepods/cladocerans ratio was always higher in the fish pens, but the difference was not significant ( $P > 0.1$ ). Phytoplankton were represented mainly by *Microcystis*, *Ulothrix*, *Oocystis*, *Cyclotella* and *Melosira*. Annual total biomass of phytoplankton was higher in the surrounding lake ( $9.74 \text{ mg L}^{-1}$ ) than in the pens ( $8.39 \text{ mg L}^{-1}$ ), but the difference also was not significant ( $P > 0.1$ ). Seasonal succession of phytoplankton was similar in both the fish pens and the surrounding lake (Fig. 3). Chlorophyta (mainly *Ulothrix*) predominated in spring, but was replaced by Cyanophyta (mainly *Microcystis*) after July. *Microcystis* biomass and intracellular MC concentration were lower in the pens than in the surrounding lake in most months, but their difference was not statistically significant (Fig. 4). Among the three analogues of microcystins, MC-RR was the most dominant, whereas MC-YR was the least.

Table 4  
Filtration rates ( $\text{L g}^{-1} \text{ h}^{-1}$ ) of silver and bighead carp for plankton calculated from the daily ration and the plankton density in the lake water

	Density of plankton in lake water ( $\text{mg/L}$ )		Filtration rate for phytoplankton ( $\text{L g}^{-1} \text{ h}^{-1}$ )		Filtration rate for zooplankton ( $\text{L g}^{-1} \text{ h}^{-1}$ )	
	Phytoplankton	Zooplankton	Silver carp	Bighead	Silver carp	Bighead
May	26.27	1.58	0.22	0.02	0.31	1.18
July	16.28	4.02	0.37	0.11	0.24	0.08
August	16.45	2.77	0.45	0.14	0.33	0.29
September	3.01	2.13	1.53	0.61	0.44	1.41
October	1.04	1.25	1.10	0.68	0.26	0.85
Average	12.61	2.35	0.73	0.31	0.32	0.76

Table 5  
Mean initial and final individual body weight and body length of silver and bighead carp

	Individual body length (cm)	Individual weight (g)	Total weight (kg)
Silver carp			
Stocking	$19.8 \pm 3.3$	$160 \pm 81.3$	24,775
Harvest	$37.8 \pm 2.02$	$1134 \pm 122.9$	98,000
Bighead carp			
Stocking	$18.5 \pm 1.9$	$139 \pm 43.8$	8005
Harvest	$40.4 \pm 1.68$	$1522 \pm 211.4$	42,000

### 3.3. Gut contents analysis

The temporal variation in biomass of the phytoplankton and zooplankton in both the lake water and the gut contents are shown in Fig. 5. The composition of the gut contents of silver and bighead carp showed similar dynamics with the community structure of plankton in the lake water. Throughout the year, phytoplankton were predominant in the gut contents of silver carp, and the average biomass contribution was 68.5% with a range of 26.4–91.6%; however, zooplankton were predominant in the gut contents of bighead carp, and the average biomass contribution was 64.7% with a range of 15.4–98.4%, except in July and August when the gut contents of bighead carp were also dominated by phytoplankton.

Fifty three phytoplankton genera were found in the gut contents of both carps. The major phytoplankton species in the gut contents were similar to those in the lake water, with *Ulothrix*, *Cyclotella* and *Oocystis* from January to June, and *Microcystis* and *Melosira* from June to December. On average, Cyanophyta, Chlorophyta, Bacillariophyta, Euglenophyta and Chrysophyta constituted 43%, 38%, 17%, 1.6% and 0.23%, respectively, of phytoplankton contents in the guts of silver carp, and 65%, 24%, 3.26%, 6% and 1.32% in the guts of bighead carp, respectively (Fig. 6). In July and

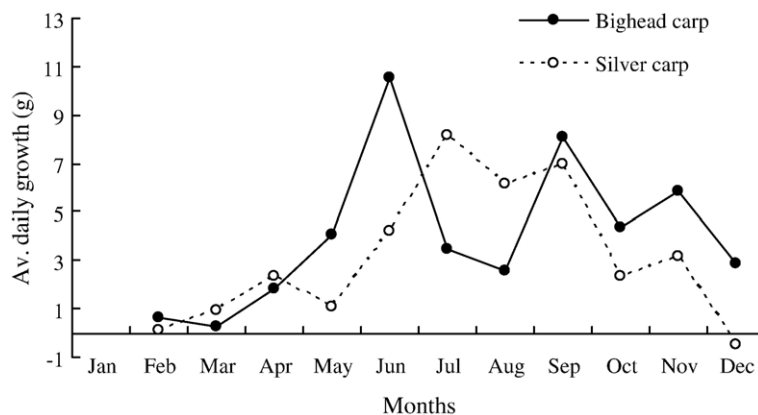


Fig. 10. Seasonal variation in average daily growth of silver and bighead carp in 2005.

August, *Microcystis* contributed more than 80% of the phytoplankton in the gut contents of silver carp and almost 100% of the phytoplankton in the gut contents of bighead carp. A total of 16 species of crustacean zooplankton and 14 species of rotifer were identified from the gut contents of both carp species, although many zooplankton could not be identified to species because of digestion. On average, *Daphnia*, *Bosmina*, *Limnoithona*, calanoids and rotifers constituted 10%, 59%, 2.6%, 1.3% and 21%, respectively, of zooplankton contents in the guts of silver carp, and 11%, 51%, 4.8%, 1.3% and 13% in the guts of bighead carp, respectively (Fig. 7). *Daphnia* and *Cyclops* were dominant only in spring and later winter. *Bosmina* and *Limnoithona* were the predominant species of the most time. The contribution of rotifers was higher in the gut contents of silver carp than in those of bighead carp.

The Ivlev's (1961) selectivity index of silver and bighead carp for plankton components varied greatly over time (Fig. 8). Silver carp had a negative selectivity for phytoplankton during January–June but a positive selectivity during June–December, while the index for zooplankton was reversed with a very low value (average 0.002) most time. Bighead carp had a relatively high selectivity index for zooplankton (average 0.31), except in July and November.

### 3.4. Feeding activity and daily ration

The most active feeding rhythm was observed at 14:00–18:00 h and the least active at 06:00–10:00 h, except in May, in which the feeding peak was at 06:00 (Fig. 9). Silver carp had the highest mean gut fullness rate in August (8.12%), and bighead carp in September (4.87%). The feeding intensity of silver carp was significantly stronger than bighead carp in every

month ( $P < 0.1$ ). Gut fullness rates and daily rations of silver and bighead carp are shown in Table 3. The filtration rate of silver carp for phytoplankton was higher than that of bighead carp; however, the filtration rate of bighead carp for zooplankton was higher than that of silver carp (Table 4). Mean filtration rate of silver and bighead carp for phytoplankton were  $0.73 \text{ L g}^{-1} \text{ h}^{-1}$  and  $0.31 \text{ L g}^{-1} \text{ h}^{-1}$ , respectively. Mean filtration rates of silver and bighead carp for zooplankton were  $0.32 \text{ L g}^{-1} \text{ h}^{-1}$  and  $0.76 \text{ L g}^{-1} \text{ h}^{-1}$ , respectively.

### 3.5. Growth

The total yields of silver and bighead carp were about  $14 \times 10^4 \text{ kg}$ , of which silver and bighead carp comprised about 70% and 30%, respectively. Mean initial and final individual body weight and length of silver and bighead carp are shown in Table 5. The pen-cultured silver and bighead carp all displayed fast growth (Fig. 10). The average growth rates of silver and bighead carp were  $3.18 \text{ g day}^{-1}$  and  $4.05 \text{ g day}^{-1}$ , respectively. The maximum daily increments of the body weight of silver carp and bighead carp were 8.14 g in June–July, and 10.5 g in May–June, respectively. The survival rates of silver and bighead carp were estimated to be 26.5% and 41.6%, respectively.

## 4. Discussion

In the present study, silver carp mainly fed on phytoplankton while bighead carp mainly fed on zooplankton. This was consistent with many previous studies (Cremer and Smitherman, 1980; Chen, 1990). Compared with previous reports, however, there was a greater proportion of zooplankton in the guts of silver and bighead carp in our study: mean zooplankton



contribution reached 31.5% and 64.7%, respectively. This greater proportion might be due to the abundant food resource in the water of Meiliang Bay, as predators tend to ignore less valuable food when more profitable ones are abundant (Krebs, 1979). In the present study, the components of food items of silver and bighead carp showed obvious temporal variation, and the selectivities of silver and bighead carp for phytoplankton increased after *Microcystis* predominated in the lake water. Miura (1990) reports that colonial *Microcystis* blooms can disturb the selective feeding of bighead carp on zooplankton, resulting in a greater consumption of the algae. Smith (1989) reports that silver and bighead carp can more efficiently filter-feed larger algae than small ones. During the outbreak of *Microcystis* blooms, the low zooplankton/phytoplankton ratio in the lake water may be responsible for the declined selectivity of fish on zooplankton (Dong and Li, 1994). On the other hand, *Microcystis* always formed large colonies and floated on the surface of the lake water, which were perhaps easily fed upon by silver and bighead carp.

The present results suggest that the outbreak of *Microcystis* blooms might have changed the feeding rhythm of silver and bighead carp. Li et al. (1980) reported that maximum feeding activity of silver and bighead carps were restricted to a few hours in the early morning and late afternoon. This observation was only consistent with our results in May. From July to October, we observed a reversed feeding rhythm: a feeding peak at 14:00–18:00 h and a low feeding rate at 00:60–10:00 h. It should be noted that the dominant phytoplankton in the lake water and guts of silver and bighead carp was *Ulothrix* in May, but *Microcystis* from July to October in our study. Li et al. (1980) suggest that feeding rhythm of silver and bighead carp may be a function of light intensity, dissolved oxygen, and temperature. Our results suggest that dominant plankton species in the lake water may be another important factor affecting feeding rhythm of silver and bighead carps. *Microcystis* has a very significant diel vertical migration with light, usually aggregating on the water surface at noon. Thus, the higher feeding intensity of silver and bighead carps at noon in Lake Taihu might be related to the higher food availability. It seems that feeding periodicity may reflect a physiological rhythm evolved as an adaptation to optimal utilization of natural food resources.

In the present study, the crustacean zooplankton biomass was significantly lower in the fish pens than in the surrounding lake water, suggesting the strong suppression of crustacean zooplankton by the pen-cultured fishes. The zooplankton were mainly dominat-

ed by small-sized crustacean zooplankton such as *Bosmina longirostris* (0.23–0.7 mm), *Ceriodaphnia cornuta* (0.2–0.45 mm), and *Limnoithona sinensis* (0.35–0.55 mm) during the outbreak of cyanobacterial blooms in Meiliang Bay. These small-sized crustacean zooplankton were less efficient consumers of *Microcystis* blooms in warm-water lakes (Domaizon and Devaux, 1999; Xie and Liu, 2001). Moreover, the area of the fish pens was still quite small compared with Meiliang Bay. Smith (1985) indicated that the negative effects of fish on herbivorous zooplankton can probably be largely eliminated by confining the fish in a restricted region. In general, although zooplankton were suppressed by these pen-cultured carp, the grazing pressure on large-size algae was not reduced in our study. In the present study, there was a lower cladocerans/copepods ratio in the fish pens than in the surrounding lake water. Drener and Hambright (1987) reports that cladocerans are more vulnerable than copepods to capture mechanisms of filter-feeding planktivores because of weaker evasive capacity. Our results support this conclusion: the cladocerans/copepods ratios were higher in the gut contents of silver and bighead carp than in the lake water of the fish pens.

In the present study, compared with crustacean zooplankton, biomass of phytoplankton was not significantly suppressed by the pen-cultured fishes except in August. According to the density and filtration rate of silver and bighead carp, the clearance time of the total phytoplankton by the pen-cultured fishes was estimated to be 0.74–22 days (average 6.2 days) and that of zooplankton was 1.2–7.1 days (average 3.6 days) during the main growing seasons of the fish. Apparently, the clearance time for phytoplankton might be too long to get a significant suppression of phytoplankton. In an enclosure experiment, Zhang et al. (2006) found that a fish stocking density of 55 g/m<sup>3</sup> was the most efficient in controlling *Microcystis* blooms and increasing water clarity. In the warm seasons, dense *Microcystis* blooms were accumulated in Meiliang Bay from the pelagic region of Lake Taihu due to wind action. The present stocking density of silver plus bighead carp (about 40 g/m<sup>3</sup> in July) might be too low to reduce the phytoplankton significantly in the fish pens. Thus, a higher stocking density of fishes is needed to more effectively control *Microcystis* blooms.

In the present study, although silver and bighead carp consumed a large amount of plankton (mainly *Microcystis*) throughout the growing season, a considerable amount of feces were also excreted into the lake water during the experiment. There have been studies to document that some phytoplankton are intact after

passing-through the guts of silver and bighead carps (Chen, 1990; Vörös et al., 1997). To some extent, the effect of biomanipulation was probably counteracted by the resuspension of feces into the lake water, which might be another reason for the lack of efficient control of phytoplankton in our study. Therefore, stocking some omnivorous fishes into the biomanipulation pens to feed on these feces is expected to solve this problem in our future study.

Microcystins (MCs) are a group of potent hepatotoxins produced by species of *Microcystis*, which are potent and specific inhibitors of protein phosphatase 1 and 2A and are considered as potent tumor promoters (Eriksson et al., 1990; Runnegar et al., 1993). In the present study, MC concentration was relatively high in the water of Meiliang Bay during the outbreak of *Microcystis* blooms, which could be risky to both the ecosystem and human health of this region. In our results, the intracellular MC concentration was lower in the fish pens than in the surrounding lake water, suggesting that silver and bighead carp could be used as an effective tool to reduce MC concentration in the lake water through controlling biomass of toxic *Microcystis*. Beveridge et al. (1993) reports that toxic *Microcystis* blooms can suppress feeding activity and growth of planktivorous fish. However, in our study, silver and bighead carp did not decrease their feeding intensity but had the highest gut fullness during the outbreak of *Microcystis* blooms; while in July and August, the decline in growth rate of bighead carp might be due to an increased proportion of *Microcystis* (>75%) in their gut contents, as *Microcystis* are usually less nutrition than zooplankton.

MCs can accumulate in the organs of fishes with ingestion of *Microcystis*, therefore posing a potential risk of toxin transference to humans through the food chain (Figueiredo et al., 2004). However, the phytoplanktivorous silver and bighead carp seem to have evolved a fast capacity to depurate MCs, probably because they have been exposed frequently to toxic cyanobacteria in natural history (Xie et al., 2004; Li et al., 2005). Chen et al. (2006, in press) reported the distribution patterns and dynamics of microcystins in silver and bighead carp cultured in the same fish pens in 2004: for silver carp, the annual mean content of MCs ( $\mu\text{g/g DW}$ ) was in the order of intestine (24.97) > liver (0.957) > kidney (0.782) > blood (0.379) > muscle (0.197) > spleen (0.159) > gallbladder (0.086) > gill (0.062); and for bighead carp, higher annual MC contents ( $\mu\text{g/g DW}$ ) were also found in intestine (19.32), liver (0.374), kidney (0.797) and spleen (0.311), while MCs were relatively low in muscle (0.124). Although both

carps had accumulated a higher MC concentration in the muscle ( $1\text{--}2 \mu\text{g g}^{-1} \text{ DW}$ ) during the heavy *Microcystis* blooms, their MC content in muscle were less than  $0.2 \mu\text{g g}^{-1} \text{ DW}$  in December, which was lower than the provisional tolerable daily intake level by world health organization (WHO). This indicates that if we choose a proper harvest time (such as in December), the fishes would be safe for human consumption.

In the past decades, silver and bighead carps have been the main species stocked in many Chinese lakes. The combination of silver and bighead carps can ensure the maximum utilization of available natural plankton food because of their different feeding habits. Recently, there is a trend to increase the stocking ratio of bighead carp in lakes because of its good price and fast growth rate. While, our results indicate that bighead carp have a weaker ability to eliminate phytoplankton than silver carp. As the decline of algal biomass is a function of fish biomass (Lazzaro et al., 1992; Lu et al., 2002), a higher stocking proportion of bighead carp probably needs a higher threshold fish biomass above which *Microcystis* blooms can be well suppressed. However, the intensive stocking density can bring a problem of ichthyoeutrophication. Datta and Jana (1998) indicated that silver carp was more suitable for controlling *Microcystis* than bighead carp and tilapia because of its minor ichthyoeutrophication effect. To get a more successful biomanipulation with minimum effect of ichthyoeutrophication, the stocking proportion of bighead carp should be reduced in the future culture practice. During our study, we did not observe obvious fish disease or death, but the survival rates of silver and bighead carp was quite low when estimated by fish yield. We extrapolate that many fishes escaped from the pens. Management of fish pens should be improved in the future pen-culture practice.

## 5. Conclusion

The present study indicates that silver and bighead carp can significantly suppress crustacean zooplankton, and decrease MC concentration in the water column through decreasing *Microcystis* biomass. However, the present stocking density of silver plus bighead carp (about  $40 \text{ g/m}^3$  in July) was likely too low to achieve a significant control of *Microcystis*. Although both carps can accumulate higher microcystins in muscle during the heavy *Microcystis* blooms, these fishes can be safe for human consumption if we choose a proper harvest time (such as in December). Silver carp had a stronger ability of eliminating phytoplankton than bighead carp. To achieve a successful biomanipulation with a minimum effect of

ichthyoeutrophication, the stocking proportion of bighead carp should be controlled in the future practice.

## Acknowledgements

We would like to express our sincere thanks to Dr. Costa-Pierce and three anonymous reviewers for their useful comments and suggestions on the manuscript. This work was supported by a fund from the National Natural Science Foundation of China (30530170) and by the Key Project of CAS titled “The effects of the regenerative organic pollutant microcystins on the safety of aquatic food” (Grant No. KSCX2-SW-129).

## References

- APHA, 1992. Standard Methods for the Examination of Water and Waster Water, 18th edn. American Public Health Association, Washington, DC.
- Bernardi, R.D., Giussani, G., 1990. Are blue-green algae a suitable food for zooplankton? An overview. *Hydrobiologia* 200/201, 29–41.
- Berthou, E.G., Amich, R.M., 2000. Food of introduced pumpkinseed sunfish: ontogenetic diet shift and seasonal variation. *J. Fish Biol.* 57, 29–40.
- Beveridge, M.C.M., Baird, D.J., Rahmatullah, S.M., Lawton, L.A., Beattie, K.A., Codd, G.A., 1993. Grazing rates on toxic and non-toxic strains of cyanobacteria by *Hypophthalmichthys molitrix* and *Oreochromis niloticus*. *J. Fish Biol.* 43, 901–907.
- Chen, S.L., 1990. Fish and its role on nutrient cycling in water. In: Liu, J.K. (Ed.), *Ecological Studies of Lake Donghu* (1). Science Press, Beijing, pp. 292–371 (in Chinese).
- Chen, J., Xie, P., Zhang, D.W., Ke, Z.X., Yang, H., 2006. In situ studies on the bioaccumulation of microcystins in the phytoplanktivorous silver carp (*Hypophthalmichthys molitrix*) stocked in Lake Taihu with dense toxic Microcystis blooms. *Aquaculture* 261, 1026–1038.
- Chen, J., Xie, P., Zhang, D.W., Lei, H.H., in press. In situ studies on the distribution patterns and dynamics of microcystins in a biomanipulation fish–bighead carp (*Aristichthys nobilis*). *Environ. Pollut.*
- Cremer, M.C., Smitherman, R.O., 1980. Food habits and growth of silver and bighead carp in cages and ponds. *Aquaculture* 20, 57–64.
- Datta (Saha), S., Jana, B.B., 1998. Control of bloom in a tropical lake: grazing efficiency of some herbivorous fishes. *J. Fish Biol.* 53, 12–24.
- Domaizon, I., Devaux, J., 1999. Impact of moderate silver carp biomass gradient on zooplankton communities in a eutrophic reservoir. Consequences for the use of silver carp in biomanipulation. *C. R. Acad. Sci. Paris, Sciences de la vie / Life Science*, vol. 322 pp. 621–628.
- Dong, S.L., Li, D.S., 1994. Comparative studies on the feeding selectivity of silver carp *Hypophthalmichthys molitrix* and bighead carp *Aristichthys nobilis*. *J. Fish Biol.* 44, 621–626.
- Drener, D.W., Hambright, K.D., 1987. Experimental study of size-selective phytoplankton grazing by a filter-feeding cichlid and cichlids effects on plankton community structure. *Limnol. Oceanogr.* 32, 1138–1144.
- Eggers, D.M., 1977. Factors interpreting data obtained by diel sampling of fish stomachs. *J. Fish. Res. Board Can.* 34, 290–294.
- Eriksson, J.E., Toivola, D., Meriluoto, J.A.O., Karaki, H., Han, Y., Hartshorne, D., 1990. Hepatocyte deformation induced by cyanobacterial toxin reflects inhibition of protein phosphatases. *Biochem. Biophys. Res. Commun.* 173, 1347–1353.
- Figueiredo, D.R., Azeiteiro, U.M., Esteves, S.M., Gonçalves, F.J.M., Pereira, M.J., 2004. Microcystin-producing blooms — a serious global public health issue. *Ecotoxicol. Environ. Saf.* 59, 151–163.
- Fukushima, M., Takamura, N., Sun, L., Nakagawa, M., Matsushige, K., Xie, P., 1999. Changes in the plankton community following introduction of filter-feeding planktivorous fish. *Freshw. Biol.* 42, 719–735.
- Huang, X.F., Chen, X.M., Wu, Z.T., Hu, C.Y., 1984. Studies on changes in abundance and biomass of zooplankton in Lake Donghu, Wuhan. *Acta Hydrobiol. Sin.* 8, 345–358 (in Chinese, with English abstract).
- Ivlev, V.S., 1961. *Experimental Ecology of the Feeding of Fishes*. Yale University Press, New Haven. 302 pp.
- Jin, X.C., Hu, X.Z., 2003. A comprehensive plan for treating the major polluted regions of lake Taihu, China. *Lakes Reserv. Res. Manage.* 8, 217–230.
- Krebs, J.B., 1979. Foraging strategies and their social significance. In: Martens, P., Vandenbergh, J.G. (Eds.), *Handbook of Behavioural Neurobiology*, Vol. 3. Plenum press, New York.
- Lazzaro, X., Drenner, R.W., Stein, R.A., Smith, J.D., 1992. Planktivores and plankton dynamics: effects of fish biomass and planktivore type. *Can. J. Fish. Aquat. Sci.* 49, 1466–1473.
- Li, S.F., Yang, H.Q., Lu, W.M., 1980. Preliminary research on diurnal feeding rhythm and the daily ration for silver carp, bighead carp and grass carp. *J. Fish. China* 4 (3), 275–283 (in Chinese, with English abstract).
- Li, L., Xie, P., Chen, J., 2005. In vivo studies on toxin accumulation in liver and ultrastructural changes of hepatocytes of the phytoplanktivorous bighead carp i.p. — injected with extracted microcystins. *Toxicol.* 46, 533–545.
- Liang, Y., Melack, J.M., Wang, J., 1981. Primary production and fish yields in Chinese ponds and lakes. *Trans. Am. Fish. Soc.* 110, 346–350.
- Lieberman, D.M., 1996. Use of silver carp (*Hypophthalmichthys molitrix*) and bighead carp (*Aristichthys nobilis*) for algae control in a small pond: changes in water quality. *J. Freshw. Ecol.* 11 (4), 391–397.
- Lu, M., Xie, P., Tang, H.J., Shao, Z.J., Xie, L.Q., 2002. Experimental study of trophic cascade effect of silver carp (*Hypophthalmichthys molitrix*) in a subtropical lake, Lake Donghu: on plankton community and underlying mechanisms of changes of crustacean community. *Hydrobiologia* 487, 19–31.
- McQueen, D.J., 1990. Manipulation lake community structure: where do we go from here? *Freshw. Biol.* 23, 613–620.
- Miura, T., 1990. The effects of planktivorous fishes on the plankton community in a eutrophic lake. *Hydrobiologia* 200, 567–579.
- OECD, 1982. *Eutrophication Waters. Monitoring, Assessment and Control*. OECD, Paris.
- Park, H.D., Lwami, C., 1998. Temporal variabilities of the concentrations of intra- and extracellular microcystin and toxic Microcystis species in a hypereutrophic lake, Lake Suwa, Japan (1991–1994). *Environ. Toxicol. Water Qual.* 13, 61–72.
- Runnegar, M.T., Kong, S., Berndt, N., 1993. Protein phosphatase inhibition and in vivo hepatotoxicity of microcystins. *Am. J. Physiol.* 265, 224–230.
- Shapiro, J., Lamarra, V., Lynch, M., 1975. Biomanipulation: an ecosystem approach to lake restoration. In: Brezonik, P.L., Fox, J.L. (Eds.), *Proceeding of a Symposium on Water Quality Management through Biological Control*. University of Florida, Gainesville, pp. 85–89.

- Smith, D.W., 1985. Biological control of excessive phytoplankton growth and the enhancement of aquaculture production. *Can. J. Fish. Aquat. Sci.* 42, 1940–1945.
- Smith, D.W., 1989. The feeding selectivity of silver carp, *Hypophthalmichthys molitrix* Val. *J. Fish Biol.* 34, 819–828.
- Starling, F.L.R.M., 1993. Control of eutrophication by silver carp (*Hypophthalmichthys molitrix*) in the tropical Paranao Reservoir (Brasilia, Brazil): a mesocosm experiment. *Hydrobiologia* 257, 143–152.
- Tucker, C.S., 2006. Low-density silver carp *Hypophthalmichthys molitrix* (valenciennes) polyculture does not prevent cyanobacterial off-flavours in channel catfish *Ictalurus punctatus* (Rafinesque). *Aquac. Res.* 37, 209–214.
- Vörös, L., Presing, L.O.M., Balogh, K.V., 1997. Size selective filtration and taxon-specific digestion of plankton algae by silver carp (*Hypophthalmichthys molitrix* Val). *Hydrobiologia* 342/343, 223–228.
- William, Y.B.C., 1996. Major environmental changes since 1950 and the onset of accelerated eutrophication in Taihu Lake, China. *Acta Palaeontol. Sin.* 35 (2), 155–174.
- Xie, P., 1999. Gut contents of silver carp, *Hypophthalmichthys molitrix*, and the disruption of a centric diatom, *Cyclotella*, on passage through the esophagus and intestine. *Aquaculture* 180, 295–305.
- Xie, P., Liu, J.C., 2001. Practical success of biomanipulation using filter-feeding fish to control cyanobacteria blooms. *Sci. World J.* 1, 337–356.
- Xie, P., Yang, Y., 2000. Long-term changes of Copepoda community (1957–1996) in a subtropical Chinese lake stocked densely with planktivorous filter-feeding silver and bighead carp. *J. Plankton Res.* 22 (9), 1757–1778.
- Xie, L., Xie, P., Ozawa, K., Honma, T., Yokoyama, A., Park, H., 2004. Dynamics of microcystins-LR and -RR in the phytoplanktivorous silver carp in a sub-chronic toxicity experiment. *Environ. Pollut.* 127, 431–439.
- Zhang, X., Xie, P., Hao, L., Guo, N.C., Gon, Y.G., Hu, X.L., Chen, J., Liang, G.D., 2006. Effects of the phytoplanktivorous silver carp (*Hypophthalmichthys molitrix*) on plankton and the hepatotoxic microcystins in an enclosure experiment in a eutrophic lake, Lake Shichahai in Beijing. *Aquaculture* 257, 173–186.