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ECOLOGY

Alteration of phytoplankton assemblages caused by changes in water hardness in Feitsui Reservoir, Taiwan

Jiunn-Tzong WU[1,2,*] and Lai-Chu KOW[1]

[1]*Biodiversity Research Center, Academia Sinica, Taipei 11529, Taiwan*
[2]*Institute of Ecology and Evolutionary Biology, National Taiwan University, Taipei 10660, Taiwan*

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ABSTRACT. Feitsui Reservoir is a subtropical water reservoir constructed in 1986 for supplying the domestic water of Taipei metropolitan area. During long-term monitoring conducted from 1990 to 2007, we observed a remarkable alteration in dominant phytoplankton groups, showing a succession of chlorophytes versus *Microcystis* spp. Analysis of such change with respect to 27 physico-chemical variables in reservoir waters showed that the either phytoplankton group displayed different nutrient demands and that the dominance of chlorophytes was closely related to the elevated level of water hardness, including calcium, magnesium, soluble solids, and sulfate, which were resulted from the pollutants discharged from the construction site of highway in the watershed. A calcium-enrichment experiment supported the belief that the higher chlorophyte density was due to elevated calcium levels. The possible way by which calcium influenced the availability of carbon- as well as phosphate-source in waters was discussed. This study suggests the possible strategy of mitigating the *Microcystis* problems in water reservoir.

Keywords: Ca-enrichment; Feitsui Reservoir; Phytoplankton; Subtropical water reservoir; Water hardness.

INTRODUCTION

In Taiwan, an increase in the input of anthropogenic pollutants to the aquatic environment has resulted in eutrophication of a number of water resources. As a result, problems associated with eutrophication such as the presence of toxins and off-flavors in drinking water have become a common occurrence. Such nuisance problems are usually related to the presence of certain harmful cyanobactaria (Wu and Hsu, 1988; Carmichael, 2001; Huisman et al., 2005). As a result, the control of cyanobacteria has received great attention and interest in recent years (Chorus and Bartram, 1999; Kim, 2008).

Feitsui Reservoir (FTR) is a water reservoir which was constructed in 1986 in order to supply domestic water to the Taipei metropolitan area. Long-term monitoring of the water quality and phytoplankton in this reservoir has been conducted since its construction (Wu and Kow, 2008). During the monitoring period, a *Microcystis* bloom occurred in 1988. Since then, high concentrations of this cyanobacterium have existed in the FTR. *M. aeruginosa* is one of species which has been shown to produce the hepatotoxin microcystin-LR (Liu, 1995). As a result, a great attention has been paid to the problems associated with the occurrence of *Microcystis* in FTR.

Over last two decades, two important events occurred in the watershed that disturbed the aquatic environment of FTR, the construction of a wastewater treatment plant in 1994 to treat the sewage waters discharged from the villages and the construction of a highway across the watershed, which began in 1996 and was completed in 2005. Both construction sites were very close (<2 km) to the major riverine inflow to the FTR. The turbid wastewater discharged from the construction sites contained clay (suspended solids), soluble solids, and a variety of nutrients including phosphorus, ammonia, and cations including calcium and magnesium (Wu and Kow, 2002). The concentrations of these compounds were particularly high in the wastewater discharged in the area in which a 12.9 km tunnel was constructed. Even though a portion of the wastewater was treated prior to being discharged, the majority of the pollutants entered the FTR. As a result, these construction projects have had a large impact on the water quality as well as the structure of the phytoplankton community in the reservoir. In this study, we attempted to identify key factors that regulate these changes in the phytoplankton community. To accomplish this, we evaluated data pertaining to 27 physio-chemical variables that were collected from 1990 to 2007 on a monthly basis. In addition, we conducted a calcium-enrichment experiment to confirm the factors identified by the data analysis.

*Corresponding author: E-mail: jtwu@gate.sinica.edu.tw; Tel:+886-2-27871073; Fax: +886-2-27871182.

MATERIALS AND METHODS

Study site

Feitsui Reservoir (N24°54', E121°34') (Figure 1), which is located in the north of Taiwan, was constructed in 1986 to supply domestic water to the Taipei metropolitan area. The reservoir has an elevation of approximately 170 m above sea level at the dam site, is situated in a valley (altitude between 450 and 1,170 m above sea level) and covers an area of 10.24 km^2, giving it an effective storage of 4.06×10^8 m^3 of water. The majority of the watershed that this reservoir is located in is covered by secondary subtropical forests. In addition, a large portion of the land in the water shed is used to cultivate tea plants on mountain slopes or terraces near the riverine regions. Therefore, tea cultivation acts as a source of non-point source pollution for the FTR. Furthermore, discharge from a village located in the vicinity of the major riverine inflow to the FTR serves as a point source of pollution. The waters in this reservoir are mesotrophic or slightly eutrophic, with Carlson's trophic state index (Carlson, 1977) values ranging from 31.2 to 61.5.

The FTR is a warm monomictic system with a maximum depth of 110 m. Thermal stratification occurs from April to November, while overturn occurs usually in February. The insolation in the reservoir fluctuates throughout the year, reaching its maximum in July and minimum in January. In addition, there were obvious seasonal fluctuations in water quality and the composition of the phytoplankton community over the course of this study. The maximum, minimum and mean values of the 27 limnological variables collected throughout the duration of this study are shown in Table 1.

Water and phytoplankton sampling methods

Water and phytoplankton samples were collected at the same time on a monthly basis near the dam site of the FTR. Samples were immediately fixed with Lugol's iodine solution, after which the phytoplankton samples were stained with Coomasie brilliant blue solution (1% Coomasie brilliant blue filtered through a nitrate cellulose filter (Sartorius, Göttingen, Germany) prepared, identified and quantified in the laboratory under a light microscope (Leitz DM, Germany). The frequency of the occurrence of each species was then calculated based on counting 1000 cells per sample.

Table 1. Limnological characteristics of waters in Feitsui Reservoir over the time of this study.

Parameter	Average	Max	Min
Secchi depth (m)	3.5±1.2	6.6	1.0
Water temperature (°C)	24.0±4.6	33.5	16.5
Turbidity (NTU)	3.1±6.1	50.0	0.6
Alkalinity (m equiv. L^{-1})	0.17±0.04	0.26	0.04
pH	7.3±0.5	8.8	6.5
Chloride (mg L^{-1})	5.0±1.7	12.0	1.0
Sulfate (mg L^{-1})	8.4±1.8	15.1	5.0
Ammonium-N (mg L^{-1})	0.04±0.04	0.41	0.001
Nitrate-N (mg L^{-1})	0.47±0.15	0.81	0.12
Dissolved oxygen (mg L^{-1})	8.1±0.6	9.2	6.0
Biochemical oxygen demand (mg L^{-1})	0.7±0.4	2.0	0.3
Chemical oxygen demand (mg L^{-1})	3.6±2.0	9.6	0.2
Dissolved solid (mg L^{-1})	39.2±9.1	59.0	25.0
Suspend solid (mg L^{-1})	2.8±3.6	22.6	0.2
Conductivity (μS cm^{-1})	65.8±8.7	81.0	50.0
Total hardness (mg L^{-1})	25.0±5.2	40.8	15.8
Ca (mg L^{-1})	5.5±1.5	10.4	3.6
Mg (mg L^{-1})	2.8±0.8	6.4	1.1
Fe (mg L^{-1})	0.15±0.41	2.84	0.01
Mn (mg L^{-1})	0.008±0.001	0.044	0.004
Total bacteria (MPN 100-mL^{-1})	1609±717	60000	2
E.coli (MPN 100-mL^{-1})	85±260	2000	0
Total organic carbon (mg L^{-1})	1.01±0.37	1.87	0.08
Total inorganic carbon (mg L^{-1})	2.42±0.42	3.12	0.84
Total carbon (mg L^{-1})	4.19±1.56	9.61	0.98
Phosphate (μg L^{-1})	5.6±4.7	26.9	0.7
Total phosphorus (μg L^{-1})	19.6±20.4	150.8	4.7

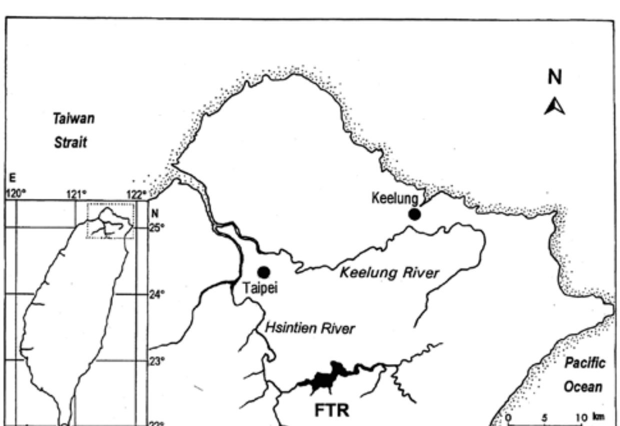

Figure 1. Map of Taiwan, showing the locality of Feitsui Reservoir (FTR).

Analyses of physico-chemical variables

The quality of the water was determined each time the phytoplankton was sampled. The transparency (Secchi depth), water temperature, dissolved oxygen, conductivity, and pH were measured *in situ* during sampling. The turbidity, alkalinity, concentrations of nitrate, ammonium, phosphate, total hardness, calcium, magnesium, manganese, iron, total inorganic and organic carbons, total phosphorus, chloride, sulfate, dissolved solids, suspended solids, total bacteria, total coli-form, chemical oxygen demand, and biochemical oxygen demand were analyzed in the laboratory, using the standard methods (APHA, 1998). Total phosphorus, chlorophyll *a*, and transparency were used to determine the trophic state of the reservoir, employing the models described by Carlson (1977).

Ca-enrichment experiment

The effects of calcium on the natural phytoplankton community in the reservoir were determined by adding calcium in the form of chloride salt reservoir samples and then evaluating the effects under laboratory conditions in the spring of 2005. Specifically, 0-16.0 mg Ca L^{-1} were added to 100 mL reservoir samples that already contained 7 mg Ca L^{-1} to give final concentrations of 7, 9, 11, 13, 15, 18, and 23 mg of calcium L^{-1}. All experiments were conducted in 250 mL culture flasks and there were 2 replicates for each treatment. The cultures were then incubated at 25°C under a 12:12 h light-dark cycle, with illumination at a level of 200 μmol photon m^{-2} s^{-1} and shaking with 60 rpm. After incubation for 1 week, the species composition in the phytoplankton assemblages in each treatment was analyzed.

Statistical analysis

The principle factors were analyzed by multiple r-square, Pearson correlation analysis and one way ANOVA using software Statistica (StatSoft, Tulsa, OK, USA, http://www.statsoft.com).

RESULTS

Changes of dominant phytoplankton over time

Phytoplankton in FTR exhibited significant seasonal succession in species composition of phytoplankton assemblages. As a rule, diatoms dominated in winter. This was followed by the dominance of chlorophytes in the spring and cyanobacteria in summer and autumn. However, this water reservoir was generally characterized by a predominance of cyanobacteria, while chlorophytes were subdominant (Figure 2).

The dominant cyanobacterial species in the FTR were of colonial form and included four *Microcystis* species, and *Aphanocapsa minutissima*. Single-celled species were rare. In the chlorophyte assemblages, the dominant species were primarily in colonial form and included *Coelastrum, Eutetramorus* (*Sphaerocystis*), *Coenocystis*, *Botryococcus*, *Oocystis*, and *Pseudoquadrigula,* which were associated with low abundance of single-celled form such as *Ankistrodesmus*, *Closteriopsis*, *Monoraphidium*, *Tetraedron, Cosmarium,* and *Staurastrum*.

There were 14 species of desmids belonging to the genera *Cosmarium, Sphaerozosma,* and *Staurastrum*. During the early stage of this study (1990 to 1995), relatively few species of desmids were observed in the FTR. However, after 1997 the number of species of

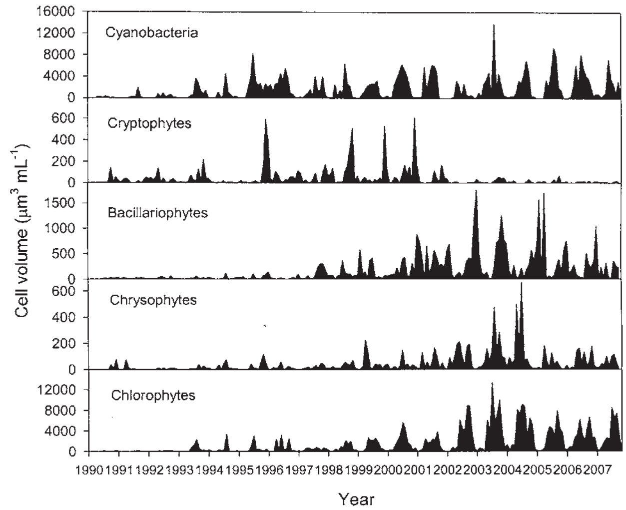

Figure 2. Alteration of the cell density (volume) of each phytoplankton group in Feitsui Reservoir over the study time.

Staurastrum (including *S. dejectum, S. excavatum, S. grallatorium,* and *S. tohopekaligense*) increased as three species of *Staurastrum* (*S. cingulum* var. *inflatum, S. perundulatum,* and *S. tetracerum*) disappeared, while the species of *Cosmarium* remained unchanged. A remarkable increase in *Staurastrum* density, particularly those of *S. dejectum, S. excavatum,* and *S. tohopekaligense* was observed.

Compared with cyanobacteria and chlorophytes, other groups of phytoplankton such as chrysophytes, cryptophytes, and dinophytes appeared only occasionally and sparsely, particularly in terms of the cell volume in waters. Figure 2 showed that the density of these phytoplankton fluctuated over time from 1990 to 2007. As a whole, the density of bacillariophytes, chrysophytes, and chlorophytes exhibited about the same tendency of change, showing increasing in density from 1990 to 2003 and decreasing after 2004, while those of cyanobacteria and cryptophytes followed a different change pattern.

Succession between Chlorophytes and *Microcystis*

Four *Microcystis* species were recorded in the FTR, *M. flos-aquae*, *M. aeruginosa*, *M. robosta*, and *M. ichthyoblade*. Sparse blooms of a mixture of these species occurred during the early stages of this study (1990 to 1995). However, the dominance of *Microcystis* spp. was replaced by chlorophytes between 1997 and 2005 (Figure 3A), which just matched with the period of time of highway construction in the watershed. The dominant chlorophytes were mainly of colonial genera such as *Eutetramorus* (*Sphaerocystis*), *Coelastrum*, and *Coenocystis*.

Prior to 1996, the density of chlorophytes was low, with yearly average of 220±85 cells·mL^{-1} or 117 $\mu m^3 \cdot L^{-1}$. However, a remarkable increase in chlorophyte density occurred over time beginning in 1997 and reaching a

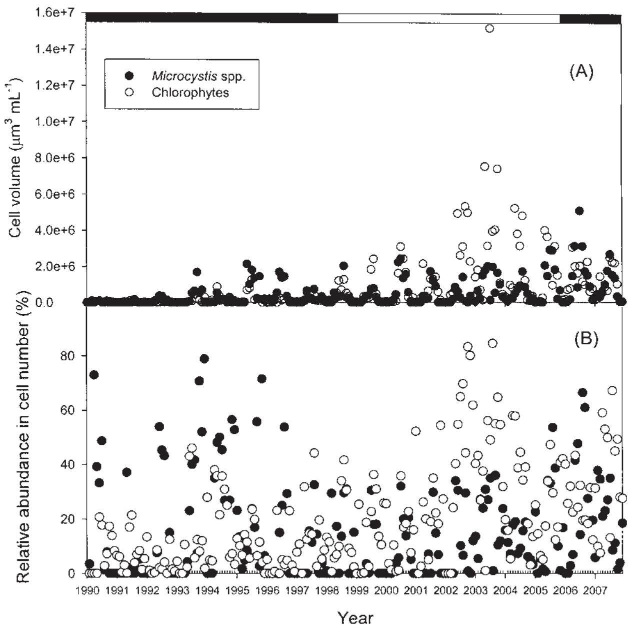

Figure 3. Succession of *Microcystis* spp. and chlorophytes in cell number (A) and cell volume (B) in Feitsui Reservoir over the study time. Filled block indicates the period of *Microcystis*-dominance, while blank one chlorophyte-dominance.

maximum of 23,063 cells·mL^{-1} or 7,541 μm^{3}·L^{-1} in the spring of 2003.

After 2005, *Microcystis* spp. again dominated over chlorophytes, particularly during the seasons of spring and summer, although chlorophytes dominated during autumn. Nevertheless, the densities of *Microcystis* spp. and chlorophytes were lower than those before 2005. Such a phenomenon of alteration in dominance was particularly pronounced when either groups of phytoplankton were plotted in terms of cell volume (Figure 3B).

Fluctuations in N- and P-resources in reservoir waters

In FTR, the concentrations of nitrite and ammonium were quite low, so that the N-nutrient for the growth of phytoplankton was contributed mainly by nitrate. The sum of nitrate, nitrite and ammonium, namely total inorganic nitrogen (TIN), fluctuated with time, displaying a seasonal variation and an increase from 1996 to 2003 and then a decline after 2004 (Figure 4). Such a change, however, did not agree with the fluctuation in the density of *Microcystis* spp. nor that of chlorophytes in FTR (cf. Figure 3).

Changes in trophic state and water quality over time

The trophic state in the FTR fluctuated throughout the study period. Specifically, the FTR was oligotrophic from 1990 to 1995, with average Carlson's TSI values of 39.2±4.5. However, the TSI values increased to an average of 45.7±5.9 from 1996 to 2004, after which they decreased to 40.5±3.7 from 2005 to 2007, suggesting that it shifted to mesotrophic state over 1996-2007.

Phosphate concentration in FTR waters was usually low (average 6.7±10.2 μg L^{-1}), though there exhibited some peaks throughout the time of this study. An increase in phosphate concentration was measured over the time of highway construction, from 1996 to 2005. After 2006, it lowered to about the same level as that prior to highway construction.

Similar to phosphate, the concentrations of total inorganic nitrogen (TIN, including nitrate, nitrite, and ammonium) before 1996 were lower than the period of highway construction, though they fluctuated to certain degrees. The ratio of TIN/P increased from 1990 to 1995 and then fluctuated after 1996 (Figure 4). Throughout the study time, the ratios of TIN/P ranged between 33 and 172 (average 107.6±31.2), suggesting that P was possibly the limiting for the growth of phytoplankton.

Total C (including inorganic and organic) in reservoir water was higher before 1996 (with average 5.08±1.54 mg L^{-1}) and lower after 1996 (average 3.74±1.36 mg L^{-1}). The ratios of TC to TIN were higher before 1995, namely prior to the construction of highway. The ratios remained low over 1996-2007.

The water hardness in the FTR occurs primarily due to the presence of calcium. The levels of water hardness

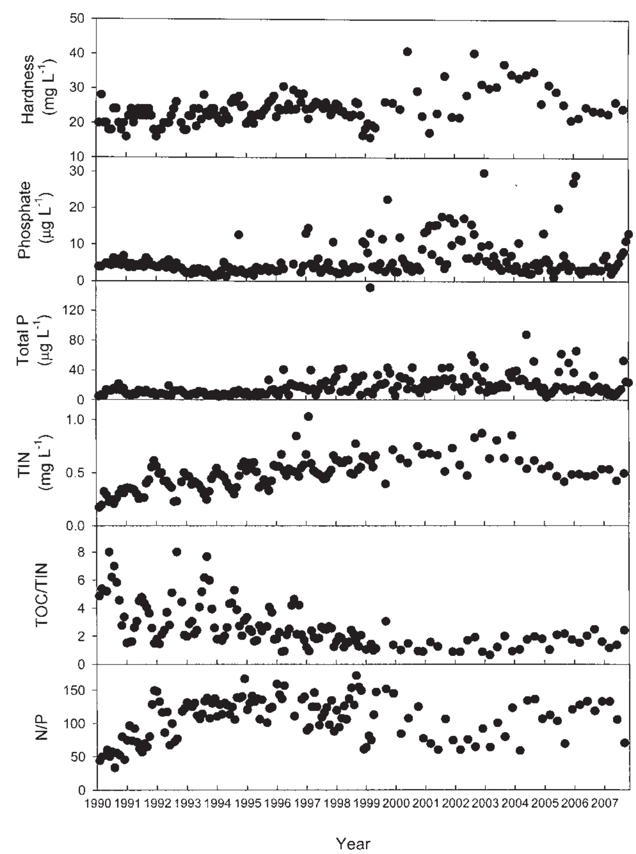

Figure 4. Changes in water hardness, concentrations of phosphate, total phosphorus, total inorganic nitrogen (TIN), the ratios of total organic carbon (TOC) to TIN and N/P in Feitsui Reservoir over the study time.

were lower from 1990 to 1995, with average values of 21.8±2.9 mg L^{-1}, being observed during this period. The water hardness increased over time from 1996 to 2004, with the highest average value (32.9±3.3 mg L^{-1}) being recorded in 2003-2004, after which the value decreased (Figure 4). The time course of changes in water hardness and calcium agreed well with the construction projects that were conducted in the watershed from 1995 to 2004.

Relationship between chlorophyte abundance and environmental factors

The abundance of *Microcystis* spp. and chlorophytes was analyzed with respect to the physico-chemical variables. The results of correlation analysis demonstrated that the abundance of chlorophytes exhibited significantly positive and higher coefficients with water hardness, calcium, sulfate, magnesium, dissolved solids, alkalinity, and total phosphorus (Table 2). These variables were all related to discharges from the construction projects, which contained clay particles and various kinds of soluble nutrients.

It was noteworthy that the abundance of chlorophytes did not display close correlation with TIN. In addition, the abundance of chlorophytes displayed a positive and significant correlation with TIC. No close correlation was measured between chlorophyte abundance and TOC (cf. Table 2).

Compared with chlorophytes, the abundance of *Microcystis* spp. neither exhibited close correlation with water hardness nor cations. Instead, it was correlated with pH, water temperature, dissolved solids, conductivity, and total organic carbon, showing different nutrient demands in either group of phytoplankton.

Ca-enrichment experiment

A nutrient enrichment experiment was conducted to determine the role that calcium played in inducing changes in the composition of the phytoplankton assemblages. As shown in Figure 5, enrichment with calcium resulted in alteration of the composition of the phytoplankton assemblages. As a result, the density of chlorophytes in terms of cell number as well as cell volume in the phytoplankton assemblages was enhanced as the calcium levels were elevated from 2 to 8 mg L^{-1}. No further enhancement was observed above 8 mg L^{-1}. In addition, there was a decline in cyanobacterial density, mainly composed of *Microcystis* spp. and *Aphanocapsa delicatissima,* that was associated with the elevation in chlorophytes density. The species of chlorophytes which were enhanced by elevated calcium concentrations were of the genera of *Eutetramorus*, *Coelastrum*, *Coenocystis*, and *Dictyosphaerium*. Enrichment with calcium also caused changes in the density of other algae, such as diatoms, cryptomonoids, dinophytes, and chrysophytes. However, these changes were too small to play an important role in the phytoplankton community.

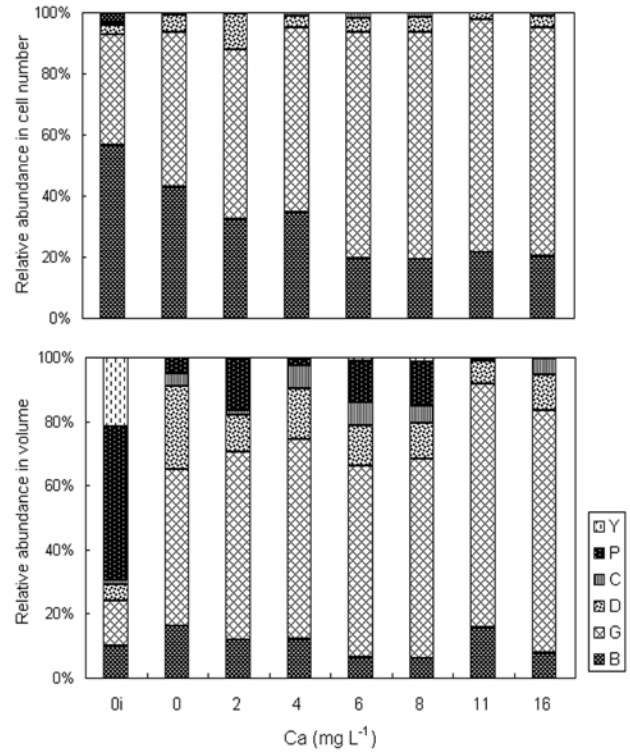

Figure 5. Changes in the relative abundance in cell number and cell volume of each phytoplankton group in response to enrichment with various concentrations of calcium in natural waters of Feitsui Reservoir and incubation under laboratory culture conditions. Ca 0_i: control set at initial of experiment; Ca 0-16: treatment sets measured at 6^{th} day with Ca in the enclosures from 0 to 16 mg L^{-1}, respectively. B: cyanobacteria; C: cryptomonoids; D: diatoms; G: green algae; Y: chrysophytes.

DISCUSSION

The growth of phytoplankton in lakes or reservoirs is dependent upon a variety of factors. Attempts have been made to demonstrate a relationship between the distribution and periodicity of the phytoplankton and variations in the characteristics of the dissolved substances present in various aquatic systems (Pearsall, 1932). In principle, nutrients in the reservoir surface could be released from the hypolimnion. FTR is a subtropical warm monomictic deep reservoir. In this type of reservoir, the atelomixis process, as mentioned by Barbosa and Padisák (2002), Tavera and Mertínez-Almeida (2005), and Souza et al. (2008), might play an important role in seasonal succession in the composition of phytoplankton assemblages. However, the density of total phytoplankton in the aquatic environment is usually determined by total amount of available nutrients. It is unlikely that the elevation of chlorophytes density in FTR is attributed to such a process.

Upon the introduction of chemical variables to the aquatic environment, macro-elements such as C, N, and P play an important role in the growth and succession of

Table 2. Correlation coefficients between the abundance of *Microcystis* spp., chlorophytes and the physico-chemical variables in Feitsui Reservoir waters over the time of this study.

	Mic	Grn	SD	Temp	Turb	Alk	pH	Cl	SO_4^{2-}	NH3	NO_3^-	DO	BOD	COD	DS	SS	EC	Hard	Ca	Mg	Fe	Mn	Bact	Ecol	TOC	PO_4^{3-}
Grn	-0.31*																									
SD	-0.16	0.04																								
Temp	0.39*	0.46*	-0.33*																							
Turb	-0.21	-0.10	-0.43*	-0.09																						
Alk	0.13	0.34*	0.28	0.30	-0.36*																					
pH	0.44*	-0.06	-0.28	0.55*	-0.11	0.08																				
Cl	0.12	-0.16	0.05	-0.05	-0.08	0.07	0.22																			
SO_4^{2-}	0.02	0.45*	-0.04	0.29	-0.12	0.31*	-0.06	0.10																		
NH_3	-0.17	0.14	0.43*	0.01	-0.09	0.02	-0.16	-0.09	-0.12																	
NO_3^-	-0.19	0.24	0.09	-0.23	0.16	0.13	-0.26	-0.18	0.17	-0.03																
DO	-0.12	-0.30	0.07	-0.42*	0.01	-0.15	0.10	0.21	-0.16	-0.08	0.00															
BOD	0.30	-0.01	-0.16	0.28	-0.04	0.05	0.26	0.01	-0.01	-0.06	0.01	0.10														
COD	-0.13	0.06	0.43*	0.01	-0.08	0.02	-0.10	-0.07	-0.11	0.78*	0.01	-0.05	-0.19													
DS	0.35*	0.66*	0.26	0.29	-0.16	0.43*	-0.21	-0.13	0.55*	0.21	0.30	-0.36*	-0.06	0.18												
SS	0.25	-0.10	-0.44*	-0.05	0.90*	-0.32*	-0.10	-0.13	-0.09	-0.11	0.15	-0.02	-0.05	-0.12	-0.13											
EC	0.42*	0.17	-0.09	0.27	-0.29	0.55*	0.18	0.10	0.43*	-0.20	0.15	-0.18	0.08	-0.22	0.31*	-0.17										
Hard	0.12	0.56*	0.08	0.27	-0.17	0.42*	0.00	0.01	0.51*	-0.09	0.31*	-0.16	0.01	-0.08	0.70*	-0.13	0.51*									
Ca	-0.04	0.53*	0.08	0.20	-0.12	0.42*	-0.11	-0.15	0.55*	-0.07	0.42*	-0.22	-0.04	-0.05	0.72	-0.13	0.38*	0.81*								
Mg	0.20	0.36*	0.05	0.24	-0.12	0.19	0.07	0.17	0.31*	-0.07	0.08	-0.07	0.05	-0.12	0.38*	-0.05	0.43*	0.68*	0.23							
Fe	0.08	-0.09	-0.36*	0.08	0.51*	-0.10	0.10	0.37*	-0.15	-0.08	0.08	0.04	0.30	-0.07	-0.14	0.47*	-0.06	-0.08	-0.09	-0.04						
Mn	0.00	0.03	0.14	-0.08	-0.01	0.20	-0.10	0.05	0.28	-0.01	0.19	-0.01	-0.06	-0.03	0.27	-0.02	0.13	0.16	0.27	-0.07	-0.09					
Bact	-0.18	-0.06	-0.16	0.03	0.07	-0.15	-0.08	0.09	-0.01	-0.06	-0.15	0.06	0.05	-0.06	-0.11	0.04	-0.13	-0.11	-0.12	-0.03	-0.03	-0.08				
Ecol	0.05	-0.01	-0.01	0.07	0.06	0.04	0.07	0.15	-0.10	-0.05	0.03	0.02	0.10	-0.07	0.06	0.05	0.12	0.13	-0.03	0.22	0.00	-0.08	0.35			
TOC	0.31*	0.16	-0.02	0.33*	-0.21	0.23	0.26	-0.09	0.24	0.00	-0.36*	-0.04	0.01	-0.07	-0.06	-0.18	0.21	0.02	0.07	0.05	-0.29	-0.11	0.04	-0.10		
PO_4^{3-}	-0.14	-0.06	-0.14	-0.20	0.08	-0.34*	-0.11	-0.10	-0.05	-0.01	0.15	0.07	-0.04	-0.04	-0.11	0.27	-0.19	-0.18	-0.07	-0.16	0.00	-0.02	0.00	-0.02	-0.04	
TP	-0.10	0.27	-0.15	-0.01	0.11	-0.18	-0.03	-0.10	0.13	-0.08	0.27	-0.03	-0.06	-0.06	0.13	0.30	-0.05	0.11	0.20	0.04	0.04	-0.04	-0.02	0.02	-0.02	0.78*

Abbreviations: Mic: *Microcystis* spp.; Grn: chlorophytes; SD: Secchi depth; Temp: water temperature; Turb: turbidity; Alk: alkalinity; BOD: biochemical oxygen demand for 5 days; COD; chemical oxygen demand; DO: dissolved oxygen; DS: dissolved solid; SS: suspended solid; EC: conductivity; Hard: water hardness; Bact: total bacteria; Ecol: *E. coli*; TOC; total organic carbon; TP: total phosphorus. *: significant at $p<0.05$, n=175.

phytoplankton (Riegman, 1998). However, it is showed in the present study that the change patterns of N- and P-resources in FTR did not well match with the time of increase in chlorophyte density (cf. Figure 3). Also the analysis of correlation did not exhibit a close correlation between chlorophyte density and N- or P-resource (cf. Table 2). Thus, it is possible that the increase in chlorophytes in FTR is not a result of elevation in N- or P-level, but water hardness in reservoir waters.

Water hardness is the sum of calcium and magnesium ions present in the water. Similar to the majority of aquatic environments, calcium is the primary contributor to water hardness in the FTR (cf. Table 1). Remarkable changes were observed in the hardness of the reservoir water over the course of the present study. These changes were attributed to construction events conducted in the watershed between 1995 and 2004. During the construction of a wastewater treatment plant and a highway in the watershed, a large amount of cement was used. It is known that cement contains approximately 60% calcium oxide (Carles-Gibergues and Cyr, 2002). Following the aggregation reaction that occurs in cement, a small portion of calcium salt dissolved in water as well as clay particles were likely discharged to the environment, thereby inducing an elevation in calcium levels in the receiving waters. Other than cement, the wastewater discharged during excavation and tunnel drilling contained a considerable amount of inorganic minerals, including calcium and magnesium. These minerals, together with those from cement, would contribute to the total calcium and hardness of receiving waters of FTR. Thus, this is the possible reason why the hardness of FTR waters increased with time during the construction of highway and decreased following its completion in 2005.

One important role of calcium is its effect on pH and the CO_2-HCO_3^- system. Calcium ions enter the lake from the inflowing rivers, after which they react with the most abundant inorganic carbon, bicarbonate, to form soluble $Ca(HCO_3)_2$. This is then converted to calcium carbonate and carbon dioxide, which is taken up by algae during photosynthesis. Therefore, any change in calcium concentration would alter the carbon dioxide supply and be favorable for the growth of those species that have higher utilization efficiency when competing for carbon dioxide. Thus, calcium can have a selective influence among phytoplankton that have different sensitivities to pH and carbon sources. The results of laboratory and field studies conducted by King (1970) suggested that the dominance of cyanobacteria/chlorophytes was controlled by CO_2 (i.e. alkalinity). Possibly, the elevated calcium level in waters would affect the availability of C-source for the growth of algae.

It is also possible that calcium affects the growth of algae through its effect on P-availability in waters. Redfield (1958) noted that phytoplankton contained C, N and P in a ratio that was fairly constant with respect to the content of C as a result of the incorporation of these elements during photosynthesis and growth. Redfield's ratio allows us to compare the importance of various elements for the growth and production of phytoplankton. Broecker (1974) found that a fairly constant relationship existed between major elements and calcium during the biotic utilization of these elements and recalculated Redfield's ratio to include calcium carbonate. As a matter of fact, the amount of calcium in waters could affect the availability of these macro-elements, particularly P, because calcium reacts with phosphate to form insoluble calcium-phosphate compound. Increase in calcium concentration in waters would lead to enhanced precipitation of phosphate. As a result, this would give rise to a reduction in phosphate availability for phytoplankton growth. Thus, change in calcium concentrations would exert a selection force for species that grow well under the prevailing conditions.

It was reported by some authors that dominance of desmids was observed in soft waters in temperate climate zones (Moss, 1973; Woelkerling and Gough, 1976; Tavera and Martinez-Almeida, 2005). FTR is also a soft water mountainous water reservoir. In the present study, we found a succession in desmid species when water hardness/calcium of reservoir waters increased. Nevertheless, the density of desmids in FTR was quite low (less than 0.7% in the phytoplankton assemblages in average), although it was observed that there was a slight increase in desmid density when water hardness was elevated. For certain desmids, calcium is known to be involved in the development of the cell envelope. During morphogenesis of the cell walls, vesicles related to primary wall formation in desmids preferentially fuse at plasma membrane sites of Ca accumulation (Kiermayer and Meindl, 1989). Thus, they require a specific and finite concentration of calcium for their growth. This might explain the change in the density of desmids observed in the FTR when the calcium levels were elevated.

Addition of calcium (in the form of calcium hydroxide) had been used to control algae in eutrophic waters (Cervenka et al., 1980). However, in the present study, elevation of calcium levels and water hardness were too low to result in the death of any algae or cyanobacteria. It is well known that changing the nutrient composition leads to the selection of species that grow well under prevailing conditions (Sakshaug et al., 1983). As a response to an elevation in water hardness (or calcium levels) in the reservoir water, the composition of the phytoplankton community was altered. The results of the enrichment experiment confirm that an elevation in calcium levels was favorable for chlorophytes in the FTR.

In calcium enrichment experiment, the abundance of some colonial chlorophyte species, particularly of genera *Eutetramorus* (*Sphaerocystis*), *Coelastrum*, and *Coenocystis*, was enhanced in response to elevating calcium concentrations. During the highway construction period, it was the same group of chlorophytes which appeared to be dominant over others in phytoplankton assemblages in reservoir waters. Thus, the enrichment experiment provides another evidence of a favoring effect of calcium on the growth of chlorophytes.

The dominance of cyanobacteria could be related to their buoyancy mechanism (Reynolds et al., 1987). *Microcystis* is known to produce gas vesicles that allow cells to migrate vertically in the water column to obtain nutrients and a favorable light source (Walsby, 1994). Thus, it is best adapted to systems with a stable water column such as the FTR. However, the turbid clay particles discharged from the construction projects in the watershed are unfavorable for buoyant species such as *Microcystis* because the clay particles are especially effective at attaching to their mucilaginous covering. This would result in flocculation of the cyanobactaria, thereby favoring chlorophytes (Cuker et al., 1990). In our study, changes in the density of chlorophytes in the FTR were closely correlated with changes in the concentration of soluble solids and total phosphorus. Thus, clay particles discharged from construction sites might have played a role in regulating the changes in the dominance of cyanobacteria and chlorophytes in the phytoplankton community, particularly during the highway construction period.

CONCLUSIONS

The results of this study indicate that the elevation in water hardness that was observed in the FTR occurred as a result of discharges from the construction of a wastewater treatment plant and a highway in the watershed. These changes were associated with succession from a *Microcystis*-dominated to a chlorophytes-dominated phytoplankton community. Furthermore, the increase in the density of chlorophytes was found to closely correlate with water hardness, soluble solids, calcium, sulfate, magnesium, and alkalinity in the water. Finally, experiments evaluating enrichment with calcium confirmed that an elevation in calcium levels would enhance the growth of chlorophytes. The present study suggested that the elevated water hardness has played a selecting force, possibly through altering C- and P-availabilities in waters, in affecting the dominance of phytoplankton.

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LITERATURE CITED

American Public Health Association (APHA). 1998. American Water Works Association Water Environmental Federation. Standard Methods for Examination of Water and Wastewater. 20th ed. Washington DC, APHA.

Barbosa, F.A.R. and J. Padisá. 2002. The forgotten lake stratification pattern: atelomixis and its ecological importance. Verh. Internat. Verein. Limnol. **28:** 1385-1395.

Broecker, W.S. 1974. Chemical Oceanography. Harcourt Brace Jovanovich.

Carles-Gibergues, A. and M. Cyr. 2002. Interpretation of expansion curves of concrete subjected to accelerated alkali-aggregate reaction (AAR) tests. Cement Concrete Res. **32:** 691-700.

Carlson, R.E. 1977. A trophic state index for lakes. Limnol. Oceanogr. **22:** 361-369.

Carmichael, W.W., S.M.F.O. Azvedo, J.S. An, R.J.R. Molica, E.M. Jochimsen, S. Lau, K.L. Rinehart, G.R. Shaw, and G.K. Eaglesham. 2001. Human fatalities from cyanobacteria: chemical and biological evidence for cyanotoxins. Environ. Health Perspect. **109:** 663-668.

Cervenka, L., M.G. Eissa, J. Barica, and L. Mur. 1980. Algenbekämpfung durch Zusatz von Kalk- und Dolomithydrat. Develop. Hydrobiol. **2:** 343-344.

Chorus, I. and J. Bartram. 1999. Toxic Cyanobacteria in Water. E. & FN Spon, London, 416 pp.

Cuker, B.E., P. Gama, and J.M. Burkholder. 1990. Type of suspended clay influences lake productivity and phytoplankton community response to phosphorus loading. Limnol. Oceanogr. **35:** 830-839.

Huisman, J., H.C.P. Matthijs, and P.M. Visser. 2005. Harmful Cyanobacteria. Springer, Dordrecht, 241 pp.

Kiermayer, O. and U. Meindl. 1989. Cellular morphogenesis: the desmid (Chlorophyceae) system. *In* A.W. Coleman, L.J. Goff, and J.R. Stein-Tayler (eds.), Algae as Experimental System. Alan R. Liss, Inc., New York, pp. 149-167.

Kim, H.G. 2008. Mitigation and controls of HABs. *In* E. Granéli and J.T. Turner (eds.), Ecology of Harmful Algae. Springer-Verlag, Berlin, pp. 327-338.

King, D.L. 1970. Role of carbon in eutrophication. J. Water Pollut. Contr. Fed. **42:** 2035-2051.

Liu, C.J. 1995. Study of microcystins isolated from local *Microcystis aeruginosa*. Master Thesis, Soochow University, Taipei, Taiwan, 97 pp.

Moss, B. 1973. The influence of environmental factors on distribution of freshwater algae: an experimental study; II. The role of pH and the carbon dioxide-bicarbonate system. J. Ecol. **61:** 172-192.

Pearsall, W.H. 1932. Phytoplankton in the English lakes: The composition of the phytoplankton in relation to dissolved substances. J. Ecol. **20:** 241-262.

Redfield, A.C. 1958. The biological control of chemical factors in the environment. Amer. Scient. **46:** 205-221.

Reynolds, C.S., R.L. Oliver, and A.E. Walsby. 1987. Cyanobacterial dominance: The role of buoyancy regulation in dynamic lake environments. New Zealand J. Mar. Freshwat. Res. **21:** 379-390.

Riegman, R. 1998. Species composition of harmful algal blooms in relation to macronutrient dynamics. *In* D.M. Anderson, A.D. Cembella, and G.M. Hallegraeff (eds.), Physiology of Harmful Algal Blooms. Vol. 41. NATO ASI Series, Springer-Verlag, Berlin, pp. 475-488.

Sakshaug, E., K. Andersen, S. Myklestad, and Y. Olsen. 1983. Nutrient status of phytoplankton communities in Norwegian waters (marine, brackish and fresh) as revealed by their chemical composition. J. Plankton Res. **5:** 175-196.

Souza, M.B., C.F.A. Barros, F. Barbosa, E. Hajnal, and J. Pádisak. 2008. Role of atelomixis in replacement of phytoplankton assemblages in Dom Helvécio Lake, South-East Brazil. Hydrobiologia **607:** 211-224.

Tavera, R. and V. Mertínez-Almeida. 2005. Atelomixis as a possible driving force in the phytoplankton composition of Zirahuén, a warm-monomictic tropical lake. Hydrobiologia **533:** 199-208.

Walsby, A.E. 1994. Gas vesicles. Microbiol. Rev. **58:** 94-144.

Woelkerling, W.J. and S.B. Gough. 1976. Wisconsin desmids. III. Desmid community composition and distribution in relation to lake type and water quality. Hydrobiologia **51:** 3-32.

Wu, J.T. and Y.M. Hsu. 1988. Relation of algae to earthy odors of fish in Taiwan. Bot. Bull. Acad. Sin. **29:** 183-188.

Wu, J.T. and L.T. Kow. 2002. Long-term monitoring of the phytoplankton and water quality in Feitsui Reservoir. Technical Report, Feitsui Reservoir Administration Bureau, Taipei, Taiwan, 86 pp.

Wu, J.T. and L.T. Kow. 2008. Monitoring of the relationship between phytoplankton and water quality in Feitsui Reservoir. Technical Report, Feitsui Reservoir Administration Bureau, Taipei, Taiwan, 141 pp.

翡翠水庫水硬度改變造成之浮游藻的群落變化

吳俊宗[1,2] 高麗珠[1]

[1] 中央研究院 生物多樣性研究中心

[2] 國立臺灣大學 生態與演化生物學研究所

位於亞熱帶之翡翠水庫興建於 1986 年，它供應大臺北地區自來水之水源。自 1990 至 2007 年間之長期監測發現，水庫浮游藻之優勢藻群，特別是綠藻和微囊藻間有明顯的變遷。由分析 27 項水質理化參數發現，藻群間之消長與其各自對營養鹽之需求不同有關。相關分析顯示，綠藻之增加與鈣、鎂、溶解固體、硫酸鹽等參數所造成之水硬度增高有關，而這些參數值之增高推測係因調查期間在水庫上游有高速公路施工所造成之污染所造成。本研究在實驗室進行鈣添加試驗，其結果支持鈣離子增高確實會造成綠藻類的增加，且所增加的優勢種類也和水庫內因水硬度增高而變多的藻種相同。本文進一步討論鈣如何影響藻類對碳和磷的利用，並提供處理水庫微囊藻問題的策略。

關鍵詞：翡翠水庫；鈣添加；浮游藻；亞熱帶水庫；水硬度。