Hydrobiologia (2008) 610:161–173
DOI 10.1007/s10750-008-9431-z

PRIMARY RESEARCH PAPER

Phytoplankton as a monitoring tool in a tropical urban shallow reservoir (Garças Pond): the assemblage index application

Luciane O. Crossetti · Carlos E. de M. Bicudo

Received: 4 August 2007 / Revised: 5 May 2008 / Accepted: 13 May 2008 / Published online: 28 June 2008

Abstract This study aimed at evaluating phytoplankton as a monitoring tool for water quality assessment in an urban shallow eutrophic reservoir considering temporal and vertical scales. Garças Reservoir is located in the Parque Estadual das Fontes do Ipiranga Biological Reserve (23°38′08″S and 23°40′18″S; 46°36′48″W and 46°38′00″W) that lies in the southeastern part of the Municipality of São Paulo, southeast Brazil. Samplings were carried out monthly during 8 consecutive years (1997–2004) following the water column vertical profile (5 depths: subsurface, 1, 2, 3 m and ~20 cm from the bottom). Abiotic variables analyzed were: water temperature, electric conductivity, DO, pH, total alkalinity, free CO_2, dissolved inorganic carbon, N series, P series and SiO_4H_4. Biological variables studied were: total density, total biomass and chlorophyll *a*, which were integrated arithmetically. At the beginning of the 8 year series, Garças Reservoir was an eutrophic ecosystem with 20% of its surface covered by *Eichhornia crassipes* (phase I: January 1997–March 1998). Water hyacinth reached 70% of pond surface coverage (phase II: April 1998–August 1999), and then it was mechanically removed (phase III: September 1999–December 2004). After this intervention, drastic alteration in the limnological features was detected, leading to the conclusion that removal of the aquatic macrophyte modified nutrient dynamics drastically reduced water transparency and led to photosynthetic productivity and phytoplankton biomass increase, the latter becoming a physical barrier to light penetration. Twenty one functional groups '*sensu*' Reynolds were identified. Cyanobacteria contribution played the main role during the drastic alterations that occurred after water hyacinth removal. Results of ecological status of reservoir using Q index showed statistical difference among the 3 limnological phases (one way ANOVA; $F = 119.4$; $P = 0.000$). Regarding Q index classification, Garças Reservoir limnological phases were characterized as follows: (1) phase I: $0 \geq Q \leq 2.9$, medium to bad; (2) phase II: $1.4 \geq Q \leq 3$, tolerable to medium; and (3) phase III: $0 \geq Q \leq 1.5$, bad to tolerable ecological states.

Keywords Assemblage index · Phytoplankton · Functional groups · Cyanobacteria · *Eichhornia crassipes* · Water quality · Eutrophication · Tropical shallow lake

---

Handling editor: J. Padisak

L. O. Crossetti
Department of Limnology, University of Pannonia,
Egyetem u. 10, 8200 H-Veszprem, Hungary
e-mail: lcrossetti@terra.com.br

L. O. Crossetti · C. E. de M. Bicudo (✉)
Ecology Section, Instituto de Botânica, Caixa Postal 3005,
01061-970 Sao Paulo, SP, Brazil
e-mail: cbicudo@terra.com.br

Introduction

Temporal variability, structure and dynamics of phytoplankton community are of utmost importance to aquatic ecosystems' metabolism. As responses to the interactive processes of physical, chemical and biological variables, abundance and specific composition reorganization of the phytoplankton community are frequently observed.

Phytoplankton community has already been very adequately identified for detection of sporadic events and also long term variations (Barbosa & Padisák, 2004) not only regarding species composition, but also in terms of biomass. Use of species or above species phytoplankton categories for water quality determination has already a long history (Padisák et al., 2006).

Reynolds et al. (2002) proposed a functional classification scheme of phytoplankton species, which sorts species with morphological, physiological and ecological similarities into functional groups labelled by alpha-numeric characters. Many studies have already proved the explanatory and predictive manner of functional groups (e.g. Marinho & Huszar, 2002; Kruk et al., 2002; Mischke & Nixdorf, 2003; Albay & Akçaalan, 2003; Morabito et al., 2003; Nixdorf et al., 2003; Naselli-Flores & Barone, 2003; Naselli-Flores et al., 2003; Mischke, 2003; Rojo & Álvarez-Cobelas, 2003; Silva, 2004; Crossetti & Bicudo, 2005; Fonseca & Bicudo, 2008) by describing the abiotic characteristics of the environments in which they occur. Following this approach, tests using phytoplankton have been developed to determine the water quality (Padisák et al., 2006).

Padisák et al. (2006) developed the Q index that provides 5 degrees of water quality through phytoplankton functional groups (Reynolds et al., 2002). Q index was developed to assess the ecological status of different lake types established by the Water Framework Directive (2000; WFD hereafter) proposed by the European Community. WFD is a broad concept that aims at developing sustainable management strategies for ground and surface waters in Europe (Padisák et al., 2006). The index takes into account the relative weight of functional groups in the total biomass, as well as a factor number determined for each type of water body. Although the proposal is very recent and until now no study has been carried on to show the Q index utilization, it can be applied for environment ecological evaluation with no geographic limitation (Padisák et al., 2006).

Differently from any other existing one, the Q index of functional groups gives no preference to any kind of human impact in particular, which considerably increases its application spectrum (Padisák et al., 2006). Finally, the Q index is also adequate to test monitoring ecosystems based on habitat concepts (Padisák et al., 2006).

The present study aimed at testing the Q index as a tool for water quality evaluation using a phytoplankton long term series (8 years) in Garças Reservoir, thus contributing to the discussion on phytoplankton as an indicator group of water quality.

Study area

Garças Reservoir is located in the Parque Estadual das Fontes do Ipiranga Biological Reserve located in the southeast region of the municipality of São Paulo (23°38′08″S and 23°40′18″S; 46°36′48″W and 46°38′00″W). Its mean altitude is 798 m and the total area is 526.4 ha (Fernandes et al., 2002). The reserve is one of the few remaining patches of Atlantic Rain Forest in the midst of a densely urbanized region of the city of São Paulo and one of the largest metropolitan green areas in Latin America.

Although locally called Garças Pond, the ecosystem is, in fact, a reservoir recently classified as eutrophic (Bicudo et al., 2002b). With its 4.7 m maximum depth, the reservoir is the most man affected ecosystem in the reserve. Its surface area is 88,156 m^2, maximum length is 512 m, maximum width is 319.5 m and the mean retention time is 71 days (Bicudo et al., 2002a). Garças Reservoir has a single exit and seven tributaries, four of which carry sewage without any kind of treatment and only one is considered oligotrophic (Carmo et al., 2002).

The Garças Reservoir's mixture type is discontinuous warm polymictic according to Lewis' (1983) model (Bicudo et al., 2002b).

A recent temporal series (8 years) study of limnological data of Garças Reservoir identified 3 phases characterized by their physical, chemical and biological features, considering the water hyacinth cover influence on the system (Bicudo et al., 2006).

1. *Phase I* (from January 1997 to March 1998): characterized by a gradual increase of the water hyacinth cover, which reached 10–20% of the water surface, with cyanobacterial blooms restricted to the spring (Fonseca & Bicudo, 2008), and high chlorophyll *a* and pH values in the surface (Bicudo et al., 2006).
2. *Phase II* (from April 1998 to August 1999): characterized by the rapid macrophyte proliferation, that covered 40–70% of the reservoir surface, favored by the increasing of allochthonous nutrients input (N, P). Extensive macrophyte banks accounted for a very serious mosquito proliferation (*Mansonia* sp.) that called for mechanical removal of the macrophytes. This phase was characterized by low algal biomass, increasing transparency and DO (dissolved oxygen), SRP (soluble reactive phosphorus) decrease and the increased physical stability of the ecosystem (Bicudo et al., 2006).
3. *Phase III* (from September 1999 to December 2004): characterized by the water hyacinth cover reduced to only 10% of the water surface, retained by wire screens. Removal totalled 3,100 m^3 of the macrophyte (Bicudo et al., 2007). During this phase, an abrupt limnological change in Garças Reservoir was registered, represented by an increase of chlorophyll *a*, pH, TP and SRP values, and a dramatic decrease of transparency, free CO_2 and low DO values, mainly at the deepest layers of the reservoir (Bicudo et al., 2006).

Material and methods

Samplings were performed monthly over eight consecutive years (1997–2004), at five depths (subsurface, 1, 2, 3 m and $\sim$20 cm from the bottom) always in the morning at the deepest site of reservoir. Water samples ($n = 2$) were collected with a van Dorn sampler and transferred to acid-rinsed bottles. Samples were filtered for the dissolved nutrients analyses under low pressure (< 0.3 atm) using Whatman GF/F membrane filters. In the field, water temperature, pH and conductivity were measured using standard electrodes. Water transparency was determined using a Secchi disk.

The following variables were also determined at the sampling day: dissolved oxygen (DO) (Winkler modified by Golterman et al., 1978), alkalinity (Golterman & Clymo, 1971), free CO_2, $HCO_3{}^-$ and CO_3^- (Mackereth et al., 1978), soluble reactive phosphorus (SRP) and total dissolved phosphorus (TDP) (Strickland & Parsons, 1960), total phosphorus (TP) (Valderrama, 1981), nitrite ($NO_2{}^-$) and nitrate ($NO_3{}^-$) (Mackereth et al., 1978), ammonium ($NH_4{}^+$) (Solorzano, 1969), total nitrogen (TN) (Valderrama, 1981) and soluble reactive silica (SRS) (Golterman et al., 1978).

Trophic state index values were taken from Bicudo et al. (2006). Indexes used were Carlson's Trophic State Index (TSI) modified by Toledo (1990) and Lamparelli (2004). The two latter authors adapted the original Carlson's TSI for tropical systems. Toledo (1990) basically promoted alteration on the transparency, chlorophyll *a* and total phosphorus equations, considering them equivalent in the mean TSI. Lamparelli (2004) altered only chlorophyll *a* and total phosphorus TSI equations. Lamparelli (2004) used a large database supplied by CETESB (State of São Paulo Environmental Technology and Sanitation Company), which refined the index (Bicudo et al., 2006) and was, consequently, used for the Garças reservoir trophic evaluation.

Biological data from the year 1997 were taken from Fonseca & Bicudo (2008).

For determination of chlorophyll *a* concentration corrected for phaeophytin, pigment extraction was accomplished with ethanol 90% as an organic solvent (Sartory & Grobbelaar, 1984; Wetzel & Likens, 2000; Pápista & Böddi, 2002; Wasmund et al., 2006; Huang & Cong, 2007).

Phytoplankton, and especially cyanoprokaryotes, were identified with the possible most recent taxonomic literature (e.g. Azevedo et al., 1996; Azevedo & Sant'Anna, 2003; Komárek & Anagnostidis, 1986, 1989, 1999, 2005; Komárek & Azevedo, 2000; Komárek & Fott, 1983; Komárková-Legnerová & Cronberg, 1994; Sant'Anna & Azevedo, 2000; Sant'Anna et al., 1989).

Phytoplankton quantification followed Utermöhl (1958) and sedimentation time was set according to Lund et al. (1958). Biomass (mg l^{-1}) was estimated using the biovolume calculated values and multiplying each species' density by the mean volume of its cells considering, whenever possible, the mean dimension of 30 individuals of each species following Sun & Liu (2003), Hillebrand et al. (1999) and Fonseca (2005).

Biological data were integrated arithmetically ($mm^3\ m^{-2}$). Reynolds' functional groups approach was applied to each species biomass ($mg\ l^{-1}$). Using biological data, Q index (Padisák et al., 2006) was applied considering the following 5 degrees classification: 0–1: bad; 1–2: tolerable; 2–3**:** medium; 3–4: good and 4–5: excellent.

For comparison among limnological phases, variance analysis (one-way ANOVA) was employed using the software MINITAB (version 14.1).

Results

Abiotic variables

Temporal and spatial study of water temperature clearly showed alternation between stratification (rainy periods) and mixing periods (dry periods) (Fig. 1). However, during phase II, in which the macrophytes almost entirely covered the reservoir surface, the greatest water transparency and depth of the mixing (Z_{mix}) and euphotic zones (Z_{eu}) were registered. From the beginning of phase III, these values considerably decreased, except for the ones registered during the winter months of 2004. Considering the other limnological features, phases were characterized as follows:

- *Phase I*: greatest DO gradients (Fig. 2) were registered during the stratification months (mainly September 1997). A substantial increase of CO_2 was observed in the entire water column in the end of phase I as well as the greatest values of $N\text{-}NH_4^+$, especially during the period of thermal stratification, and anoxia occurred at the bottom of the reservoir. Molar total N:P and dissolved N:P ratios oscillated and long periods of P limitation were observed.
- *Phase II*: DO distribution was more homogeneous; however, during the mixing period and the entire phase II, the lowest values of $N\text{-}NH_4^+$ were registered. SRP concentrations were, almost always, below the detection limit of the used analytical method during phases I and II. CO_2 increase was observed in the entire water column in the beginning of this phase.
- *Phase III*: in spite of the greatest DO values observed during the spring months ($16.5\ mg\ l^{-1}$) at the surface in September 2001 (Fig. 2), long lasting periods of anoxia at the deepest layers of reservoir were evident. With the increase of pH, there was also a drastic decrease in the free CO_2 availability and an increase in CO_3^- and HCO_3^- concentrations. TN substantially increased, especially at the deepest layers of reservoir, its greatest concentration being detected at the bottom of the lake in April 2001 ($74{,}207\ \mu g\ l^{-1}$). After macrophyte removal, a steady increase of SRP was registered in the deepest layers of the reservoir, mainly during the thermal stratification periods (Fig. 3). The highest concentrations of the nutrients above were 703.1 and $549.9\ \mu g\ l^{-1}$, respectively, at the bottom of reservoir in December 2003 and at 3 m depth in November 2003. TP followed a similar trend, however, with an increase in its concentration at the upper layers of the reservoir during the thermal stratification periods. The highest TP concentration was measured at the bottom of the reservoir in December 2000 and at its surface in July 2000 (respectively, $1{,}588\ \mu g\ l^{-1}$ and $979.7\ \mu g\ l^{-1}$). During some stratification months of phase III and mostly

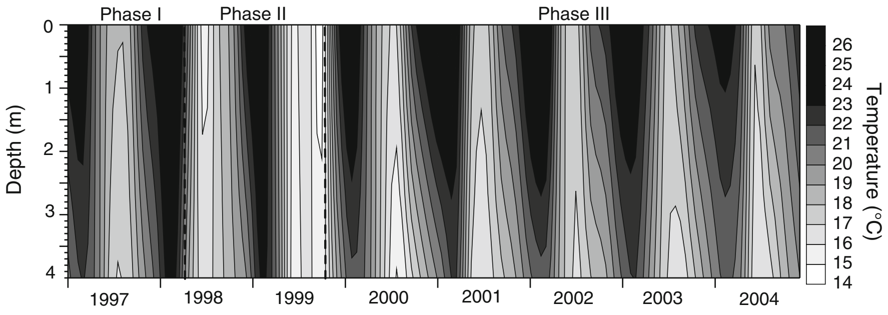

Fig. 1 Depth–time diagram of water temperature (°C) during the period 1997–2004 at Garças Reservoir. Dashed lines indicate limits of the three limnological phases

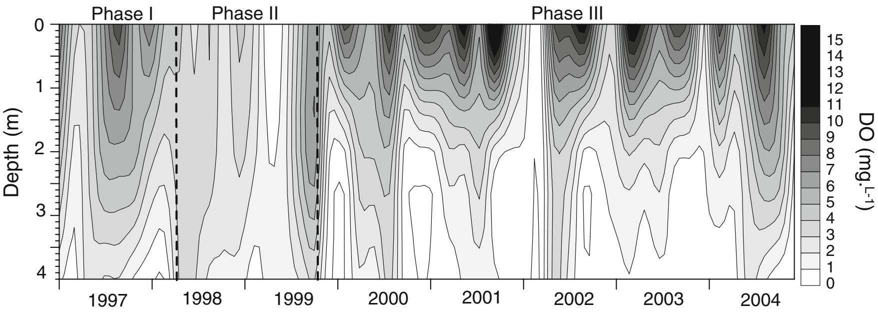

Fig. 2 Depth–time diagram of dissolved oxygen (mg l^{-1}) during the period 1997–2004 at Garças Reservoir. Dashed lines indicate limits of the three limnological phases

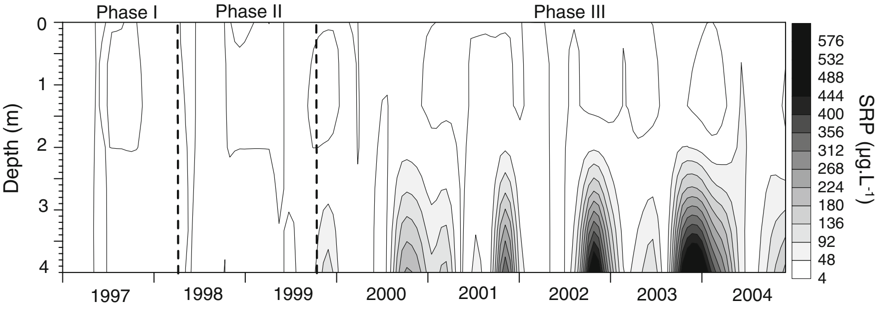

Fig. 3 Depth–time diagram of soluble reactive phosphorus (µg l^{-1}) during the period 1997–2004 at Garças Reservoir. Dashed lines indicate limits of the three limnological phases

during the spring at the surface of reservoir, N limitation was observed (Table 1).

Biological variables

Chlorophyll *a* values showed a clear temporal pattern along the reservoir limnological phases, increasing markedly after intervention in the entire water column, and ranging from 218 in phase I to 1,324 µg l^{-1} in phase III. Chlorophyll *a*:wet weight ratio among the phases were similar considering the whole water column values (0.31, 0.25 and 0.30, respectively). However, considering the surface values, the third phase presented the lower mean value (0.16) (Table 1). Integrated values of chlorophyll *a* (Table 1) increased 2 times from phase I to phase II, and 3 times from phase II to phase III. Chlorophyll *a* values were considered significantly different among the three limnological phases (ANOVA 'one way'; $F = 22.8$; $P < 0.05$) (Table 1, Fig. 4).

Total biomass also showed a huge increase after water hyacinth removal, ranging from 148 mg l^{-1} in phase I to 4,236 mg l^{-1} in phase III. The greatest value of integrated biomass was documented in November 2000 (5,539.537 mm^3 m^{-2}). The three limnological phases were considered significantly different in relation to the integrated total biomass values (ANOVA 'one way'; $F = 6.3$; $P = 0.000$) (Table 1, Fig. 4).

Cyanobacterial biomass increment was observed during phase III. Maximum values of its total biomass increased almost 29 times from phase I to phase III, and almost 58 times from phase II to phase III (Table 1, Fig. 4). Cyanobacterial integrated biomass increased almost eight times from phase I to phase III and about 21 times from the period the reservoir was covered by macrophytes to the one after their removal (Table 1). During the first phase, the greatest contribution was registered in September 1997, reaching 330.104 mm^3 m^{-2}. In the presence of the macrophyte, other algal groups contributed most to the biomass observed. After intervention and beginning of phase III, blue greens dominated during the entire period, reaching its maximum in November 2000 (5,523.969 mm^3 m^{-2}). Considering the cyanobacterial integrated biomass values, the three phases were considered significantly different (ANOVA 'one way'; $F = 6.8$; $P < 0.05$).

According to Reynolds et al. (2002) assemblages approach, 21 functional groups were identified during the entire temporal series study. During phase I, despite the contribution of many functional groups, group **M** (*Sphaerocavum brasiliense*) was the most representative in the bloom period (September 1997),

Table 1 Minimum, maximum and mean values, standard error (SE) and variation coefficient (VC, %) of abiotic and biological variables during phase I ($n = 75$), phase II ($n = 85$) and phase III ($n = 320$) and Q Index (phase I, $n = 15$; phase II, $n = 17$; phase III, $n = 64$) at Garças Reservoir

	Phase I					Phase II					Phase III				
	Min	Max	Mean	SE	VC	Min	Max	Mean	SE	VC	Min	Max	Mean	SE	VC
pH	5.7	8.8	6.8	0.1	8	6.4	7.0	6.7	0.0	2	6.4	10.7	7.4	0.0	11
Conductivity (µS cm^{-1})	110	259	146	3.3	20	74	190	156	1.9	11	138	614	235	4.2	32
Water temperature (°C)	17	27	21	0.3	14	15	25	19	0.3	15	12	27	20	0.2	14
DO (mg l^{-1})	0.0	10.6	4.1	0.3	63	0.1	5.5	2.6	0.2	53	0.0	16.5	3.6	0.2	102
NO_2^- (µg l^{-1})	5	55	21	1.5	64	5	60	20	1.5	68	5	84	18	1.0	96
NO_3^- (µg l^{-1})	8	588	174	16.4	82	8	667	114	15.0	121	8	571	76	5.7	135
NH_4^+ (µg l^{-1})	10	11862	1347	271.0	174	13	1883	551	43.4	73	4	39120	3879	324.0	150
TN (µg l^{-1})	32	15388	2095	342.0	142	243	5276	1555	118.0	70	159	74207	9224	513.0	99
TN:TP (molar)	0	448	43	7.87	157	7	192	37	3.45	86	2	559	73	4.02	99
N:P dissolved (molar)	6	2050	486	53.6	95	32	886	326	19.7	56	1	21744	707	104	264
SRP (µg l^{-1})	4	41	7	0.6	85	4	15	5	0.2	40	4	703	57	6.3	197
TDP (µg L^{-1})	9	55	18	1.0	49	6	29	15	0.6	36	4	709	84	6.5	140
TP (µg l^{-1})	60	269	121	5.3	38	38	243	100	3.9	36	39	1588	321	13.7	76
SRSi (mg l^{-1})	1	5	2	0.1	34	1	3	2	0.1	33	1	9	3	0.1	50
free CO_2 (mg l^{-1})	0.1	190.8	26.3	4.5	147	8.6	37.2	22.6	0.8	34	0.0	188.9	13.6	1.2	157
HCO_3^- (mg l^{-1})	32.3	123.2	49.9	2.3	40	41.0	68.4	61.0	0.7	10	0.0	358.5	70.8	2.3	57
CO_3^{-2} (mg l^{-1})	0.0	1.1	0.0	0.0	384	0.0	0.0	0.0	0.0	43	0.0	12.7	0.7	0.1	322
Alcalinity (mEq l^{-1})	0.5	2.0	0.8	0.0	40	0.3	1.7	0.7	0.0	36	0.2	5.9	1.2	0.0	56
Chlorophyll *a* (µg l^{-1})	9	218	60	4.7	68	10	141	42	2.8	60	0	1324	120	7.5	112
Total biomass (mg l^{-1})	3	148	23	2.5	96	2	74	20	1.3	62	0	4236	120	16.4	246
Chlo -*a*: Wet weight (%)	0.03	1.46	0.31	0.02	71	0.08	1.11	0.25	0.02	63	0.00	6.84	0.30	0.03	203
Chlorophyll a (µg l^{-1})*	21.1	218.3	74.8	14.7	76	11.8	140.8	40.9	7.8	78	36.2	1323.6	252.0	26.3	83
Total biomass (mg l^{-1})*	8.2	147.7	29.6	9.1	119	5.6	63.7	18.4	3.2	72	16.0	4235.9	315.4	73.1	185

Table 1 continued

	Phase I					Phase II					Phase III				
	Min	Max	Mean	SE	VC	Min	Max	Mean	SE	VC	Min	Max	Mean	SE	VC
Chlo -*a*: Wet weight (%)*	0.1	0.5	0.3	0.0	39	0.1	0.7	0.2	0.0	60	0.0	0.6	0.2	0.0	75
Cyanobacteria (mg l^{-1})	1	146	14	2.5	156	1	26	5	0.6	96	0	4229	111	16.3	262
Chlorophyll - *a* (µg m^{-2})	139136	614213	300277	37133	48	80814	427935	212228	26620	52	135109	1643890	599882	34764	46
Total biomass (mm^3 m^{-2})	48941	337254	114420	18848	64	48264	239313	98917	12501	52	69083	5539537	597642	97124	130
Cyanobacteria (mm^3 m^{-2})	7400	330104	70169	20735	114	8545	91549	26971	5539	85	29501	5523969	555141	96964	140
Q index	0.1	2.9	1.5	0.2	49	1.4	3.0	2.4	0.1	20	0.0	1.5	0.4	0.0	96

Variables marked with * were considered in surface depth (phase I, $n = 15$; phase II, $n = 17$; phase III, $n = 64$)

meanwhile $\mathbf{S_N}$ (*Cylindrospermopsis raciborskii*) presented greater contribution during the other stratification months. At the end of the first phase and during almost the whole second one the functional group **Y** (*Cryptomonas curvata* and *Cryptomonas erosa*) presented higher contribution. In that period, $\mathbf{L_O}$ (*Peridinium* spp. and *Merismopedia* spp.) and **K** (*Aphanocapsa elachista*, *Aphanocapsa delicatissima* and *Aphanothece smithii*) were also important. Despite **W2** (*Trachelomonas sculpta* and *Trachelomonas volvocinopsis*) appearance at the end of the studied period, this group was as well registered at the beginning of phase II. **H1** (*Aphanizomenon gracile* and *Anabaena planctonica*), $\mathbf{S_N}$ (*Cylindrospermopsis raciborskii*) and **S1** (*Planktothrix agardhii*, *Pseudanabaena galeata* and *Geitlerinema unigranulatum*) presented important biomass contribution during the entire third phase. However, **M** functional group (*Sphaerocavum brasiliense*, *Microcystis panniformis* and *M. aeruginosa*) was responsible for the greatest biomass values registered in that period (Fig. 5).

Q Index and trophic status index

Following the steps recommended by Padisák et al. (2006) for the Q Index application, the factor F was determined for each functional group occurring at Garças Reservoir (Table 2).

The Q Index showed differences between the three limological phases. Phase I classification varied from *bad* to *medium*. Under the macrophyte influence, Q index pointed to *tolerable* and *medium* classifications. After intervention, Garças Reservoir ecological status was during most of the period under *bad* classification, with exception of the last few months at the end of phase III, in which the classification was *tolerable*. The three limnological phases were considered significantly different considering the Q Index (ANOVA 'one way'; $F = 119.4$; $P = 0.000$). Table 1 shows the Q Index variation amplitude at Garças Reservoir phases (Fig. 6).

Different trophic status indices applied to Garças Reservoir by Bicudo et al. (2006) evidenced the three limnological phases. Not only through Carlson's modified by Toledo (1990) TSI, but also Lamparelli's (2004) indicated changes among the three phases, considering chlorophyll *a*, Secchi disk and TP data set or the mean values themselves, especially when comparing phases I and III to phase II (Fig. 7).

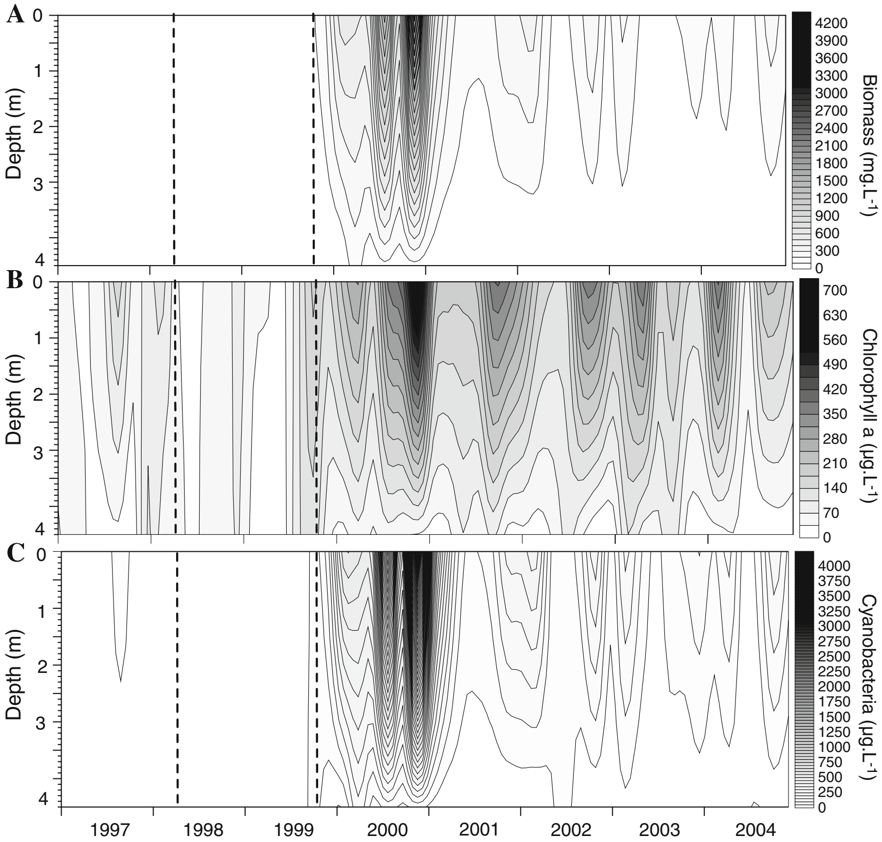

Fig. 4 Depth–time diagrams of (**A**) total biomass, (**B**) chlorophyll *a* and (**C**) Cyanobacteria during the period 1997–2004 at Garças Reservoir. Dashed lines indicate limits of the three limnological phases

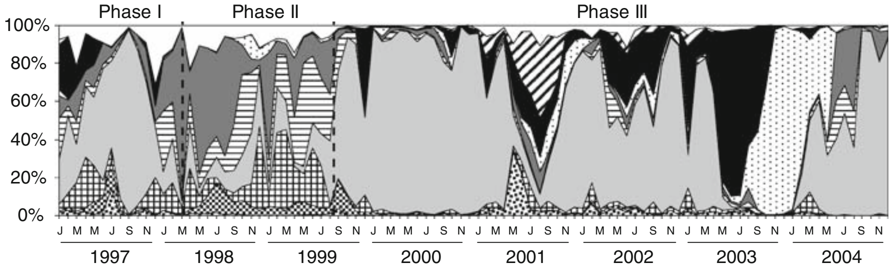

Fig. 5 Relative integrated biomass (%) of functional groups (*sensu* Reynolds, 1997 and Reynolds et al., 2002) during the three limnological phases at Garças Reservoir. Legend: ▩ **D**, ▦ **L_O**, ▤ **W2**, ☐ **S1**, ▨ **H1**, ▩ **K**, ☐ **M**, ▩ **Y**, ■ **S_N** and ☐ others

Discussion

Temporal series analyses done by Bicudo et al. (2007) aiming at determining if intervention had a significant effect on the limnological characteristics of Garças Reservoir classified the limnological responses to the macrophyte removal a 'permanent abrupt impact' (sensu McDowall et al., 1980).

On the basis of all data obtained during the long term study of Garças Reservoir, a continuous degradation of the ecosystem after the water hyacinth removal was evidenced. Cyclic anoxic periods previously observed during springs and summers were replaced by a more persistent period of anoxic conditions in the sediment-overlying water (Bicudo et al., 2007). P dynamics, initially driven by allochthonous sources, was replaced by an internal loading. Water hyacinth removal triggered self-eutrophication leading to an irreversible turbid phase (Bicudo et al., 2007).

Alterations above were clearly demonstrated by the phytoplankton responses and the Q Index. Oscillations in the ecological status of Garças Reservoir during phase I evidenced that, although eutrophic, there was a *tolerable* period in which the ecosystem was inhabited by many 'good' functional groups, and

Table 2 Factor F to the functional groups of Garças Reservoir

Functional group	Factor F
D	2
E	5
F	5
H1	1
J	5
K	3
L$_\mathbf{O}$	5
M	0
N	5
P	2
Q	4
S1	0
S$_\mathbf{N}$	0
T	5
V	1.5
W1	0
W2	1
X1	5
X2	5
X3	4
Y	3

another period when the **M** group dominated, driving the reservoir ecological status to *bad*.

Q index pointed for a better ecological status during the macrophyte presence, classified in between *tolerable* and *medium*. This was due to the contribution of a more diverse composition of functional groups and greatest values of the F factor.

After macrophyte removal, worst ecological status, varying from *bad* to *tolerable*, was registered due to the overwhelming dominance of functional groups of bloom forming cyanobacteria.

In 2000, *Microcystis panniformis* bloom was responsible for the high biomass values (4,236 mg l^{-1}) due to its big colonies. These are characterized by having cells densely aggregated in all the mucilage surface (Sant'Anna et al., 2004) and were already registered as tending to preserve their arrangement after Lugol's solution addition (Naselli-Flores & Barone, 2003). Also, in that period, high value of chlorophyll *a* was registered (1,197 μg l^{-1}). In 2004, the highest chlorophyll *a* value was observed (1,324 μg l^{-1}), with the contribution of *M. aeruginosa* to almost 100% of the total biomass registered. These bloom episodes were reflected directly in chlorophyll *a*:wet weight ratio variance observed in phase III. Although the average value found in all phases are under the common ones found in world literature (Vörös & Padisák, 1991; Temponeras et al., 2000), the huge biomass of cyanobacteria found in the third phase would explain not only the ratio's wide variation (variation coefficient = 203%, $n = 320$; 75%, $n = 64$) but also its extreme values observed. Such values of chlorophyll *a* and biomass were also verified in very impacted lakes and reservoirs in Brazil (Huszar et al., 2000, 2006; Da Silva, 2005) and in other ecosystems around the world (Zohary & Breen, 1989; Zohary et al., 1996; Kotut et al., 1998). In Brazilian fishponds in the metropolitan region of São Paulo, Da Silva (2005) registered a biomass peak of *M. aeruginosa* reaching almost 20,000 mg l^{-1}. Huszar and collaborators (2006) found a maximum value of 556 μg l^{-1} chlorophyll *a* in a set of 192 (among them 79 Brazilian) tropical and subtropical ecosystems. In the Garças reservoir, despite those peaks mentioned above, the mean values of chlorophyll *a* and total biomass are comparable to the aforementioned selection of lakes (Table 1). Extreme values of chlorophyll *a* were also registered by Zohary & Breen (1989) when studying *Microcystis aeruginosa* hyperscums in a hypertrophic South African lake (Hartbeespoor Dam). These authors registered a high peak of chlorophyll *a* exceeding 100,000 mg m^{-3}.

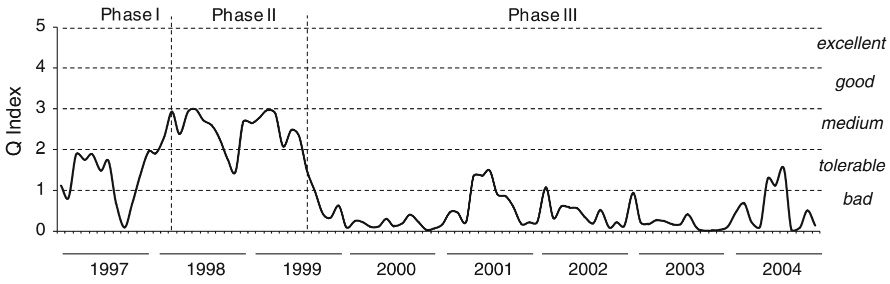

Fig. 6 Ecological status evaluation by Q Index Relative during the three limnological phases at Garças Reservoir

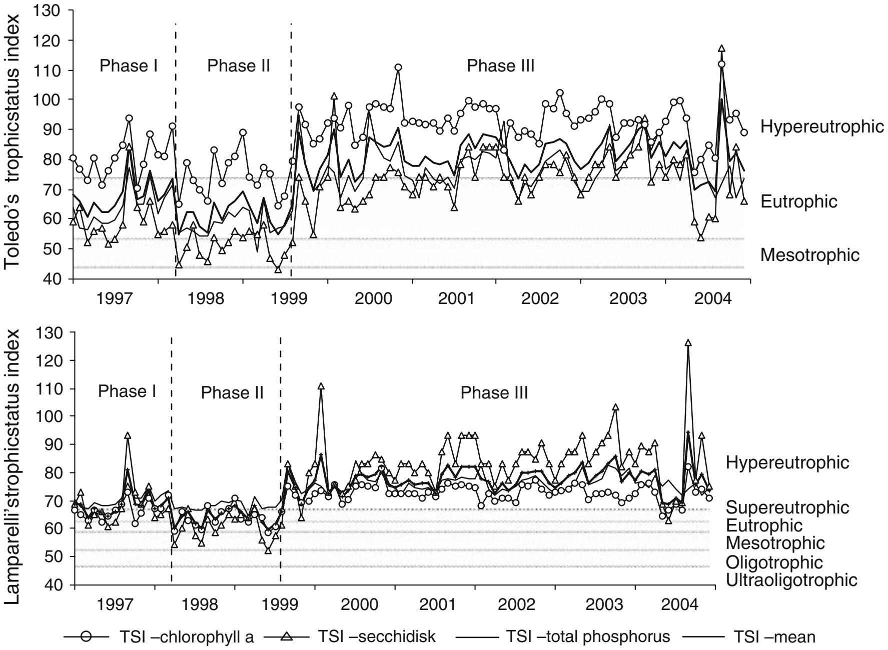

Fig. 7 Garças Reservoir trophic classification according to Carlson's trophic status index modified by Toledo (1990) and Lamparelli (2004) (from Bicudo et al., 2006)

Cyanobacterial blooms were important integrating factors in the self-establishing ecosystem state (stable degraded state) during phase III, acting in the feedback mechanisms already identified in the lake (Bicudo et al., 2007). In a general way, Q index was inversely proportional to the dominance of **M**, **S$_N$**, **S1** and **H1** functional groups.

Padisák et al. (2006) concluded that, after discussing the Q index application to Balaton Lake with an evaluation made through some categories based on algal fresh weight by Mischke et al. (2002 in Padisák et al., 2006), responses of the first one were more realistic than those given by chlorophyll *a* and biomass. Latter authors also evidenced that the trophic gradient of Balaton Lake were registered by the index.

Comparing responses of both Garças Reservoir TSI methods applied by Bicudo et al. (2006) to the Q index ones, alterations registered in the three limnological phases could be identified in all of them. Although TSI have shown different results on the trophic classification, both showed better water conditions during phases I and II and a worse one during phase III, as well as did the Q index. However, the only biological characteristic considered in the TSI was chlorophyll *a*. Q index takes into account not only the algal biomass through the functional groups contribution, but also the species themselves. According to Padisák et al. (2006), the method is differently sensitive to taxonomical misidentifications and cyanobacterial identification requires special skills and profuse taxonomic literature.

Among the weaknesses pointed by the Q Index's authors is the fact that the factor F is determined exclusively by previous knowledge and experience. Determining factor F was the most difficult step during the present study. However, it was our first intention not to let the eutrophic conditions of the Garças Reservoir influence the weight of each functional group. It was also attempted to imagine what those functional groups would be during the pristine status of the system, although there is no scientific or historical reference about them. In this regard, considering the morphological similarities between the Garças Reservoir (hypereutrophic urban shallow lake) and IAG Reservoir (oligotrophic urban shallow lake), both located in the same park, it was suggested that the same factor F could be applied to the last reservoir that could be considered the opposite or the pristine state of Garças Reservoir.

About the IAG Reservoir, phytoplankton studies were performed (Oliveira, 2004; Ferragut, 2004; Lopes et al., 2005). Lopes et al. (2005) postulated, after evaluating the short term and temporal variation of phytoplankton in this lake, that during the dry period under constant mixture and least nutrient availability functional groups **X2** (*Chlamydomonas planctogloea*, *Chlamydomonas* sp.) and **F** (*Oocystis lacustris* and *Kirchneriella pseudoaperta*) were relevant. Meanwhile, in the rainy season, period of daily stratification and highest nutrient availability, the same species of functional groups **X2** (*Chlamydomonas planctogloea*, *Chlamydomonas* sp.), **Z** (*Synechococcus nidulans*, *Chroococcus minor*), **F** (*Oocystis lacustris*, *Elakatothrix gelatinosa*), **J** (*Crucigenia tetrapedia*, *Tetraedron caudatum*) and **X1** (small Chlorococcales) were the most representative ones. In the present study, Lopes et al.'s (2005) contribution on functional groups was taken into account regarding the factor F determination. To those assemblages that are supposed to occur under a pristine state in the Garças Reservoir, a higher *F* value was ascribed.

Padisák et al. (2006) described eight types of lakes in Hungary and for each one ascribed different factor F weights. However, according to Garças Reservoir characteristics, it does not seem adequate to use the same weights defined to the Hungarian lakes. Considering its typology and the fact that the Garças Reservoir is a man made, tropical, urban, small, not exposed to wind action, with a sedimentary rocks ground, and considering the long term phytoplankton expertise and knowledge, it seems that F factors are well determined by phytoplankton of IAG Reservoir examples, since this last one has the same characteristics and is under no substantial human impact.

Among the Hungarian lakes described in Padisák et al. (2006), the closest one to Garças Reservoir is the type 7, formed by oxbows. Nevertheless, when evaluating the factor F of this sort, it seems not reasonable to ascribe weights 3 and 3.5 to, respectively, **X1** and **X2** functional groups, since previous studies showed the important contribution of these assemblages to what we consider the pristine state of the Garças Reservoir. Consequently, in the present study, factor F has different weights compared to those of the oxbows' sort.

Finally, Q index presented very good results as the trophic index applied to the Garças Reservoir, indicating that phytoplankton could be successfully used as an indicator of ecological status of the system in monitoring processes.

Acknowledgments The authors thank Dr. Judit Padisák for fruitful discussions about the Q index method.

References

Albay, M. & R. Akçaalan, 2003. Factors influencing the phytoplankton steady state assemblages in a drinking-water Reservoir (Ömerli Reservoir, Istanbul). Hydrobiologia 502: 85–95.

Azevedo, M. T. P., N. M. C. Nogueira & C. L. Sant'Anna, 1996. Criptógamos do Parque Estadual das Fontes do Ipiranga, São Paulo, SP. Algas, 8: Cyanophyceae. Hoehnea 23: 1–38.

Azevedo, M. T. P. & C. L. Sant'Anna, 2003. *Sphaerocavum*, a new genus of Cyanobacteria from continental water bodies in Brazil. Algological Studies 109 (Cyanobacterial Research 4): 79–92.

Barbosa, F. A. R. & J. Padisák, 2004. Algumas considerações sobre desenho amostral de longa duração. In Bicudo, C. & D. Bicudo (eds), Amostragem em Limnologia. RiMa, São Carlos: 343–351.

Bicudo, C. E. M., C. F. Carmo, D. C. Bicudo, A. C. S. Pião, C. M. Santos & M. R. M. Lopes, 2002a. Morfologia e morfometria de três reservatórios do PEFI. In Bicudo, D. C., M. C. Forti & C. E. M. Bicudo (eds), Parque Estadual das Fontes do Ipiranga (PEFI): unidade de conservação que resiste à urbanização de São Paulo. Editora Secretaria do Meio Ambiente do Estado de São Paulo, São Paulo: 352 pp.

Bicudo, D. C., M. C. Forti, C. F. Carmo, C. Bourotte, C. E. M. Bicudo, A. J. Melfi & Y. Lucas, 2002b. A atmosfera, as águas superficiais e os reservatórios no PEFI: caracterização química. In Bicudo, D. C., M. C. Forti & C. E. M. Bicudo (eds), Parque Estadual das Fontes do Ipiranga (PEFI): unidade de conservação que resiste à urbanização de São Paulo. Editora Secretaria do Meio Ambiente do Estado de São Paulo, São Paulo: 143–160.

Bicudo, D. C., B. M. Fonseca, C. E. M. Bicudo, L. M. Bini & T. Araújo-Jesus, 2006. Remoção de *Eichhornia crassipes* em um reservatório tropical raso e suas implicações na classificação trófica do sistema: estudo de longa duração no Lago das Garças, São Paulo, Brasil. In Tundisi, J. G., T. Matsumura-Tundisi & C.V. Sidagis Galli (eds), Eutrofização na América do Sul: causas, conseqüências e tecnologias para gerenciamento e controle. Instituto Internacional de Ecologia, São Carlos: 413–438

Bicudo, D. C., B. M. Fonseca, L. M. Bini, L. O. Crossetti, C. E. M. Bicudo & T. Araújo-Jesus, 2007. Undesirable side-effects of water hyacinth control in a shallow tropical Reservoir. Freshwater Biology 51: 1120–1133.

Carmo, C. F., R. Henry, D. C. Bicudo & C. E. M. Bicudo, 2002. A degradação nos reservatórios do PEFI. In Bicudo, D. C., M. C. Forti & C. E. M. Bicudo (eds), Parque Estadual das Fontes do Ipiranga (PEFI): unidade de conservação que resiste à urbanização de São Paulo. Editora Secretaria do Meio Ambiente do Estado de São Paulo, São Paulo: 271–296.

Crossetti, L. O. & C. E. M. Bicudo, 2005. Structural and functional phytoplankton responses to nutrient impoverishment in mesocosms placed in a shallow eutrophic Reservoir (Garças Pond), São Paulo, Brazil. Hydrobiologia 541: 71–85.

Da Silva, D., 2005. Dynamic of microcystis populations (Cyanobacteria) in fishing ponds in the Metropolitan Region of São Paulo, SP, Brazil. Master Thesis. Instituto de Botânica de São Paulo, São Paulo: 146 pp.

Fernandes, A. J., L. A. M. Reis & A. Carvalho, 2002. Caracterização do meio físico. In Bicudo, D. C., M. C. Forti & C. E. M. Bicudo (eds), Parque Estadual das Fontes do Ipiranga (PEFI): unidade de conservação que resiste à urbanização de São Paulo. Secretaria do Meio Ambiente do Estado de São Paulo, São Paulo: 49–62.

Ferragut, C., 2004. Respostas das algas perifíticas e planctônicas à manipulação de nutrientes (N e P) em reservatório urbano (Lago do IAG, São Paulo). Doctoral Dissertation. Universidade Estadual Paulista, Rio Claro: 184 pp.

Fonseca, B. M., 2005. Diversidade fitoplanctônica como discriminador ambiental em dois reservatórios rasos com diferentes estados tróficos no Parque Estadual das Fontes do Ipiranga, São Paulo, SP. Doctoral Dissertation. Universidade de São Paulo, São Paulo: 208 pp.

Fonseca, B. M. & C. E. M. Bicudo, 2008. Phytoplankton seasonal variation in a shallow stratified eutrophic reservoir (Garças Pond, Brazil). Hydrobiologia 600: 267–282.

Golterman, H. L. & R. S. Clymo, 1971. Methods for chemical analysis of freshwaters. International Biological Programme. Blackwell Scientific Publications, Oxford & Edinburg: 161 pp.

Golterman, H. L., R. S. Clymo & M. A. M. Ohmstad, 1978. Methods for Chemical Analysis of Freshwaters. Blackwell Scientific Publications, Oxford & Edinburg: 213 pp.

Hillebrand, H., D. Dürseken, D. Kirschiel, U. Pollingher & T. Zohary, 1999. Biovolume calculation for pelagic and benthic microalgae. Journal of Phycology 35: 403–424.

Huang, T. L. & H.-B. Cong, 2007. A new method for determination of chlorophylls in freshwater algae. Environmental Monitoring and Assessment 129: 1–7.

Huszar, V. L. M., L. H. S. Silva, M. Marinho, P. Domingos & C. L. Sant'Anna, 2000. Cyanoprokaryote assemblages in eight productive tropical Brazilian Waters. Hydrobiologia 424: 67–77.

Huszar, V. L. M., N. Caraco, F. Roland & J. Cole, 2006. Nutrient-chlorophyll relationships in tropical-subtropical lakes: do temperate models fit? Biogeochemistry 79: 239–250.

Komárek, J. & K. Anagnostidis, 1986. Modern approach to the classification system of cyanophytes, 2: Chroococcales. Algological Studies 43: 157–226.

Komárek, J. & K. Anagnostidis, 1989. Modern approach to the classification system of cyanophytes, 4: Nostocales. Algological Studies 56: 247–345.

Komárek, J. & K. Anagnostidis, 1999. Cyanoprokaryota, 1: Chroococcales. In Ettl, H., G. Gärtner, H. Heynig & D. Möllenhauer (eds), Süßwasserflora von Mitteleuropa, Vol. 19(1). Gustav Fischer Verlag, Stuttgart: 1–548.

Komárek, J. & M. T. P. Azevedo, 2000. *Geitlerinema unigranulatum*, a common tropical cyanoprokaryote from freshwater Reservoirs in Brazil. Algological Studies 99: 39–52.

Komárek, J. & K. Anagnostidis, 2005. Cyanoprokaryota, 2: Oscillatoriales. In Ettl, H., G. Gärtner, H. Heynig & D. Möllenhauer (eds), Süßwasserflora von Mitteleuropa, Vol. 19(2). Gustav Fischer Verlag, Stuttgart: 1–759.

Komárek, J. & B. Fott, 1983. Chlorophyta (Grünalgen) Ordnung: Chlorococcales. In: Huber-Pestalozzi, G. (ed.), Das phytoplankton des Süßwassers: Systematik und Biologie, Vol. 7(1). E. Schweizerbart'sche Verlagsbuchhandlung (Nägele u. Obermiller), Stuttgart: 1044 pp.

Komárková-Legnerová, J. & G. Cronberg, 1994. Planktic blue-green algae from lakes in South Scania, Sweden, 1: Chroococcales. Algological Studies 72: 13–51.

Kotut, K., L. Krienitz & M. Muthuri, 1998. Temporal changes in phytoplankton structure and composition at the Turkwel Gorge Reservoir, Kenya. Hydrobiologia 368: 41–59.

Kruk, C., N. Mazzeo, G. Lacerot & C. S. Reynolds, 2002. Classification schemes for phytoplankton: a local validation of a functional approach to the analysis of species temporal replacement. Journal of Plankton Research 24: 901–912.

Lamparelli, M. C., 2004. Graus de trofia em corpos d'água do Estado de São Paulo: avaliação dos métodos de monitoramento. Doctoral Dissertation. Universidade de São Paulo, São Paulo: 235 pp.

Lewis, W. M., Jr., 1983. A revised classification of lakes based on mixing. Canadian Journal of Fisheries and Aquatic Sciences 40: 1779–1787.

Lopes, M. R. M., C. E. M. Bicudo & C. Ferragut, 2005. Short term spatial and temporal variation of phytoplankton in a shallow tropical oligotrophic reservoir, southeast Brazil. Hydrobiologia 542: 235–247.

Lund, J. W. G., C. Kipling & E. D. LeCren, 1958. The inverted microscope method of estimating algal numbers and the statistical basis of estimations by counting. Hydrobiologia 11: 143–170.

Mackereth, F. J. H., J. Heron & J. F. Talling, 1978. Water analysis: some revised methods for limnologists. Titus Wilson & Son Ltd., Kendall: 117 pp. (Freshwater Biological Association Scientific Publication, n^{o} 39)

Marinho, M. M. & V. L. M. Huszar, 2002. Nutrient availability and physical conditions as controlling factors of phytoplankton composition and biomass in a tropical Reservoir (southeastern Brazil). Archiv für Hydrobiologie 153: 443–468.

McDowall, D., R. McCleury, E. E. Meidinger & R. A. Hay, 1980. Interrupted Time Series Analysis. Sage Publications, Beverly Hills: 96 pp.

Mischke, U., 2003. Cyanobacteria associations in shallow polytrophic lakes: influence of environmental factors. Acta Oecologica 24: S11–S23.

Mischke, U. & B. Nixdorf, 2003. Equilibrium phase conditions in shallow German lakes: how Cyanoprokaryota species establish a steady state phase in late summer. Hydrobiologia 502: 123–132.

Mischke, U., B. Nixdorf, E. Hoehn & U. Riedmüller, 2002. Möglichkeiten zur Bewertung von Seen anhand des Phytoplanktons. Aktueller Stend in Deutschland. Aktuelle Reihe, Vol. 5/02., Brandenburgische Technische Universität, Cottbus: 25–37.

Morabito, G., A. Oggioni & P. Panzani, 2003. Phytoplankton assemblage at equilibrium in large and deep subalpine lakes: a case study from Lago Maggiore (N. Italy). Hydrobiologia 502: 37–48.

Naselli-Flores, L. & R. Barone, 2003. Steady-state assemblages in a Mediterranean hypertrophic Reservoir. The role of

Microcystis ecomorphological variability in maintaining an apparent equilibrium. Hydrobiologia 502: 133–143.

Naselli-Flores, L., J. Padisák, M. T. Dokulil & I. Chorus, 2003. Equilibrium/steady-state in phytoplankton ecology. Hydrobiologia 502: 395–403.

Nixdorf, B., U. Mischke & J. Rücker, 2003. Phytoplankton assemblages and steady state in deep and shallow eutrophic lakes: an approach to differentiate the habitat properties of Oscillatoriales. Hydrobiologia 502: 111–121.

Oliveira, A. M., 2004. Distribuição vertical da comunidade fitoplanctônica em dois períodos climáticos (seco e chuvoso) em reservatório oligotrófico (Lago do IAG, São Paulo). Graduation paper. Universidade Metodista de São Paulo, São Bernardo do Campo: 56 pp.

Padisák, J., G. Borics, I. Grigorszky & E. Soróczki-Pintér, 2006. Use of phytoplankton assemblages for monitoring ecological status of lakes within the Water Framework Directive: the assemblage index. Hydrobiologia 553: 1–14.

Pápista, É., É. Ács & B. Böddi, 2002. Chlorophyll-a determination with ethanol—a critical test. Hydrobiologia 485: 191–198.

Reynolds, C. S., V. Huszar, C. Kruk, L. Naselli-Flores & S. Melo, 2002. Towards a functional classification of the freshwater phytoplankton. Journal of Plankton Research 24: 417–428.

Rojo, C. & M. Álvarez-Cobelas, 2003. Are there steady-state phytoplankton assemblages in the field? Hydrobiologia 502: 3–12.

Sant'Anna, C. L. & M. T. P. Azevedo, 2000. Contribution to the knowledge of potentially toxic Cyanobacteria from Brazil. Nova Hedwigia 71: 359–385.

Sant'Anna, C. L., M. T. P. Azevedo & L. Sormus, 1989. Fitoplâncton do Lago das Garças, Parque Estadual das Fontes do Ipiranga, São Paulo, Brasil: estudo taxonômico e aspectos ecológicos. Hoehnea 16: 89–131.

Sant'Anna, C. L., M. T. P. Azevedo, P. A. C. Senna, J. Komárková, & J. Komárek, 2004. Planktic Cyanobacteria from São Paulo State, Brazil: Chroococcales. Brazilian Journal of Botany 27: 213–227.

Sartory, D. P. & J. U. Grobbelaar, 1984. Extraction of chlorophyll *a* from freshwater phytoplankton for spectrophotometric analysis. Hydrobiologia 114: 177–187.

Silva, L. H. S., 2004. Fitoplâncton de um pequeno reservatório eutrófico (Lago Monte Alegre, Ribeirão Preto, SP): dinâmica temporal e respostas à ação do zooplâncton e peixes. Doctoral Dissertation. Universidade Federal do Rio de Janeiro, Rio de Janeiro: 219 pp.

Solorzano, L., 1969. Determination of ammonia in natural waters by the phenolhypochlorite method. Limnology and Oceanography 14: 799–801.

Strickland, J. D. H. & T. R. Parsons, 1960. A manual of seawater analysis. Fisheries Research Board of Canada Bulletin 125: 1–185.

Sun, J. & D. Liu, 2003. Geometric models for calculating cell biovolume and surface area for phytoplankton. Journal of Plankton Research 25: 1331–1346.

Temponeras, M., J. Kristiansen & M. Moustaka-Gouni, 2000. Seasonal variation in phytoplankton composition and physical-chemical features of the shallow Lake Doïrani, Macedonia, Greece. Hydrobiologia 424: 109–122.

Toledo A. P. T., Jr., 1990. Informe preliminar sobre os estudos para a obtenção de um índice para a avaliação simplificada do estado trófico de reservatórios de regiões quentes tropicais. Companhia de Tecnologia de Saneamento Ambiental, São Paulo: 12 pp. (CETESB Internal Report).

Utermöhl, H., 1958. Zur Vervolkomnung der quantitative Phytoplankton: Methodik Mitteilungen der internationale Vereinigung für theoretische und angewandte Limnologie 9: 1–38.

Valderrama, J. C., 1981. The simultaneous analysis of total nitrogen and total phosphorous in natural waters. Marine Chemistry 10: 109–122.

Vöros, L. & J. Padisák, 1991. Phytoplankton biomass and chlorophyll-*a* in some shallow lakes in central Europe. Hydrobiologia 215: 111–119.

Wasmund, N., I. Topp & D. Schories, 2006. Optimising the storage and extraction of chlorophyll samples. Oceanologia 48: 125–144.

Water Framework Directive, 2000. Directive 2000/60/ec of the European Parliament and of the Council 22.12.2000. Official Journal of the European Communities L327: 1–72.

Wetzel, R. G. & G. E. Likens, 2000. Limnological Analyses. Springer-Verlag, New York: 229 pp.

Zohary, T. & C. M. Breen, 1989. Environmental factors favouring the formation of *Microcystis aeruginosa* hyperscums in a hypereutrophic lake. Hydrobiologia 178: 179–192.

Zohary, T., A. M. Pais-Madeira, R. Robarts & K. D. Hambright, 1996. Interannual phytoplankton dynamics of a hypereutrophic African lake. Archiv für Hydrobiologie 136: 105–126.