### Responses of periphytic diatoms to mechanical removal of *Pistia* stratiotes L. in a hypereutrophic subtropical reservoir: dynamics and tolerance

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Received March 20, 2012 - Accepted October 18, 2012 - Distributed November 29, 2013 (With 3 figures)

#### Abstract

The Itaqui reservoir in Paraná state, southern Brazil, is dominated by the floating macrophyte Pistia stratiotes L. and is used for recreation and irrigation. The reservoir's excessive plant cover suggests an extreme trophic state and interferes with multiple uses. The aims of this study were to determine the trophic state of the reservoir water and to document the limnological conditions and the composition of the periphytic diatom community before and after the mechanical removal of macrophytes. As each diatom species has certain autoecological requirements in a given geographic area, another objective of the study was to identify diatoms that can be considered tolerant of the reservoir's trophic state in a substropical environment. Local water samples collected for physical and chemical analyses, including estimates of chlorophyll a, showed the hypereutrophic status of the reservoir before and after macrophyte removal. Environmental conditions exceeded acceptable values for fishing and irrigation, providing a clear example of how the inadequate management of water resources can directly reduce their usefulness. Trimestral sampling was carried out between May 2008 and February 2009. For quantitative analyses, biofilms were scrubbed off glass slides submerged for 30 days at a depth of approximately 40 cm. Diatom samples were cleaned with potassium permanganate and hydrochloric acid and mounted on permanent slides with Naphrax. All individuals found in random transects under three replicates were identified and counted up to a minimum of 600 valves. Thirteen species tolerant of eutrophication were selected. Four species mostly known from low-nutrient sites may be considered tolerant of eutrophic conditions. The composition of the diatom community was influenced by seasonal changes in temperature and rainfall. Canonical Correspondence Analyses confirmed a correlation between higher diatom densities and the increased photic zone following macrophyte removal.

Keywords: Itaqui Reservoir, PR, artificial substrate, eutrophication.

# Respostas das diatomáceas perifíticas à remoção mecânica de *Pistia stratiotes* L. num reservatório subtropical hipereurófico: dinâmica e tolerância

#### Resumo

A represa Itaqui, localizada no estado do Paraná, sul do Brasil, é dominada pela macrófita flutuante *Pistia stratiotes* L. e utilizada para recreação e irrigação. Esta excessiva massa vegetal sugere um elevado estado de trofía e interfere nos múltiplos usos do corpo d'água. Os objetivos deste estudo foram determinar o estado trófico da represa e documentar suas condições limnológicas e a composição da comunidade de diatomáceas perifíticas antes e após a remoção mecânica da massa de macrófitas. As espécies apresentam autoecologias específicas em diferentes áreas geográficas, desta forma outro objetivo foi identificar diatomáceas em ambiente subtropical que possam ser consideradas tolerantes ao estado trófico da represa. Amostras de água foram coletadas para analises físicas e químicas, incluindo estimativas de clorofila-a que evidenciou o estado hipereutrófico da represa antes e depois da remoção das macrófitas. As condições ambientais excederam os valores aceitáveis para pesca e irrigação evidenciando como o manejo inadequado dos recursos hídricos pode diretamente reduzir sua utilidade. Amostragem trimestral foi realizada entre maio de 2008 e

fevereiro de 2009. O biofilme desenvolvido sobre lâminas de vidro submersas foram raspados para análise quantitativa, contando-se 600 valvas em triplicatas de material oxidado. Treze espécies tolerantes a eutrofização foram selecionadas. Quatro espécies comumente identificadas em ambientes com baixa disponibilidade de nutrientes mostraram-se tolerantes às condições eutróficas. As diatomáceas responderam qualitativamente à sazonalidade como temperatura e pluviosidade. Análise de Corresponência Canônica confirmou que densidades mais elevadas coincidiram com o aumento da zona fótica oriunda da remoção das macrófitas.

Palavras-chave: represa Itaqui, PR, substrato artificial, eutrofização.

#### 1. Introduction

Diatoms are unicellular algae characterized by complex taxonomy and specialised structures by which the cells adhere to a variety of substrates (Round et al., 1990). This strategy gives them a competitive advantage in colonising natural and artificial substrates. Both diatom growth and community composition vary with environmental heterogeneity, and their sessile habit and inability to migrate away from adverse conditions make them reliable proxies for their environmental surroundings and limnological conditions (Stevenson, 1997). As a result, diatom communities are increasingly used worldwide as indicators of environmental disturbance (Wetzel, 1993; Stevenson, 1997; Stoermer and Smol, 1999).

Studies that use diatom assemblages to classify trophic states have focused to date on lotic environments in the temperate zone (Wunsam et al., 2002; Rimet, 2009). Because lentic environments have been much less studied, documenting relationships between nutrient concentrations and the composition and structure of diatom communities in lakes (e.g., Kitner, 2003: Denicola et al., 2004; Blanco et al., 2004; Àcs et al., 2005; Stenger-Kovacs et al., 2007) remains a priority. Diatom species have different autoecological requirements in different geographical areas (Álvarez-Blanco et al., 2011), and little is known about their environmental tolerances in subtropical systems, and especially hypereutrophic ones. In Brazil, studies using diatoms as indicators of organic pollution and eutrophication have also focused on lotic environments (Lobo and Ben da Costa, 1997; Oliveira et al., 2001; Lobo et al., 2002, 2004a,b,c, 2010; Salomoni et al., 2006; Hermany et al., 2006). Few Brazilian studies have focused on periphytic diatoms in typical eutrophic lentic environments (Rodrigues and Bicudo, 2001; Cetto et al., 2004; Fonseca and Rodrigues, 2005; Faria et al., 2010; Silva et al., 2010).

In this context, assessments of the response and tolerance of diatom species to environmental conditions in tropical and subtropical regions remain a high priority. This study, carried out in a reservoir in the Itaqui River watershed that is dominated by the floating macrophyte *Pistia stratiotes* L., used glass slides as colonisation substrates. Both the diatom community and limnological conditions were sampled before and after the mechanical removal of the macrophytes. Common species were identified and selected as potential bioindicators of strongly eutrophic environments.

#### 2. Materials and Methods

Paraná's Itaqui River drains an area of 39.80 km<sup>2</sup>. The Itaqui reservoir is located in São José dos Pinhais (25°29'49.9" S, 49°07'44.3" W), has a mean size of 13-20 ha, and is used for recreation. Starting in 2002, the reservoir began to get overgrown with the floating macrophyte *Pistia stratiotes* L. The excessive plant cover suggested strongly eutrophic conditions and interfered with multiple uses. Mechanical removal of the plants was carried out between November 2008 and February 2009 by technicians from the Paraná water company (SANEPAR). The reservoir's eutrophic conditions reflect the high input of industrial and domestic wastes throughout the river basin, in addition to agricultural areas along the Itaqui River (SANEPAR 2006; unpublished data).

Local water samples were collected from the reservoir for analyses of physical, chemical, and microbiological variables. Nitrate, total Kjeldahl nitrogen, total phosphorus, orthophosphate, chemical oxygen demand (COD), biochemical oxygen demand (BOD<sub>5</sub>), and total and thermotolerants coliforms were analysed at the Laboratório de Pesquisas Hidrogeológicas (Universidade Federal do Paraná), following the methodologies described in APHA (1995). Water and air temperature, pH, and dissolved oxygen in the field were measured with a CONSORT C535 meter. The eutrophic zone was determined by a Secchi Disk. Rainfall data were provided by the Paraná state weather service (SIMEPAR) throughout the sampling period (see Table 1). The Trophic State In-

 Table 1 - Accumulated monthly rainfall at the Curitiba

 weather station from May 2008 to February 2009.

| Month          | Rainfall (mm) |  |  |
|----------------|---------------|--|--|
| May 2008       | 46.0          |  |  |
| June 2008      | 97.6          |  |  |
| July 2008      | 26.6          |  |  |
| August 2008    | 109.8         |  |  |
| September 2008 | 31.4          |  |  |
| October 2008   | 194.8         |  |  |
| November 2008  | 49.8          |  |  |
| December 2008  | 42.6          |  |  |
| January 2009   | 146.8         |  |  |
| February 2009  | 114.2         |  |  |

dex (TSI) (Lamparelli, 2004) was used in order to characterise the trophic status of the Itaqui River and the reservoir before and after macrophyte removal. Chlorophyll-a was extracted with 90% alkaline acetone, estimated by spectrophotometry, and calculated using Jeffrey and Humphrey's (1975) equation.

Glass slides were submerged for 30 days at a depth of 40 cm for periphytic diatom sampling. Sampling was performed every three months between May 2008 and February 2009, in order to sample a full seasonal cycle. Biofilm was removed by scraping, washing, and oxidised with potassium permanganate (KMnO<sub>4</sub>) and hydrochloric acid (HCl). Permanent slides were mounted with Naphrax® (R.I. = 1.74) resin.

Taxonomic identifications were made using the following literature: Hustedt (1930); Cleve-Euler (1953); Patrick and Reimer (1966; 1975); Metzeltin and Lange-Bertalot (1998; 2007); Rumrich et al. (2000); Metzeltin et al. (2005) and analysed samples deposited in the herbarium of the Universidade Federal do Paraná (UPCB; see Table 2).

Quantitative analysis of the cleaned material was based on three replicates, each consisting of 600-valve counts (Kobayasi and Mayama, 1982). Counting efficiency followed Pappas and Stoermer (1996). Abundant and dominant species selection followed Lobo and Leighton (1986) and diatom densities (valves/cm<sup>2</sup>) were based on Batarbee (1986).

Bartlett's test was applied to check for homogeneity of variances across samples, revealing significant population-level differences between replicates. Bray-Curtis cluster analysis was performed based on quantitative data from the total diatom community, characterised using the Biodiversity Pro® Program. Total diatom densities calculated before and after the mechanical removal of macrophytes were presented in a boxplot graph (mean,  $\pm$ standard error and  $\pm$  standard deviation). Total density differences were tested via a Mann-Whitney test (Mon-

**Table 2** - Sampling dates of the Itaqui reservoir, with corresponding herbarium record numbers of diatom samples and pH values of water samples.

| Herbarium record<br>number | Sampling date    | рН   |
|----------------------------|------------------|------|
| UPCB 63475                 | 15 May 2008      | 5.71 |
| UPCB 63476                 | 15 August 2008   | 6.62 |
| UPCB 63477                 | 15 November 2008 | 5.86 |
| UPCB 63478                 | 15 February 2009 | 6.57 |
|                            |                  |      |

te-Carlo permutation test) using PAST software (Hammer et al., 2008).

Canonical correspondence analysis (CCA) was used to elucidate the relationships between the absolute frequencies of species and the corresponding collection and environmental data, and the statistical significance of specific relationships was tested via Chi-squared tests in the XLStat2009® software program (Addinsoft, 2009).

#### 3. Results

## 3.1. Trophic state and physical and chemical water conditions

Water of the Itaqui reservoir was classified as hypereutrophic by the TSI, with high levels of phosphorus ( $80.42 \ \mu g \ L^{-1}$  in 2008 and 79.90  $\ \mu g \ L^{-1}$  in 2009) and chlorophyll a ( $67.70 \ \mu g \ L^{-1}$ ). Water that enters the reservoir from the Itaqui River has high total phosphorus (79.18  $\ \mu g \ L^{-1}$  in 2008 and  $68.85 \ \mu g \ L^{-1}$  in 2009) and high biomass content ( $61.54 \ \mu g \ L^{-1}$  in 2008 and  $66.72 \ \mu g \ L^{-1}$  in 2009), and these conditions worsen in the reservoir. Reservoir water does not meet Brazilian water body quality standards for human consumption, fishing, and the irrigation of crops and orchards (Table 3).

| 2008 February 200 |
|-------------------|
|                   |

Table 3 - Abiotic and bacteriological variables measured in water samples from the Itaqui reservoir between May 2008 and

| Water attributes   | May 2008 | August 2008 | November 2008 | February 2009 |
|--|----------|-------------|---------------|---------------|
| Nitrate (NO <sub>3</sub> ; mg L <sup>-1</sup> )                    | 2.40     | 0.05        | < 0.01        | 0.45          |
| Total Phosphorus (P; mg L <sup>-1</sup> )                          | 0.82     | 1.49        | 0.54          | 0.50          |
| Orthophosphate (mg L <sup>-1</sup> )                               | 0.09     | 0.02        | 0.04          | 0.15          |
| Biochemical oxygen demand (BOD <sub>5</sub> ; mg L <sup>-1</sup> ) | 6.2      | < 10        | < 10          | 16            |
| pH   | 5.71     | 6.62        | 5.86          | 6.57          |
| Dissolved oxygen (mg L <sup>-1</sup> )                             | 3.50     | 0.85        | 2.82          | 5.25          |
| Air temperature (°C)   | 12.8     | 16.1        | 18.1          | 24.3          |
| Water temperature (°C)   | 14.0     | 17.5        | 20.8          | 24.7          |
| Secchi (cm)  | 44.8     | 44.8        | 67.2          | 134.4         |
| Total coliforms  | 2419.6   | 2419.6      | 1906.3        | > 2419.6      |
| Thermotolerants coliforms  | 579.4    | 579.4       | 10.8          | 1553.1        |

BOD<sub>5</sub> ranged between < 10 and 16 mg L<sup>-1</sup>. In May 2008 and February 2009 BOD5 values scored far above the established water quality standards for the proposed uses, which are 5 mg L<sup>-1</sup> according to CONAMA 367/2005 legislation. However, observed oxygen concentrations (between 0.85 and 3.50 mg L<sup>-1</sup>) were far lower than those permitted by Brazilian norms  $(\geq 5 \text{ mg L}^{-1})$ , except in February 2009 (5.65 mg L<sup>-1</sup>). Because the highest temperatures and BOD<sub>5</sub> were recorded in February, the high OD values recorded in that month were unexpected (5.25 mg  $L^{-1}$ ). On the other hand, the measurement was taken after a rainy week during a rainy summer, which suggests that rainfall may have increased water turbulence and led to greater oxygenation of the water. Chlorophyll a ranged 65.93 µg L<sup>-1</sup> (limits proposed by Brazilian norms are  $\leq 30 \ \mu g \ L^{-1}$ ), which reflect the production of algal biomass and total phosphorus was also higher, ranging from 0.50 to 1.49 mg L<sup>-1</sup> (the proposed limit is  $\leq 0.05 \text{ mg L}^{-1}$ ), both following CONAMA 357/2005.

#### 3.2. Analysis of the Itaqui reservoir diatom community

78 diatom taxa were identified, 13 of which formed the abundant diatom assemblage (Table 4). Subsamples (replicates) were not homogeneous, and were thus treated as independent samples in the grouping analysis. The primary clustering separated samples into two large groups: one consisting of samples from May and August 2008 (before macrophyte removal), which showed lower diatom densities, and other consisting of samples from November 2008 and February 2009 (after macrophyte removal), which showed higher diatom densities (see Figure 1). Despite replicate heteroscedasticity, the replicates of a given sampling period clustered together. The samples in the latter group corresponded to a period of increased temperature and light availability, as well as a deeper photic zone due to a reduction of shade from



**Figure 1** - Dendrogram generated using the Bray-Curtis coefficient (0.952 of similarity). Key: C1 = May 2008, C2 = August 2008, C3 = November 2008, C4 = February 2009. T1, T2, and T3 represent the replicates for each sampling date.

 Table 4 - Mean and standard deviation of densities of abundant diatom species over four seasons in the Itaqui reservoir (valves/cm<sup>2</sup>). Abundant species in each sample are highlighted (\*).

| Abundant species                  | May/08              | Aug/08              | Nov/08                | Feb/09                |
|-----------------------------------|---------------------|---------------------|-----------------------|-----------------------|
|                                   | Mean (± s)          | Mean (± s)          | Mean (± s)            | Mean (± s)            |
| Achnanthidium minutissimum        |                     |                     |                       | *4,616 ± 2,809        |
| Aulacoseira italica               | *7,479 ± 2,422      | *9,396 ± 3,003      | $1,\!585\pm1,\!123$   | $2,392 \pm 1,436$     |
| Cocconeis placentula var. lineata | 6,640* ± 2,617      | $348 \pm 110$       | $266\pm460$           | $1,200 \pm 487$       |
| Encyonema silesiacum              | $320\pm360$         |                     | $374\pm327$           | *10,335 ± 1,901       |
| Eolimna minima                    | $522 \pm 62$        | $158 \pm 274$       | $1,\!996\pm1,\!038$   | *33,138 ± 8,905       |
| Eunotia bilunaris                 | $*26,799 \pm 6,983$ | $*28,495 \pm 4,381$ | *36,936 ± 7,426       | $1,507\pm763$         |
| Fragilaria parva                  | $1,770\pm514$       | *54,581 ± 21,066    | $971 \pm 1,069$       | *27,692 ± 5,593       |
| G. parvulum var. saprophilum      | $359\pm424$         | $316\pm547$         | *14,313 ± 5,450       | *5,325 ± 1,815        |
| Gomphonema gracile                | $1,\!131\pm399$     | $728 \pm 221$       | *8,251 ± 731          | $751\pm847$           |
| Gomphonema parvulum               | *12,990 ± 1,335     | *7,195 ± 4,286      | *80,677 ± 22,810      | *61,662 ± 2,779       |
| Lemnicola hungarica               | *3,770 ± 214        | $1,043\pm328$       | $2,286 \pm 1,204$     | $*17,190 \pm 2,737$   |
| Navicula cryptotenella            | $1,\!205\pm125$     | $186 \pm 322$       |                       | *11,014 ± 1,252       |
| Nitzschia palea                   | *4,424 ± 865        | $1,\!796\pm990$     | $19,924 \pm 11815$    | $8{,}695 \pm 4{,}425$ |
| Sellaphora seminulum              | $974\pm458$         |                     | $2,\!839 \pm 1,\!361$ | *12,351 ± 2,447       |
| Total density of abundant species | 68,383              | 104,242             | 170,418               | 197,868               |
| Total density                     | 78,620              | 108,072             | 203,526               | 241,761               |

macrophytes. Figure 2 shows that total diatom densities were lower before macrophyte removal and increased afterwards ( $p \le 0.01$ ).

The canonical correspondence analysis (CCA) was significant ( $\chi^2 = 243.727$ ; GL = 209; p < 0.0001), with the first two axes capturing 85.10% of variation in the dataset (Eigenvalue F1 = 0.373; F2 = 0.294). The first axis (47.61%) was related to variation in species abundance, while the second (37.49%) was related to species distributions with regard to temperature, light, and rainfall. Eunotia bilunaris (Ehr.) Mills (EUBI), Cocconeis placentula var. lineata (Ehr.) Van Heurk (CPLA), and Aulacoseira italica (Ehr.) Simonsen (AUIT) were the most abundant species in the May 2008 sampling period (C1), and were associated with turbidity and low water temperature. Fragilaria parva (Gru.) Tuji and Williams (FFAM) dominated the August 2008 sampling period (C2) and was associated with high levels of available phosphorus, following a period of heavy rainfall. The species Nitzschia palea (Kut) W.Smith (NPAL), Gomphonema parvulum (Kut) Kutzing (GPAR), and G. parvulum var. saprophilum Lange-Bertalot and Reichardt (GSAP) were associated with the November 2008 sampling period (C3), with lower phosphorus availability (even though November 2008 did not have the lowest phosphorus levels), and with low rainfall. The species Encyonema silesiacum (Bleish) Mann (ENSY), Eolimna minima (Gru) Lange-Bertalot (EOMI), Sellaphora seminulum (Gru) Mann (SSEM), Navicula cryptotenella Lange-Bertalot (NCRY), Lemnicola hungarica (Gru) Round and Basson (LHUN), and Achnanthidium minutissimum (Kut) Czarnecki (AMIN) formed an assemblage that was associated with the February 2009 sampling period (C4), with increased water temperature, high rainfall, and a deeper photic zone (see Figure 3).

#### 4. Discussion

In the case of the Itaqui reservoir, hypereutrophic conditions prompted the excessive growth of the macrophyte *Pistia stratiotes*, which colonised the surface of the



Figure 2 - Boxplot comparing diatom densities before and after the macrophytes mechanical removal in Itaqui Reservoir.



Figure 3 - Ordination of abundant diatom species in the seasonal samples taken between May 2008 and February 2009.

reservoir throughout 2008. The macrophyte cover had both aesthetic and economic impacts, preventing fishing and recreational activities and causing unpleasant smells due to rotting plant matter. After macrophyte removal in 2009, the environment was still classified as hypereutrophic, reflecting the high input of industrial and domestic wastes in the region. In hypereutrophic environments like the Itaqui reservoir, seasonality is the leading driver of variation in diatom species distributions. In addition to temperature and rainfall, the increased photic zone following the mechanical removal of the macrophytes had an important effect on diatom development and led to higher diatom densities.

Abundant and dominant species were considered the most suitable for characterising the physical and chemical conditions of the reservoir's water, since they occurred at the highest densities in the reservoir's eutrophic conditions. The quantitative abundance of a taxon can be a good indicator of sampling station conditions and thus a good indicator of environmental conditions (Descy, 1979). The reservoir had high phosphorus levels and was classified as hypereutrophic throughout the study. The assemblages of abundant species observed in every sampling period can be assumed to be tolerant of highly eutrophic conditions.

*Eunotia bilunaris*, *Cocconeis placentula* var. *lineata*, and *Aulacoseira italica* were associated with lower temperatures and shading caused by plant cover. The genus *Eunotia* has been commonly recorded in more acidic environments (Denicola, 2000). However, *Eunotia* species have rarely been mentioned in studies of trophic states, apart from reports of occurrence in low-nutrient (Gómez and Licursi, 2001) and mesotrophic sites (Stenger-Kovács et al., 2007), and their characterisation as typical of oligosaprobic environments (Patrick and Reimer, 1966). In Brazil, *E. bilunaris* is also considered indicative of less polluted environments (Lobo et al., 2004a; Bere and Tundisi, 2010). However, Salomoni et al. (2006; 2011) studied epilithic diatoms in Gravataí River (RS, Brazil)

and found the species tolerant of eutrophic conditions and an indicator of organic pollution. In our study *E. bilunaris* was common in May, August, and November 2008 and showed high tolerance of eutrophic environments.

*A. italica* was common in May and August 2008 and has been reported as tolerant of mesoeutrophic conditions (Yang and Dickman, 1993; Van Dam et al., 1994). *C. placentula* var. *lineata*, common in May 2008, is considered sensitive to organic pollution (Krammer and Lange-Bertalot, 1986; Salomoni and Torgan, 2008) but has also been reported as broadly tolerant of nutrient inputs across a range from unpolluted sites to moderately and strongly eutrophic ones (Lobo et al., 2004a,b).

Fragilaria parva dominated the August 2008 sampling period with the highest phosphorus concentrations and high rainfall, and was abundant in February 2009, showing a good tolerance of eutrophic conditions. The species has a somewhat confusing taxonomy, which may hinder an understanding of its autecology. Fragilaria parva (Grunow) Tuji and Williams is a new combination based on Synedra familiaris f. parva Grunow lectotype for Tuji and Williams (2008). The authors cited F. parva as an important taxon for freshwater ecological periphyton studies. Van Dam et al. (1994) recorded it as Synedra rumpens var. familiaris and considered it typical of oligo-mesotrophic conditions, while Hoffman (1994) considered it tolerant of eutrophic conditions. While this taxon has been recorded in taxonomic studies, little mention of its environmental preferences has been made by Brazilian studies like Lozovei and Shirata (1986) and Contin (1990), which recorded it in polluted rivers in Paraná state, Brazil.

Nitzschia palea, Gomphonema parvulum, and G. parvulum var. saprophilum were associated with lower phosphorus availability, lower rainfall, and the beginning of macrophyte removal. G. parvulm f. saprophilum was common in November 2008 and February 2009, months after macrophyte removal, and is considered tolerant of eutrophic conditions (Blanco et al., 2004; Hofmann, 1994) and characteristic of hypereutrophic environments (Van Dam et al., 1994). G. parvulum is considered typical of eutrophic conditions (Van Dam et al., 1994; Blanco et al., 2004; Stenger-Kovács et al., 2007; Phiri et al., 2007;) and highly tolerant of organic pollution (Lange-Bertalot, 1979; Kobayasi and Mayama, 1989). In Brazil it is commonly recorded in eutrophic environments (Oliveira et al., 2001; Lobo et al., 2004a,c; Cetto et al.; 2004; Salomoni et al., 2006). In our study, the species was present during all sample periods, but its maximum density peaks were associated with lower phosphorus levels. Similarly, Souza (2002) and Salomoni et al. (2011) recorded this species in oligotrophic and mesotrophic lotic environments in Brazil.

*N. palea*, which was common in May and November 2008, is commonly cited as an indicator of eutrophic conditions (Lange-Bertalot, 1979; Gómez and Licursi, 2001; Lobo et al., 2004a,b,c; Salomoni et al., 2006; Potapova and Charles, 2007; Stenger-Kovács et al., 2007), β-meso-

saprobic to polysaprobic environments (Salomoni et al., 2011), and polysaprobic environments (Descy, 1979), and is often reported to occur in water with low concentrations of dissolved oxygen (Van Dam, et al., 1994; Hofmann, 1994; Duong et al., 2006). *N. palea*, like *G. parvulum*, was considered an indicator of high phosphorus levels in a study that examined a gradient of phosphorus concentrations in a protected area in Florida (Cooper et al., 1999) and highly polluted sites in Brazilian rivers (Bere and Tundisi, 2010). By contrast, Salomoni and Torgan (2008) reported that these species were present in every sample of their study on Guaíba Lake (RS, Brazil) and did not serve as indicator species.

Encyonema silesiacum, Eolimna minima, Sellaphora seminulum, Navicula cryptotenella, Lemnicola hungarica, and Achnanthidium minutissimum formed an assemblage that was associated with higher water temperature, high rainfall, and a deeper photic zone after macrophyte removal. S. seminulum and E. minima were common in February 2009. The former's tolerance of eutrophic conditions has been documented by Lobo et al. (2002) in a subtropical river in southern Brazil and by Gómez and Licursi (2001) using epipelic diatoms and the Diatom Pampean Index (DPI) in benthic samples from lotic subtropical environments in Argentina. S. seminulum has been noted as typical of eutrophic and polysaprobic environments (Van Dam, et al., 1994; Hofmann, 1994; Potapova and Charles, 2007). The species has also been considered an indicator of eutrophication in rivers of southern Brazil (Lobo et al., 2004a). The authors of this latter study, as well as Stenger-Kovács et al. (2007), also characterised E. minima as tolerant of eutrophic conditions. The species' high tolerance of pollution has also been noted (Lange-Bertalot, 1979; Duong et al., 2006).

A. minutissimum was abundant in February 2009. Ács et al. (2005) also found this species to be dominant during summer in Hungary's eutrophic Lake Velence. In Brazil, the taxon has been recorded by Lobo et al. (2002; 2004a) in southern rivers and streams, and by Cetto et al. (2004) and Silva (2010) in Paraná's Iraí reservoir. Both habitats are considered eutrophic, which corroborates our results. N. cryptotenella, common in February 2009, is depicted in the literature as tolerant of highly eutrophic conditions (Lobo et al., 2002, 2004a,c; Salomoni et al., 2006; Stenger-Kovács et al., 2007; Potapova and Charles, 2007), characteristic of mesoeutrophic and β-mesosaprobic conditions (Blanco et al., 2004), and tolerant of high nutrient levels (Van Dam and Mertens, 1993; Van Dam et al., 1994; Hofmann, 1994). However, Salomoni et al. (2008) considered it an indicator of oligossaprobic conditions in a lake in southern Brazil.

*L. hungarica* was common in May 2008 and February 2009. The species was considered tolerant of eutrophic conditions (Van Dam et al., 1994; Hofmann, 1994; Lobo et al., 2004c; Stenger-Kovács et al., 2007). Lange-Bertalot (1979) mentioned it as one of various species that are tolerant of  $\alpha$ -mesosaprobic conditions, but which

do not thrive in polysaprobic conditions. However, it was characterised as an indicator of β-mesosaprobic to polysaprobic environments by Salomoni et al. (2011). In our study *E. silesiacum* was common in February 2009. In other studies it has been recorded in low nutrient environments, especially phosphorus (Blanco et al., 2004; Lobo et al., 2004a,c; Stenger-Kovács et al., 2007).

This study contributes to increase the knowledge of periphytic diatoms that tolerate highly eutrophic environments in subtropical climate. F. parva showed high tolerance to hypereutrophication. A. italica, C. placentula var. lineata, and E. silesiacum have not been previously recorded from environments with trophic state similar to the Itaqui reservoir, but based on their quantitative importance to the reservoir's diatom community they can be considered tolerant to eutrophic conditions. E. bilunaris was abundant and showed high tolerance of eutrophic environments. E. minima, S. seminulum, A. minutissimum, N. cryptotenella, and L. hungarica are commonly recorded in literature in eutrophic and polluted environments. N. palea, G. parvulum and G. parvulum f. saprophilum are also commonly recorded as tolerant to eutrophication, however in this study they cannot be considered good indicators of the trophic state, since they were correlated with lower phosphorus levels.

In this study, abundant diatoms were found in the hypereutrophic conditions of the reservoir, showing high tolerance to high nutrient levels, and may thus be considered as potential bioindicators of eutrophic sites in subtropical environments. In this context, these data are suitable as a foundation for future studies and can help to calibrate models that search to use diatoms as bioindicators of water quality.

#### Acknowledgments

We thank the Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (CAPES) for a Master's scholarship awarded to DMF, and CNPq for financial support (CTHidro - 555397/2006-8) and for a productivity grant to TAVL.

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