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REVIEW OF THE USE OF MICROALGAE IN SOUTH AMERICA FOR MONITORING RIVERS. WITH SPECIAL REFERENCE TO DIATOMS

E. A. LOBO^{a*}, V. L. M. CALLEGARO^b, G. HERMANY^c, N. GÓMEZ^d, L. ECTOR^e

^a Laboratory of Limnology, University of Santa Cruz do Sul (UNISC),

Av. Independência, 2293 Santa Cruz do Sul, RS Brazil

^b Natural Sciences Museum, Zoobotanical Foundation of Rio Grande do Sul (FZB),

Av. Salvador França, 1427 Porto Alegre, RS Brazil

^c Graduate Course on Ecology. Federal University of Rio Grande do Sul (UFRGS), Av. Bento Gonçalves, 9500, Campus do Vale, Porto Alegre RS Brazil

Institute of Limnology Dr. Raúl A. Ringuelet (CONICET-UNLP),

P. O. Box 712-1900 La Plata, Buenos Aires, Argentina

^e Public Research Center - Gabriel Lippmann, CREBS, Avenue de la Faïencerie, 162a 1511 Luxembourg

*Author to whom correspondence should be addressed: lobo@unisc.br

WATER QUALITY ASSESSMENT BIOMONITORING BIOTIC INDEX MICROALGAE EPILITHIC DIATOMS EPIPELIC DIATOMS ARGENTINA BRAZIL

EVALUATION DE LA QUALITÉ DE L'EAU INDICE BIOTIQUE MICROALGUES DIATOMÉES ÉPILITHIQUES DIATOMÉES ÉPIPÉLIQUES ARGENTINE BRÉSII

ABSTRACT. - Microalgae communities have been used for monitoring continental freshwaters in South America, mainly in Argentina and Brazil. Considering that the epipelic diatom community dominates the sediments of running waters from the Argentinean Pampean plain, and index named Diatom Pampean Index (DPI) was formulated, aiming to assess organic pollution and eutrophication. Based on the tolerance of epilithic diatom species to water organic pollution and eutrophication in rivers and streams of Southern Brazil, the Biological Water Quality Index (BWQI) was formulated, integrating the effects of both stressors.

RÉSUMÉ. – Les communautés de microalgues ont été utilisées pour le monitoring des eaux douces continentales en Amérique du Sud, principalement en Argentine et au Brésil. Etant donné qu'en Argentine les eaux courantes de la plaine de la Pampa sont dominées par les Diatomées épipéliques, un indice appelé Îndice Diatomique de la Pampa (Diatom Pampean Index DPI) a été mis au point dans le but d'estimer la pollution organique et l'eutrophisation. Basé sur la tolérance des espèces de Diatomées épilithiques vis-à-vis de la pollution organique et de l'eutrophisation dans les rivières et ruisseaux du Sud du Brésil, l'Indice Biologique de Qualité de l'Eau (Biological Water Quality Index BWQI) a été défini en tenant compte de l'effet de ces deux facteurs.

Introduction

In South America, microalgal communities have been utilized for monitoring and evaluating water quality in continental waters. However, the first studies were published only in the second half of the 20th century and were restricted to research groups located, basically, in Argentina and Brazil.

In Argentina, the first study to relate microalgae with contamination of lotic systems was carried out by Del Giorgio et al. (1991) in the Luján River. In a review of the use of microalgae for water quality assessment in Argentina, Loez & Topalián (1999) pointed out that the most recent studies were related mainly with the phytoplankton assemblages of the Pampean Plain. The singularities of these aquatic systems require a particular methodology for its monitoring with algae, and the use of epilithon, frequently applied in biomonitoring elsewhere, had to be replaced by the use of epipelon in some lotic system of the Pampean Plain.

In Brazil, the research on phytoplankton ecology started in 1950's, even though limnological studies of sanitary emphasis took place since the thirties. Although the data obtained were empirical and qualitative, these studies are rather important, representing the studies carried out in hydrographic basins, reservoirs and water supplies (Rocha 1992). From the end of the nineties onwards, the number of studies in this area increased considerably; lotic systems, however, are still not thoroughly studied, despite the large hydrographical network in the country.

In Central America, Michels-Estrada (2003) working with the species composition and ecological requirements of benthic diatom assemblages from various Costa Rica's streams and rivers, pointed out that baseline information on the ecology of aquatic systems is urgently needed in the tropics in order to develop new and effective biological methods of monitoring water quality. Limnological and ecological concepts developed for temperate zones have to be checked to see if they can be applied in tropical conditions.

Microalgae and pollution: research in Argentina

Argentina has extensive hydric networks, spread throughout its territory, with different preservation status. The most important stresses on rivers and streams are organic enrichment, nutrients, heavy metals, pesticides, herbicides and physical changes produced by dredging and canalisation.

The geographical area known as "Pampa" comprises the plains of the southern half of Rio Grande do Sul State (Brazil), Uruguay and East of Argentina, between the latitudes of 30 and 39° South. This zone is intensely exploited due to the fertility of its soil. The most important urban centre in Argentina is located in this area, with more than 14 million inhabitants. Agriculture and cattle-raising are the main activities in most parts of the area but urbanization has promoted industrial activity. The intensive exploitation of the area has impacted some of the aquatic systems with different intensities, leading to deterioration of water quality and destruction of habitats.

Streams and rivers in the Pampa originate in shallow depressions of the plain or on the hills that occupy a small area. Running waters have a slow current due to the low slope (0.25-1.3 m km⁻¹). The headwaters lack riparian vegetation and the grass-lands represent the characteristic biome. The bottom substrate is mostly composed of slime-clay with low proportions of gravel and sand.

In these lotic systems, phytoplankton is favored by low current velocities; epilithon develops in the rocky headwaters of some streams, while epipelic communities are very well represented along the watercourses. Epiphytic algae, on the other hand, develop on a diverse community of aquatic macrophytes (emergent, rooted and floating species), unequally distributed along the rivers.

Since 1997 a series of projects were implemented, funded by several Argentinean institutions (National Council of Scientific Research – CONICET, National University of La Plata – UNLP, National Agency for the Promotion of Science and Technology – ANPCyT), aiming to use different biological associations (phytoplankton, phytobenthos, macroinvertebrates, aquatic macrophytes and fish), for monitoring biological and ecological status of lotic systems of the Pampean Plain. In order to achieve these aims, the response of phytoplanctonic and phytobenthic algae to different factors causing environmental stress was explored in an area of 50.000 km^2 , located in the Buenos Aires area and coastal zone of the La Plata River ($34^\circ 51' - 37^\circ 59' \text{ S}$; $57^\circ 21' - 59^\circ 08' \text{ W}$). Both intensive and extensive studies were carried out (Table I).

Epipelic communities turned out to be a very useful alternative for biomonitoring lotic systems on these plains, whereas epilithic communities are virtually inexistent. Artificial substrata were not considered a practical alternative, since they were frequently stolen or destroyed. An index named Diatom Pampean Index (DPI) was formulated, aiming to evaluate the organic pollution and eutrophication in rivers and streams of the Pampa (Gomez & Licursi 2001). In this work, 164 seasonal epipelon samples were collected by pipetting from the superficial sediment layer, at 50 sampling sites along twelve lotic systems. Also conductivity, pH, temperature, dissolved oxygen, nitrate (NO₃⁻⁻ N), nitrite $(NO_2^{-}-N)$, ammonium $(NH_4^{+}-N)$, phosphate ($PO_4^{3-}-P$), chemical oxygen demand (COD) and biochemical oxygen demand (BOD) were measured. To formulate the index, 210 species were classified according to their sensitivity to organic pollution and eutrophication, taking into account their responses to the concentrations of $PO_4^{3-}P$, NH₄⁺-N and BOD (Table II). The IDP can be calculated as follows:

$$IDP = \frac{\sum (I_{IDP} \cdot A)}{\sum (A)}$$

where I_{IDP} is the specific index value, according to the classification of Gomez & Licursi (2001) and A the relative abundance of each species in the sample. This index ranges from 0 to 4 and determines 5 levels of water quality (Table III).

This regional index based on the use of epipelon community is an important tool for the study of rivers and streams of the Pampean Plain, where the use of indices developed for other latitudes not always gives adequate results in terms of water quality evaluation.

Perspectives of the work with microalgae in Argentina

Results of studies with microalgae have contributed to the diagnosis of water quality in some lotic systems (Gómez 1998, Gómez 1999, Gómez & Licursi 2001, Bauer *et al.* 2002*a*, Licursi & Gómez 2002, Tolcach & Gómez 2002). Their integration with a macroinvertebrate index (Rodrigues Capítulo *et al.* 2001) and physical-chemical variables allowed the first assessments of the ecological and biological status of some hydrographic basins on the Pampean Plain (Bauer *et al.* 2002*b*). Tables I and II. – Top, study alternatives with phytoplankton and epipelic communities for monitoring rivers and streams in the Pampean Plain (Argentina). Bottom, characterisation of water quality classes in relation to BOD_5 , NH_4^+ -N, PO_4^{3-} -P (mg l⁻¹) and Interpretation of DPI (Diatom Pampean Index)

	Extensive studies	Intensive	studies				
Study strategies	Sampling periodicity: Seasonal	Sampling periodicity: Weekly-daily					
	In references and polluted sites of rivers and streams	In Situ In some sampling sites of rivers and streams Incubations of monoculture Translocation of microbenthic communities Reference sites Polluted sites	Ex Situ In laboratory Phytoplankton species and biofilms (colonised in field) exposed to stress factors (heavy metals, organic compounds, etc)				
Results	*Regional characterisation *Selection of biotics integrity metrics	*Response to stress factors: stru *Resilience and resistance	ctural and functional changes. of algae assemblages.				
Water	Colour BOD ₅ NH ₄ ⁺ -N PC Code	D ₄ ³⁻ -P IDP Significance	Degree of disturbance				

quality	Code						disturbance
0	Blue	<3	<0.1	<0.05	0-0.5	Very good: without pollution	Very slight
I	Green	>3-8	>0.1-0.5	>0.05-01	>0.5-1.5	Good: slightly polluted	Slight: extensive cattle-raising and agriculture
H	Yellow	>8-15	>0.5-0.9	>0.1-0.5	>1.5-2	Acceptable: moderately polluted and eutrophicated	<i>Moderate</i> : agricultural activity and/or intensive ranching.
111	Orange	>15-25	>0.9-2	>0.5-1	>2-3	<i>Bad</i> : strongly polluted and eutrophicated	Strong: intensive agriculture and cattle raising, moderate industrial activities, and population densities
IV	Red	>25	>2	>1	>3-4	Very bad: very strongly polluted and high concentration of organic matter	Very strong: intensive industrial activities and high population densities

Nevertheless, it is necessary to intensify phycological studies, particularly those with a functional approach, to obtain a better choice of biotic integrity measures, for an efficient diagnosis of water quality. Finally, the experience acquired in the Argentinean Pampa area should be integrated with that obtained by research groups in Brazil. This is an essential step towards the establishment of common protocols.

Microalgae and pollution: research in Brazil

In Brazil, the use of algae as pollution indicators started in the second half of last century (Rocha 1992), with several studies carried out in small lakes and reservoirs. The main studies on phytoplankton related to pollution published in Brazil are listed in Table IV.

biotic indices based Work with on phytoplankton began in the mid eighties. At the Sanitation Technology Company of São Paulo State (CETESB), the Saprobic Index was used under the supervision of Dr Sládecek. This system, however, was abandoned in 1991, since studies about the ecological valences of diatom species to tropical climate were necessary. From 1983 the Trophic State Index was adopted in São Paulo State for use in reservoirs. At the same time, the Institute for the Environment of Paraná developed a water quality matrix, including parameters of phytoTable III. – Diatom species frequently found in the epipelon of Pampean streams and rivers and ecological preferences according to quality of the water (Gomez & Licursi 2001).

TAXON	Water quality class	Specific index value (I_{IDP})
Achnanthidium minutissimum (Kützing) Czarnecki	0-I	1
Amphora coffeaeformis (Agardh) Kützing	III-IV	3,75
Amphora copulata (Kützing) Schoeman & Archibald	II-III	2,5
Amphora ovalis (Kützing) Kützing	II-III	2,25
Amphora perpusilla (Grunow) Grunow	I-II	1,75
Amphora veneta Kützing	III-IV	3,5
Anomoeoneis sphaerophora (Ehrenberg) Pfitzer	III-IV	3,25
Bacillaria paradoxa Gmelin	I-II	1,75
Caloneis amphisbaena (Bory) Cleve	I-II	1,5
Caloneis bacillum (Grunow) Cleve	I-II	1,5
Caloneis ventricosa (Ehrenberg) Meister	I-II	1,5
Cocconeis placentula Ehrenberg	I-III	2
Cocconeis placentula Ehrenberg var. euglypta (Ehrenberg) Grunow	II-III	2,25
Cocconeis placentula Ehrenberg var. lineata (Ehrenberg) Van Heurck	I-III	2
Craticula accomoda (Hustedt) Mann	III-IV	3,5
Craticula cuspidata (Kützing) Mann	II-IV	3
Craticula halophila (Grunow ex Van Heurck) Mann	п	2
Cymbella excisa Kützing	I-II	1,75
Cymbella neocistula Krammer	I-II	1.25
Cymbella lanceolata (Ehrenberg) Van Heurck	I-II	1,25
Denticula kuetzingii Grunow var. kuetzingii	I-II	1.5
Diadesmis confervacea Kützing	II-III	2,75
Diatoma vulgaris Bory	I-II	1.5
Diploneis ovalis (Hilse ex Rabenhorst) Cleve	I-II	1.25
Diploneis puella (Schumann) Cleve	I-II	1.25
Encvonema minutum (Hilse in Rabenhorst) D.G. Mann	I-0	0,5
Encyonema silesiacum (Bleisch in Rabenhorst) D.G. Mann	I-II	1,75
Eolimna subminuscula (Manguin) Moser Lange-Bertalot & Metzeltin	III-IV	3,75
Epithemia adnata (Kützing) Brébisson	I-II	1,25
Epithemia sorex Kützing	I-II	1,75
Eunotia monodon Ehrenberg	I	1
Eunotia pectinalis (Dyllwyn) Rabenhorst var. pectinalis	I	1
Fallacia pyemaea (Kützing) Stickle & Mann	II-III	2.75
Fragilaria capucina Desmazières	0-I	0.5
Gomphonema angustum Agardh	0-11	1
Gomphonema clavatum Ehrenberg	I-II	1.25
Gomphonema parvulum (Kützing) Kützing	II-IV	3.25
Gomphonema truncatum Ehrenberg	I-II	1.25
Gyrosigma acuminatum (Kützing) Rabenhorst	TI I	2
Hantzschia amphiorys (Ehrenberg) Grunow	 1-IU	2
Hippodonta capitata (Ehrenberg) Lange-Bertalot, Metzeltin & Witkowski	I-III	2.75
Hippodonta hungarica (Grunow) Lange-Bertalot Metzeltin & Witkowski	I-III	2
Lemnicola hungarica (Grunow) Round & Basson	11-111	2.5
Luticola goeppertiana (Bleisch in Rahenhorst) D.G. Mann	III-IV	3.75
Melosira varians Agardh	I-III	2
Meridion circulare (Greville) C.A. Avardh	0-1	0.25
Navicula canitatoradiata Germain	I-II	1 25
Navicula cryntocenhala Kützing	LIV	3
Navicula gregaria Donkin	1-11	2 75
Navicula tripunctata (Ω F Möller) Bory	I-III	2,72
Nitzschia acicularis (Kützing) Smith	111-17	3 75

Table III. – *Continued*.

Nitzschia amphibia Grunow	1-111	2,5
Nitzschia semirobusta Lange-Bertalot	I-III	2,5
Nitzschia angustata (W. Smith) Grunow	II-III	2,5
Nitzschia brevissima Grunow	п	2
Nitzschia dissipata (Kützing) Grunow	I-II	1,25
Nitzschia draveillensis Coste & Ricard	I-II	2
Nitzschia filiformis var. conferta (Richter) Lange-Bertalot	I-III	2,25
Nitzschia fonticola Grunow	I	1
Nitzschia frustulum (Kutzing) Grunow	I-II	1,75
Nitzschia gracilis Hantzch ex. Rabenhorst	I-II	1,5
Nitzschia heufleriana Grunow	I-II	1,25
Nitzschia linearis (Agardh) W.M. Smith	II-III	2,5
Nitzschia palea (Kützing) W. Smith	II-IV	3,75
Nitzschia recta Hantzch ex Rabenhorst	I-II	1,75
Nitzschia sigma (Kützing) W. M. Smith	II-IV	3
Nitzschia sigmoidea (Nitzsch.) W.M. Smith	II-IV	3
Nitzschia umbonata (Ehr.) Lange-Bertalot	III-IV	3,75
Pinnularia gibba Ehrenberg	I-III	2,75
Pinnularia microstauron (Ehrenberg) Cleve	1-111	2,5
Pinnularia subcapitata Gregory	I-II	1,75
Pinnularia viridis (Nitzsch) Ehrenberg	I-II	1,75
Planothidium hauckianum (Grunow) Round & Bukhtiyarova	II	2
Planothidium lanceolatum (Brébisson) Round & Bukhtiyarova	I-II	1,5
Pleurosira laevis (Ehrenberg) Compere	I-II	1,75
Rhopalodia gibba (Ehrenberg) O. Müller	I-II	1,25
Rhopalodia musculus (Kützing) O. Müller	I-II	1,5
Sellaphora pupula (Kützing) Meresckowsky	II-IV	3
Stauroneis phoenicenteron (Nitzsch) Ehrenberg	II-III	2,25
Surirella angusta Kützing	II-III	2,5
Surirella biseriata Brebisson	I-II	1,25
Surirella brebissonii Krammer & Lange-Bertalot	I-III	2
Surirella ovalis Brebisson	II-III	2,5
Surirella tenera Gregory	I-II	1,75
Tryblionella apiculata Gregory	I-IV	3
Tryblionella hungarica (Grunow) D.G. Mann	II-III	2,75
Ulnaria ulna (Nitzsch) Compère	I-III	2

Table IV. M	ost important	Brazilian	publications	on	phytoplankton	and	their	relation to	pollution,	classified by	y Brazi-
lian region.											

Region	Authors
North	Sioli (1965)
Northeast	Wright (1935)
Middle-West	Palmer (1960, 1969); Oliveira & Krau (1970)
Southeast	Kleerekoper (1944); Branco (1964); Azevedo et. al. (1967); Bicudo & Bicudo (1967); Rocha & Branco (1970)
South	Souza (1970); Kleerekoper (1944); Pellin & Zini (1971)

plankton community structure. In the last ten years there was a significant increase in the number of studies using periphytic communities for water quality evaluation, mainly in urban reservoirs of São Paulo. In a review of Brazilian phytoplankton research, Huszar & Silva (1999) pointed out the small number of studies on lotic environments, despite the large hydrographic network of the country. They also mentioned the proliferation of studies on Table V. Saprobic values (s) and differential groups for epilithic diatoms in the Guaíba Hydrographic Basin, RS, according to Lobo *et al.* (2002). A (most pollution tolerant species), B (pollution tolerant species), C (less pollution tolerant species).

Species	S	Group
Achnanthidium minutissimum (Kützing) Czarnecki	4.0	A
Adlafia bryophila (Petersen) Moser. Lange-Bertalot & Metzeltin	4.0	Α
Amphipleura lindheimeri Grunow	4.0	Α
Fragilaria ulna (Nitzsch.) Lange-Bertalot var. ulna	4.0	Α
Gomphonema cf. clevei .Fricke	4.0	Α
Gomphonema pseudoaugur Lange-Bertalot	4.0	Α
Gyrosigma scalproides (Rabenhorst) Cleve	4.0	Α
Luticola goeppertiana (Bleisch) D.G. Mann	4.0	Α
Melosira varians Agardh	4.0	Α
Navicula cryptotenella Lange-Bertalot	4.0	Α
Navicula schroeterii Meister	4.0	Α
Nitzschia amphibia Grunow	4.0	Α
Nitzschia linearis (Agardh) W. Smith var. linearis	4.0	Α
Nitzschia nana Grunow in Van Heurck	4.0	Α
Nitzschia palea (Kützing) W. Smith	4.0	Α
Pinnularia gibba Ehrenberg	4.0	А
Pinnularia maveri Krammer	4.0	Α
Planothidium rostratum (Oestrup) Lange-Bertalot	4.0	А
Sellanhora pupula (Kützing) Mereschkowsky	4.0	Α
Sellanhora rectangularis (Gregory) Lange-Bertalot & Metzeltin	4.0	A
Surirella ovata (Kützing) Cleve var. smithii Cleve-Euler	4.0	A
Achnanthes exigua Grunow var exigua	2.5	B
Achnanthes inflata (Kützing) Grunow	2.5	B
Amphora montana Krasske	2.5	B
Cocconeis placentula Ehrenberg var euglypta (Ehrenberg) Grunow	2.5	B
Cocconeis placentula Ehrenberg var placentula	2.5	B
Cyclotella meneohiniana Kiitzing	2.5	B
Cymbella aff hustedtii Krasske	2.5	B
Cymbella affinis Kützing	2.5	B
Cymbella tumida (Bréhisson) Van Heurck	2.5	B
Diadesmis confervacea Kützing	2.5	B
Diadesmis contenta (Grunow ex Van Heurck) D. G. Mann	2.5	B
Encyonema silesiacum (Bleisch ex Rabenhorst) D.G.Mann	2.5	B
Fragilaria canucina Desmazières var rumpens (Kützing) Lange-		-
Bertalot	2.5	в
Geissleria, aikenensis (Patrick) Torgan & Oliveira	2.5	B
Gomphonema angustatum (Kützing) Rabenhorst	2.5	B
Gomphonema argcile Ehrenberg	2.5	B
Gomphonema parvulum (Kützing) Kützing v. parvulum	2.5	B
Gyrosiama acuminatum Fhrenherg	2.5	B
Mayamea atomus (Kützing) Lange-Bertalot	2.5	B
Navicula rostellata Kützing var rostellata (Kützing) Cleve	2.5	B
Navicula cryptocenhala Kützing	2.5	B
Navicula gregaria Donkin	2.5	B
Nitzschia siama (Kützing) W Smith	2.5	B
Planothidium lanceolatum (Bréhisson) Round & Bukhtivarova	2.5	B
Sellanhora seminulum (Grunow) D. G. Mann	2.5	B
Surirella angusta Kützing	2.5	B
Surirella linearis W Smith	2.5	B
Ulnaria ulna (Nitzsch) Compère	2.5	B
Frustulia rhomboides (Fhrenberg) De Toni	10	Č
Comphonema angustum Agardh	1.0	č
Gomphonema neonasutum Lange-Bertalot & Rechardt	1.0	č

phytoplankton ecology in the nineties, with 111 studies published until December 1998, and the increasing occurrence of toxic cyanobacteria in Brazilian waters. Hence, the Ministry of Health issued an edict in 2000, determining that cyanotoxin analyses will be necessary every time cyanobacterial densities become higher than 20.000 cells mL^{-1} at water treatment stations.

Regarding to the use of epilithic diatom communities for water quality evaluation in continental

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Table VI. TWINSPAN groups with respective abundant species and indicative values related to eutrophication, according to Lobo *et al.* (2004).

Species	Twinspan	Tolerance to	Indicative	
A web and we of the Company	Group	Eutrophication		
Emistilia of anxished di Unoto di	C	very Low	1	
<i>Frustutia</i> c1. <i>weinnotati</i> Husteut				
Luticola goeppertiana (Bleisch in Rabennorst)				
D. G. Mann				
Geissleria aikenensis (Patrick) Torgan &				
Oliveira				
Rhopalodia gibberula (Ehrenberg) O. Muller				
Surirella angusta Kützing				
Ulnaria ulna (Nitzsch) Compère	_	_		
Cocconeis placentula Ehrenberg var. placentula	D	Low	2	
Encyonema silesiacum (Bleisch) D. G. Mann				
Gomphonema angustum Agardh				
Nitzschia amphibia Grunow				
Planothidium rostratum (Oestrup) Lange-				
Bertalot				
Adlafia bryophila (Petersen) Moser. Lange-	Α	Medium	3	
Bertalot & Metzeltin				
Amphipleura lindheimeri Grunow				
Cocconeis placentula Ehrenberg var. euglypta				
(Ehrenberg) Grunow				
Cyclotella meneghiniana Kützing				
Cymbella affinis Kützing				
Cymbella aff, hustedti Krasske				
Diadesmis contenta (Grunow ex Van Heurck)				
D.G. Mann				
Gomphonema cf. clevei Fricke				
Melosira varians Agardh				
Navicula cryptotenella Lange-Bertalot				
Navicula gregaria Donkin				
Navicula symmetrica Patrick				
Nitzschia linearis (Agardh) W Smith				
Nitzschia nalea (Kützing) W. Smith				
Folimna minima (Grunow) Lange-Bertalot	в	High	4	
Eragilaria capucina Desmazières var rumpens	Б	Tign	-	
(Kützing) Lange-Bertalat				
(Kutzing) Lange-Dertaiot				
Bahanharat				
Gomphonema parvulum Kutzing				
Navicula rostellata Kutzing	F	V	E	
Acnnantnes exigua Grunow var. exigua	E	very High	5	
Achnanthidium minutissimum (Kutzing)				
Czarnecki				
Mayamea atomus (Kützing) Lange-Bertalot				
Sellaphora seminulum (Grunow) D. G. Mann				

freshwaters, little attention has been paid to the use of this group as bioindicator. Only a few related studies have been carried out, especially in the South of Brazil (i.e. Lobo *et al.* 1996, Lobo & Bender 1998, Oliveira *et al.* 2001). A detailed review of the use of epilithic diatoms as bioindicator organisms in lotic systems of Southern Brazil between 1988 and 1998 was published by Lobo & Callegaro (2000).

The first attempt to classify diatom species according to their tolerance to organic pollution was made in Southern-Brazilian rivers in 1996 (Lobo *et al.* 1996). Continuing this research, Lobo *et al.* (2002) published the first Brazilian saprobic system. In this work, in order to describe the pattern of occurrence of diatom species to water pollution, a total of 183 samples of epilithic diatoms and water samples for determination of BOD₅ were collected in 31 stations from 18 lotic systems (including rivers and streams), belonging to the Guaíba Hydrographical Basin. This region comprises about 50 % of the municipal districts of Rio Grande do Sul State, including the capital and the metropolitan region, with an approximate area of 86.000 km². Its main course, named Jacuí River, has a length of about 720 km with a depth ranging

Table VII. Summary of the main chemical characteristics of the sample units that form the TWINSPAN groups of Lobo *et al.* (2004). VL= Very low tolerance to eutrophication; L= Low tolerance to eutrophication; M= Medium tolerance to eutrophication; H= High tolerance to eutrophication; VH= Very high tolerance to eutrophication.

		TWINSPAN Group						
		С	D	Α	B	E		
		VL	L	Μ	Η	VH		
Cond (μ S cm ⁻¹)	Mean	0.083	0.122	0.130	0.118	0.078		
	Standard deviation	0.041	0.025	0.48	0.130	0.032		
NID (mg L^{-1})	Mean	2.54	2.23	3.53	1.88	1.66		
	Standard deviation	1.35	0.08	5.12	1.82	1.32		
BOD (mg L^{-1})	Mean	34.6	24.3	11.9	4.7	6.1		
	Standard deviation	18.1	17.0	4.8	4.0	5.8		
$PO_4^{3}-P (mg L^{-1})$	Mean	0.059	0.070	0.058	0.172	0.251		
	Standard deviation	0.063	0.023	0.024	0.123	0.273		
* Percentage <	: 0.025 (mgL ⁻¹) PO ₄ ⁻³ -P	18%	0%	14%	0%	0%		
** Percentage <	$(0.037 (mgL^{-1}) PO_4^{-3} - P)$	36%	10%	21%	0%	0%		
*** Percentage <	$(0.050 (mgL^{-1}) PO_4^{-3} - P)$	82%	20%	29%	10%	13%		
**** Percentage >	$0.100 (mgL^{-1}) PO_4^{3-}-P$	9%	10%	0%	60%	56%		
***** Percentage	$> 0.200 (mgL^{-1}) PO_4^{3} - P$	0%	0%	0%	40%	44%		
******Percentage	$> 0.400 (mgL^{-1}) PO_4^{-3} P$	0%	0%	0%	0%	25%		

* Percentage of sampling units with total phosphate concentration less than 0.025 (mg L⁻¹) PO₄³-P ** Percentage of sampling units with total phosphate concentration less than 0.037 (mg L⁻¹) PO₄³-P

*** Percentage of sampling units with total phosphate concentration less than 0.057 (mg L⁻¹) PO_4^{-1} -P

**** Percentage of sampling units with total phosphate concentration higher than 0.100 (mg L⁻¹) PO_4^{-1} -P ***** Percentage of sampling units with total phosphate concentration higher than 0.200 (mg L⁻¹) PO_4^{-1} -P

***** Percentage of sampling units with total phosphate concentration higher than $0.200 \text{ (mg L}^{-1)} \text{PO}_4^{-2}\text{-P}$ ****** Percentage of sampling units with total phosphate concentration higher than $0.400 \text{ (mg L}^{-1)} \text{PO}_4^{-3}\text{-P}$

from 12 to 14 m and an average annual discharge ca. 714 m³ s⁻¹ (Lobo *et al.*1996). The relative abundance of each species was plotted against the logtransformed BOD₅ values of the water from which it was collected. The height of the mode for the distribution pattern of each species was determined by considering the weighted average between relative abundance and BOD₅. The different levels of pollution used to characterize the distribution pattern of each diatom species were determined following Hamm's (1969) chemical classification of water quality. The results indicated the occurrence of three differential diatom groups: Group A (highly pollution-tolerant species), Group B (pollution-tolerant species) and Group C (less pollution-tolerant species). The groups are listed in Table V.

Lobo et al. (2004) proposed the use of the Biological Water Quality Index (BWQI), integrating the effects of organic enrichment, from the classification proposed by Lobo et al. (2002), and eutrophication, from the indicative values determined using multivariate analyses techniques. Physical, chemical and biological data were obtained from the project "Water quality evaluation of Sampaio, Grande and Bonito Streams, Mato Leitão, RS, Brazil" carried out between 1993 and 1998. Sampling consisted of seasonal field trips to 12 sites. Water temperature (°C), pH, conductivity (Cond), turbidity (Turb), dissolved oxygen (DO), biochemical oxygen demand (BOD₅), dissolved inorganic nitrogen (DIN), phosphate (PO₄), and total solids (TS) were used as parameters for organic pollution and eutrophication assessment.

For an integrated analysis, firstly, species and samples were grouped using TWINSPAN - Twoway Indicator Species Analysis (Hill 1979). CCA -Canonical Correspondence Analysis (Ter Braak 1986) was then applied in order to evaluate the relationship between the abiotic variables and diatom species composition patterns, within the groups defined by TWINSPAN, using the covariance matrices. For each diatom species from the five TWINSPAN groups distributed along the eutrophication gradient detected, operational indicative values from one to five were given, based on their tolerances to eutrophication. These corresponded, respectively, to very low, low, medium, high, and very high tolerance levels. Table VI summarises this information, highlighting the indicative values for each species. Using these values, the Biological Water Quality Index (BWQI) can be calculated, according to the equation given by Wegl (1983), modified by the authors:

$$BWQI = \frac{\sum (s \cdot h \cdot vi)}{\sum (h \cdot vi)}$$

where s is the species saprobic value, according to the classification of Lobo *et al.* (2002); *h* is the percentage of occurrence (abundance) of each species in the sample and *vi* is the species indicative value. Values of BWQI vary from 1 to 4 in aquatic environments: 0-0.9 (pollution absent), 1.0-1.4 (low pollution), 1.5-2.0 (moderate pollution), 2.1-2.7 (heavy pollution) and 2.8-4.0 (very heavy pollution).

The characterisation of the trophic spectrum of this research was done on the basis of the percent-

ages of sampling units – for each TWINSPAN group – which were found to contain total phosphate values of less than 0.025, 0.032 and 0.050 (mg L⁻¹) PO₄³⁻-P, and values higher than 0.100, 0.200 and 0.400 (mg L⁻¹) PO₄³⁻-P (Table VI). These values corresponded, respectively, to: the limit established by the Brazilian National Environment Council for waters free of eutrophication; the phosphate concentration considered normal for freshwaters in Rio Grande do Sul State (Haase *et al.* 1997); the phosphate concentration necessary to cause eutrophication problems in streams that flow into lentic water bodies (Train 1979); the phosphate concentration considered excessive and extremely excessive according to Lobo *et al.* (2004).

The results of Lobo *et al.* (2004) generally confirmed conclusions by other studies in streams and rivers of Europe. *Sellaphora seminulum* and *Mayamea atomus*, grouped among the species with high tolerance to eutrophication, were described by Van Dam *et al.* (1994) as species characteristic of respectively eutrophic and hypereutrophic waters. The same authors described *Achnanthes exigua* var. *exigua* and *Achnantidium minutissimum* as species with broad tolerance ranges, occurring successfully from oligotrophic to eutrophic environments.

Kelly and Whitton (1995) assigned the indicative value five to *Gomphonema parvulum* and small (< 12 µm) species of the genera *Navicula* and *Sellaphora*, which were qualified as typical species of environments with phosphate values (≥ 0.3 mg L⁻¹. In the present study the same trend was found, with these species also showing high and very high tolerances to eutrophication. Kelly *et al.* (1996) reported *Cocconeis placentula* as dominant species in less eutrophic environments of the River Kennet, England; a similar situation was found in the present study, where it showed low tolerance to eutrophication.

The differences observed in the tolerances of some species such as *Ulnaria ulna* and *Luticola goeppertiana*, widely cited as eutrophic species, however classified as "less tolerant to eutrophication" taxa in Brazilian rivers, can be ascribed to specific adaptations to the regional ecotones, confirming that trophic indices developed in other countries are not always adequate to be used successfully in South American freshwater environments.

Perspectives of the work with microalgae in Brazil

In this country, there is a general lack of specialised human resources, mostly regarding taxonomical work. The majority of technicians and researchers in Brazil study industrial and domestic effluents for public water supply, where the need for immediate action makes accurate taxonomical work very difficult to carry out.

These aspects reinforce the importance of the creation of research groups dedicated to taxonomy, and the development of a National Centre for Taxonomy, serving as a data reporting for microflora, allowing knowledge transfer between Brazilian researchers and institutions.

Final remarks

It is clear that there are regional differences determining the use of distinct methodologies, such as the use of the epipelic community in Argentina for the assessment of the quality of surface freshwaters on the Pampean Plain.

Monitoring studies have to be approached with a holistic perspective, incorporating saprobic and trophic indices, as well as diatom and macroinvertebrate communities and, more recently, biological and ecological status of water bodies. A more specific analysis revealed the need to unify methodological criteria and technical information in order to establish unified investigation protocols.

Moreover, the development of biomonitoring Environmental Sciences in South America using microalgae and particularly diatoms is still incipient and restricted, basically, to Argentina and Brazil. Efforts should be put into increasing the number of researchers and countries involved in such studies, through programs of human resource development, especially in diatom taxonomy, as well as the permanent review of the tolerances of organic diatom species to pollution and eutrophication. Such efforts will allow the biological systems for environmental assessment to be kept up-to-date, aiming to develop a tested model adapted to the characteristics of the water systems of each country, able to fulfill the needs of routine water quality evaluation in freshwater ecosystems.

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