

Applicability of borrowed diatom-based water quality assessment indices in streams around São Carlos-SP, Brazil

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Abstract Diatom-based indices are increasingly becoming important tools for the assessment of ecological conditions in lotic systems. The applicability of regional and foreign diatom-based water quality assessment indices to streams around São Carlos-SP, Brazil, is discussed. The relationship between measured water quality variables and diatom index scores was assessed. The indices, when compared to chemical analyses, proved useful in providing an indication of the quality of the investigated waters. Though all borrowed indices were applicable to the study area because many widely distributed diatom species have similar environmental tolerances to those recorded for these species elsewhere, ecological requirements of some diatom species from Brazil need to be clarified and incorporated in a diatom-based water quality assessment protocol unique to the region.

Keywords Diatoms · Indices · Water quality · Biomonitoring

Introduction

Patterns of benthic diatom communities are responsive to the nature of the physical and chemical characteristics of lotic systems (Stevenson et al., 1996; Wehr & Sheath, 2003; Azim et al., 2005). They respond rapidly to degradation of water quality, often changing in both taxonomic composition and biomass where even slight contamination occurs (Round, 1991; Pan et al., 1996; Stevenson et al., 1996; Biggs & Kilroy, 2000; Potapova & Charles, 2003; Wehr & Sheath, 2003; Azim et al., 2005). Therefore, the integrity of these communities provides a direct, holistic and integrated measure of the integrity of lotic systems. Thus, pollution control and monitoring programmes routinely include the examination of diatoms to investigate the ecological status of lotic systems (e.g. Round, 1991; Pan et al., 1996; Gómez & Licursi, 2001; Lobo et al., 2002; Potapova & Charles, 2002, 2003). They have gained momentum in their usage as alternatives to chemical analyses because the latter techniques provide, at best, a fragmented overview of the state of lotic systems as sporadic or periodic sampling cannot reflect fluxes of effluent discharge. In contrast, diatom community structure in lotic systems gives a time-integrated indication of the water quality components (Taylor

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et al., 2007b). The unique composite picture of ecosystem conditions provided by the diatoms can only be replicated by intensive chemical monitoring studies.

Numerous studies focusing on the application of standardised methods based on diatom assemblages for water quality assessment have been carried out, especially in the northern hemisphere and in particular in European countries (e.g. Descy & Coste, 1991; Kelly & Whitton, 1995; Prygiel et al., 1999). Several diatom-based indices have been developed most of which are based on the weighted average equation of Zelinka & Marvan (1961) and are general pollution indices. There are as many indices as there are the number of researchers working in the field (Rimet et al., 2005). A discussion of the manner in which diatom indices function may be found in Harding et al. (2005).

The wide geographical distribution and well-studied ecology of most diatom species are cited as major advantages of using diatoms as indicator organisms (McCormick & Cairns, 1994). These assumptions imply that diatom-based water quality assessment tools should have universal applicability across geographical areas and environments. For this reason, due to lack of information on ecological preferences and tolerances of diatoms in some regions, indices developed in other regions are often borrowed. However, there is evidence that diatom metrics or indices developed in one geographical area are less successful when applied in other areas (Pipp, 2002). This is due not only to the floristic differences among regions (Pan et al., 1996; Taylor et al., 2007b) but also to the environmental differences that modify species responses to water quality characteristics (Potapova & Charles, 2005). Taylor et al. (2007b) recommended that borrowed diatom indices can be used for gaining support and recognition for diatom-based approaches to water quality monitoring allowing for sample and data collection, which can then be used later in the formulation of a unique diatom index. Strict testing of these borrowed indices is required to ensure that diatom index scores give a realistic reflection of the specific type of environmental pollution being tested.

The use of diatoms as indicators of water quality changes has relatively fewer precedents in South America compared to North America and Europe. In Brazil, the first studies on the use of aquatic biota,

particularly phytoplankton, for monitoring of the ecological status of lotic systems were carried out in the catchment areas of São Paulo City by a French researcher, Henric Charles Potel, between 1907 and 1910, based on empirical and qualitative data (Rocha, 1992). Other studies followed up on this work based on quantitative data. All these studies confirmed diatoms as excellent indicators of environmental conditions in lotic systems (e.g. Lobo & Torgan, 1988; Rosa et al., 1988; Lobo et al., 1991, 1996, 1999; Lobo & Callegaro, 2000). However, in most cases, the assessment of the ecological conditions of lotic systems was determined by foreign methods (e.g. Lange-Bertalot, 1979; Watanabe et al., 1986; Kobayasi & Mayama, 1989), because no information on diatom ecological preferences was present. This prompted the first attempts to classify diatoms in terms of tolerance of species to organic pollution in rivers in southern Brazil by Lobo et al. (1996). Subsequently, Lobo et al. (2002) determined the tolerance of diatom species to organic pollution. Based on this information, Lobo et al. (2002) developed the first saprobic system in the country, which uses epilithic diatoms for water quality assessment in southern Brazil. This study was completed by Lobo et al. (2004) leading to the formation of the first diatom-based water quality assessment index, called the Biological Index of Water Quality (BIWQ) trophic index, that incorporates the effects of organic contamination and eutrophication.

However, this index was developed in the southern part of the country and very little was done in other lotic systems. Given the high variations in climatic conditions among different regions in Brazil due to its geographical positioning, there is need for strict testing of the BIWQ to ensure that its scores give a realistic reflection of the specific type of environmental pollution being tested in other regions of Brazil. Thus, the objective of the present study was to test the applicability of the BIWQ, together with other indices developed in other regions and calculated by the OMNIDIA version 5.3 software, to the study area. This study presents the relationship between measured water quality variables in the Monjolinho River and its tributaries and diatom index scores. Diatom index scores were calculated and correlated to concurrent physical and chemical water quality data. The results of these correlation analyses

were compared to results obtained in similar studies carried out elsewhere, e.g. in Europe and South Africa. This paper discusses the limitations of the Brazilian-based index (BIWQ) compared to other indices.

Materials and methods

Study area

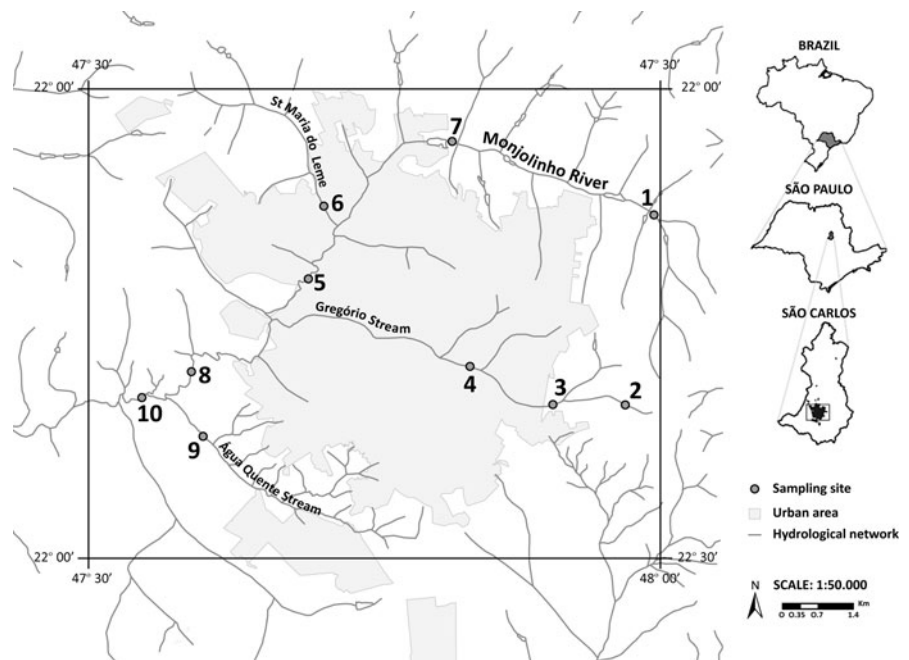
The study area is shown in Fig. 1. The headwaters of the Monjolinho River and the tributaries studied fall mainly within an agricultural area. From the agricultural area, the streams pass through urban area of the city of São Carlos, which covers a total area of 1,143.9 km². The area is characterised by a rugged topography and an average annual temperature of around 19.5°C, with mean monthly maximum of around 21.9°C recorded in January and February and the mean monthly minimum of around 15.9°C recorded in July.

In 2008, the population of São Carlos was estimated at 218,080 inhabitants by Instituto Brasileiro de Geografia e Estatística. Now, the expansion of the city does not meet the technical standards that go with it in terms of sewage treatment, collection of garbage, urban drainage and so on. Streams in the

study area, therefore, receive untreated or semi-treated effluent from various domestic and industrial sources as well as other diffuse sources as they pass through the city. This disorderly growth of the city results in stream health deterioration, loss of the remaining primary vegetation, organic pollution and eutrophication amongst other problems.

Ten sites were established along the Monjolinho river and its tributaries: four sites (1, 2, 3 and 7) in the relatively less impacted agricultural and forested headwaters to act as reference sites; three sites (4, 5 and 6) in the moderately polluted urban area; and three sites (8, 9 and 10) in highly polluted downstream area after the urban area (Fig. 1). The rationale for choosing the sampling sites was to obtain a pollution gradient of all the stream systems from relatively unpolluted agricultural headwaters to highly polluted urban downstream sites. Substrate assessment, diatom, and water quality sampling were done during dry seasons (autumn and winter) when flow was stable. Four samplings were carried out, two in September and October 2008 and two in May and June 2009. Sampling was done during the dry season to avoid variable effects of rainy season such as great variations in water level and velocity, floods and inundations. These factors affect diatom development, especially growth rates and relative abundance of different species (Round, 1991).

Fig. 1 Location of sampling sites in the study area



Environmental variables

At each site, dissolved oxygen (DO), electrical conductivity (EC), temperature, pH, and turbidity were measured using a Horiba U-23 and W-23XD Water Quality Meter (Horiba, Japan). Depth and current velocity were measured at each site with an FP 201 global flow probe (Global Water Instrumentation, AK, USA). The percentage riparian vegetation cover was visually estimated at each site. Altitude was determined at each site using a GPS (Northport Systems, Toronto, Canada).

Water samples for metals, ions, total nitrogen (TN), total phosphorus (TP) and biological oxygen demand (BOD₅) analysis were collected at each site into acid-cleaned polyethylene containers (APHA, 1988). In the laboratory, the concentrations of TN, TP and BOD₅ in the water samples were determined following standard methods (APHA, 1988). The following metals were analysed in water samples using flame atomic absorption spectrometry analytical methods (Varian Australia, Victoria, Australia): cadmium (Cd), lead (Pb), chromium (Cr), copper (Cu) and iron (Fe). The concentrations of fluoride (F⁻), chloride (Cl⁻), nitrate (NO₃⁻), phosphate (PO₄³⁻), sulphate (SO₄²⁻), sodium (Na⁺), ammonium (NH₄⁺), potassium (K⁺), calcium (Ca²⁺) and magnesium (Mg²⁺) were analysed by isocratic ion analysis using a suppressed conductivity detection ion chromatography method using a Dionex DX-80 Ion Analyser (Dionex Corporation, 2001).

Biological elements

At each site, epilithic diatom samples were collected as outlined in Bere & Tundisi (2010). Dead wood was used as a substrate in the absence of boulders as suggested by Kelly et al. (1995). In the laboratory, subsamples of the diatom suspensions were cleaned of organic material using wet combustion with concentrated sulphuric acid and mounted in Naphrax (RI = 1.74; Northern Biological Supplies, UK), following Biggs & Kilroy (2000). Three replicate slides were prepared for each sample. A total of 250–600 valves per sample (based on counting efficiency determination method by Pappas & Stoermer, 1996) were identified and counted using a phase-contrast light microscope (×1,000; Type—020-519.503 LB30T; Leica Microsystems, Wetzlar, Germany).

The diatoms were identified to species level based on studies by Metzeltin et al. (2005), Bicudo & Menezes (2006) and Metzeltin & Lange-Bertalot (1998, 2007).

Indices

The diatom species counts were entered into the diatom database and index calculation tool OMNIDIA version 5.3 (Lecoite et al., 1993) and the following indices were calculated and tested: the Artois-Picardie Diatom Index (APDI; Prygiel et al., 1996); the Eutrophication/Pollution Index (EPI; Dell'Uomo, 1996); the Biological Diatom Index (BDI; Lenoir & Coste, 1996); Schiefele and Schreiner's index (SHE; Schiefele & Schreiner, 1991); the Saprobic Index (SI; Rott et al., 1997); the Trophic Index (TI; Rott et al., 1999); the Watanabe index (WAT; Watanabe et al., 1986); the Specific Pollution Sensitivity Index (SPI; Coste in Cemagref, 1982); the Sládeček's Index (SLA; Sládeček, 1986); Descy's Pollution Index (DES; Descy, 1979); Leclerq (IDSE; Leclerq & Maquet, 1987); the Generic Diatom Index (GDI; Coste & Ayplassorho, 1991); the Commission of Economical Community Index (CEC; Descy & Coste, 1991); the Trophic Diatom Index (TDI; Kelly & Whitton, 1995); the Pampean Diatom Index (PDI; Gómez & Licursi, 2001) and the Biological Index of Water Quality trophic index (BIWQ; Lobo et al., 2004). All these indices, except CEC, SHE, TDI and WAT, are based on the formula of Zelinka & Marvan (1961). The last two indices (BIWQ and PDI) were developed in South America and are here classified as regional indices while all the other indices were developed outside South America and are here classified as foreign indices. The percentage of species used in calculation of the indices, as indicated by the OMNIDIA, was also recorded.

Statistical analysis

The available environmental data consisted of 27 environmental variables (Table 1). Environmental variables that were not normally distributed (Shapiro-Wilk, $P \leq 0.05$) or had no equal variance (Levene's test, $P \leq 0.05$) were transformed as described in Bere & Tundisi (2010). Two-way analysis of variance (ANOVA) was used to compare means of transformed environmental variables among the three sites categories and among the four sampling periods (see "Study

area”). Distribution of dominant taxa among the three site categories (see “Study area”) is given here briefly. Full discussion on diatom assemblages and their relationships with measured environmental variables and the multivariate techniques used can be found in Bere & Tundisi (2010, 2011a, b).

Pearson correlation was used to determine the relationship between the calculated index scores and measured physical and chemical water quality data. One-way ANOVA with Tukey’s pairwise comparisons was used to compare values of correlations between the calculated index scores and the measured water quality variables among different indices. The same method was used to compare percentages of species used in calculation of different indices. Forward stepwise multiple regression analysis was

performed on the data to determine the indices that gave the best reflection of general water quality (Lenoir & Coste, 1996). This method can give important additional information about the factors that influence the various index scores over and above pure correlations (Lenoir & Coste, 1996). Adjusted R^2 values were used as indicators of the level of success with which the variables were able to explain the variation in the index values; the higher the value, the more accurate are the indices as indicators of the measured water quality variables. Pearson correlation, ANOVA, Levene’s test and Shapiro–Wilk were performed using Palaeontological Statistics (PAST) software v.1.95 (Hammer et al., 2009). Forward stepwise multiple regression was performed with STATISTICA Version 7.

Table 1 The mean ($n = 4$) values of physical and chemical variables measured at 10 sites during 4 sampling periods

Site	1	2	3	4	5	6	7	8	9	10
Temperature (°C)	18.3	20.9	20.6	21.2	21.2	20.39	24	24.8	23	21.3
Altitude (m)	761	837	831	794	745	761	774	724	630	627
Canopy cover (%)	80	95	60	50	4	45	20	20	50	5
BOD ₅ (mg l ⁻¹)	0.9	1	2.6	6.9	1.2	7.2	1.6	19.5	24.5	26.2
DO (mg l ⁻¹)	7.3	8.2	7.6	6.9	7.6	7.2	6.8	1.9	2.1	0.4
Conductivity (µScm ⁻¹)	45	20	53	89	103	30	28	715	322	283
pH	6.6	6.4	6.3	6.8	7.2	6.8	6.7	7.2	7.2	7.1
Turbidity (NTU)	5.1	4.2	4.7	19.5	11.1	13.2	7.3	45.3	53.2	60.4
TN (mg l ⁻¹)	0.65	0.18	0.24	1.29	1.41	0.93	1.72	38.32	14.87	10.17
TP (mg l ⁻¹)	0.007	0.008	0.012	0.16	0.062	0.017	0.034	2.965	1.106	0.746
Nitrate (µg l ⁻¹)	27.46	175.05	524.86	964.3	1,473.58	242.49	96.69	2,140.22	316.26	714.4
Phosphate (µg l ⁻¹)	<2	<2	4.07	83.81	12.77	<2	<2	19.88	142.53	248.66
Ammonium (µg l ⁻¹)	7.84	91.62	12.05	757.66	629.27	28.63	11.75	609.38	5,492.55	2,547.22
Sulphate (mg l ⁻¹)	0.07	1.99	0.69	6.32	6.18	1.81	0.27	2.69	14.43	5.48
Sodium (mg l ⁻¹)	1.12	2.2	1.22	6.64	4.99	1.64	1.49	6.02	12.81	15.08
Potassium (mg l ⁻¹)	0.89	0.63	0.98	2.17	1.67	0.57	0.59	1.43	4.26	2.88
Magnesium (mg l ⁻¹)	0.56	0.49	0.66	1.06	1.4	0.59	0.58	1.36	2.42	1.31
Calcium (mg l ⁻¹)	1.03	1.55	1.07	4.33	7.07	2.41	1.87	5.92	10.99	6.70
Fluoride (µg l ⁻¹)	38.84	46.58	28.93	57.79	125.21	35.67	24.29	175.37	212.61	131.56
Chloride (mg l ⁻¹)	1.51	4.32	2.34	16.54	9.34	2.39	2.12	11.29	18.3	22.21
Depth (m)	0.2	0.3	0.4	0.4	0.3	0.4	0.2	0.5	0.3	0.3
Velocity (m s ⁻¹)	2.5	2.8	2.6	2.7	1.4	2.9	2.23	3.5	2.4	2.34
Cr (mg l ⁻¹)	0.01	0.01	0.01	0.01	<0.007	0.01	0.02	0.03	0.03	0.03
Cu (mg l ⁻¹)	0.002	0.003	0.004	0.006	0.003	0.002	0.005	0.029	0.008	0.012
Fe (mg l ⁻¹)	3.04	0.35	0.5	0.26	0.48	0.29	0.43	0.45	1.02	0.79
Cd (mg l ⁻¹)	0.001	0.001	0.002	0.002	<0.001	<0.001	0.002	<0.001	0.005	0.004
Pb (mg l ⁻¹)	<0.01	<0.01	0.02	0.01	<0.01	<0.01	<0.01	0.02	0.01	0.02

Results

Physico-chemical variables

The values of physical and chemical variables measured in the study area during the study period are shown in Table 1. The pH increased slightly down the agricultural to urban gradient being slightly acidic at upstream sites and slightly alkaline/neutral at downstream sites. However, the difference in pH among the three site categories (see “Study area”) was not statistically significant (ANOVA, $P > 0.05$). Temperature increased downstream, but as in the case of pH, the increase was not significant (ANOVA, $P > 0.05$). On the other hand, conductivity, BOD₅, turbidity, TN, TP and most of metals increased significantly downstream (ANOVA, $P < 0.05$) while DO and percentage riparian vegetation cover decreased significantly downstream (ANOVA, $P < 0.05$). The concentrations of all the ions in water increased significantly downstream (ANOVA, $P < 0.05$) (Table 1). No significant differences were observed in the means of environmental variables among the four sampling periods (ANOVA, $P > 0.05$). This is expected since all sampling was confined to stable base flow periods when variations in water chemistry are low compared to the rainy season.

Community composition

All the sites were subject to some form of pollution (agricultural or urban); hence, species distribution was strongly biased towards those that are cosmopolitan and tolerant of elevated or slightly elevated levels of pollution. A total of 208 diatom species belonging to 63 genera were recorded. Species richness, diversity and evenness differed significantly ($P < 0.05$) among sampling sites, tending to be higher in upstream, relatively unpolluted mainly agricultural and forest area compared to downstream polluted mainly urban sites (Bere & Tundisi, 2010, 2011a, b).

Forested and agricultural sites (1, 2, 3 and 7), with mean good water quality (Bere & Tundisi, 2011b), were associated with such species as *Aulacoseira alpigena* (Grunow) Krammer, *A. ambigua* (Grunow) Simonsen, *Brachysira vitrea* (Grunow) Ross, *Cymbopleura naviculiformis* (Auerswald) Krammer,

Diatoma spp., *Diadlesmis contenta* (Grunow) Mann, *Eunotia bilunaris* (Ehrenberg) Mills, *E. intermedia* (Hustedt) Nörpel and Lange-Bertalot, *E. papillo* (Ehrenberg) Hustedt, *E. sudetica* Müller, *Meridion anceps* (Ehrenberg) Williams, *Melosira varians* Agardh, *Orthoseira dentroteres* (Ehrenberg) Crawford, *Stauroneis phoenicenteron* (Nitzsch) Ehrenberg and *Thalassiosira weissflogii* (Grunow) Fryxell and Hasle.

Urban sites (4, 5 and 6), with bad to medium water quality, were associated with such species as *Achnanthis minutissimum* (Kützing) Czarnecki, *Aulacoseira granulata* (Ehrenberg) Simonsen, *Cyclotella pseudostelligera* Hustedt, *Diadlesmis dissimilis* Moser, Lange-Bertalot & Metzeltin, *Encyonema neomesianum* Krammer, *E. silesiacum* (Bleisch) Mann, *Frustulia rhomboides* (Rabenhorst) De Toni, *Navicula cryptocephala* (Grunow) Cleve, *Neidium ampliatum* (Ehrenberg) Krammer, *N. affine* (Ehrenberg) Pfitzer, *Nitzschia linearis* (Agardh) Smith, *Pinnularia gibba* Ehrenberg, *P. divergens* Krammer, *Surirella linearis* Smith and *S. robusta* Enrenberg.

Downstream sites (8, 9 and 10), with very bad water quality, were associated with such species as *Fallacia monoculata* (Hust) Mann, *Gomphonema accuminatum* Ehrenberg, *G. parvulum* (Kützing) Kützing, *Nitzschia palea* (Kützing) Smith, *Nupela praecipua* (Reichardt) Reichardt, *Planothidium lanceolatum* (Brébisson) Grunow and *Sellaphora pupula* (Kützing) Mereschkowsky. These species have been reported to be pollution tolerant (Biggs & Kilroy, 2000; Potapova & Charles, 2003; Duong et al., 2006).

Indices

All the diatom index scores show significant correlations ($P < 0.05$) to water quality variables (Table 2). With the exceptions of metals, correlations between water quality variables and all index scores ranged from $R^2 \pm 0.62$ to $R^2 \pm 0.99$ indicating strong correlations. Significantly low correlation ($P < 0.05$) was observed between metal levels, except Cr, and all index scores compared to that between other variables (especially those related to eutrophication and organic pollution) and all index scores. Fe levels did not correlate significantly with any of the indices. Correlation coefficients between the calculated index scores and the measured water quality variables differed

significantly ($P < 0.05$) among different indices. These values did not differ significantly ($P > 0.05$) between the two ‘regional indices’ (BIWQ and PDI) whilst they tended to differ between regional and some foreign indices like DES, IDSE, SHE, WAT, TDI, GDI, CEE, SPI and BDI. Correlation between the BIWQ scores and water quality variables was generally significantly low ($P < 0.05$) compared to that between some foreign index scores like DES, IDSE, SHE, WAT, GDI, SPI and BDI scores and water quality variables. Foreign index scores like IDSE, GDI, BDI, EPI, CEE, TI and SPI were calculated based on significantly more species ($P < 0.05$) compared to regional indices (Fig. 2).

Based on forward stepwise multiple regression analysis performed to determine the indices that gave best reflection of water quality, measured water quality variables significantly ($P < 0.05$) account for the variation in all index scores. Adjusted R^2 values (which reflect the amount of variation in index scores explained by the measured water quality variables, i.e. the degree of applicability of index scores to assess water quality) ranged from 50% (CEE) to 85% (DES) (Fig. 3). The higher the adjusted R^2 value, the more accurate are the indices as indicators of the measured water quality variables. Adjusted R^2 value for measured water quality variables and BIWQ scores (78%) was slightly low compared to those based on water quality variables and some index scores from some indices developed in Europe, USA, Argentina and other countries such as DES (85%), PDI (80%), IDSE (79%), GDI (79%) and SPI (79%). The rest of the indices had lower adjusted R^2 values compared to the BIWQ (Fig. 3).

Discussion

Discussion on diatom assemblages and their relationships with measured environmental variables is found in Bere & Tundisi (2010, 2011a, b). Significant correlations between diatom-based index scores and physical and chemical characteristics of streams recorded in this study indicate the success with which diatom indices may be used to reflect general changes in water quality variables in lotic systems. Results of Pearson correlation coefficients between water quality variables and diatom indices obtained

in the present study compared favourably with results between indices and water quality variables from some European and South African authors (Prygiel & Coste, 1993; Kwadrans et al., 1998; Taylor et al., 2007a, b) and in some cases better than the correlations demonstrated in Europe.

Although there may be concerns as to the feasibility of transferring data concerning the ecological tolerance limits of diatoms between the northern and southern hemispheres (Round, 1991), most of the dominant diatom species encountered in this study (detailed in Bere & Tundisi 2010, 2011a, b) are cosmopolitan species well-documented in international literature, especially from the northern hemisphere (e.g. Krammer & Lange-Bertalot, 1986–1991). This is supported by the observation that foreign index scores like IDSE, GDI, BDI, EPI, CEE, TI and SPI were calculated based on significantly more species compared to regional indices (Fig. 2). These results are in agreement with Bate et al. (2004) who found that most dominant diatom species found in South African rivers were already recorded in the international literature. For that reason, most European diatom indices may be used in the study area as they are based on the ecology of widely distributed or cosmopolitan taxa.

From the forward stepwise multiple regression analysis results, measured water quality variables significantly accounted for the variation in all index scores (Fig. 3) indicating the success with which these indices can be used in the study region. The amount of variation in index scores accounted for by measured water quality variables compared favourably with results from some European and South African authors (Prygiel & Coste, 1993; Kwadrans et al., 1998; Taylor et al., 2007a, b) and was, in some cases, better than the amount of variation observed in Europe. For example, Lenoir & Coste (1996) showed that the overall water quality analysis explained 72% of the variation in the BDI, whilst in this study measured water quality variables accounted for 75% of the variation in BDI (Fig. 3). Thus, it can be concluded that this index is both applicable and useful for monitoring water quality in the study region. The fact that most of the variation in other index scores (in most cases above 60%; Fig. 3) was significantly accounted for by the physical and chemical variables in the study region also indicates the applicability and usefulness of these indices.

Table 2 Pearson correlation coefficients between measured environmental variables and diatom index scores generated for sites in the Monjolinho River and its tributaries

	SLA	DES	IDSE	SHE	WAT	TDI	GDI	CEE	SPI	BDI	APDI	EPI	DI-CH	PDI	BIWQ	SI	TI
BOD ₅	0.95	-0.94	-0.96	-0.97	-0.92	0.96	-0.96	-0.81	-0.97	-0.94	-0.88	0.94	0.90	0.94	0.89	0.97	0.84
DO	-0.92	0.92	0.92	0.94	0.85	-0.95	0.93	0.71	0.94	0.91	0.81	-0.91	-0.85	-0.89	-0.86	-0.94	-0.83
Conductivity	0.78	-0.76	-0.78	-0.77	-0.66	0.81	-0.77	-0.62	-0.79	-0.77	-0.67	0.75	0.69	0.71	0.73	0.79	0.73
pH	0.76	-0.86	-0.82	-0.78	-0.72	0.82	-0.78	-0.74	-0.80	-0.83	-0.73	0.78	0.86	0.82	0.85	0.82	0.82
Turbidity	0.95	-0.96	-0.97	-0.98	-0.92	0.97	-0.97	-0.82	-0.98	-0.96	-0.90	0.95	0.92	0.95	0.91	0.98	0.87
TN	0.74	-0.70	-0.72	-0.70	-0.59	0.77	-0.72	-0.56	-0.73	-0.71	-0.58	0.69	0.62	0.64	0.67	0.72	0.65
TP	0.73	-0.70	-0.72	-0.70	-0.59	0.76	-0.71	-0.57	-0.73	-0.71	-0.59	0.69	0.62	0.64	0.67	0.72	0.65
PO ₄ ³⁻	0.68	-0.75	-0.74	-0.79	-0.75	0.72	-0.74	-0.66	-0.76	-0.75	-0.79	0.76	0.75	0.78	0.69	0.76	0.70
NH ₄ ⁺	0.84	-0.80	-0.80	-0.79	-0.93	0.78	-0.84	-0.63	-0.81	-0.81	-0.82	0.86	0.77	0.83	0.77	0.80	0.69
SO ₄ ²⁻	0.73	-0.75	-0.74	-0.70	-0.87	0.68	-0.76	-0.70	-0.73	-0.77	-0.84	0.79	0.76	0.79	0.75	0.72	0.70
Na ⁺	0.84	-0.89	-0.88	-0.90	-0.89	0.85	-0.89	-0.79	-0.90	-0.91	-0.92	0.90	0.88	0.90	0.85	0.89	0.84
K ⁺	0.80	-0.84	-0.82	-0.83	-0.93	0.78	-0.85	-0.70	-0.83	-0.85	-0.91	0.88	0.84	0.88	0.81	0.83	0.79
Mg ²⁺	0.85	-0.87	-0.86	-0.82	-0.92	0.83	-0.87	-0.70	-0.85	-0.88	-0.88	0.90	0.85	0.88	0.86	0.86	0.83
Ca ²⁺	0.85	-0.90	-0.88	-0.83	-0.91	0.85	-0.88	-0.77	-0.87	-0.90	-0.89	0.90	0.88	0.89	0.89	0.87	0.85
F ⁻	0.90	-0.90	-0.90	-0.86	-0.91	0.89	-0.90	-0.73	-0.89	-0.92	-0.86	0.91	0.86	0.87	0.86	0.90	0.82
Cl ⁻	0.76	-0.86	-0.85	-0.87	-0.84	0.80	-0.84	-0.84	-0.85	-0.88	-0.94	0.86	0.87	0.88	0.83	0.85	0.86
Cr	0.85	-0.77	-0.78	-0.79	-0.73	0.85	-0.82	-0.53	-0.82	-0.77	-0.63	0.80	0.64	0.74	0.74	0.80	0.68
Cu	0.65	-0.64	-0.66	-0.65	-	0.70	-0.64	-0.53	-0.67	-0.65	-0.53	0.62	0.55	0.58	0.63	0.66	0.65
Fe	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
Cd	0.56	-0.53	-0.52	-0.55	-0.66	0.52	-0.59	-	-0.56	-0.53	-0.58	0.63	-	0.59	0.52	0.54	0.50
Pb	0.51	-0.54	-0.58	-0.61	-	0.58	-0.57	-	-0.59	-0.55	-0.58	0.60	-	0.57	0.53	0.61	0.63

Numerical values indicate significant correlations at $P \leq 0.05$

APDI Artois-Picardie Diatom Index, EPI Eutrophication/Pollution Index, BDI Biological Diatom Index, SHE Schiefele and Schreiner's Index, SI Saprobic Index, TI Trophic Index, WAT Watanabe Index, SPI Specific Pollution Sensitivity Index, SLA Sládeček's Index, DES Desey's Pollution Index, IDSE Leclercq, GDI Generic Diatom Index, CEC Commission of Economical Community Index, TDI Trophic Diatom Index, PDI Pampean Diatom Index, BIWQ Biological Index of Water Quality Trophic Index

Fig. 2 The means ($n = 4$) and standard deviations of the percentage of species used in calculation of different indices score in this study

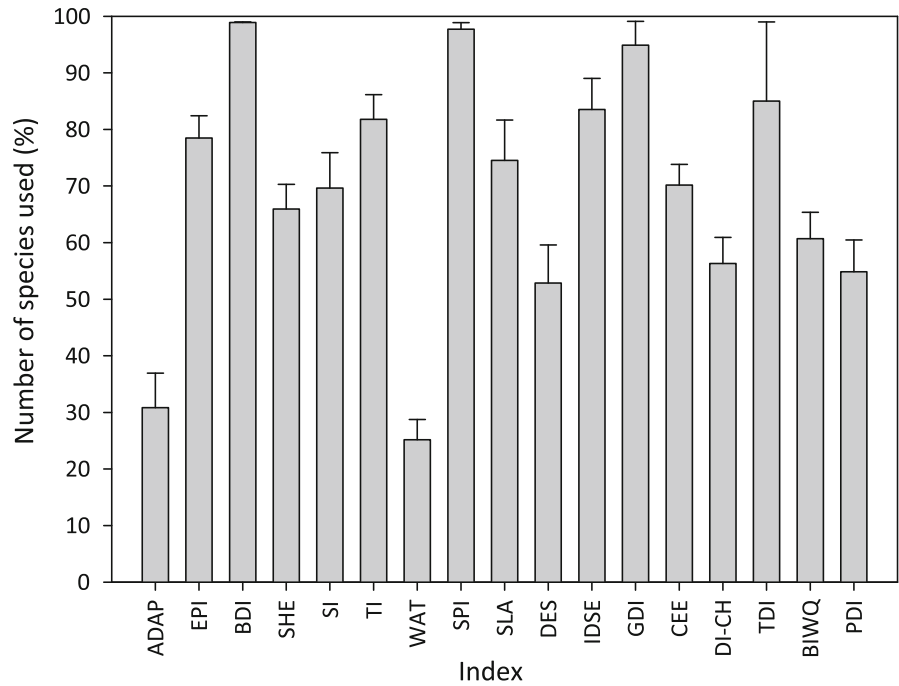
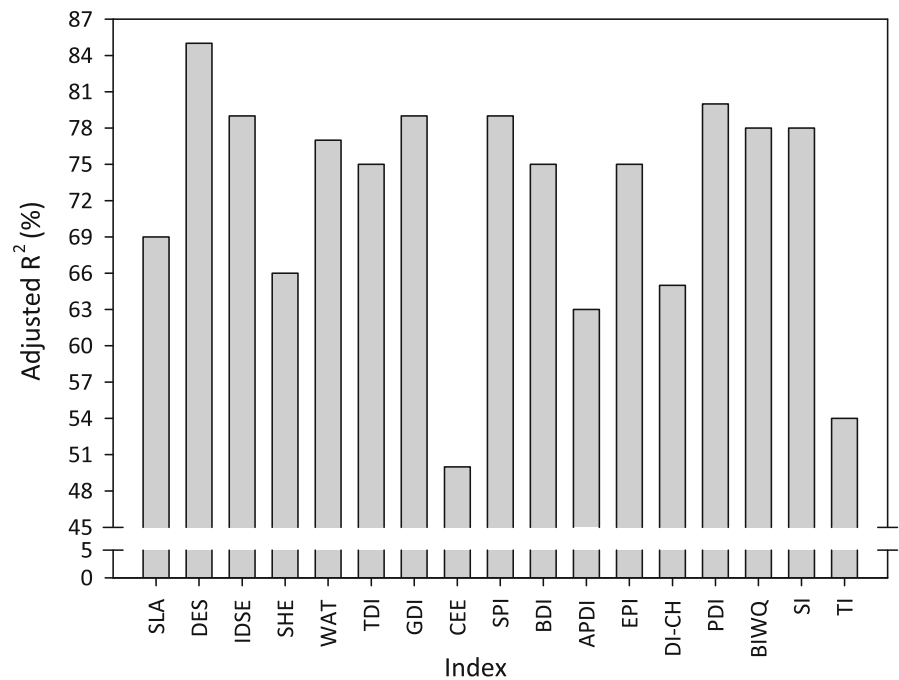


Fig. 3 Adjusted R^2 values obtained from forward stepwise multiple regression analysis performed on index scores and measured water quality variables. Index abbreviations as in Table 2



Generally, significantly low correlation was observed between metal levels, and all index scores compared to that between other variables (especially those related to eutrophication and organic pollution) and all index scores. This is expected since most of

these indices are designed to monitor organic pollution and eutrophication with no provision for assessment of metal contamination, underlining a clear vision to develop diatom-based water quality monitoring protocols that incorporate the effects of metals.

It could also be due to the overriding effects of organic pollution and eutrophication on metal pollution.

Weaker correlation of the BIWQ scores and water quality variables compared to that between water quality variables and some foreign index scores like DES, IDSE, SHE, WAT, GDI, SPI and BDI scores was observed. Additionally, adjusted R^2 value for measured water quality variables and BIWQ scores was slightly low compared to those based on water quality variables and some index scores from indices developed in Europe, USA, Argentina and other countries like DES, PDI, IDSE, GDI and SPI. This indicates better applicability of these indices to the study region compared to the BIWQ. Potapova & Charles (2005) showed that diatom metrics created specifically for U.S. rivers were better suited for water-quality assessment of those rivers than metrics developed for other geographic areas. Logically, therefore, the BIWQ, developed specifically for Brazilian waters, should have provided a better perspective of the water quality in the study region compared to indices developed in Europe, USA, Argentina and other countries, but this was not the case in this study.

These discrepancies can be attributed to differences in ecological characterisation of many, even common, diatom species in the BIWQ classification. One striking difference is the classification of *Nitzschia palea* and *Gomphonema parvulum*. Although these diatoms are reported from almost every survey of freshwater algae, the information about their distribution in relation to eutrophication and organic pollution or about their responses to eutrophication and organic pollution is often inconsistent. In their calculation of the BIWQ scores, Lobo et al. (2004) described *N. palea* as a medium pollution-tolerant species. In this study, however, this species had high relative abundance (around 90%) at hypereutrophic sites 8, 9 and 10 with very bad water quality (Bere & Tundisi, 2011b), an indication that it is tolerant of high pollution. This is supported by several studies (e.g. Lange-Bertalot, 1979; Kilham et al., 1986; Kobayasi & Mayama, 1989; Van Dam et al., 1994; Kelly & Whitton, 1995). However, recent evidence from the study by Trobajo et al. (2009) suggests that *N. palea* is taxonomically problematic. Their work illustrated that *N. palea* is almost certainly a complex of several or many separate species, which may differ

subtly in morphology and may not all share the same ecology. Thus, differences in pollution tolerance designated to *N. palea* by Lobo et al. (2004) and what we observed in our study can possibly be attributed to different species in the two regions.

The work by Lobo et al. (2002) upon which the BIWQ is based considered *G. parvulum*, a relatively abundant species at downstream eutrophic sites with high organic pollution as α -mesosaprobic. There is a lot of controversy in the literature regarding the classification of this species in relation to its pollution tolerance. In streams located in the Municipal District of Mato Leitão (Brazil), Lobo et al. (1999) classified this species as belonging to both α -mesosaprobic and polysaprobic environments. In the same streams, Rodrigues & Lobo (2000) registered the occurrence of this species in moderately polluted, β -mesosaprobic waters. In a study carried out in the same study area as the current study, Souza (2002) encountered this species in oligosaprobic environments. However, in studies carried out in rivers of Japan, Kobayasi & Mayama (1989) and Lobo et al. (1996) classified *G. parvulum* as highly tolerant to organic pollution in agreement with the results of the current study. Similarly, Kelly & Whitton (1995), working in rivers of UK described this species as highly tolerant to eutrophication (indicative value = 3 and sensitivity value = 5) in their calculation of the Trophic Diatom index (TDI). Diverse morphotypes of *G. parvulum*, however, exist, probably corresponding to different varieties (Moreles & Jasinski, 2002), which would explain the variety of responses attributed to this species. Diatom morphology has also been shown to vary because of both genetic variability and ecological variation, which may result in the formation of ecotypes (Salomoni et al., 2006). Detailed study on the autecology of *N. palea* and *G. parvulum* are called for in order to clarify their taxonomy and ecological requirements. Round (2004) discovered that lumping of several similar looking taxa into one “morphospecies” diminishes discriminative ability of diatom metrics, while detailed taxonomic and ecological studies allow recognition of taxa with good indicator properties. With diatom taxonomy undergoing rapid changes, especially at the genus level, diatom identification guides and methods to be used in South America must be consistent and updated.

Weak correlation of the BIWQ scores and the water quality and discrepancies in classification of

sites based on BIWQ compared to other indices may also be attributed to the fact that many species that were abundant in the present study area, such as *Brachysira vitrea*, *Eunotia bilunaris*, *Frustulia saxonica*, *Lemnicola hungarica*, *Navicula radiosa* Kützing, *Nitzschia clausii* (Hantzsch) Grunow, *Pinnularia braunii* and *P. microstauron* (Ehrenberg) Cleve, were classified as indicators of oligotrophic and oligosaprobic conditions. However, most of these species are known to be tolerant of eutrophication and organic pollution (Van Dam et al., 1994). The ongoing discussion demonstrates the inconsistency and lack of appropriate information on ecological requirements of diatom species from South America hampering formulation of sound diatom-based water quality management protocols. In addition, the ecological characteristics of indicator species and the uniqueness of each stream are not well documented. Thus, more work is needed to elucidate the ecological requirements of diatoms in Brazil, something that is far from being understood.

Besides ecological characterisation, another source of criticism of the BIWQ is the database from which it was derived, that is 12 sampling sites in a small geographical area. This is barely enough to characterise the distribution of the most common taxa, and clearly not sufficient to determine ecological properties of relatively rare diatoms across diverse and extensive geographical regions typical of Brazil. Indices from other countries have proven to be more applicable to the study area than our own Brazilian index because these indices are based on a large number of sampling sites encompassing wide and varied geographical regions with a variety of environmental conditions. Thus, the species base of the BIWQ is too narrow for widespread use in Brazil and needs to be broadened. This underlines a clear vision for developing a diatom-based water quality monitoring protocol that encompasses all the regions of Brazil. This sounds too cumbersome a task, but Potapova & Charles (2005) have demonstrated that it is possible by developing diatom matrices to monitor eutrophication based on 1,240 sampling sites covering the entire United States of America. In addition, data from 1,332 sites were used to develop diatom metrics and indices in France (Prygiel & Coste, 1993), 671 sites in Austria (Rott et al., 2003), and 553 sites in Japan (Watanabe et al., 1988). Van Dam et al. (1994) combined data from hundreds of literature sources and their own

numerous observations made in The Netherlands, to assign diatom taxa to ecological categories. Better coverage (more sites per area) would further improve the autecological information badly needed in Brazil for production of a sound diatom-based water quality assessment protocol as the autecology of more diatom species could be quantified.

Meanwhile, as recommended by Taylor et al. (2007b), BIWQ, as well as other foreign indices can be used to (1) gain support and recognition for diatom-based approaches to water quality monitoring in other regions, (2) provide an indication of water quality and allowing for the dissemination of simplified useful information to resource managers, conservationists and the general public, and (3) allow for sample and data collection which can then be used later in the formulation of a Brazilian diatom index tailored for different regions. Combining several datasets produced by different institutions or researches might provide a good way to increase the number of samples available for particular geographical regions and types of rivers. Such work, however, would be complicated by the taxonomic inconsistencies that exist between different datasets.

Conclusions

Assessment of water quality based on diatom-based indices is deemed useful in Brazil to provide information on water quality impacts on rivers and streams for management purposes. Although all borrowed indices were applicable to the study area because many widely distributed diatom species have similar environmental tolerances to those recorded for these species elsewhere, ecological requirements of some diatom species from Brazil need to be clarified and incorporated in a diatom-based water quality assessment protocol unique to the region. These species include *Brachysira vitrea*, *Eunotia bilunaris*, *Frustulia saxonica*, *Gomphonema parvulum*, *L. hungarica*, *Navicula radiosa*, *Nitzschia clausii*, *N. palea*, *Pinnularia braunii* and *P. microstauron*. To improve diatom-based water quality assessments in Brazil, autecology-based diatom metrics should be developed based on datasets representative of the areas or river types where the metrics will be applied and by assuring high-quality taxonomic identifications.

References

- APHA, 1988. Standard Methods for the Examination of Water and Waste Water, 20th ed. American Public Health association, Washington, DC.
- Azim, M. E., M. C. J. Verdegem, A. A. Van Dam & M. C. M. Bederige (eds), 2005. Periphyton Ecology, Exploitation and Management. CABI Publishing, Cambridge.
- Bate, G., P. Smailes & J. Adams, 2004. A water quality index for use with diatoms in the assessment. *Water SA* 30: 493–498.
- Bere, T. & J. G. Tundisi, 2010. The effects of substrate type on diatom-based multivariate water quality assessment in a tropical river (Monjolinho), São Carlos-SP, Brazil. *Water Air Soil Pollution* 216: 391–409.
- Bere, T. & J. G. Tundisi, 2011a. Influence of ionic strength and conductivity on benthic diatom communities in a tropical river (Monjolinho), São Carlos-SP, Brazil. *Hydrobiologia* 661: 261–276.
- Bere, T. & J. G. Tundisi, 2011b. Influence of land-use patterns on benthic diatom communities and water quality in the tropical Monjolinho hydrological basin, São Carlos-SP, Brazil. *Water SA* 37: 93–102.
- Bicudo, C. E. M. & M. Menezes, 2006. Gêneros de água de águas continentais do Brasil: chave para identificação e descrições. Rima Editora, São Carlos, SP, Brazil: 391–339.
- Biggs, B. J. F. & C. Kilroy, 2000. Stream Periphyton Monitoring Manual. NIWA, Christchurch, New Zealand.
- Cemagref, 1982. Etude des methods biologiques d'appréciation quantitative de la qualité des eaux. Rapport Q. E. Lyon, Agence de l'eau Rhône-Méditerranée-Corse-Cemagref, Lyon, France.
- Coste, M. & H. Ayphassorho, 1991. Étude de la qualité des eaux du Bassin Artois-Picardie à l'aide des communautés de diatomées benthiques (application des indices diatomiques). Rapport Cemagref. Bordeaux – Agence de l'Eau Artois-Picardie, Douai.
- Dell'Uomo, A., 1996. Assessment of water quality of an Apennine river as a pilot study. In Whitton, B. A. & E. Rott (eds), *Use of Algae for Monitoring Rivers II*. Institut für Botanik, Universität Innsbruck, Innsbruck: 65–73.
- Descy, J. P., 1979. A new approach to water quality estimation using diatoms. *Nova Hedwigia* 64: 305–323.
- Descy, J. P. & M. Coste, 1991. A test of methods for assessing water quality based on diatoms. *Verhandlungen der Internationalen Vereinigung für Theoretische und Angewandte Limnologie* 24: 2112–2116.
- Dionex Corporation, 2001. Dionex DX-80 Ion Analyzer Operator's Manual. Dionex Corporation, USA.
- Duong, T., Coste, M., Feurtet-mazel, A., Dang, D., Gold, C., Park, Y. et al, 2006. Impact of urban pollution from the Hanoi area on benthic diatom communities collected from the Red, Nhue and Tolich Rivers (Vietnam). *Hydrobiologia* 563:201–216.
- Gómez, N. & M. Licursi, 2001. The Pampean Diatom Index (IDP) for assessment of rivers and streams in Argentina. *Aquatic Ecology* 35: 173–181.
- Hammer, O., D. A. T. Harper & P. D. Ryan, 2009. PAST – Palaeontological Statistics, Version 1.90. <http://folk.uio.no/ohammer/past>.
- Harding, W. R., C. G. M. Archibald & J. C. Taylor, 2005. The relevance of diatoms for water quality assessment in South Africa: a position paper. *Water SA* 31: 41–46.
- Kelly, M. G. & B. A. Whitton, 1995. The trophic diatom index: a new index for monitoring eutrophication in rivers. *Journal of Applied Phycology* 7: 433–444.
- Kelly, M. G., C. J. Penny & B. A. Whitton, 1995. Comparative performance of benthic diatom indices used to assess river water quality. *Hydrobiologia* 302: 179–188.
- Kilham, P., S. S. Kilham & R. E. Hecky, 1986. Hypothesized resources relationships among African plankton diatoms. *Limnology and Oceanography* 31: 1169–1181.
- Kobayasi, H. & S. Mayama, 1989. Most pollution-tolerant diatoms of severely polluted rivers in the vicinity of Tokyo. *Japanese Journal of Phycology* 30: 188–196.
- Krammer, K. & H. Lange-Bertalot, 1986–1991. Süßwasserflora von Mitteleuropa. Band 2. Bacillariophyceae. Teil 1–4. Gustav Fischer, Stuttgart, Germany.
- Kwandrans, J., P. Eloranta, B. Kawecka & W. Kryzysztzof, 1998. Use of benthic diatom communities to evaluate water quality in rivers of southern Poland. *Journal of Applied Phycology* 10: 193–201.
- Lange-Bertalot, H., 1979. Pollution tolerance of diatoms as a criterion for water quality estimation. *Nova Hedwigia Beiheft* 64: 285–304.
- Leclerq, L. & B. Maquet, 1987. Deux nouveaux indices chimique et diatomique de qualité d'eau courante. Application au Samson et à ses affluents (bassin de la Meuse belge). Comparaison avec d'autres indices chimiques, biocéc notiques et diatomiques. Institut Royal des Sciences Naturelles de Belgique, document de travail 28.
- Lecointe, C., M. Coste & J. Prygiel, 1993. "Omnidia": software for taxonomy, calculation of diatom indices and inventories management. *Hydrobiology* 269(270): 509–513.
- Lenoir, A. & M. Coste, 1996. Development of a practical diatom index of overall water quality applicable to the French National Water Board network. In Whitton, B. A. & E. Rott (eds), *Use of Algae for Monitoring Rivers II*. Institut für Botanik, Universität Innsbruck, Innsbruck: 29–43.
- Lobo, E. A. & V. L. Callegaro, 2000. Avaliação da qualidade de águas doces continentais com base em algas diatomáceas epilíticas: Enfoque metodológico. In Tucci, C. E. M. & D. M. Marques (eds), *Avaliação e Controle da Drenagem Urbana*. Ed. Universidade/UFRGS, Porto Alegre: 277–300.
- Lobo, E. A. & L. C. Torgan, 1988. Análise da estrutura da comunidade de diatomáceas (Bacillariophyceae) em duas estações do sistema Guaíba, RS, Brasil. *Acta Botânica Brasílica* 1: 103–119.
- Lobo, E. A., M. A. Oliveira, M. T. Neves & S. Schuler, 1991. Caracterização de ambientes de terras úmidas, no Estado do Rio Grande do Sul, onde ocorrem espécies de anátídeos com valor cinético. *Acta Biológica Leopoldensia* 13: 19–60.
- Lobo, E. A., V. L. M. Callegaro, M. A. Oliveira, S. E. Salomoni, S. Schuler & K. Asai, 1996. Pollution tolerant diatoms from lotic systems in the Jacui Basin, Rio Grande do Sul, Brasil. *Iheringia Série Botânica* 47: 45–72.
- Lobo, E. A., D. A. Bem, A. Costa & A. Kirst, 1999. Avaliação da qualidade da água dos arroios Sampaio, Bonito e Grande, Município de Mato Leitão, RS, Brasil, segundo a

- resolução do CONAMA 20/86, Vol. 4. Revista Redes, Santa Cruz do Sul: 129–146.
- Lobo, E. A., V. L. Callegaro & P. Bender, 2002. Utilização de algas diatomáceas epilíticas como indicadoras da qualidade da água em rios e arroios da Região Hidrográfica do Guaíba, RS. EDUNISC, Santa Cruz do Sul, Brasil.
- Lobo, E. A., V. L. Callegaro, G. Hermany, D. Bes, C. E. Wetzel & M. A. Oliveira, 2004. Use of epilithic diatoms as bioindicator from lotic systems in southern Brazil, with special emphasis on eutrophication. *Acta Limnologica Brasiliensia* 16: 25–40.
- McCormick, P. V. & J. J. Cairns, 1994. Algae as indicators of environmental change. *Journal of Applied Phycology* 6: 509–526.
- Metzeltin, D. & H. Lange-Bertalot, 1998. Tropical diatoms of South America I. *Iconographia Diatomologica* 5: 1–695.
- Metzeltin, D. & H. Lange-Bertalot, 2007. Tropical diatoms of South America II. *Iconographia Diatomologica* 18: 1–877.
- Metzeltin, D., H. Lange-Bertalot & F. García-Rodríguez, 2005. Diatoms of Uruguay. *Iconographia Diatomologica* 15: 1–736.
- Moreles, E. A. & A. Jasinski, 2002. Morphological studies in *Gomphonema parvulum* complex: evidence of cryptic species? In 17th International Diatom Symposium, Ottawa, Canada, 25–31 August, 2002, Book of Abstracts: 90 pp.
- Pan, Y., R. J. Stevenson, B. H. Hill, A. T. Herlihy & G. B. Collins, 1996. Using diatoms as indicators of ecological conditions in lotic systems: a regional assessment. *Journal of North American Benthological Society* 15: 481–495.
- Pappas, J. L. & E. F. F. Stoermer, 1996. Quantitative method for determining a representative algal sample count. *Journal of Phycology* 32: 693–696.
- Pipp, E., 2002. A regional diatom-based trophic state indication system for running water sites in Upper Austria and its overregional applicability. *Verhandlungen der Internationalen Vereinigung für Theoretische und Angewandte Limnologie* 27: 3376–3380.
- Potapova, M. G. & D. F. Charles, 2002. Benthic diatoms in USA Rivers: distributions along speciation and environmental gradients. *Journal of Biogeography* 29: 167–187.
- Potapova, M. & D. F. Charles, 2003. Distribution of benthic diatoms in U.S. rivers in relation to conductivity and ionic composition. *Freshwater Biology* 48: 1311–1328.
- Potapova, M. & D. F. Charles, 2005. Choice of substrate in algae-based water quality assessment. *Journal of North American Benthological Society* 24: 415–427.
- Prygiel, J. & M. Coste, 1993. The assessment of water quality in the Artois-Picardie water basin (France) by the use of diatom indices. *Hydrobiologia* 269(279): 343–349.
- Prygiel, J., L. Lévêque & R. Iserentant, 1996. Un nouvel indice diatomique pratique pour l'évaluation de La qualité des eaux en réseau de surveillance. *Revue des Sciences de l'Eau* 1: 97–113.
- Prygiel, J., B. A. Whitton & J. Bukowska, 1999. Use of Algae for Monitoring Rivers III. *Agence de L'Eau Artois-Picardie, Douai*: 271 pp.
- Rimet, F., H. M. Cauchie, L. Hoffmann & L. Ector, 2005. Response of diatom indices to simulated water quality improvements in a river. *Journal of Applied Phycology* 17: 119–128.
- Rocha, A. A., 1992. Algae as indicators of water pollution. In Cordeiro-Marino, M., M. T. P. Azevedo, C. L. Sant'anna, N. Y. Tomita & E. M. Pastino (eds), *Algae and Environment: A General Approach*. Sociedade Brasileira de Ficologia, CETESB, São Paulo: 34–55.
- Rodrigues, L. M. & E. A. Lobo 2000. Análise da estrutura de comunidades de diatomáceas epilíticas no Arroio Sampaio, Município de Mato Leitão, RS, Brasil, Vol. 2. *Caderno de Pesquisa Série Botânica, Santa Cruz do Sul*: 5–27.
- Rosa, Z. M., L. C. Torgan & L. A. W. Herzog, 1988. Análise da estrutura de comunidades fitoplanctônicas e de alguns fatores abióticos em trecho do Rio Jacuí, Rio Grande do Sul, Brasil. *Acta Botanica Brasílica* 2: 31–46.
- Rott, E., G. Hofmann, K. Pall, P. Pfister & E. Pipp, 1997. Indikationslisten für Aufwuchsalgen. Teil 1. Saprobielle Indikation. Bundesministerium für Land- und Forstwirtschaft, Wien: 1–73.
- Rott, E., E. Pipp, P. Pfister, H. van Dam, K. Ortler, N. Binder & K. Pall, 1999. Indikationslisten für Aufwuchsalgen in österreichischen Fließgewässern. Teil 2: Trophieindikation (sowie geochemische Präferenzen; taxonomische und toxikologische Anmerkungen). *Wasserwirtschaftskataster herasgegeben vom Bundesministerium f. Land- u. Forstwirtschaft, Wien*. ISBN 3-85 174-25-4: 248 pp.
- Rott, E., E. Pipp & P. Pfister, 2003. Diatom methods developed for river quality assessment in Austria and a cross-check against numerical trophic indication methods used in Europe. *Algological Studies* 110: 91–115.
- Round, F. E., 1991. Diatoms in river water-monitoring studies. *Journal of Applied Phycology* 3: 129–145.
- Round, F. E., 2004. pH scaling and diatom distribution. *Diatom Research* 20: 9–12.
- Salomoni, S. E., O. Rocha, V. L. Callegaro & E. A. Lobo, 2006. Epilithic diatoms as indicators of water quality in the Gravataí river, Rio Grande do Sul, Brazil. *Hydrobiologia* 559: 233–246.
- Schiefele, S. & C. Schreiner, 1991. Use of diatoms for monitoring nutrient enrichment acidification and impact salts in Germany and Austria. In Whitton, B. A., E. Rott & G. Friedrich (eds), *Use of Algae for Monitoring Rivers*. Institut für Botanik, Universität Innsbruck, Innsbruck.
- Slàdeček, V., 1986. Diatoms as indicators of organic pollution. *Acta Hydrochimica et Hydrobiologica* 14: 555–566.
- Souza, M. G. M., 2002. Variação da comunidade de diatomáceas epilíticas ao longo de um rio impactado no município de São Carlos – SP e sua relação com variáveis físicas e químicas. Tese (Doutorado e Ciências Biológicas) Ecologia a Recursos Naturais, Universidade Federal de São Carlos, São Carlos, SP, Brasil.
- Stevenson, R. J., M. L. Bothwell & R. L. Lowe, 1996. *Algal Ecology—Freshwater Benthic Ecosystems*. Academic Press, San Diego: 750 pp.
- Taylor, J. C., M. C. Janse Van Vuuren & A. J. H. Pieterse, 2007a. The application and testing of diatom-based indices in the Vaal and Wilge Rivers, South Africa. *Water SA* 33: 51–59.
- Taylor, C. J., A. V. Prygiel, P. A. De La Rey & S. Van Rensburg, 2007b. Can diatom-based pollution indices be used for biological monitoring in SA? A case study of the Crocodile West and Marico water management area. *Hydrobiologia* 592: 455–464.

- Trobajo, R., E. Clavero, V. A. Chepurnov, K. Sabbe, D. G. Mann, S. Ishihar & E. J. Cox, 2009. Morphological, genetic and mating diversity within the widespread bio-indicator *Nitzschia palea* (Bacillariophyceae). *Phycologia* 48: 443–459.
- Van Dam, H., A. Mertens & J. Sinkeldam, 1994. A coded checklist and ecological indicator values of freshwater diatoms from the Netherlands. *Aquatic Ecology* 28: 117–133.
- Watanabe, T., K. Asai & A. Houki, 1986. Numerical estimation of organic pollution of flowing waters by using the epilithic diatom assemblage – Diatom Assemblage Index (DIApo). *Science of the Total Environment* 55: 209–218.
- Watanabe, T., K. Asai & A. Houki, 1988. Numerical water quality monitoring of organic pollution using diatom assemblages. In Round, F. E. (ed.), *Proceedings of the Ninth International Diatom Symposium*. Koeltz Scientific Books, Koenigstein, Germany: 123–141.
- Wehr, J. D. & R. G. Sheath, 2003. *Freshwater Algae of North America, Ecology and Classification*. Academic Press, San Diego, CA, USA: 918 pp.
- Zelinka, M. & P. Marvan, 1961. Zur praxisierung der biologischen klassifikation der reinheit fließender gewässer. *Archiv Hydrobiologia* 57: 389–407.