Epilithic diatom community in a high altitude stream impacted by fish farming in southern Brazil.

SCHNECK[1], F., TORGAN[2], L.C. & SCHWARZBOLD[1], A.

[1] Laboratório de Limnologia, Departamento de Ecologia da Universidade Federal do Rio Grande do Sul, Caixa Postal 15007, Cep 91501-970, Porto Alegre, RS, Brazil. fabischneck@yahoo.com.br; albano.schwarzbold@ufrgs.br

[2] Museu de Ciências Naturais, Fundação Zoobotânica do Rio Grande do Sul, Rua Dr. Salvador França 1427, Cep 90690-000, Porto Alegre, RS, Brazil. lezilda.torgan@fzb.rs.gov.br

ABSTRACT: Epilithic diatom community in a high altitude stream impacted by fish farming in southern Brazil. The aim of this study was to determine the community structure of epilithic diatom in a 3rd order stream in southern Brazil, at an altitude of approximately 1,000m, in a stretch impacted by fish farming effluents. From July 2005 to February 2006, samples of diatoms on stones were collected monthly, with the physical and chemical water variables, in four sampling stations, two were located upstream and two downstream of the fish farming. The epilithic diatom community was represented by 48 taxa and the results revealed the occurrence of an impact gradient associated to the fish farming effluents, with a significant increase in the levels of nutrients and total solids downstream. The diatom community responded to trophic enrichment with a clear substitution of species characteristic to oligotrophic environments by indicator species of eutrophic environments. Throughout the Indicator Species Analysis, it was possible to identify two groups of species. The first group, composed by Gomphoneis rhombica, Navicula angusta, and Planothidium frequentissimum indicated environments with little impact, low availability of nutrients, small quantity of total solids, and low turbidity. The second composed, by Luticola goeppertiana, Melosira varians, Nitzschia palea, Gomphonema parvulum, Navicula cryptotenella, and Psammothidium subatomoides indicated eutrophic waters. So, epilithic diatoms demonstrated be a useful tool in biomonitoring impacts caused by eutrophication.
Key-words: biomonitoring, eutrophication, indicator species, periphyton.

RESUMO: Comunidade de diatomáceas epilíticas em um riacho de altitude no sul do Brasil impactado por piscicultura. Objetivou-se determinar a estrutura da comunidade de diatomáceas epilíticas em um riacho de 3a. ordem no sul do Brasil, a uma altitude de aproximadamente 1,000 m, em trecho impactado por efluentes de piscicultura. De julho de 2005 a fevereiro de 2006 foram coletadas, mensalmente, amostras de diatomáceas sobre seixos e feitas medições de variáveis físicas e químicas da água, em quatro estações amostrais, sendo duas a montante e duas a jusante da piscicultura. A comunidade de diatomáceas epilíticas esteve representada por 48 táxons e os resultados demonstraram a ocorrência de um gradiente de impacto associado aos efluentes da piscicultura, com significativo incremento nos teores de nutrientes e sólidos totais a jusante. A comunidade de diatomáceas respondeu ao enriquecimento trófico através de uma nítida substituição de espécies características de ambientes oligotróficos por espécies indicadoras de ambientes eutróficos. A partir da Análise de Espécies Indicadoras foi possível identificar dois grupos de espécies. O primeiro grupo, composto por Gomphoneis rhombica, Navicula angusta e Planothidium frequentissimum indicou ambientes pouco impactados, com baixa disponibilidade de nutrientes, pouca quantidade de sólidos totais e baixa turbidez. O segundo, constituído por Luticola goeppertiana, Melosira varians, Nitzschia palea, Gomphonema parvulum, Navicula cryptotenella e Psammothidium subatomoides indicou águas eutróficas. Assim, as diatomáceas epilíticas demonstraram ser uma útil ferramenta no biomonitoramento de impactos causados por eutrofização.
Palavras-chave: biomonitoramento, espécies indicadoras, eutrofização, perifíton.

Introduction

Diatoms are widely known for their potential as biondicators of aquatic environments, since they respond rapidly to environmental alterations due to differences in species ecological tolerance and competition ability (Round, 1993).

Furthermore, they have been used with success as a tool in monitoring water quality and in the evaluation of the ecological integrity of lotic ecosystems (Whitton et al., 1991; Kelly & Whitton, 1995; Jüttner et al., 1996).

Significant alterations in species composition when faced to pollution, both trophic enrichment and organic pollution, have been reported by many authors (e.g. Pringle, 1990; Lobo et al., 1995; Salomoni et al., 2006). However, other factors also play a relevant function in community structure, like water velocity (Hermany et al., 2006), grazing (Rosemond et al., 1993), and light availability (Kawecka, 1986).

The information existing on periphytic diatoms in Brazilian streams, especially those located in high altitude regions, are very scarce. Mendes (2003), evaluated the effect of enrichment on the diatom community in a stream in the Cipó Range, and Canani (2005), studied the diatom flora in the Ibitipoca State Park, both carried out in Minas Gerais State.

Effluents from fish farming activities mainly contain metabolic and feed residues, rich in ammonia and phosphorus (Kendra, 1991). Thus, some studies have evaluated the impacts caused by fish farming effluents in lotic environment biota, like the study by Szluha (1974), which observed a seven-time increment in the production of periphyton downstream to a culture of fish in a North American river. Villanueva et al. (2000), in an Andean river, found a reduced algal biomass downstream the fish farm despite the phosphorus enrichment, and they attributed it to the high input of organic matter from the fish farm, decreasing light availability, and increasing the amount of heterotrophic organisms. The same authors observed alterations in the periphytic diatom community. Kendra (1991) evaluated the impact of the effluent using benthic invertebrates, while Oberdorff & Porcher (1994) analyzed the influence in the fish assembly. Both studies showed alterations in species composition after the effluent discharge.

This study aimed to determine the longitudinal variation of the epilithic diatom community structure under fish farming influence, in a stretch of the upper course of the Antas River, comparing abundance and species composition patterns with the physical and chemical water variables, as well as identifying indicator species.

Material and methods

Study area

The region of the Campos de Cima da Serra is the highest and coldest region in the state of Rio Grande do Sul, southern Brazil, reaching altitudes higher than 1,400 m and temperatures of - 8°C in the winter (Moreno, 1961).

The present study was carried out on a third order stretch (sensu Strahler; Allan, 2001) in the upper course of the Antas River (altitude of 1,030 to 1,005 m), characterized by a riparian vegetation forming mosaics of bush and field and by a basaltic rocky formation. In this stretch, rainbow trout (Oncorhynchus mykiss Walbaum) fish farming takes place in tanks and dams, discharging effluents directly into the river.

The samplings were carried out monthly, from July 2005 to February 2006, in four sampling stations, two located upstream (S1 and S2) and two downstream (S3 and S4) from the fish farming (Fig. 1).

Environmental variables

Oxygen saturation, pH, electrical conductivity, biochemical oxygen demand (BOD; azide modification method), turbidity (nephelometric method), nitrate (NO_3^-; ultraviolet spectrophotometric screening method), ammoniacal nitrogen (NH_4) (absorciometric method), total nitrogen (Kjeldahl method), total phosphorus (stannous chloride method), total solids, and silicon (SiO_2^{-2}; molybdosilicate method) were analysed according to standardized procedures (APHA, 1998). Absorbance at 320 nm, a dissolved organic carbon indicator, followed Edwards & Cresser (1987), and the periphyton chlorophyll a concentration was measured using the method described by Jespersen & Christoffersen (1987). Current velocity and depth of the stream were also evaluated. The sampling stations were, in addition, classified according the Trophic State Index (Carlson, 1977 modified by Toledo et al., 1983), using total phosphorus as the criterious for classification.

Epilithic diatom sampling

In each sampling station, an area of 25 cm² of material was scraped off the upper surface of five submerged stones using a toothbrush, totalizing a composed sample of 125 cm² preserved with Lugol (Round, 1993).

The quantification and preliminary

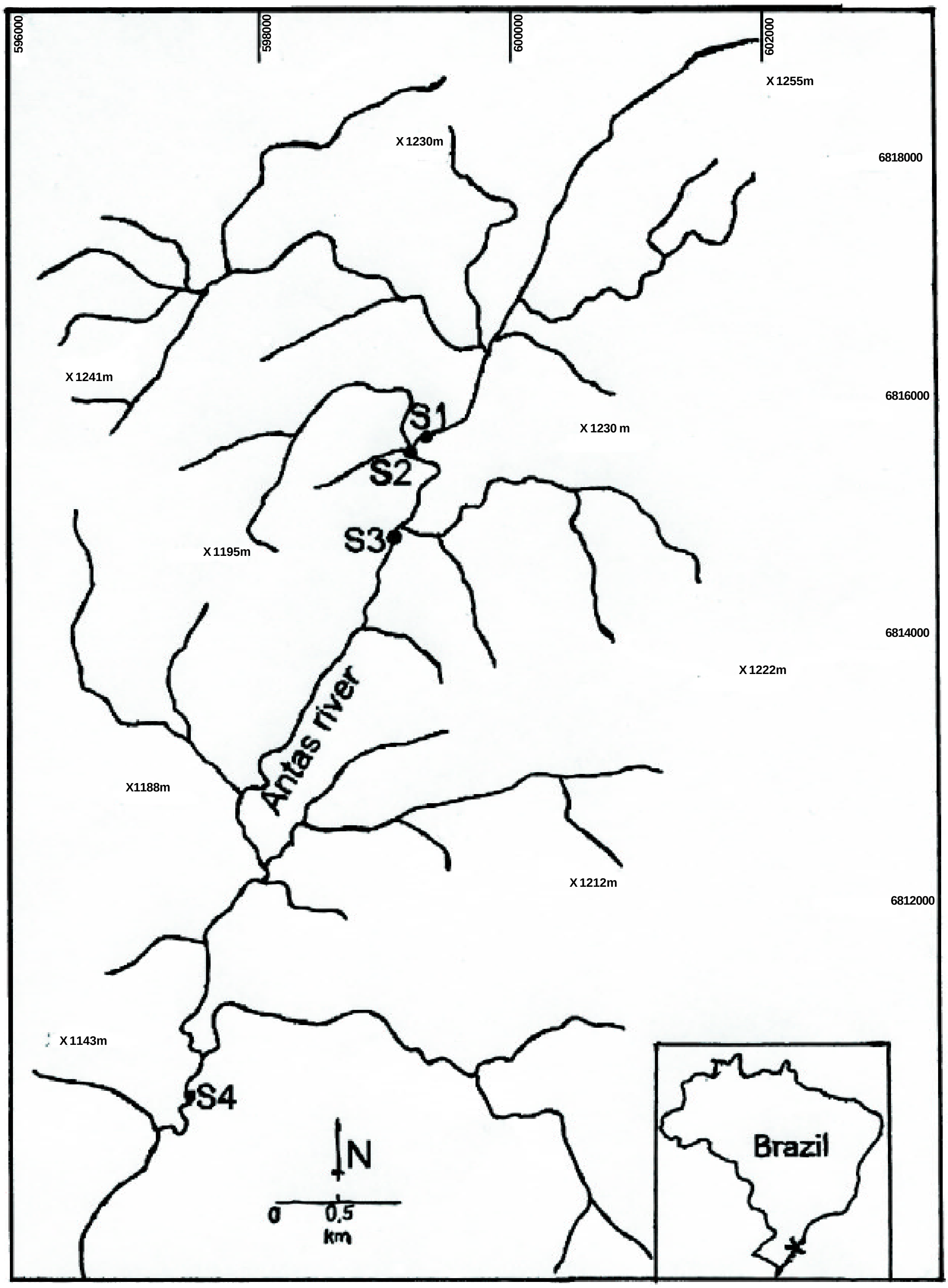

Figure 1: Location of the study area in the state of Rio Grande do Sul and of the sampling stations (S1, S2, S3, S4) in the upper course of the Antas River, São José dos Ausentes, RS, Brazil.

identification of the organisms followed Utermöhl method (1958), using an inverted microscope at 400x magnification. Only the individuals with chloroplasts were considered, where at least 300 viable cells were counted, until reaching a counting efficiency of 90%, according to Pappas & Stoermer (1996). The results were expressed in number of individuals by cm^2. Species' richness, and Shannon-Wiener Diversity Index (H') were calculated (Krebs, 1989). The determination of the dominant and abundant species followed the criteria set by Lobo & Leighton (1986).

A quantified aliquot was oxidized with hydrogen peroxide and potassium dichromate for confection of glass slides using Naphrax, and precise species identification was carried out using a light microscope with 1,000x magnification. Taxa identification was based on specialised bibliography (Patrick & Reimer, 1966; Krammer & Lange-Bertalot, 1986; 1988; 1991a; 1991b; Lange-Bertalot & Metzeltin, 1996; Lange-Bertalot, 1999; Metzeltin & Lange-Bertalot, 1998, 2002; Rumrich et al., 2000; Metzeltin et al., 2005) and journals. The classification system followed Round et al. (1990). Samples and slides are registered in the Prof. Dr. Alarich Schultz Herbarium (HAS) of the Fundação Zoobotânica do Rio Grande do Sul (HAS 107146 to 107177).

Statistical analyses

The sampling stations described by the physical and chemical data, richness, Shannon-Wiener diversity, and species density, were compared probabilistically, separately for each variable, through randomization tests (Pillar & Orlóci, 1996). The variables were log transformed (log (x+1)), except pH, and H'. Delineation in blocks was carried out in all analyses, with the sampling dates forming the blocks, thus isolating the temporal effect. The null hypothesis (H_0) tested was that there are no significant differences (a = 0.05) between comparisons in 1,000 iterations (Pillar, 2006), using as criterion the sum of the squares of Euclidean distances among sampling units. Randomization tests were carried out with the software MULTIV 2.3.20 (Pillar, 2006).

Indicator Species Analysis (Dufrêne & Legendre, 1997) was used to explore species distribution and abundance patterns, applied on the 48 species density matrix, and using sampling stations as categorical variable.

Subsequently, Canonical Correspondence Analysis (CCA) (Ter Braak, 1986) was applied to identify the environmental variables that better explain species distribution. The indicator species density data and the environmental variables were log transformed (log(x+1)), encompassing, respectively, the biotic and the abiotic data matrices. Monte Carlo Test was applied in order to verify the significance of the results (999 permutations; p £0.05). Indicator Species Analysis, and the CCA were performed using PCOrd version 4.10 for Windows (McCune & Mefford, 1999).

Results

Environmental Variables

Table I shows the results of the environmental variables averages in the four sampling stations. The results of the randomization tests are presented in Table II.

The smallest stream depth was observed in S1, in contrast with S4, which showed the highest depth. S2 figured as the environment with the greatest water velocity.

Biochemical oxygen demand only showed detectable values in S3, with an average of 2.0 mg L^{-1}. S3 was also the sampling station with the lowest percentage of O_2 saturation. The electrical conductivity was low in all sampling stations (means between 16.3 and 17.9 mS cm^{-1}) and the pH values were close to neutral. Higher average of total solids and turbidity in S3 (84 mg L^{-1} and 4.2 NTU, respectively), and lower in S2 (41 mg L^{-1} and 1.8 NTU, respectively) were detected.

Regarding nutrients, total phosphorus had the lowest average concentrations in S1 (25.36 mg L^{-1}) and in S2 (32.36 mg L^{-1}), and the highest in S3 (72.88 mg L^{-1}). The same was observed for nitrate and total nitrogen, with the highest concentrations in S3. Periphyton chlorophyll a followed the same nutrient trend, with the higher mean values in S3 (0.94 mg cm^{-2}), lower values in S1 and S2 (0.40 and 0.30 mg cm^{-2}, respectively) and intermediate values in S4 (0.59 mg cm^{-2}). The Trophic State Index (TSI) defined the sampling stations S1 and S2 as oligotrophic, S3 as eutrophic, and S4 as mesotrophic.

Composition and distribution of the epilithic diatom community

The epilithic diatom community was represented by 48 taxa, in a specific and infra-specific level, distributed in 26 genera (Tab. III). Sampling station 3 showed the highest richness, with an average of 22 taxa (Tab. I), differing from the other sampling stations (p£0.05; Tab. II). In this same sampling station, the highest algal density was also registered, which reached the peak of 26,237 ind cm^{-2} and average of 10,765 ind cm^{-2}, showing significant difference with S1 (p£0.05).

Figure 2 shows the variation in relative abundance of the eight species that better represent the gradient of environmental variations observed, evidencing the substitution of species along this gradient. Gomphoneis rhombica was abundant in S1 and S2, representing 33% of the individuals in each sampling station, however in S3 the species represented less than 1%, increasing again its contribution in S4 (6%). The same occurred with Cocconeis placentula var. acuta. Inverse situation was observed for Luticola goeppertiana, Navicula cryptotenella, Nitzschia palea, and Psammothidium subatomoides, which showed higher densities in S3 (5%, 6%, 3%, and 8%, respectively). Gomphonema parvulum morph. 1 and Gomphonema

Table I: Mean and standard deviation (±) of environmental variables and of biotic variables at the sampling stations in the upper course of the Antas River, RS, Brazil. Coord(UTM) = Universal Transverse Mercator coordinate system.

Variables	S1	S2	S3	S4
Coord (UTM)	22J 0599354	22J 0599163	22J 0599065	22J 0597448
	6815858	6815734	6815020	6810506
Oxygen saturation (% O_2)	75.2 (±8.1)	76.5 (±9.0)	73.9 (±9.1)	78.7(±7.7)
BOD (mg l^{-1})	nd	nd	2.0 (±0.8)	nd
pH	6.9 (±0.2)	6.9 (±0.2)	6.7 (±0.2)	6.8 (±0.1)
Conductivity (mS cm^{-1})	16.5 (±5.1)	16.3 (±5.2)	17.4 (±4.7)	17.9 (±4.1)
Total phosphorus (mg l^{-1})	25.36 (±10.37)	29.37 (±13.52)	72.88 (±34.68)	52.26 (±24.07)
NO_3 (mg l^{-1})	0.07 (±0.03)	0.06 (±0.04)	0.13 (±0.05)	0.09 (±0.04)
NH_4 (mg l^{-1})	0.27 (±0.08)	0.29 (±0.08)	0.29 (±0.06)	0.31 (±0.08)
Total nitrogen (mg l^{-1})	0.41 (±0.09)	0.41 (±0.10)	0.49 (±0.16)	0.41 (±0.07)
Total solids (mg l^{-1})	53 (±22)	41 (±12)	84 (±15)	51 (±32)
Turbidity (NTU)	1.9 (±0.7)	1.8 (±0.6)	4.2 (±1.7)	3.9 (±1.9)
SiO_2 (mg l^{-1})	13.78 (±3.91)	13.84 (±3.86)	13.18 (±3.63)	12.44 (±2.78)
Absorbance 320 nm	0.074 (±0.044)	0.079 (±0.045)	0.079 (±0.046)	0.080 (±0.042)
River depth (cm)	21.8 (±6.7)	25.2 (±8.2)	27.3 (±9.7)	28.3 (±8.8)
Current velocity (m s^{-1})	0.41 (±0.16)	0.46 (±0.16)	0.39 (±0.13)	0.38 (±0.12)
Chlorophyll a (mg cm^{-2})	0.40 (±0.27)	0.30 (±0.14)	0.94 (±0.51)	0.59 (±0.37)
Trophic State Index	oligotrophic	oligotrophic	eutrophic	mesotrophic
Density (ind cm^{-2})	5,327 (±4,073)	3,803 (±3,104)	10,765 (±9,806)	6,012 (±5,929)
Richness (n° taxa)	19 (±3.5)	17 (±4.9)	22 (±1.6)	18 (±2.6)
H' (nats ind)	1.940 (±0.466)	1.742 (±0.571)	2.148 (±0.203)	2.059 (±0.275)

Table II: Results of randomization tests of the environmental and biotic variables among sampling stations. P = probability that the sum of the squares (Q) obtained by randomization (Q_b^{o3}) be as extreme as the sum of the squares of the observed data (Q_b) in 1000 iterations. * = significant results with a £ 0.05. § = raw data below detection level.

Contrasts	S1XS2	S1XS3	S1XS4	S2XS3	S2XS4	S3XS4
P (Q >Q)						
%O2	0,512	0,401	0,133	0,084	0,300	0,019*
BOD	§	§	§	§	§	§
pH	0,716	0,216	0,313	0,049*	0,173	0,651
Cond	0,120	0,152	0,178	0,080	0,148	0,423
TP	0,393	0,013*	0,009*	0,012*	0,016*	0,008*
NO3	0,844	0,008*	0,070	0,006*	0,018*	0,006*
NH4	0,253	0,247	0,039*	0,972	0,465	0,222
TN	1,000	0,060	0,963	0,085	0,974	0,233
TS	0,263	0,014*	0,832	0,007*	0,678	0,026*
Turb	0,297	0,012*	0,005*	0,013*	0,011*	0,109
SiO2	0,618	0,123	0,080	0,057	0,064	0,278
Abs320	0,380	0,016*	0,070	0,966	0,589	0,819
Depth	0,038*	0,019*	0,010*	0,128	0,052	0,314
Vel	0,040*	0,596	0,318	0,024*	0,038*	0,959
Cloro-a	0,224	0,016*	0,007*	0,006*	0,019*	0,021*
Density	0,498	0,042*	0,904	0,060	0,230	0,188
Richness	0,158	0,049*	0,462	0,032*	0,506	0,032*
H'	0,354	0,391	0,558	0,101	0,307	0,481

Table III: Results of Indicator Species Analysis. IV = indicator value. p = probability that IV obtained by randomization be as extreme as IV of observed data in 1000 permutations. ¨ = indicator species (p £ 0.05); * = abundant species.

Taxon	IV	p	Station
cf. Achnanthidium sp. *	34.4	0.665	S4
Achnanthidium microcephalum (Kützing) Czarnecki *	32.0	0.840	S3
Aulacoseira granulata (Ehrenberg) Simonsen var. angustissima (O. Müller) Simonsen	9.1	1.000	S2
Brachysira brebissonii Ross	33.7	0.101	S3
Cocconeis placentula Ehrenberg var. acuta Meister *	33.2	0.449	S1
Cocconeis placentula Ehrenberg var. lineata (Ehrenberg) Van Heurck *	30.8	0.715	S1
Cyclotella meneghiniana Kützing	25.0	0.253	S1
Encyonema minutum (Hilse) Mann *	53.6	0.059	S3
Encyonema silesiacum (Bleisch in Rabenhorst) Mann *	28.2	0.672	S1
Eunotia bilunaris (Ehrenberg) Souza	34.7	0.210	S3
Eunotia cf. incisa Gregory	11.0	1.000	S1
Eunotia pseudosudetica Metzeltin, Lange-Bertalot & García-Rodríguez *	36.2	0.428	S3
Fragilaria capucina Desmazieres *	32.5	0.577	S3
Fragilaria capucina Desmazieres var. mesolepta Rabenhorst	19.7	0.236	S2

Table III: Cont.

Taxon	IV	p	Station
Fragilaria sp.1 *	31.0	0.360	S3
Fragilaria sp.2	37.4	0.077	S3
Frustulia crassinervia (Brèbisson) Lange-Bertalot & Krammer	13.9	0.953	S3
Frustulia saxonica Rabenhorst	25.0	0.226	S3
Gomphonema affine Kützing *	34.1	0.176	S3
Gomphonema parvulum (Kützing) Kützing morfotipo 1 *	46.8	0.298	S3
Gomphonema parvulum (Kützing) Kützing morfotipo 2 ¨ *	50.5	0.047	S3
Gomphonema parvulum (Kützing) Kützing morfotipo 3 *	34.8	0.267	S3
Gomphonema pseudoaugur Lange-Bertalot	25.0	0.215	S3
Gomphonema tenuissimum Fricke ¨ *	50.7	0.036	S1
Gomphonema sp.1	15.9	0.758	S3
Gomphonema sp.2	33.1	0.072	S3
Gyrosigma sp.	15.9	0.426	S1
Lemnicola hungarica (Grunow) Round & Basson	6.8	0.741	S2
Luticola costei Metzeltin & Lange-Bertalot	25.0	0.208	S2
Luticola goeppertiana (Bleisch) Mann ¨ *	71.9	0.001	S3
Luticola saxophila (Bock ex Hustedt) Mann	25.0	0.222	S3
Melosira varians Agardh ¨	32.5	0.050	S3
Meridion circulare (Greville) Agardh var. constrictum (Ralfs) Van Heurck *	12.4	0.936	S3
Navicula angusta Grunow ¨	35.7	0.046	S1
Navicula cryptocephala Kützing *	23.6	0.613	S4
Navicula cryptotenella Lange-Bertalot ¨ *	79.9	0.002	S3
Navicula sp.	7.5	1.000	S2
Neidium sp.1	24.5	0.244	S3
Neidium sp.2	27.7	0.295	S1
Nitzschia palea (Kützing) Smith ¨ *	90.0	0.002	S3
Nitzschia sp.	35.7	0.092	S3
Nupela praecipua (Reichardt) Reichardt	31.2	0.355	S3
Pinnularia parvulissima Krammer	25.8	0.168	S3
Planothidium frequentissimum (Lange-Bertalot) Round & Bukhtiyarova ¨	37.5	0.046	S2
Psammothidium subatomoides (Hustedt) Bukhtiyarova & Round ¨ *	64.2	0.004	S3
Sellaphora pupula (Kützing) Mereschkowsky	12.5	1.000	S4
Surirella sp.	25.0	0.244	S3
Ulnaria ulna (Nitzsch) Compère	11.9	0.698	S4

parvulum morph. 2 occurred in large quantity in all sampling stations, although with a higher abundance in S3 (8% and 30%, respectively) and S4 (11% and 30%, respectively).

Table III shows the result of the Indicator Species Analysis, which pointed nine species (Fig. 3) that had its abundances and frequencies statistically associated to some of the sampling stations.

Relation between diatoms and the environmental variables

The CCA performed with the nine indicator species revealed a significant

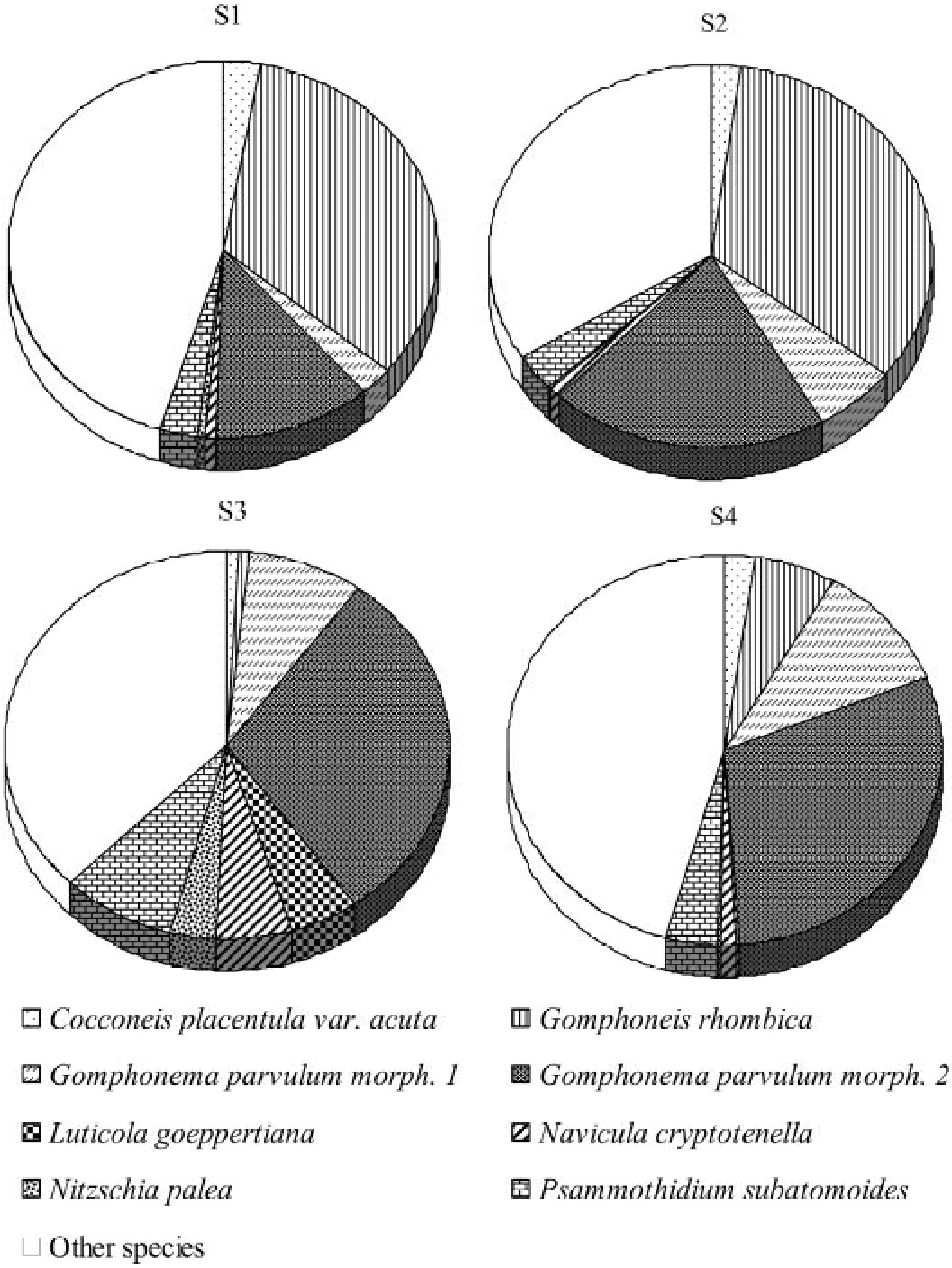

Figure 2: Relative abundance (%) of the epilithic diatom species in each sampling station (S1, S2, S3, S4) of the upper course of the Antas River, RS, Brazil.

correlation between the diatom community and the environmental gradient, reflecting the physical and chemical alterations in the water (Fig. 4). The two first ordination axes explained 42% of the data variance and were statistical significant (eigenvalue of axis 1 = 0.17, p = 0.002; axis 2 = 0.10, p = 0.042). The species-environment correlations were high and significant for the two axes (r = 0.918, p = 0.011; r = 0.878, p = 0.014, respectively), indicating a strong association between the environmental variables and the species distribution. The intra-set correlations showed that the first axis was negatively correlated to chlorophyll a (-0,726), total solids (-0,616),

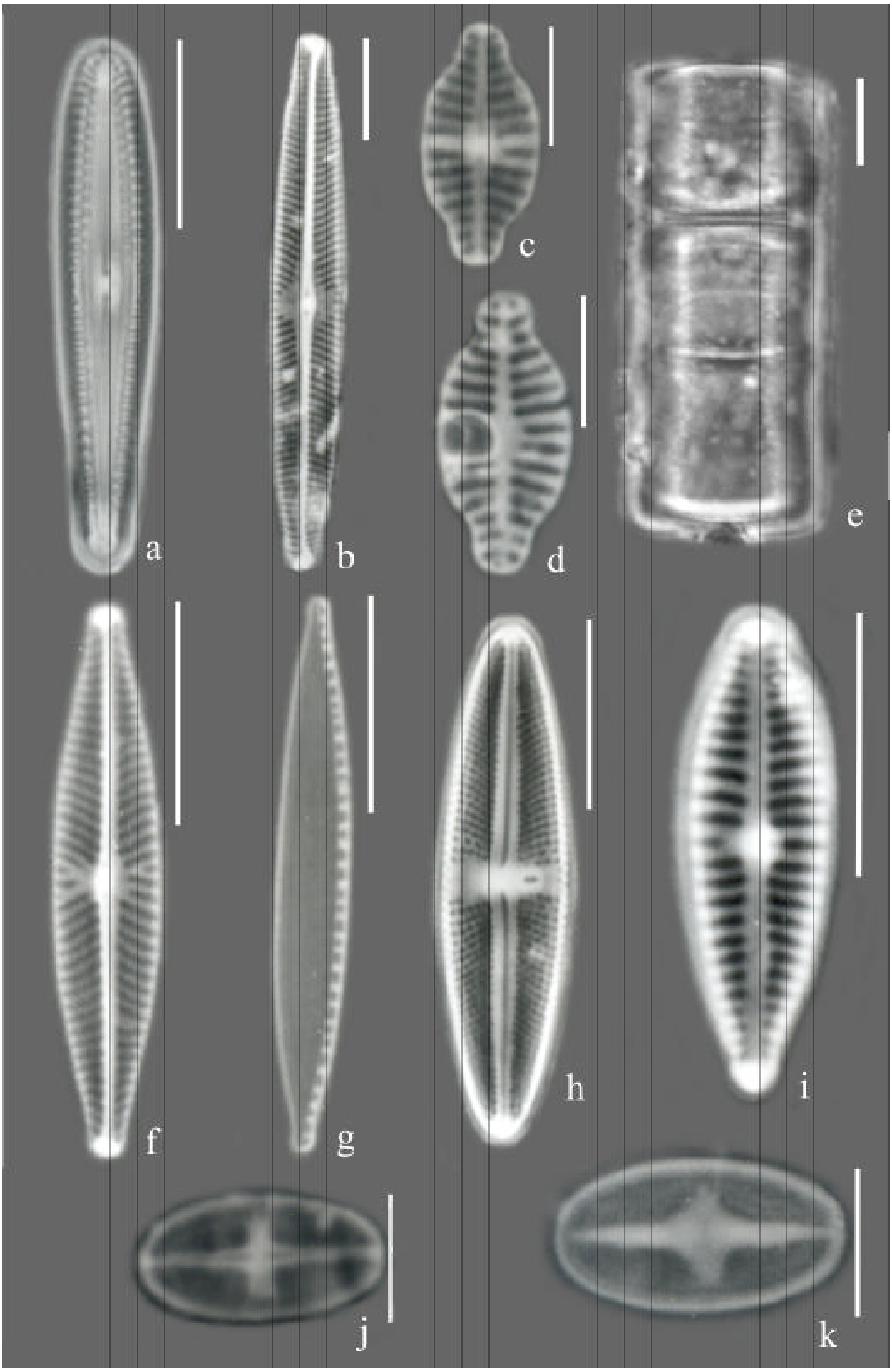

Figure 3: Indicator species in the upper course of the Antas River, RS, Brazil. a. Gomphoneis rhombica; b. Navicula angusta; c. Planothidium frequentissimum valve with raphe; d. P. frequentissimum valve without raphe; e. Melosira varians; f. Navicula cryptotenella; g. Nitzschia palea; h. Luticola goeppertiana; i. Gomphonema parvulum morph.2; j. Psammothidium subatomoides valve with raphe; k. P. subatomoides valve without raphe. Scale: c, d, j and k = 5 mm; a, b, e, f, g, h and i = 10 mm.

turbidity (-0,579), total phosphorus (-0,540), and nitrate (-0,396), and positively correlated to pH (0,547). On the positive side of this axis are located oligotrophic and oligossaprobic species, such as Gomphoneis rhombica and Navicula angusta, while there are eutrophic and polissaprobic species on the negative sphere, such as Luticola goeppertiana, Melosira varians, and Nitzschia palea. Axis 2 was correlated mainly with O_2 saturation (-0,571).

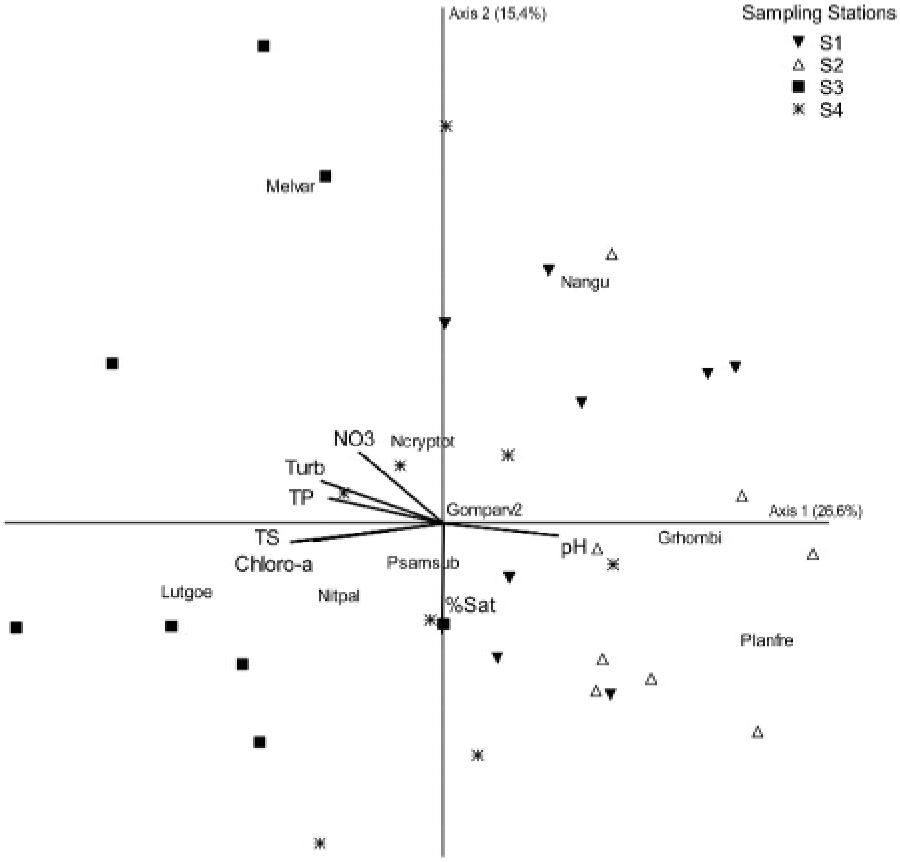

Figure 4: Scatter plot of the Canonical Correspondence Analysis based on the indicator species. Axes 1 and 2. Grhombi: Gomphoneis rhombica; Gomparv2: Gomphonema parvulum morph.2; Lutgoe: Luticola goeppertiana; Melvar: Melosira varians; Nang: Navicula angusta; Ncryptot: N. cryptotenella; Nitpal: Nitzschia palea; Planfre: Planothidium frequentissimum; Psambub: Psammothidium subatomoides. %Sat: oxigen saturation; Chloro-a: chlorophyll a; NO3: nitrate; TP: total phosphorus; TS: total solids; Turb: turbidity.

Discussion

Environmental Variables

Sampling stations upstream the fish farming influence are characterized by waters with low nutrient and chlorophyll a concentrations, low electric conductivity and despicable values of BOD, besides being the shallowest and fastest environments, characteristics typically associated to headwater streams naturally not very productive (Vannote et al., 1980). These attributes evidence the integrity of these environments, with natural characteristics still preserved.

The most pronounced pattern in the physical and chemical water composition, along the studied stretch, was related to those variables directly associated to the fish farming effluents, such as nutrients, solids, and turbidity. Total phosphorus, nitrate, chlorophyll a, total solids and turbidity reached values approximately two to three times higher in S3 than in S1 and S2, enabling to observe a clear longitudinal vector of decrease in the water quality, with a higher trophic compromise in S3. Other authors related processes of eutrophication in water bodies receiving fish farming effluents (Kendra, 1991; Oberdorff & Porcher, 1994).

The main course of the Antas River receives water from a series of affluents between the sampling stations S3 and S4, contributing to the dilution of nutrients and solids originated from the dumping of the fish farming effluent. Even so, S4 did not show the same conditions as the upstream sampling stations, probably due to the rundown of materials from adjacent terrestrial areas, mainly small rural properties, and also due to the remaining effects of the fish farming effluents.

Structure of the diatom community and its association with environmental variables

The diatom richness observed in the upper course of the Antas River (48 taxa) is low when compared to other lotic systems in the state of Rio Grande do Sul. Hermany et al. (2006) registered 147 taxons of epilithic diatoms in a stream belonging to the Guaíba Basin and Salomoni et al. (2006) observed 166 taxa in the Gravataí River.

However, it was demonstrated that high altitude streams show taxocenoses poor in species, probably due to the more austere natural conditions. These environments tend to be poor in nutrients and to show faster and colder waters, increasing the competition for resources and limiting the number of available niches for the different species of algae. Jüttner et al. (1996), studying a high altitude oligotrophic river in Nepal, found 36 species of epilithic diatoms. Mendes (2003) and Canani (2005), cited 38 and 30 taxa, respectively, in streams in the Southeast of Brazil located in altitudes between 1,200 and 1,450 m.

This study focused on upstream waters characterized by low nutrient concentrations, where the autotrophic production acts as a component of lower magnitude in relation to the heterotrophic production. Streams with such characteristics are particularly sensitive to the enrichment of nutrients arising from allochthonous sources, which can induce an alteration in the energetic base of the system, causing autotrophic processes typical of medium courses of rivers (Vannote et al., 1980). The result of this trophic enrichment and consequent water eutrophication is the increase in primary production and the alteration of the algal community structure, offering advantages to more competitive species and excluding less tolerant species. According to Rott et al. (2006), the composition of benthic algae communities in non-shaded altitude streams is affected mainly by the chemical characteristics and by water flow.

Stevenson (1999) points out that the high richness of species indicate high biotic integrity, while the increase in pollution leads to a decrease in richness since it breaks the stability of the system, where many species find themselves in situations of environmental stress. However, the richness of the Antas River showed an inverse trend, with the sampling station with the most compromised quality of water (S3) showing the highest richness. As it was pointed by Jüttner et al. (1996), and Bahls et al. (1992) a quick increase in the concentration of nutrients can lead to an increase of richness in springs and oligotrophic streams.

Alterations in the algal density might reflect the effect of other environmental variables than nutrients, such as current velocity, grazing, and luminosity. Ghosh & Gaur (1998) observed an inverse correlation between the density of periphytic algae and current velocity. On the other hand, Hermany et al. (2006) did not find a relation between the density of epilithic diatoms and the velocity; however, they attributed the fluctuations in the density to the availability of light. In the Antas River, no significant differences were found in the water velocity between S1 and S3, the only sampling stations with density statistically different, and all sampling stations were located in an open area (fields), not having significant riparian shading.

It is also assumed that grazing does not act as a predominant factor, from the assumption that the strongly pastured environments are characterized by a flora with prostrated growth, apically flattened, which is not the case of Antas River, where the best represented taxa showed vertical growth, such as Gomphoneis rhombica and Gomphonema parvulum.

The majority of the abundant species found are considered cosmopolitan and with large environmental amplitude. Other species are described as having preference for oligotrophic environments and/or environments of altitude, however not being restricted to these environments, such as Cocconeis placentula var. acuta, Meridion circulare var. constrictum, Psammothidium subatomoides, and Gomphoneis rhombica (Van Dam et al., 1994; Silva-Benavides, 1996).

Outstanding structural alterations were established, strongly governed by the observed eutrophication gradient, with the substitution, immediately due to discharge of fish farming effluents, by species that are tolerant to pollution, like Luticola goeppertiana, Melosira varians, Nitzschia palea, and Gomphonema parvulum, broadly described in the literature as typical of eutrophic environments (Denys, 1991; Van Dam et al., 1994; Salomoni et al., 2006) and that in the Antas River show preference for waters enriched with nutrients and with higher concentrations of solids. These taxa, together with Navicula cryptotenella and Psammothidium subatomoides, were pointed out as indicators of greater anthropogenic impact.

Kelly & Whitton (1995) attributed an indicative value of 4 for Melosira varians and Nitzschia palea, in a growing scale of eutrophication with a maximum of 5, in order to calculate the Trophic Diatom Index (TDI). For Gomphonema parvulum and Navicula cryptotenella a value of 5 was attributed, correspondent to environments of strong eutrophication. However, Lobo et al. (2004), studying three streams in the state of Rio Grande do Sul (Brazil) with PO_4 concentrations between 0.018 and 0.6 mgL^{-1}, classified Navicula cryptotenella as having intermediate tolerance to eutrophication. It is known that Navicula cryptotenella shows wide ecological amplitude, being described by many authors as common both in oligotrophic and eutrophic environments (Denys, 1991; Van Dam et al., 1994; Silva-Benavides, 1996; Salomoni et al., 2006). In the Antas River, Navicula cryptotenella showed a high indicator value for eutrophic waters, reaching densities of eight to 20 times higher than in mesotrophic and oligotrophic waters, respectively.

In Holland, Van Dam et al. (1994) referred Psammothidium subatomoides as oligotrophic to mesotrophic, as well as Silva-Benavides (1996) in the Savegre River in Costa Rica. Planothidium frequentissimum was mentioned as an indicator of eutrophic environments (Rimet et al., 2004) and tolerant to organic pollution (Van Dam et al., 1994). In a study carried out by Wetzel et al. (2002), the species stood out in the middle course of the Pardo River hydrographical basin (RS, Brazil), characterized by moderate pollution. However, in the Antas River Psammothidium subatomoides was related to eutrophic waters, while Planothidium frequentissimum was associated to sampling station S2. Gomphoneis rhombica is common in cold waters and non-polluted mountainous areas, as in the region of Pirineus in France and in the Himalayas, Pakistan (Krammer & Lange-Bertalot, 1991b). Merino et al. (1994) cite this species occurring in non-polluted waters, characterized by high concentrations of O_2, low conductivity and low concentration of nutrients, in a high altitude stream in the Andorras. Similar results were related by Lam & Lei (1999) in Hong Kong and by Silva-Benavides (1996) in Costa Rica, which found Gomphoneis rhombica dominating the diatom flora in rivers with total phosphorus contents of 7 to 26 mg L^{-1} and of 20 mg L^{-1}, respectively. Also Navicula angusta is extensively referred in the literature as not very tolerant to eutrophication and with high oxygen requirements (Denys, 1991; Van Dam et al., 1994). Furthermore, Patrick & Reimer (1966) mentioned the species as characteristically of montane environments. These trends were confirmed for the Antas River, with Gomphoneis rhombica and Navicula angusta showing affinity for environments with total phosphorus concentrations between 25 and 29 mg L^{-1}.

Conclusions

The results of this study indicate the occurrence of a strong gradient of water quality associated to fish farming effluents, causing the trophic compromise of downstream environments and affecting the distribution of epilithic diatom species. An evident substitution of species characteristic of oligotrophic environments by species indicators of eutrophic environments was observed.

Two water quality indicator groups were outlined. The first group, composed by Gomphoneis rhombica, Navicula angusta, and Planothidium frequentissimum indicated environments with low nutrient availability, low quantity of total solids, and low turbidity. Luticola goeppertiana, Melosira varians, Nitzschia palea, Gomphonema parvulum, Navicula cryptotenella, and Psammothidium subatomoides indicated eutrophic waters, with greater anthropogenic impact. The capacity of bioindication of the highlighted species is confirmed for the study region, except Psammothidium subatomoides and Planothidium frequentissimum, which indicated eutrophic and oligotrophic conditions, respectively.

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