

Structural Characteristics and Oxygen Consumption of the Epipelic Biofilm in Three Lowland Streams Exposed to Different Land Uses

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Abstract The structural features and O₂ consumption of the epipelic biofilm in streams of the Pampean plain were explored. The study was conducted in three lowland streams subjected to different anthropic disturbances. Three sampling sites were selected in different sectors of these streams considering land use intensity (high, moderate, and low). Samples of the water and of the epipelic biofilm were taken seasonally. El Pescado stream is subjected to a low level of human impact and showed lower organic matter and nutrient contents than the Rodríguez and Don Carlos streams which are subjected to moderate and high levels of human impact. The biofilm composition of the three streams was represented by cyanophytes and diatoms but with different species composition and dominance; protozoans and nematodes were the characteristic heterotrophic groups in the three streams. The Rodríguez and Don Carlos streams showed the highest abundance of organisms. Multiple regression showed that O₂ consumption, chlorophyll *a* and trophic index were significantly correlated with the oxygen demands. On the other hand, the Rodríguez and Don Carlos streams exhibited significant differences with the El Pescado stream in O₂ consumption,

trophic index, and chlorophyll *a* content. Our results demonstrated that the different biological descriptors responded to environmental variables that are influenced by the different land use intensities, being chlorophyll *a*, abundance of organisms, and O₂ consumption the most sensitive variables to the changes water quality.

Keywords Epipelon · Pampean plain · Water quality · Anthropogenic disturbance · Biological descriptors

1 Introduction

Stream biofilms are microbial populations of bacteria, algae, fungi, protozoans, and micrometazoans, embedded in a polysaccharide matrix and inhabiting rocks, gravel, wood, and sediments. The organic matter accumulated in stream sediments is decomposed by bacteria, fungi and occasionally algae, which convert high-molecular-weight molecules to low-molecular-weight ones by extracellular enzymatic hydrolysis (Romaní et al. 1998). As a consequence, biofilms play a key role in the energetic dynamics of the streams and knowing the way and efficiency in which organic matter and nutrients are processed, implies knowing the self-depuration capacity of lotic systems (Sabater et al. 1993).

Stream systems running through the Pampean plain are affected by the activities and products of agriculture, cattle-raising and industry. In addition, the most

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important urban centre in Argentina is located in this area. Water quality deterioration is mainly caused by organic enrichment, nutrients' input, heavy metals, pathogenic agents, and pesticides. The bottom substrate of these streams is mostly composed of slime and clay with low proportions of gravel and sand, except at the mouth of the rivers and streams where sand can be dominant. Limestone concretions can be found in the mid or low sections of certain streams (Gómez and Licursi 2001). As a consequence, the epipelon is the most represented benthic community in these running waters (Licursi and Gómez 2002).

Few studies have analysed the characteristics of biofilm development on soft substrates (epipsammic–epipelic biofilm) (Hedin 1990; Hill et al. 1998, 2000; Admiraal et al. 1999; Scinto and Reddy 2003; Velasco et al. 2003; Rasheed et al. 2004; Romani et al. 2004; Fellows et al. 2006). Other studies carried out in lowland streams of the Pampean plain have analyzed structural parameters of the epipelic community (Giorgi and Malacalza 1994, 2002; Claps 1996; Solari and Claps 1996; Gómez 1998; Gómez and Licursi 2001; Licursi and Gómez 2002; Tolcach and Gómez 2002; Giorgi et al. 2003, 2004), and only Vilches (2005) explored the metabolism of autotrophic organisms in one stream of this area.

The aim of this work was to study the structural features and O₂ consumption of the biofilm, comparing different water qualities in lowland streams of the Pampean plain.

This information is of interest for the diagnosis of the ecological quality, and the monitoring and management of lowland streams.

2 Materials and Methods

2.1 Study Area

Running waters of the Pampean plain run through a vast grassy plain and have slow current. The climate is temperate humid with mean annual precipitation between 600 and 1,200 mm, and a mean annual temperature of 16°C; even though precipitation is distributed all along the year, maximum rainfall generally occurs in spring and autumn. The study was carried out in three Pampean plain streams called Don Carlos (DC), Rodríguez (R), and El Pescado (P), which are subjected to different anthropic disturbances

(Fig. 1). These systems are located in the La Plata city surroundings, and flow into the Río de la Plata estuary. We selected three sampling sites in different sections of these streams: site 1 (upstream section), site 2 (medium section) and site 3 (downstream section; Table 1).

El Pescado stream runs through a rural area with agricultural and cattle-rearing activities. Different studies carried out in the Pampean plain (Macluf et al. 1998; Graça et al. 2002; Licursi and Gómez 2002) have pointed out that El Pescado stream is subjected to a low level of human impact therefore we have considered it as a reference stream.

The Rodríguez stream runs across suburban areas. Site 1 is located upstream where the main activities are horticulture and cattle farming. Its medium section, site 2, is exposed to effluents from a meat factory and downstream, in site 3, the most important impact is due to urban activities such as insufficiently treated sewage discharges.

The Don Carlos stream runs across a suburban area where site 1 is located near the source and exposed to horticultural activities; sites 2 and 3 are exposed to sewage effluents and outflows coming from a textile and a metallurgical industry, respectively. Hydraulic engineering practices, such as canalization and the modification of its bed and bank occur downstream from site 2.

Studies carried out in the Don Carlos and Rodríguez streams (Gómez and Licursi 2001, 2003; Tolcach and Gómez 2002; Bauer et al. 2002) showed that the sites 2 and 3 of these streams are subjected to a moderate and high level of human impact respectively.

2.2 Water Analysis

Seasonally at each site the following physical and chemical variables were measured: dissolved oxygen (DO; YSI 52 dissolved oxygen meter), conductivity (Lutron CD-4303), temperature, and pH (Hanna HI 8633). Water samples to be analysed for the dissolved inorganic nutrients were filtered through glass fiber filters (Whatman GF/C) and, together with samples for biochemical oxygen demand and chemical oxygen demand, were stored at 4°C until arrival at the laboratory. Nitrite and ammoniacal nitrogen were determined colorimetrically, nitrate was reduced to nitrite before colorimetrically measurement. BOD₅ was determined after 5 days incubation at 20°C and COD by oxidation with potassium dichromate in

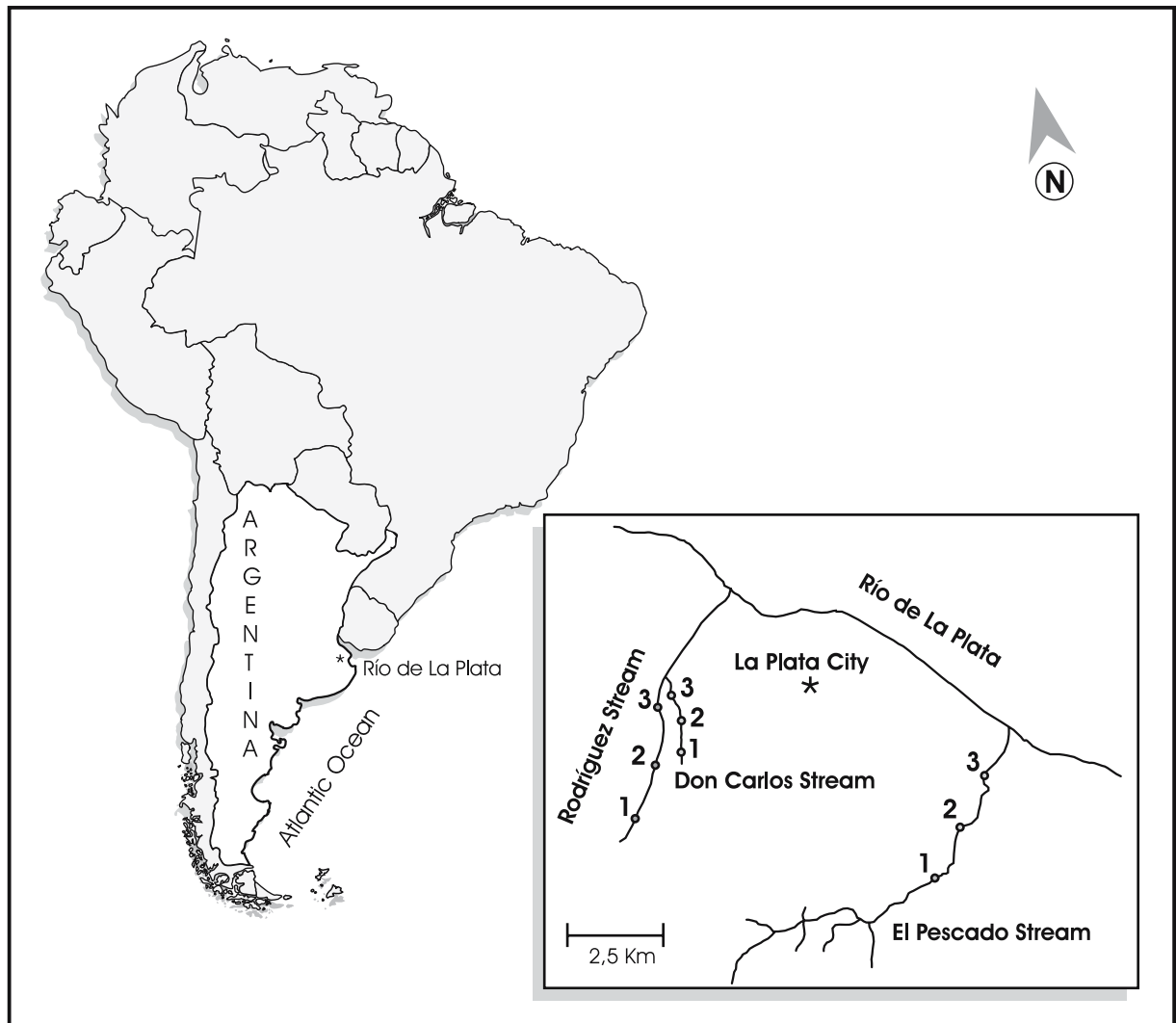


Fig. 1 Locations of the studied streams and sampling sites

acid medium (Tabatabai 1974; Mackereth et al. 1978; APHA 1998).

2.3 Sample Collection and Preparation

The microbenthos was sampled in winter and spring 2004, and in summer and autumn 2005. At each sampling site 1 cm² of the epipelic biofilm was collected by pipetting of 5 ml of the superficial layer of the bottom (Gómez and Licursi 2001). The samples obtained were kept cold and in the dark during transport to the laboratory. Two replicates were taken for chlorophyll *a* and ash-free dry weight (AFDW) determinations. For chlorophyll analysis two aliquots

5 ml were filtered using Whatman GF/C glass fiber filters and immersed in 90% acetone for 24 h in the dark at 4°C. The extract was read with a spectrophotometer and the chlorophyll *a* concentration was obtained according to Steinman and Lamberti (1996). AFDW was measured as the difference in weight between the dried mass at 60°C for 24 h and combusted at 550°C for 4 h (Bourasa and Cataneo 1998). The trophic index (TI), which provides an estimate of amount of autotrophic biomass relative to the total biomass, was calculated as the ratio between chlorophyll *a* and AFDW (Clark et al. 1979). Five samples were fixed with formaldehyde (4%) and used for the study of community composition (except

Table 1 Average values of the morphometric and hydraulic characteristics of the sampling sites

	El Pescado stream			Rodríguez stream			Don Carlos stream		
	1	2	3	1	2	3	1	2	3
Depth (m)	0.83	1.5	1.5	0.31	0.21	0.22	0.5	0.35	0.25
Width (m)	6.81	15	19.83	2.5	2.8	4.5	1.5	1	5
Flow (m s ⁻¹)	0.11	0.16	0.48	0.22	0.14	0.3	0.11	1.8	0.46
Discharge (m ³ s ⁻¹)	0.9	10.56	8.25	0.15	0.06	0.38	0.15	0.1	0.3

bacteria and fungi), using standard keys, and quantified in a Sedgwick-Rafter chamber through an optical microscope (Olympus BX 50).

Oxygen consumption was measured in triplicate in the laboratory in the dark at constant temperature (20–22°C). Two aliquots were collected and carefully placed in glass bottles (100 ml), which were filled with filtered stream water (Whatman GF/C glass fiber filters) and then sealed airtight and wrapped in aluminium foil. Microbenthic oxygen consumption was determined before the bottles were closed and again after 1.5 to 2 h of incubation (Sabater et al. 1998). The samples were not stirred during the incubation except 1 min before each O₂ measurement to homogenize the bottle content. Dissolved oxygen levels in each bottle were measured with an oxygen meter (Rasheed et al. 2004).

2.4 Statistical Analysis

In order to compare some variables and to know if there were differences between the streams and among the sampling sites, a two way analysis of variance was carried out. Two way ANOVA is a parametric test that assumes that all the samples were drawn from normally distributed populations with the same variances. Therefore, data which were not normally distributed were normalized, temperature, dissolved oxygen, NO₂⁻, PO₄³⁻, BOD₅, O₂ consumption, AFDW, chl *a* and TI values were log₁₀ transformed, and conductivity and NO₃⁻ were square root transformed.

Multiple regression analysis was used to test for the influence of the O₂ demand, and nutrients (independent variables) on the dependent variables (O₂ consumption, AFDW, chl *a* and TI).

The ordination of the samples, based on the physical and chemical variables, was performed using principal component analysis (PCA).

3 Results

3.1 Water Quality

Water in the three streams had uniform pH and temperature, but differed markedly in nutrients and organic matter (Table 2). ANOVA revealed significant differences ($P < 0.05$) between the Rodríguez and El Pescado streams for PO₄³⁻, NO₂⁻, BOD₅, and conductivity. The Don Carlos and El Pescado streams only showed significant differences ($P < 0.05$) in dissolved oxygen, BOD₅, and conductivity values. In Don Carlos stream the PO₄³⁻ amount showed significant differences ($P < 0.05$) between at site 1 and at sites 2 and 3, while in the Rodríguez stream the conductivity showed significant differences ($P < 0.05$) between at site 1 and at sites 2 and 3. On the other hand we didn't observe significant differences between at sites of the El Pescado stream for none of the variables analysed. The first two factors of the PCA carried out on water quality variables, accounted for 46.7% of the total variance (Fig. 2). The variables that weighted heavily were, in the first factor BOD₅ (-), COD (-) and DO (+) and in the second factor were NH₄⁺ (-) and PO₄³⁻ (-). The Fig. 2b show the ordination of the sampling sites according to the first two factors. The sites 2 and 3 of the Don Carlos stream were associated with a greater content of organic matter and, otherwise, most of the sites of El Pescado stream were associated with a greater content of dissolved oxygen. The sites 2 and 3 of Rodríguez stream were associated to waters enriched by nutrients.

3.2 Epipelic Biofilm

Taxa with densities greater 0.2% in the samples analysed are listed in Table 3. Biofilm composition in the three streams was represented by cyanophytes and diatoms but with different species composition

Table 2 Physicochemical characteristics of the sampling sites

	T (°C)	DO (mg l ⁻¹)	Conductivity (μS cm ⁻¹)	pH	NO ₃ -N (mg l ⁻¹)	NO ₂ -N (mg l ⁻¹)	NH ₄ ⁺ -N (mg l ⁻¹)	PO ₄ ³⁻ -P (mg l ⁻¹)	BOD ₅ (mg l ⁻¹)	COD (mg l ⁻¹)
P1	17.12±8.49	5.92±2	335.85±161.58	7.43±0.33	1.22±1.13	0.04±0.03	0.17±0.18	0.28±0.12	16.97±19.3	66±40.91
P2	16.1±7.97	6.65±2.12	600.65±226.39	7.78±0.32	1.66±1.57	0.03±0.03	0.52±0.9	0.41±0.24	8±4.69	57.25±22.9
P3	18.03±9.15	5.9±2.95	513.87±271.85	7.66±0.21	2.32±0.89	0.04±0.04	0.58±0.89	0.37±0.2	7±3.16	43±17.57
R1	15.87±5.95	5.45±2.35	745.37±62.54	7.80±0.11	2.95±4.27	0.41±0.38	0.93±0.95	1.60±0.7	19±10.23	32±8.87
R2	17.02±6.09	2.52±0.94	1,995±468.67	7.84±0.08	1.97±2.3	0.51±0.55	0.62±0.91	2.49±0.74	48.25±16.13	102±90.4
R3	17.1±5.52	3.82±0.93	1,544.5±257.91	7.86±0.04	2.51±1.18	0.77±0.65	1.56±0.81	2.89±0.94	29.25±9.54	64.5±49.7
DC1	15.47±1.45	3.45±1.95	893±46.49	7.67±0.32	2.32±1.92	0.22±0.13	0.03±0.04	0.92±0.08	21.5 ±15.46	31±13.32
DC2	19±1.44	3.3±1.26	987.25±86.16	7.53±0.09	1.14±1.84	0.10±0.16	0.92±0.68	0.15±0.11	71±6.16	141±75.17
DC3	18.35±1.87	2.85±1.71	1,080.5±64.8	7.64±0.32	1.36±1.39	0.26±0.26	1.13±0.33	0.28±0.08	88.25±92.9	163.5±139.84

Means and standard deviations are shown.

and dominance. While euglenoids, protozoans, and nematodes in Don Carlos stream showed a trend to increase from site 1 to site 3, chlorophytes abundance was higher at sites 1 and 3 than at site 2 (Fig. 3). At sites 2 and 3 of the Don Carlos stream thick mats of filamentous bacteria (*Beggiatoa* sp.) were noticed, in which ciliate protozoans cohabited. In the Rodriguez stream chlorophytes abundance showed the same trend that in the Don Carlos stream and in relation to the heterotrophic organisms, while density of protozoans increased toward site 3, nematodes decreased toward that site. In the El Pescado stream cyanophytes decreased towards site 3 and diatoms reached similar values of abundance at sites 1 and 3 that were higher than at site 2. Both groups, protozoans and nematodes, showed a clear trend to increase from site 1 to site 3. The abundance of all taxa in El Pescado stream was lower than the other streams.

Significant differences ($P<0.05$) were found in O₂ consumption, trophic index, and chlorophyll *a* amount between the Rodriguez and Don Carlos streams vs the El Pescado stream (Fig. 4). The O₂ consumption was linked with the gradient of pollution of the most impacted streams. In the Don Carlos and Rodriguez stream this descriptor showed a tendency to increase downstream from the industrial discharges (textile effluent, and meat factory effluent respectively). It was six times higher at sites 2 and 3 of the Don Carlos stream than at sites 2 and 3 of El Pescado stream while at sites 2 and 3 of the Rodríguez stream it tripled the values registered at sites 2 and 3 of El Pescado stream. In relation to the trophic index it was between three and eight times higher at sites 2 and 3 of the Don Carlos stream than at sites 2 and 3 of the El Pescado stream while at sites 2 and 3 of the Rodríguez stream it tripled the values registered at sites 2 and 3 of the El Pescado stream.

Despite the lack of statistical significance, AFDW had the highest mean values at sampling sites of Don Carlos and Rodriguez stream. Results of multiple regression analysis between water quality variables and biological descriptors of biofilm are showed in Table 4. O₂ consumption, chlorophyll *a*, and trophic index were significantly correlated with the oxygen demand. The biological descriptors showed a large range variation along the seasons being the lowest values of them in spring and/or autumn. This is coincident with the period of maximum rainfall that generally occurs in spring and autumn (Fig. 5).

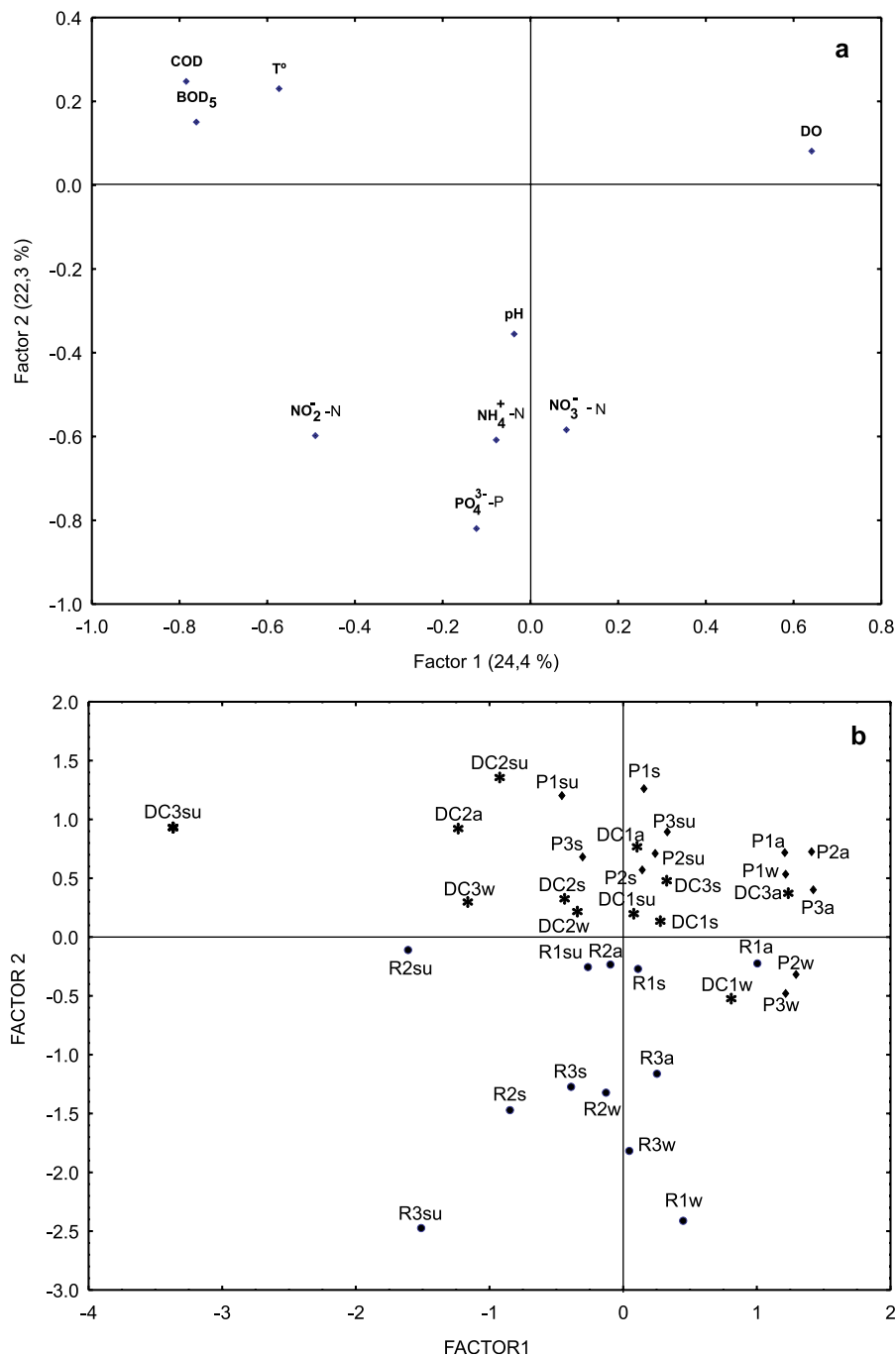


Fig. 2 **a** Representation of two first factors of PCA showing the physical and chemical variables of sampling sites. **b** Scores from physical and chemical variables (asterisk DC Don Carlos stream, filled circle R Rodríguez stream, filled diamond P El Pescado stream)

4 Discussion

Epipellic algae growing upon organic sediments often develop into dense communities several millimetres thick (Wetzel 1996). This is coincident with our

observations at sites with high amounts of organic matter as the sites 2 and 3 of the Don Carlos stream. Cyanophytes and diatoms are common in epipellic communities (Goldsborough and Robinson 1996; Solari and Claps 1996; Giorgi et al. 2004) and these

Table 3 Taxonomic composition of the epipelic biofilm and range of relative abundance

	El Pescado stream			Rodriguez stream			Don Carlos stream		
	1	2	3	1	2	3	1	2	3
Cyanophytes									
<i>Anabaena</i> sp.		*					*		
<i>Lyngbya</i> sp.								*	
<i>Merismopedia convoluta</i>	*								
<i>M. glauca</i>	*****								
<i>M. punctata</i>	****								
<i>Merismopedia</i> sp.							*		
<i>Oscillatoria articulata</i>							***		
<i>O. breve</i>								****	***
<i>O. formosa</i>				****					
<i>O. granulata</i>						*****			
<i>O. limosa</i>							****		
<i>O. subbrevis</i>				*****					
<i>O. tenuis</i>		****						*	
<i>Oscillatoria</i> spp.			****			***			
<i>Phormidium chalybeum</i>		****	***	*	*****	*		***	
Chlorophytes									
<i>Monorraphidium arcuatum</i>									*
<i>Oedogonium</i> sp.	*								
<i>Scenedesmus ecornis</i>						*			*
<i>S. obtusus</i>									*
<i>Schroederia</i> sp.			*						
<i>Spirogyra</i> sp.				*		*	*		
Euglenoids									
<i>Euglena acus</i>					*	*			
<i>Euglena</i> sp.		*						*	**
<i>Phacus</i> sp.		*							
<i>Trachelomonas</i> sp.					*				
Diatoms									
<i>Amphora veneta</i>								*	
<i>Cocconeis placentula</i>		*							
<i>Craticula cuspidata</i>							*	*	
<i>Cymbella silesiaca</i>							*		
<i>Diademsis confervacea</i>			*				*		
<i>Eunotia bilunaris</i>	*								
<i>Fragilaria ulna</i>				*			*		
<i>Gomphonem clavatum</i>				*	*		*		
<i>G. parvulum</i>	*			*	**	***	**	*	**
<i>Gyrosigma spencerii</i>	*		*	*					
<i>Melosira varians</i>	*	*	*	*					
<i>Navicula</i> spp.	*	*	***	*			*		
<i>N. capitata</i>			*				*		
<i>N. cryptocephala</i>								*	
<i>N. pupula</i>					*				
<i>N. pygmaea</i>						*			
<i>Nitzschia amphibia</i>							*		
<i>N. frustulum</i>		*			*		***		
<i>N. linearis</i>		*		*			*		
<i>N. palea</i>		*	*	*	*		*	*	*
<i>N. reversa</i>	*	*							
<i>N. sigma</i>	*	*	*				*		

Table 3 (continued)

	El Pescado stream			Rodriguez stream			Don Carlos stream		
	1	2	3	1	2	3	1	2	3
<i>N. umbonata</i>					*		*	*	
<i>Pinnularia gibba</i>					*			*	*
<i>P. microstauron</i>				*					
<i>Pinnularia</i> sp.	*								
<i>Rhoicosphenia abbreviata</i>		*							
<i>Rhopalodia musculus</i>			*						
<i>R. gibberula</i>			*						
<i>Sellaphora pupula</i>							*		
<i>Surirella angusta</i>				*	*				
<i>Surirella brebissonii</i>	*		*		*		**	**	*
Others		**	****						*
Nematodes		*	*	*					*
Protozoans									
<i>Euplotes patella</i>									*
<i>Glaucoma</i> sp.								*	*
<i>Paramecium caudatum</i>				*	*	*	*	*	**
<i>Paramecium</i> spp.		*	*	*	*	*	*	*	*
Bacteria									
<i>Beggiatoa</i> sp.								+	+

* > 0.2–5%

** > 5–10%

*** > 10–25%

**** > 25–50%

***** > 50% + mats of *Beggiatoa* sp.

were the groups that characterized the biofilm of the streams studied. At site 2 of the Don Carlos stream there are mats of the sulphur-reducing bacteria *Beggiatoa* spp. that, joined with filaments of cyanophytes, constitute a thick layer that cover the bottom substrate in which ciliate protozoans cohabited. Hagen and Nelson (1997) reported that *Beggiatoa* spp. occur at sites of organic pollution in soft anoxic mud. Bernhard et al. (2000) also noted several species of ciliates, nematodes and euglenoids feeding on organic matter, in relation to the mats of *Beggiatoa* spp. in polluted sites.

Leland (1995) pointed out that streams receiving agricultural and urban sewage waters have a surplus of nutrients that can produce high growths of benthic algae. Dodds et al. (1997) also quantified the nutrient threshold that causes nuisance algal growths to be at 350 $\mu\text{g l}^{-1}$ total N and 30 $\mu\text{g l}^{-1}$ total P. Above these nutrient concentrations, mean benthic algal

chlorophyll *a* could surpass 100 mg m^{-2} . The amount of nutrients, mainly NO_3^- and PO_4^{3-} , in the Rodriguez and Don Carlos streams was higher than the values pointed out by these authors. More than 65% of the chlorophyll *a* values were above 100 mg m^{-2} , with peaks of 360 and 443 mg m^{-2} in summer and autumn respectively in the Don Carlos stream. In the Rodriguez stream the chlorophyll *a* reached a peak of 665 mg m^{-2} in winter. In addition Bombowna (1972) found values of up to 1,600 mg m^{-2} of chl *a* in the Raba river (Poland) which is subject to pollution. On the other hand, we noticed that 66% of the AFDW values were higher than 50 g m^{-2} at sites with moderate to high land use intensity, mainly horticultural activity. The AFDW values were higher than those reported by Biggs (1996) who cited values above 15 g m^{-2} for streams in New Zealand located in areas with intensive agricultural activities.

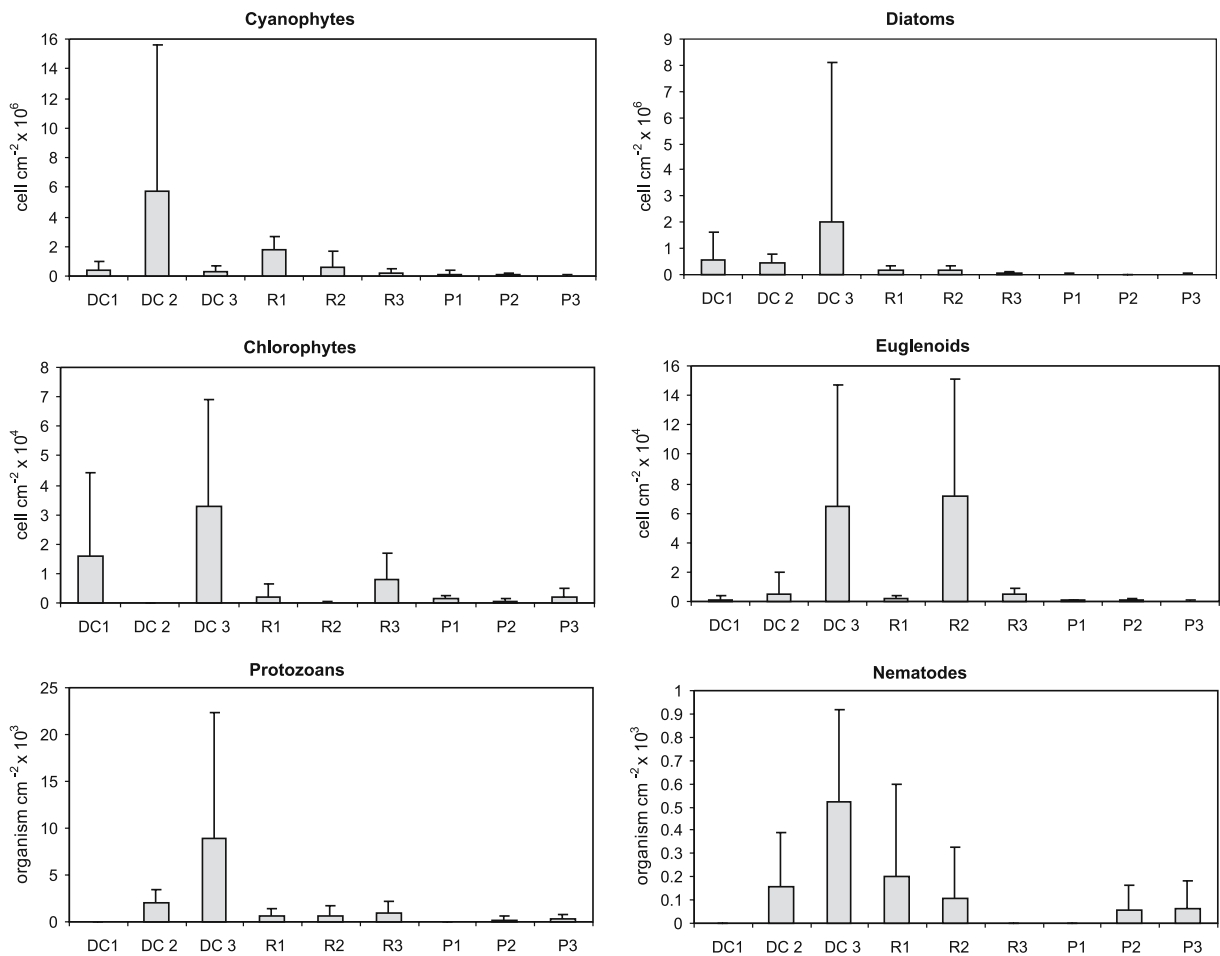


Fig. 3 Mean (\pm SD) abundance of organisms at each sampling site

Because both Chl *a* and AFDW values were the highest at sites with major impact trophic index, which is the ratio of the two, followed the same trend. Trophic index revealed that the streams with strong land use had a lesser dominance of the heterotrophic and/or detritic component, in favour of the autotrophic component of the biofilm. In the Don Carlos and Rodriguez streams, the TI values were between 3 and 8 times higher than in the El Pescado stream. Crossey and La Point (1988) argued that the Trophic index would probably be a better indicator in cases where a pollutant caused a shift from an autotrophic to a heterotrophic community, such as sewage discharge. The lack of agreement with these authors may be due to the fact that our study

was carried out in streams subjected to other kinds of pollution, which propitiate the development of the autotrophic organisms.

According to Bunn et al. (1999), benthic community respiration increases with increasing disturbance. They reported maximum respiration values of $1,550 \text{ mg C m}^{-2} \text{ day}^{-1}$ ($= 4.13 \text{ mg O}_2 \text{ m}^{-2} \text{ day}^{-1}$) at sites of the Mary River in Australia with varying amounts of grazing land-use. The O_2 consumption values estimated in our study were higher than those cited by these authors and by Fellows et al. (2006), for streams of the Moreton region in Australia ($2,340 \text{ mg C m}^{-2} \text{ day}^{-1} = 6.24 \text{ g O}_2 \text{ m}^{-2} \text{ day}^{-1}$) which had been heavily disturbed. On the other hand, our results were similar to those cited by Wiley et al. (1990) who reported a

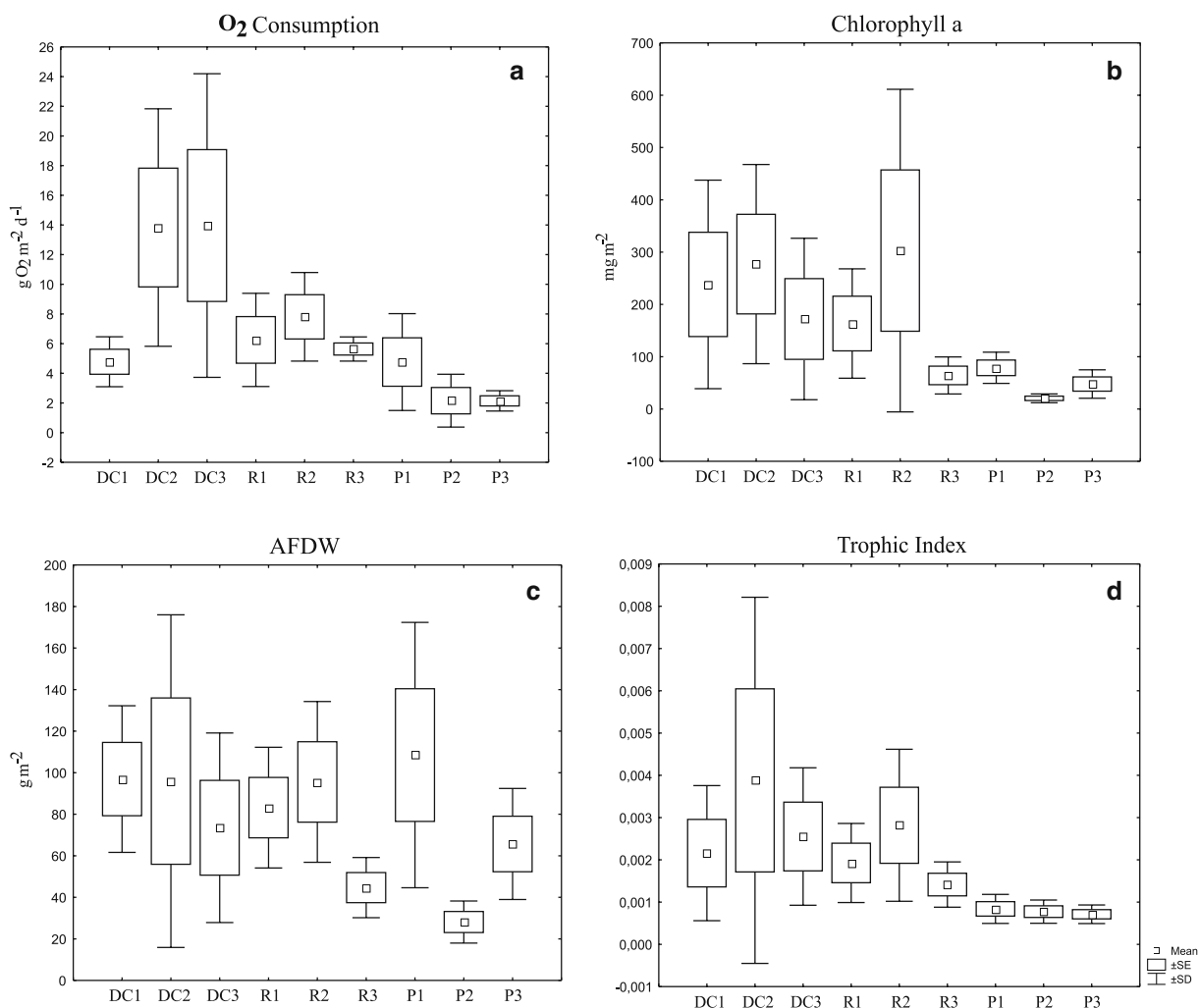


Fig. 4 Box and whisker plots of the biological variables studied, mean (small squares), SE (boxes), SD (bars)

respiration rate of $16.6 \text{ g O}_2 \text{ m}^{-2} \text{ day}^{-1}$ for a first order grassland stream in Illinois.

Few researches have looked at chemical variables in relation to community respiration (Hill et al. 2000). While some studies found a strong relationship

between O_2 consumption and nutrients (Bott et al. 1985; Fuss and Smock 1996), results of the regression analyses in our study showed that O_2 consumption was significantly associated with organic matter but not with nutrients.

Table 4 Results of the multiple regression analysis for the biological variables measured versus oxygen demands and nutrients

	BOD ₅ , COD				PO ₄ ³⁻ -P, NO ₃ ⁻ -N, NO ₂ ⁻ -N, NH ₄ ⁺ -N			
	Multiple R	R ²	P	F	Multiple R	R ²	P	F
O ₂ Consumption	0.59	0.35	0.0007	9.10	0.35	0.12	0.35	1.15
AFDW	0.21	0.046	0.45	0.80	0.16	0.026	0.93	0.209
Chlorophyll a	0.56	0.31	0.001	7.71	0.25	0.06	0.72	0.51
Trophic index	0.45	0.21	0.02	4.40	0.33	0.11	0.43	0.98

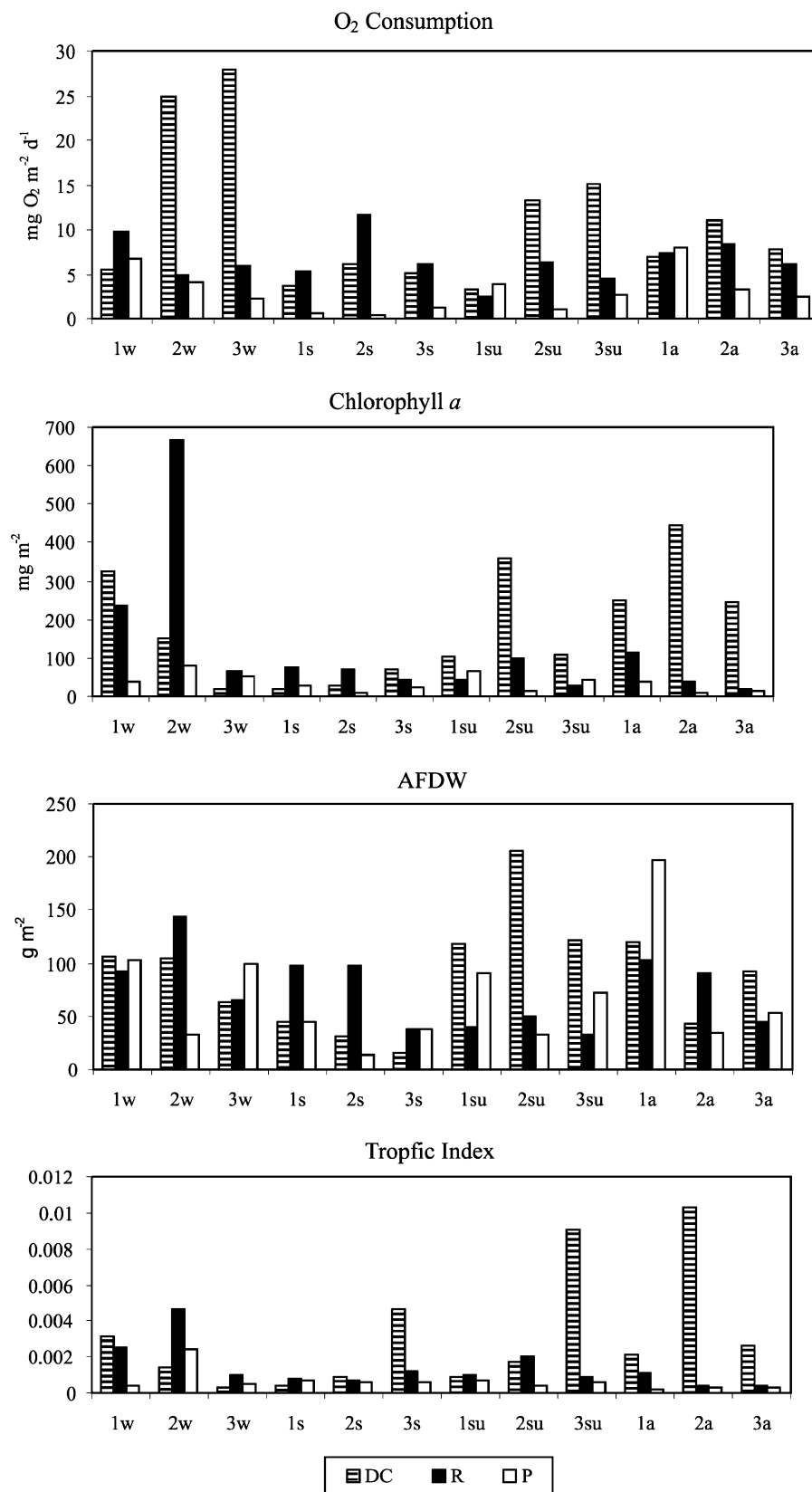


Fig. 5 Spatial and temporal changes in O₂ consumption, chlorophyll *a*, AFDW and trophic index

5 Conclusions

The results presented here demonstrated that the different biological descriptors studied respond to environmental variables that are influenced by human activities. The streams with moderate to high land use showed high contents of nutrient and organic matter and those features were clearly mirrored by high organism densities, chlorophyll *a* amount, and O₂ consumption; therefore these were the variables most sensitive to the changes of water quality.

Given that it is widely recognized that human are simplifying the physical structure of streams and rivers in ways that this may weaken their ability to perform their vital ecological functions (Cardinale et al. 2002), much additional researches are needed to understand the link between the structural and functional responses of the aquatic systems and the different anthropic disturbances.

Finally, structural and functional parameters should be an integral component of the routine assessment of stream health and can be used to establish baseline values for undisturbed and disturbed systems to be incorporated into monitoring and compliance guidelines.

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