

Effects of fish farm effluents on the periphyton of an Andean stream

Verónica Díaz Villanueva; Claudia Queimaliños; Beatriz Modenutti; Julia Ayala

Laboratorio de Limnología, Centro Regional Universitario Bariloche, Universidad Nacional del Comahue, Argentina

Abstract

In this study we analyse the structure and composition of the periphyton, upstream and downstream of a fish farm located at an Andean stream. The effluents discharge large quantities of phosphorus and organic matter from the feed remnants and the faeces. The input of organic matter caused a reduction in algal biomass and a change in the relative abundance of some species. The ratio between chlorophyll *a* and total organic matter concentrations indicated that the periphyton consisted of a high photosynthetic fraction at upstream sites, while downstream heterotrophic conditions were favoured. The community, which was composed mainly of diatoms, was dominated by *Achnanthes minutissima* at every site, except at the mouth of an effluent channel of the fish farm, where *Nitzschia supralitorea* dominated. When there are high organic matter inputs, the prostrate motile form acquires advantages over the rosette, adnate and filamentous forms, with a consequent change in community structure.

Zusammenfassung

Auswirkungen von Fischfarmabwässern auf das Periphyton eines Andenflusses

In dieser Studie wird die Struktur und Artenzusammensetzung des Periphytons eines Andenflusses oberhalb und unterhalb einer Forellen-Farm untersucht. Mit dem Wasser aus der Fischfarm gelangten hohe Konzentrationen an Phosphorverbindungen und organischer Substanz, die aus den Futterresten und Faeces herstammte, in den Fluß. Der erhöhte Gehalt an organischer Substanz führte zu einem Rückgang der Algenbiomasse und einer Veränderung der relativen Häufigkeit einiger Arten. Das Verhältnis zwischen Chlorophyll *a* und dem Gehalt an organischer Substanz auf den ausgelegten Bewuchsplatten zeigte, daß das Periphyton stromaufwärts einen hohen Anteil an photosynthetischen Arten aufwies, während stromabwärts heterotrophe Lebensbedingungen vorherrschten. Die Algengemeinschaft, die hauptsächlich aus Diatomeen bestand, wurde an jeder Untersuchungsstelle von *Achnanthes minutissima* dominiert mit der Ausnahme eines Abflußkanals, an dessen Mündung *Nitzschia supralitorea* vorherrschte. Bei der Einleitung von hohen Gehalten an organischer Substanz sind daniederliegende, bewegliche Algenarten begünstigt gegenüber solchen mit einer rosettenförmigen, festverwachsenen oder fädigen Wachstumsform. Dementsprechend ändert sich auch die strukturelle Zusammensetzung der Algengemeinschaft.

Resumen

Efectos de los efluentes de una piscicultura sobre el perifiton de un arroyo andino

Se analizó la estructura y composición del perifiton en aguas arriba y abajo de una piscicultura localizada a orillas de un arroyo andino. Los efluentes contribuyeron con grandes cantidades de fósforo y de materia orgánica, provenientes de los restos del alimento de los peces y de sus heces. Este aumento en la materia orgánica causó una reducción de la biomasa algal y un cambio en las abundancias relativas de algunas especies. La relación entre las concentraciones de clorofila *a* y de materia orgánica indicó que la fracción fotosintética del perifiton fue mayor en los sitios aguas arriba de la descarga, mientras que aguas abajo se favorecieron las condiciones heterotróficas. La comunidad, principalmente compuesta por diatomeas, estuvo dominada por *Achnanthes minutissima* en todos los sitios de muestreo, menos en la boca del drenaje de la piscicultura, donde dominó *Nitzschia supralitorrea*. Cuando hay un ingreso alto de materia orgánica, la forma postrada y móvil de esta especie, adquiere ventajas frente a las formas en roseta, adnadas y filamentosas, con el consecuente cambio en la estructura comunitaria.

Introduction

The Andean sector of the Patagonian region of Argentina has numerous mountain streams. These rithronic lotic environments are characterised by high levels, high transparency, and low temperatures (Modenutti *et al.* 1998); in addition they are poor in nutrients and have neutral to slightly acid pH values (Pedrozo *et al.* 1993). As these characteristics are suitable for salmonids (Billard 1990), these exotic fishes were repeatedly introduced to Patagonia since the early 1900s. During the last decades, commercial farming of rainbow trout has become an important economic activity, with significant capacity for further growth.

The effluents of a trout farm consist of metabolic wastes and uneaten feeds, that are continuously discharged into the receiving stream (Loch *et al.* 1996). Fish feeds are rich in phosphorus, and the effects of stream enrichment through nutrient inputs thus directly affect the primary producers (Borchardt 1996).

The periphyton is responsible for most of the primary production that occurs within a lotic system (McIntire 1973). Its biomass is controlled by various factors, such as nutrients, light, current velocity, **grazing intensity**, siltation and substratum type (McIntire 1973; Stevenson 1984; Kutka and Richards 1996). In Andean rithronic environments, epilithic communities are mainly composed of diatoms, which represent the most important primary producers (Gaglioti 1992, 1995). Current velocity and light exposure are regarded as factors controlling the community (Gaglioti 1992, 1995). This control of algal growth can be drastically modified when a trout farm is built at a stream and releases its wastes into it. One of the consequences of this phosphorus enrichment is an increase in periphytic biomass (Marcus 1980; Giorgi 1995; McCormick and O'Dell 1996).

In the Andean Patagonian region, the effects of trout farm wastes released into the system have only been tested for cage culture at reservoirs (Baffico and Pedrozo 1996). Up to now, there is no published information for this region on the effects of a raceway trout farm on stream communities.

In this contribution we analyse the effects of a trout farm on the periphyton of an Andean stream. Diatoms, the dominant group in the epilithic communities, are commonly used as quantitative indicators of environmental conditions (Kutka and Richards

1996; McCormick *et al.* 1996; Pan *et al.* 1996). The major purpose of our study was to determine the structure and composition of epilithic diatom communities upstream and downstream of a trout farm.

Study area

The study was conducted on Gutiérrez stream (41° 07' S; 71° 25' W). This Andean stream is located in North Patagonia and flows along 6 km from Lake Gutiérrez into Lake Nahuel Huapi (Figure 1). It is regulated by three dams, of which the last one is used to provide water to a fish farm by a channel that feeds a series of tanks where mostly rainbow trout (*Oncorhynchus mykiss* Walbaum) are grown (approximately 11 tons of fresh biomass per year). The water is returned to the stream through five effluent channels (Figure 1). The distance from the extraction point to the last effluent channel is 200 m. This water carries fish wastes and feed remnants. The fishes are fed daily with 132 kg of feed pellets, which contain 1.8 % phosphorus. The fish ponds are cleaned every ten days, each one on a different day.

The canopy of the stream has drastically changed since autochthonous trees were replaced by exotic species. The hydrological regime is pluvio-nival, with high levels in November and low levels in April. The stream averaged a width of 6 m and a depth of 30 cm during the study, which was conducted in late summer and early autumn.

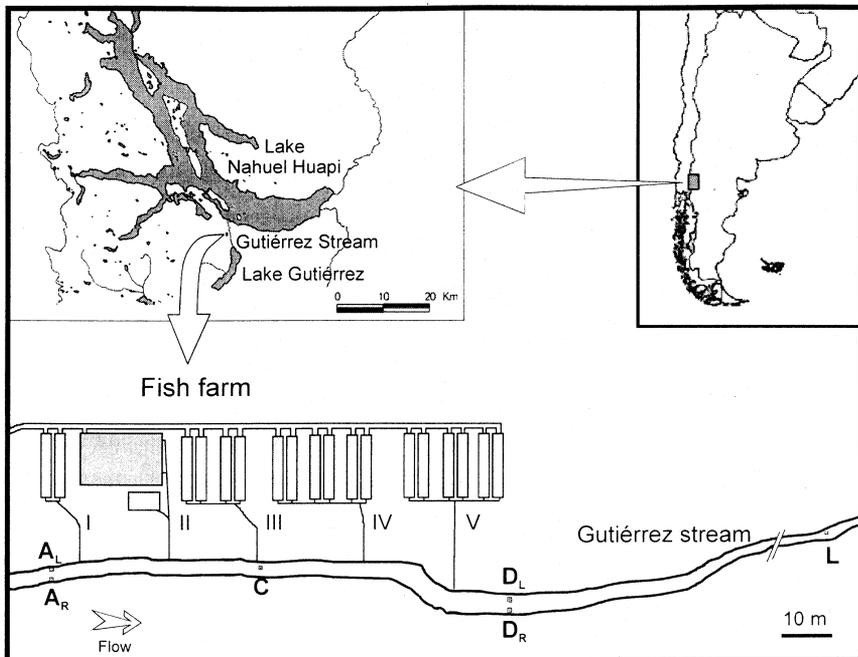


Figure 1: Location of Gutiérrez stream and layout of the fish farm. A_L, A_R, C, D_L, D_R and L represent the sampling sites; I-V represent the effluent channels.

Material and methods

The experiments were carried out in February and March 1998. In experiment 1 (February 1998), the periphyton community upstream was compared to that of an effluent channel of the fish farm. Experiment 2 (March 1998), which was more extensive, was designed to evaluate the impact of the fish farm on different sites downstream.

Unglazed ceramic tiles (8 cm × 8 cm) were placed into the stream as artificial substrates for periphyton colonisation and growth. Each tile was considered one sampling unit. In the first experiment (February 1998), a set of 6 replicates tiles was placed 5 m upstream of the first effluent channel (site A_L, Figure 1) and another set of tiles was placed at the mouth of the third effluent channel (site C, Figure 1). In the second experiment (March 1998), the four sets of tiles were placed at four sites located at different positions relative to the waste discharge. Two sets of 15 tiles were arranged 5 m upstream of the first effluent channel (A_L at the left bank and A_R at the right bank, Figure 1). Another set of 15 tiles was placed at the mouth of the third waste drain channel (C, Figure 1), and the other two sets 10 m downstream of the last waste drain channel (D_L at the left bank and D_R at the right bank, Figure 1). Finally the last set (L) was located 100 m downstream of the same point (Figure 1).

The first set of artificial substrates was placed in the stream (experiment 1) on February 17, 1998. On February 24 and March 2, three replicates were removed and brought immediately to the laboratory, each one in an individual plastic container, in darkness and thermally isolated. In experiment 2, the artificial substrates were deployed on March 2 1998. Five replicates from each set of 15 were removed on March 9, March 16 and April 1, and carried to the laboratory in the manner described.

Temperature, conductivity, dissolved oxygen concentration and current velocities were measured on each sampling date and at each site with a thermistor, conductimeter, oxymeter and flowmeter, respectively. During experiment 2, samples of 250 ml of water were taken at each experimental site in order to determine soluble reactive phosphorus (SRP), total dissolved phosphorus (TDP) and total phosphorus (TP) concentrations.

SRP and TDP concentrations were determined in the laboratory, from subsamples filtered through Whatman GF/C filters. To measure TP and TDP, subsamples were digested with potassium persulphate at 125 °C and 1.5 atm for 1 h. SRP, TP and TDP concentrations were determined by the ascorbate-reduced molybdenum blue method (APHA 1989).

Periphyton was obtained by scraping each tile with a razor blade and rinsed with distilled water. The samples were carefully homogenised and fractionated into three subsamples of 20 to 30 ml each in order to determine chlorophyll *a* (Chl *a*) concentration, ash-free dry mass (AFDM) and species composition. The concentrations were calculated per square centimetre assuming that the entire substratum was divided into three equal parts during the scraping procedure and subsequent fractionation.

Chlorophyll *a* subsamples were filtered through Whatman GF/C filters and extracted with hot 90 % ethanol, following Nusch (1980). Afterwards, spectrophotometric readings were carried out at 665 nm and 750 nm. Corrections for phaeophytin were performed by acidification with HCl. AFDM subsamples were filtered onto precombusted and preweighed Whatman GF/C filters and dried at 80 °C for 1.5 h. The filters were weighed and combusted at 550 °C for 1 h, then re-weighed, and AFDM was computed as the difference in mass before and after incineration (APHA 1989).

The subsample obtained for the analysis of algal assemblage was preserved in 4 % formalin. 10 or 20 ml were taken and three aliquots were examined in a chamber of 18 μl volume under a direct microscope at 400 \times magnification. Another 10 ml of the subsample were treated with hydrogen peroxide to oxidize the organic matter and identify, count and measure the diatoms. The slides were mounted in Naphrax[®] and examined under a direct microscope at 1000 \times magnification. A minimum of 300 valves was counted in each sample. Identifications were performed according to Krammer and Lange-Bertalot (1986, 1988, 1991). Following identification, the total density (total number of cells per square centimetre), species richness and species relative abundance (percent density of individual species per total density) were calculated.

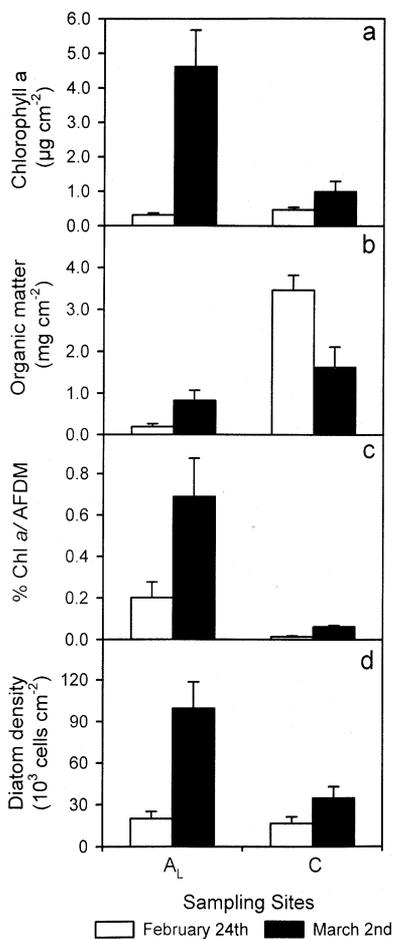


Figure 2: Responses of periphytic variables during experiment 1 (February 1998). Error bars indicate standard errors. Sampling sites as in Figure 1.

The relationship between chlorophyll *a* and organic matter (AFDM) concentrations was calculated to estimate the relative importance of the photosynthetic fraction in the community (Weber and McFarland 1969). Statistical differences between treatments were assessed using the Kruskal Wallis and t tests.

Results

During experiment 1 (February 1998), the water temperature was 18 °C, the conductivity was 60 $\mu\text{S}\cdot\text{cm}^{-1}$ and the dissolved oxygen was 12 $\text{mg}\cdot\text{l}^{-1}$ at both sites. The current velocity was 73 $\text{cm}\cdot\text{s}^{-1}$ at the upstream site (A_L) and 69 $\text{cm}\cdot\text{s}^{-1}$ at the effluent channel (C).

The chlorophyll *a* concentration was lower at the effluent channel than upstream (Figure 2a). This result was observed after two weeks of colonisation, and the statistical difference between the upstream site and the effluent channel was significant (t test, $P < 0.05$). With regard to the variation in time, there was a significant increase in the chlorophyll concentration at the upstream site between the first and the second week (t test, $P < 0.05$), whereas at the effluent site the increase in chlorophyll was not significant (t test, $P > 0.05$) (Figure 2a). The accumulation of organic matter on the other hand was significantly higher (t test, $P < 0.05$) at the effluent channel than at the upstream site after the first week, but the difference was not significant after the second week (t test, $P > 0.05$) (Figure 2b).

The ratio between chlorophyll *a* concentration and total organic matter indicates the photosynthetic fraction of the organic matter (Weber and McFarland 1969). This relationship was always consider-

ably higher at the upstream site than at the effluent site (Figure 2c). This indicates that heterotrophic conditions were favoured at the site of the discharge.

The periphyton was mainly composed of diatoms, which were more abundant on the tiles upstream after the second week of colonisation (Figure 2d). There was a noticeable change in the relative abundance of some diatom species (Figure 3). *Achnanthes minutissima* Kütz. was the dominant species at the upstream site, both after one and two weeks of colonisation. *Nitzschia supralitorea* Lange-Bertalot, which was absent upstream, was co-dominant together with *A. minutissima* at the effluent channel. Other species such as *Cocconeis placentula* Ehr. and *Fragilaria capucina* Desm. were frequent (> 5 %) but not much affected by the discharge, since the differences between sites were not significant (t-test, $P > 0.05$). *Melosira varians* Ag. and *Gomphonema angustatum* (Kütz.) Rabh. were scarce, but their densities were affected by the fish farm discharge, negatively in the former and positively in the latter (Figure 3).

The different types of growth forms were analysed, because they may be affected by environmental factors. The prostrate habit was more important at the effluent channel (site C, Figure 4), not only because of the dominance of *N. supralitorea*, but also because of other species with low relative abundance, such as *Navicula cryptocephala* Kütz. (4 %) and *Nitzschia inconspicua* Grun. (3 %), which were absent from the upstream site. Rosette-forming diatoms, such as the dominant *Achnanthes minutissima*, and other species such as *Hannaea arcus* (Ehr.) Patr. and *Synedra ulna* (Nitz.) Ehr., were more abundant at the upstream site (site A_L, Figure 4). The adnate habit became more frequent at the effluent site after two weeks of colonisation. The arborescent and filamentous growth forms did not differ between the sites or exposure periods (Figure 4).

During experiment 2 (March, 1998), the temperature varied between 13 °C and 11 °C. Conductivity and dissolved oxygen concentrations remained almost constant throughout the experimental period at all stream sites; conductivity was 60 $\mu\text{S}\cdot\text{cm}^{-1}$ and dissolved oxygen ranged between 11 and 12 $\text{mg}\cdot\text{l}^{-1}$. Current velocity was lower at site A_R on all sampling dates (15 to 73 $\text{cm}\cdot\text{s}^{-1}$), higher at sites C (29 to 164 $\text{cm}\cdot\text{s}^{-1}$) and L (102 to 108 $\text{cm}\cdot\text{s}^{-1}$) and intermediate at sites A_L (19 to 97 $\text{cm}\cdot\text{s}^{-1}$), D_L (38 to 78 $\text{cm}\cdot\text{s}^{-1}$) and D_R (54 to 98 $\text{cm}\cdot\text{s}^{-1}$). On the last date of the study the current velocity decreased strongly at sites A_L and C, but remained constant at L, and decreased slightly at D_L and D_R.

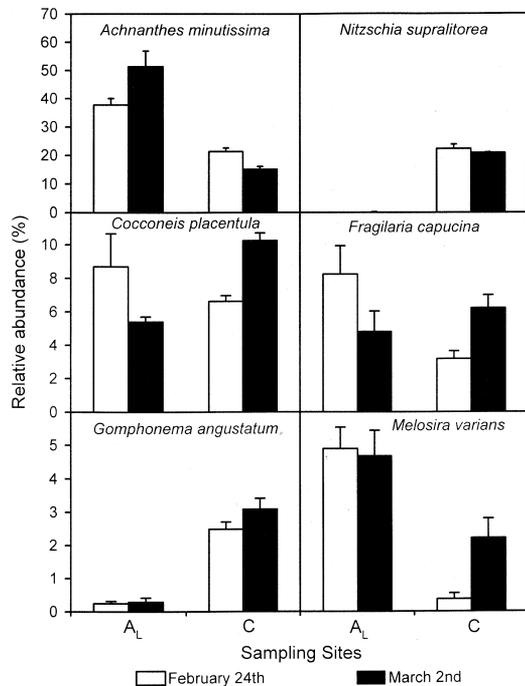


Figure 3: Relative abundance of the most abundant diatom species during experiment 1 as in Figure 1.

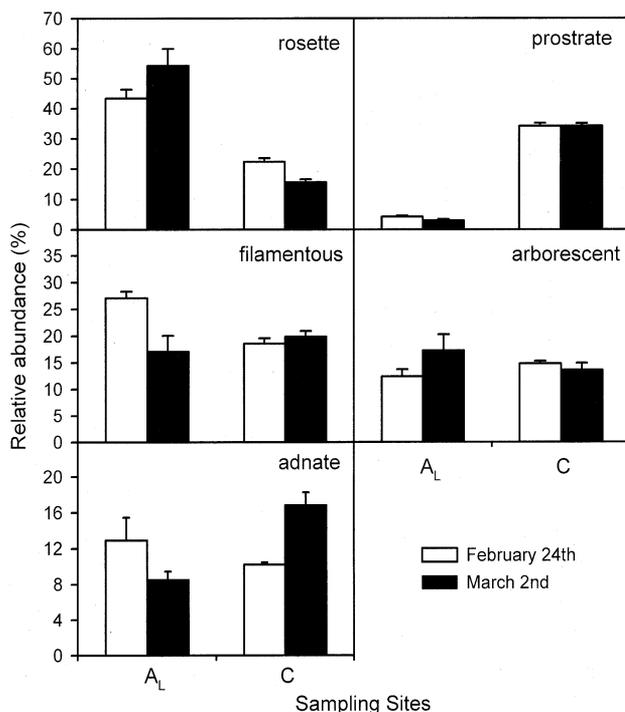


Figure 4: Relative abundance of the different diatom growth forms during experiment 1 in Figure 1.

The levels of the three phosphorus fractions were always lower at the sites upstream of the discharge channels (sites A_L and A_R) than downstream (sites C, D_L, D_R and L) (Figure 5). However, at the downstream sites there was no clear spatial pattern, with concentration peaks on different sampling dates. On the first sampling date (March 2), the highest values were recorded at D_L (Figure 5). On March 9, the highest TP concentration was determined at site D_R, while on the next sampling date it was recorded at site C (Figure 5). On the last sampling date, the phosphorus values were similar at all sites downstream of the fish farm (Figure 5). This pattern can be explained by the nature of the phosphorus enrichment, *i. e.* the pulses of feeding and of cleaning of the fish tanks.

Chlorophyll *a* concentration was always higher at A_L and A_R than at the downstream sites (Figure 6a) (K-W, $P < 0.05$). Colonisation proceeded more rapidly at A_R during the first week, but after the second week, the periphyton at site A_L overgrew during all the other samples (Figure 6a). The accumulation of organic matter (AFDM) was almost equal at all sites after the first week (Figure 6b). After two weeks, sites A_L and A_R exhibited the highest values, and the levels gradually decreased downstream. On the fourth week (April 1), the values of AFDM were highest at site C (K-W, $P < 0.05$).

The ratio between chlorophyll *a* concentration and total organic matter was almost always higher than 0.5 % at sites A_L and A_R, reaching a maximum of 1.0 % at A_R in the first week of colonisation, and at A_L in the second one (Figure 6c). It was always less than 0.1 % at the sites downstream of the discharges, except at site L on the second sampling date (ratio = 0.2 %) (Figure 6c). This indicates that the periphyton at the upstream sites contained the highest proportion of photosynthetic organic matter.

Diatoms comprised over 85 % of the total periphyton cells on all of the artificial substrates. Seventy-four diatom species were identified. Species richness on the substrates ranged from 12 to 44, but there was no significant difference in the number of species between exposure periods or between sites (K-W, $P > 0.05$). After the first week of colonisation (March 9), the periphyton had developed only on the tiles at the sites upstream of the effluent channels (Figure 6d). On March 16 and April 1 the tiles downstream of the

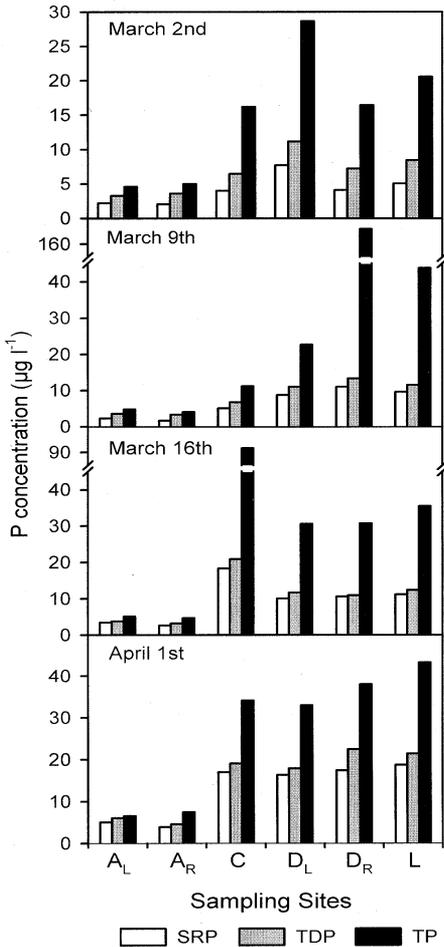


Figure 5: Phosphorus concentration as soluble reactive phosphorus (SRP), total dissolved phosphorus (TDP) and total phosphorus (TP) during experiment 2 as in Figure 1.

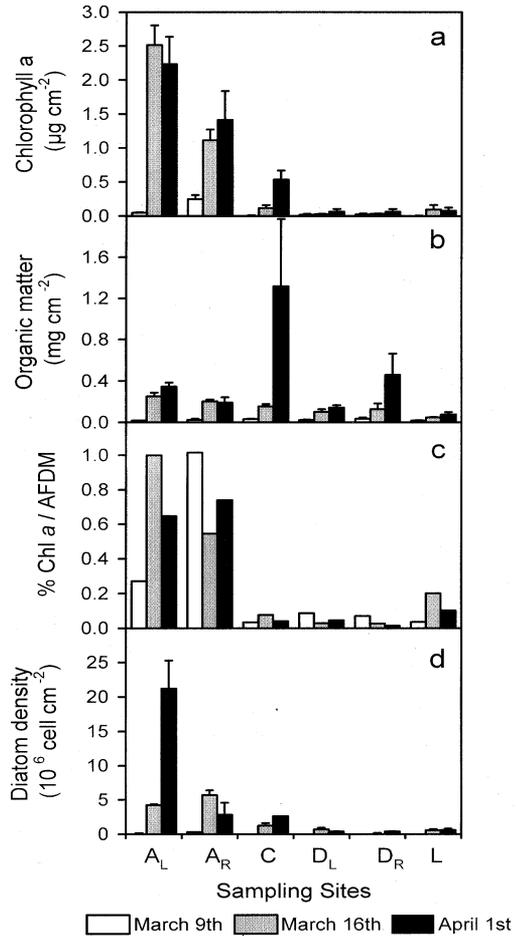


Figure 6: Responses of periphytic variables during experiment 2 (March 1998). Error bars indicate standard errors as in Figure 1.

discharges had low densities of diatom cells. At site A_L the number of diatom cells increased on the last sampling date (April 1) (Figure 6d).

On March 9, colonisation was only recorded at the upstream sites (A_L and A_R, Figure 7). The most abundant species were *Hannaea arcus*, *Cymbella silesiaca* Bleish (not included in Fig. 7), *Synedra ulna* and *Achnanthes minutissima* at site A_L, and *Fragilaria pinnata* Ehr., *Cocconeis placentula* and *A. minutissima* at site A_R. After two weeks of colonisation, *Melosira varians*, *A. minutissima*, *C. placentula* and *Fragilaria capucina* were present with more than 10 % relative abundance at sites A_L and A_R. At site C, *A. minutissima* decreased, *M. varians* almost disappeared and *C. placentula* became more abundant. At the downstream sites (D_L, D_R and L), *A. minutissima* co-dominated with *C. placentula*

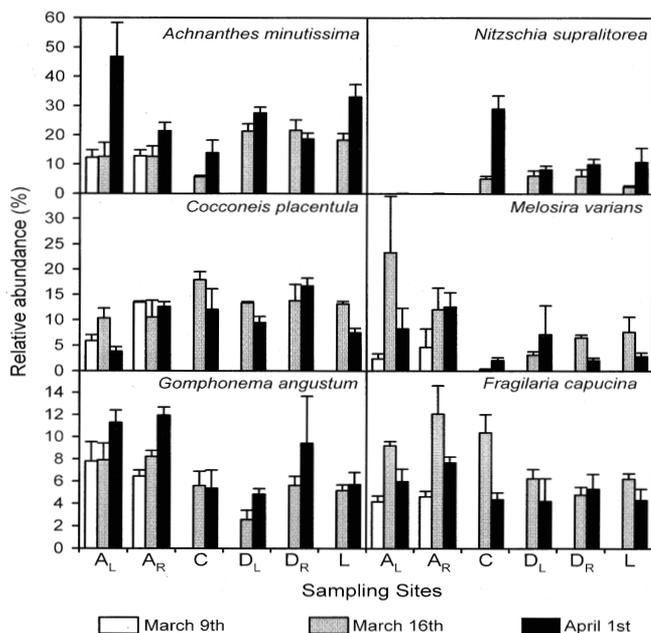


Figure 7: Relative abundance of the most abundant diatom species during experiment 2 as in Figure 1.

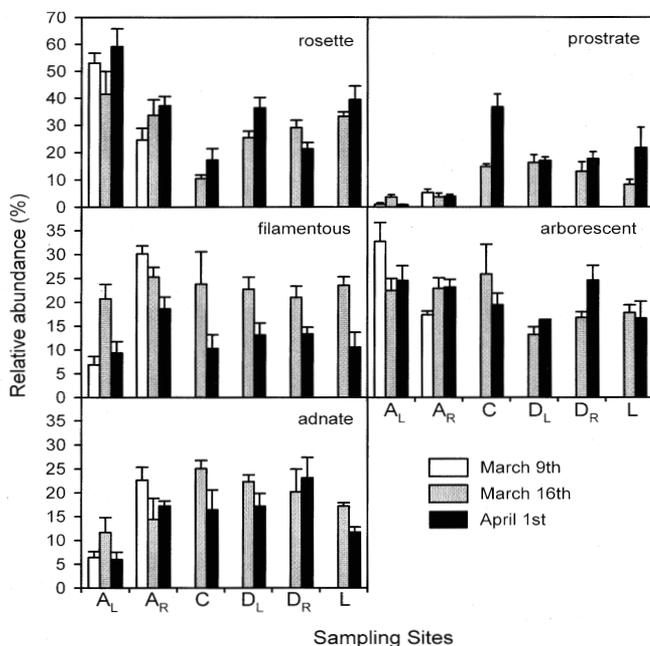


Figure 8: Relative abundance of the different diatom growth forms during experiment 2 as in Figure 1.

and *F. capucina*, and *M. varians* reappeared. It is important to note that *Nitzschia supralitorea* was present with almost 10 % relative abundance at the mouth of the effluent channel and the sites downstream. On the last sampling date (April 1), *A. minutissima* became the dominant species at all sites except site C, where it was abundant, but overgrown by *N. supralitorea*. This species was also abundant at sites D_L, D_R and L, but absent at sites A_L and A_R.

As regards the different growth forms, the rosette-forming species were dominant because of the high density of *Achmanthes minutissima*. This form decreased downstream (Figure 8). All prostrate species, however, such as *Nitzschia supralitorea* and *Navicula cryptocephala*, increased downstream of the discharge (Figure 8). The other habits were less affected by the discharge (Figure 8).

Discussion

The present study showed that the colonisation rate of the artificial substrates is lower as an effect of the discharges from the fish farm at Gutiérrez stream, although phosphorus measurements indicated that the

effluent water is indeed enriched by phosphorus. Mulholland and Rosemond (1992) also failed to find a corresponding pattern in periphyton biomass in relation to depletion of SRP concentration, but they found effects on species composition.

Not all of the phosphorus entering the stream plays an equal role in the biogeochemical cycles of the ecosystem. Most of the input released during periods of high discharge may be flushed away without entering biological or geochemical pathways (Meyer and Likens 1979). According to Newbold *et al.* (1983) periphyton accounted only for 5 % of the total uptake of phosphorus. If the phosphorus released by the fish farm is not absorbed and utilised by the algae, then it will be available downstream, as is the case in Gutiérrez stream, where the phosphorus concentration (SRP, TDP and TP) was still high 100 m downstream at site L.

The fact that the periphytic biomass was reduced downstream of the fish farm despite the phosphorus enrichment may be attributed to the high input of organic matter from the fish farm. Loch *et al.* (1996) noted that long-term waste accumulation may result in a blanketing of the stream bed and that this deposit supports a diverse community of microscopic organisms that utilise oxygen, creating a habitat deleterious to other aquatic organisms. Weber and McFarland (1969) suggested that when chlorophyll *a* constitutes less than 1 % of the natural accumulation of periphytic organic matter, heterotrophic organisms comprise a large portion of the periphyton, or algal chlorophyll production is limited by environmental factors. In this study, the Chl *a*/AFDM ratio was very low at the downstream sites (< 0.1 %), while it was always higher at the sites upstream (Figures 2c and 6c). Therefore, the periphytic community downstream was more heterotrophic. Moreover, an elevated number of ciliates was observed on the substrates at the mouth of the effluent channel, probably indicating high densities of bacteria.

The aggregation of organic matter may have produced a shading effect upon the periphytic algae, limiting their development to the surface of the organic matter deposit. This shading effect was produced only by the organic matter deposited on the substrate since no change in water turbidity was observed. Pringle (1990) has stated that light may also become a limiting factor when biomass increases with fertilisation.

As regards the change in periphytic composition, it is assumed that when several species compete for the same resource, a competitive displacement occurs due to the depletion of resource availability (Tilman 1982). The capability of nutrient uptake differs between diatom species, and the physiological and morphological characteristics of the different species of benthic diatoms respond to natural selection caused by competition for resource (Stevenson *et al.* 1991). The rise of *Nitzschia supralitorea* as the dominant species at the waste discharge would support the hypothesis that motile taxa have a competitive advantage when biomass development is limited by light. Members of the genus *Nitzschia* generally reach their greatest abundances in eutrophic waters (Van Dam *et al.* 1994) and also show the most pronounced heterotrophic capabilities among diatoms (Hellebust and Lewin 1977). *Nitzschia supralitorea*, in particular, is characterized by requiring elevated concentrations of organic compounds and facultative heterotrophy (Van Dam *et al.* 1994).

Another species affected by the discharge was *Achnanthes minutissima*, which is an entirely autotrophic species with high requirements of dissolved oxygen (Van Dam *et al.* 1994). Pringle (1990) pointed out that the abundance of this species decreases in en-

riched environments due to the continuous skin-like layer of *Navicula pelliculosa* (Bréb. ex Kütz.) Hilse, which inhibits turbulent diffusion of nutrients and oxygen into lower layers, allowing potentially toxic metabolic end products to accumulate. In the present case, the cells of *Nitzschia supralitorea* could play the same role as *Navicula pelliculosa*, and even organic matter layers could negatively affect the colonisation of *Achnanthes minutissima* in the same way. Thus, the accumulation of organic matter could represent an advantage for motile cells capable of climbing to the surface. The increase of all prostrate motile species, such as *Nitzschia supralitorea* and *Navicula cryptocephala*, supports this hypothesis.

The effluents from the fish farm contained not only phosphorus, but a high load of organic matter as well. Therefore, heterotrophic conditions increased downstream of the discharge as shown by the reduced Chl *a*/AFDM ratio. The high quantities of organic matter caused a reduction of the algal biomass by light limitation so that the phosphorus could not be retained by the periphyton and was transported downstream by the water flow.

References

- American Public Health Association, 1989: Standard methods for the examination of water, sewage, and wastewater. Washington D.C.: American Public Health Association. 715 pp.
- Baffico, G. D.; Pedrozo, F. L., 1996: Growth factors controlling periphyton production in a temperate reservoir in Patagonia used for fish farming. *Lakes & Reservoirs, Res. Manag.* 2: 243–249.
- Billard, R., 1990: Culture of salmonids in fresh water. In: Barnabé, G. (ed.): *Aquaculture*. Vol. 2. Chichester: Ellis Horwood, p. 549–592.
- Borchardt, M. A., 1996: Nutrients. In: Stevenson, R. J.; Bothwell, M. L.; Lowe, R. L. (eds.): *Algal Ecology. Freshwater Benthic Ecosystems*. New York: Academic Press, p. 184–227.
- Gaglioti, P. V., 1992: Variación espacial y estacional en la estructura de las comunidades de diatomeas epífitas de un arroyo andino. Su relación con factores abióticos. *Ecología Austral* 2: 77–86.
- Gaglioti, P. V., 1995: Secuencia de colonización de diatomeas adheridas sobre sustratos artificiales en un arroyo andino. *Medio Ambiente* 12: 67–75.
- Giorgi, A. D. N., 1995: Response of periphyton biomass to high phosphorus concentration in laboratory experiments. *Bull. Envir. Contam. Toxicol.* 55: 825–832.
- Hellebust, J. A.; Lewin, J., 1977: Heterotrophic nutrition. In: Werner, D. (ed.): *The biology of diatoms*. London: Blackwell Scientific, p. 169–197.
- Krammer, K.; Lange-Bertalot, H., 1986: Bacillariophyceae, 1. In: Ettl, H.; Gerloff, J.; Heynig, H.; Mollenhauer, D. (eds): *Süßwasserflora von Mitteleuropa*. Jena: Fischer Verlag G., 876 pp.
- Krammer, K.; Lange-Bertalot, H., 1988: Bacillariophyceae, 2. In: Ettl, H.; Gerloff, J.; Heynig, H.; Mollenhauer, D. (eds.): *Süßwasserflora von Mitteleuropa*. Jena: Fischer Verlag G., 596 pp.
- Krammer, K.; Lange-Bertalot, H., 1991: Bacillariophyceae, 3. In: Ettl, H.; Gerloff, J.; Heynig, H.; Mollenhauer, D. (eds): *Süßwasserflora von Mitteleuropa*. Jena: Fischer Verlag G., 574 pp.
- Kutka, F. J.; Richards, C., 1996: Relating diatom assemblage structure to stream habitat quality. *J. N. Am. Benthol. Soc.* 15 (4): 469–480.
- Loch, D. D.; West, J. L.; Perlmutter, D. G., 1996: The effect of trout farm effluent on the tax richness of benthic macroinvertebrates. *Aquaculture* 147: 37–55.
- Marcus, M. D., 1980: Periphytic community response to chronic nutrient enrichment by a reservoir discharge. *Ecology* 61: 387–399.
- McCormick, P. V.; O'Dell, M. B., 1996: Quantifying periphyton responses to phosphorus in the Florida Everglades: a synoptic-experimental approach. *J. N. Am. Benthol. Soc.* 15: 450–468.

- McCormick, P. V.; Rawlik, P. S.; Lurding, K.; Smith, E. P.; Sklar, F. H., 1996: Periphyton-water quality relationship along a nutrient gradient in the northern Florida Everglades. *J. N. Am. Benthol. Soc.* 15: 433–449.
- McIntire, C. D., 1973: Periphyton dynamics in laboratory streams: a simulation model and its implications. *Ecology* 43: 399–420.
- Meyer, J. L.; Likens, G. E., 1979: Transport and transformation of phosphorus in a forest stream ecosystem. *Ecology* 60: 1255–1269.
- Modenutti, B. E.; Balseiro, E.G.; Diéguez, M.C.; Queimaliños, C.P.; Albariño, R.J., 1998: Heterogeneity of fresh-water Patagonian ecosystems. *Ecología Austral* 8: 155–165.
- Mulholland, P. J.; Rosemond, A. D., 1992: Periphyton response to longitudinal nutrient depletion in a woodland stream: evidence of upstream-downstream linkage. *J. N. Am. Benthol. Soc.* 11: 405–419.
- Newbold, J. D.; Elwood, J. W.; O'Neill, R. V.; Sheldon, A. L., 1983: Phosphorus dynamics in a woodland stream ecosystem: a study of nutrient spiralling. *Ecology* 64: 1249–1265.
- Nusch, E. A., 1980: Comparison of different methods for chlorophyll and phaeopigment determination. *Arch. Hydrobiol. Beih.* 14: 14–36.
- Pan, Y.; Stevenson, R. J.; Hill, B. H.; Herlihy, A. T.; Collins, G. B., 1996: Using diatoms as indicators of ecological conditions in lotic systems: a regional assessment. *J. N. Am. Benthol. Soc.* 15: 481–495.
- Pedrozo, F.; Chillrud, S.; Temporetti, P.; Díaz, M., 1993: Chemical composition and nutrient limitation in rivers and lakes of northern Patagonian Andes (39.5°–42° S; 71° W) (Rep. Argentina). *Verh. Int. Ver. Limnol.* 25: 205–214.
- Pringle, C. M., 1990: Nutrient spatial heterogeneity: effects on community structure, physiognomy, and diversity of stream algae. *Ecology* 71: 905–920.
- Stevenson, R. J., 1984: Mathematical model of epilithic diatom accumulation. *Diatom-Symposium* p. 323–335.
- Stevenson, R. J.; Peterson, C. G.; Kirschtel, D. B.; King, C. C.; Tuchman, N. C., 1991: Density-dependent growth, ecological strategies, and effects of nutrients and shading on benthic diatom succession in streams. *J. Phycol.* 27: 59–69.
- Tilman, D., 1982: *Resource competition and community structure*. Princeton: University Press. 295 pp.
- Van Dam, H.; Mertens, A.; Sinkeldam, J., 1994: A coded checklist and ecological indicator values of freshwater diatoms from the Netherlands. *Neth. J. Aquat. Ecol.* 28: 117–133.
- Weber, C. I.; McFarland, B. H., 1969: Periphyton biomass-chlorophyll ratio as an index of surface waters and effluents. *Environmental Monitoring Series EPA-670/4-73-001*. Cincinnati, Ohio, USA: United States Environmental Protection Agency, 186 pp.

Acknowledgements

We are very grateful to Téc. Víctor Báez, Director of the Centre of Salmoniculture of the University of Comahue, who kindly permitted to us work at Gutiérrez stream. We also thank Dr. E. Balseiro for his useful comments and his critical suggestions. This work was supported by FONCyT PICT 01-00000-01194, CONICET-PIP 0739/98 and UNC B701.

Address of authors: Verónica Díaz Villanueva; Claudia Queimaliños; Beatriz Modenutti; Julia Ayala, Laboratorio de Limnología, Centro Regional Universitario Bariloche, Unidad Postal Universidad, 8400 - Bariloche, Argentina. Fax: ■, e-mail: ■

Communicated by: W. Arntz, received: 29 October 1999, accepted: 8 September 2000, print proof received from author(s):