To link to this article: DOI:10.1002/rra.675
http://dx.doi.org/10.1002/rra.675

EPILITHIC BIOMASS IN A LARGE GRAVEL-BED RIVER (THE GARONNE, FRANCE): A MANIFESTATION OF EUTROPHICATION?

THIERRY AMEZIANE,* FREDERIC GARABETIAN, DANIEL DALGER, SABINE SAUVAGE, ALAIN DAUTA and JACQUES CAPBLANCQ

Université Paul Sabatier, Toulouse III, Bat IV R 3, Centre d'Ecologie des Systèmes Aquatiques Continentaux (CESAC), UMR C5576, 118 route de Narbonne, 31062 Toulouse Cedex 04, France

ABSTRACT

In order to evaluate the impact of outputs of the city of Toulouse (740 000 inhabitants) on the epilithic communities colonizing pebble banks in the river Garonne, a large gravel-bed river (eighth order), dry mass (DM), ash-free dry mass (AFDM) and chlorophyll-*a* (chl*a*) epilithic biomass per unit area were measured and autotrophic index (AI) (i.e. ratio AFDM/chl*a*) was calculated at four stations. This river is morphologically characterized by a succession of pools and riffles and by highly fluctuating hydraulic conditions. At the four stations studied (223 km apart), the means of AFDM values varied between 17.1 and 31.1 g m^{-2} of colonized surface and the chl*a* concentration varied between 112 and 254 mg m^{-2}. However, there were no significant differences in AFDM per unit area between the parts of the river upstream and downstream of the Toulouse area (Mann–Whitney U-test statistic), nor between the four stations (Kruskal–Wallis test statistic), and the AI did not allow the description of changes in periphyton communities between sampling locations. This study showed that epilithic biomass should be considered as the typical microbial community of the river rather than as a manifestation of eutrophication.

KEY WORDS: periphyton; biofilm; AFDM; chlorophyll; nutrients; trophic status

INTRODUCTION

The river Garonne is the largest river in south west France, sometimes more than 250 m wide in the lowlands. Periphyton in large rivers mostly grows on gravel beds (i.e. epilithic biofilm) and is only found widely distributed over the river bed in the upstream sections (second to fourth order) of these rivers. Downstream, with an increase in depth, turbidity and fine sediments, it is found mainly near the banks. One of the characteristics of the Garonne is the presence of gravel banks colonized by periphyton in channels of up to the seventh order. The Garonne undergoes a severe degradation of water quality downstream of Toulouse (720 000 inhabitants) (Adour-Garonne Water Agency, 1997). The Garonne is characterized by a low concentration of chlorophyll-*a* (chl*a*) measured in the water column (Dauta, 1978a; Améziane and Dauta, 1997; Eulin, 1997). In addition, we have shown that chl*a* concentrations come mainly from diatom benthic drift (Améziane and Dauta, 1997).

The river Garonne is morphologically characterized by a succession of pool and riffle zones from upstream to downstream with variable alternation frequencies. This morphological aspect does not correspond to the fluvial continuum of Vannote *et al.* (1980). Variations of pH and dissolved oxygen around a gravel bar covered by a periphytic biofilm, after Toulouse, show the intensity of metabolic activities associated with the biofilm growing in the very shallow littoral areas; this demonstrates the major role of the functioning of the gravel bars in the river Garonne (Dauta *et al.*, 1999). Algal growth has been used to assist in the stabilization of untreated sewage and for the removal of mineral nutrients, especially nitrogen and phosphorus, from the aquatic environment for a long time (Vymazal, 1988). However, seen from a different angle, could periphyton be a manifestation of eutrophication in the river? The aims of this study are (i) to observe *in situ* the effect

* Correspondence to: Thierry Ameziane, Université Paul Sabatier, Toulouse III, Bat IV R 3, Centre d'Ecologie des Systèmes Aquatiques Continentaux (CESAC), UMR C5576, 118 route de Narbonne, 31062 Toulouse Cedex 04, France. E-mail: amezian@cict.fr

of outputs of a great urban centre (Toulouse, 740 000 inhabitants) on the biomass of biofilms in a sixth-order river by comparing values of dry mass, ash-free dry mass and chlorophyll-*a* at four stations, two of which are within Toulouse city; (ii) to calculate the autotrophic index at these different stations; and (iii) to discuss biofilm as a possible indicator of the trophic status of the river.

STUDY AREA AND SITES

Located in southwest France, the river Garonne (Figure 1a) starts in the Spanish Pyrenees, and after a distance of 647 km, enters the Atlantic Ocean. During the low water period, the river is characterized by shallow water depths (around 1 to 1.5 m), with a wide bed (around 100 to 200 m) and a mean water velocity of 0.5 m s^{-1}. The river Garonne is characterized by two low water periods, in winter and summer, and by sudden and violent floods. The annual mean discharge is around 200 m^3 s^{-1} at Toulouse. In the low water period, discharges may fall to less than 30 m^3 s^{-1} and reach more than 4500 m^3 s^{-1} during exceptional flood peaks. The hydrological regime is nival upstream from Toulouse and pluvio-nival downstream. The four stations studied are Valentine (V) at the upper part of the river Garonne (in a fourth-order river), Pinsaguel (P) and Gagnac (G) upstream and downstream respectively of Toulouse (in a sixth-order river) and Sauveterre Saint-Denis (S) in the alluvial plain (in a seventh-order river) (Figure 1b). Principal hydraulic and geomorphological variables defined by Petts and Bravard (1993) were used to characterize the four stations (Table I). The shear force and the slope characterize the three groups of stations (V, P/G and S) with a width which doubles in the sector from V to Toulouse and doubles again in the sector from Toulouse to S. The mean pebble diameter decreases by about half from upstream to downstream of the river sector studied. The mean current velocity and mean depth do not differ at each station.

From a general perspective, with the variables considered, the two stations within the Toulouse area were hydraulically and geomorphologically of the same group. As on both cross-sections the canopy is widely open, they are only 20 km apart and have a similar mean depth, the importance of the influence of light and

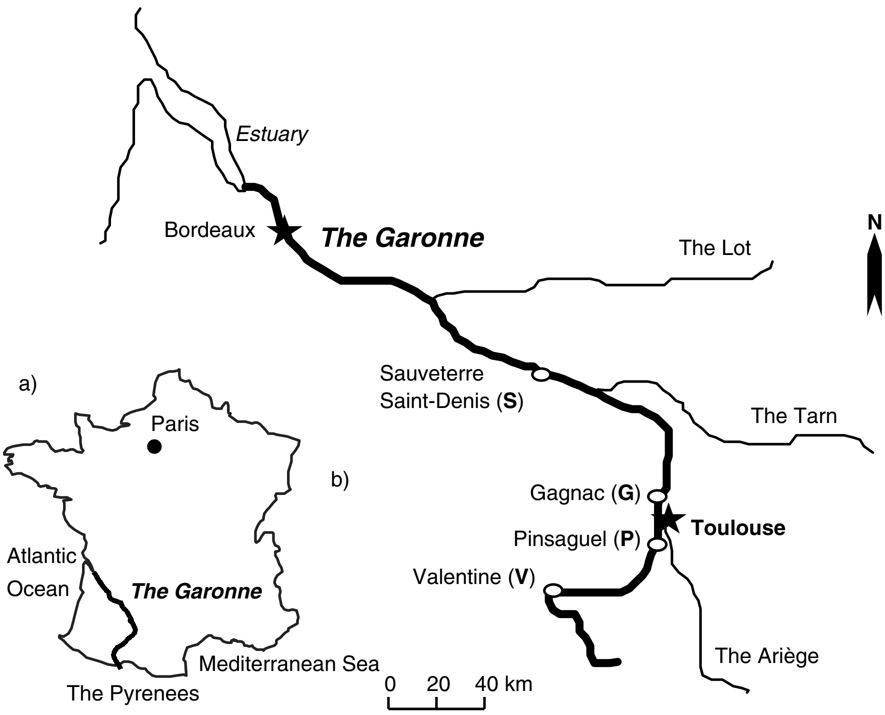


Figure 1. Map of the catchment area of the river Garonne. (a) Study area and (b) location of sampling sites

Table I. Hydraulic and geomorphologic characteristic variables at Valentine (V), Pinsaguel (P), Gagnac (G) and Sauveterre Saint Denis (S) stations

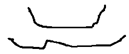
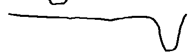
Station	Current velocity (cm s^{-1})	Shear force (cm s^{-1})	Froud number	Slope (‰)	Mean depth (cm)	Distance across (m)	Pebbles mean ϕ (cm)	Bed morphology
V	81	0.158	0.37	5.2	48	61	14	
P	71	0.059	0.33	0.8	47	126	10	
G	77	0.061	0.32	0.6	60	115	9	
S	63	0.026	0.32	0.2	39	223	8	

Table II. Water quality category upstream and downstream of the Toulouse area defined by the Adour-Garonne Water Agency (1997)

	Upstream of Toulouse	Downstream of Toulouse
Dissolved O_2 (mg l^{-1})	5 to 7	<3 to 5
% saturation	70 to 90	<50 to 70
NH_4 (mg l^{-1})	0.1 to 0.5	0.5 to 8
NO_3-N (μg l^{-1})	1200 to 6000	600 to 24000
PO_4-P (μg l^{-1})	65 to 160	160 to 1600
P total (mg l^{-1})	0.1 to 0.25	0.25 to 2.5

temperature on biofilm growth at these two sites could be similar and so discounted. Then, it would seem that water quality best differentiates these two stations. In fact, the Adour-Garonne Water Agency (1997) classified the river sector upstream of Toulouse as a good water quality sector and the downstream sector as a tolerable to mediocre water quality sector (Table II).

MATERIALS AND METHODS

The investigations took place during the summer low water period (September 1997, with a flow rate of about 65 m^3 s^{-1}) because this period is very favourable for the growth of epilithic biofilms. Firstly, we sampled epilithic biofilms at one-week intervals at the station upstream of Toulouse (Pinsaguel = $P_{(1)}$ (08/09/97) and $P_{(2)}$ (16/09/97)) and at the station downstream of Toulouse (Gagnac = $G_{(1)}$ (05/09/97) and $G_{(2)}$ (15/09/97). Our investigation was completed by sampling at around 100 km upstream from P (at Valentine = V (12/09/97)) and at around 100 km downstream from G (at Sauveterre Saint Denis = S (11/09/97)) (Figure 1b). All this sampling was carried out outside periods of hydraulic disturbance (Figure 2).

Few studies have been carried out on stream bed cross-sections (Cazaubon, 1986; Reynolds *et al*., 1991 (total suspended algae and from the river bed); Cazaubon *et al*., 1995; Rolland *et al*., 1997; Smolar *et al*., 1998) and these studies have shown irregular patterns of algal distribution. Consequently, in order to have a good representation of biofilm biomass in the wide river Garonne, at each station we sampled at 10 m intervals on transverse profiles and 20 to 40 m on longitudinal profiles (upstream and/or downstream of the transversal section, at the mean depth). All samples were compared between the different stations. Three pebbles (100 to 200 cm^2 of colonized surface) were sampled at each sampling point on the profiles. This periphyton was sampled by scraping gravel with a knife blade and a toothbrush in 500 ml of stream water *in situ*. The washings obtained were frozen immediately in a cool bag and taken to the laboratory.

During the sampling period, the current velocity was measured by SEBA Mini Current Meter MI at a distance of 7 cm from the substratum. The depth was measured with a graduated ruler.

The periphyton washings obtained after scraping were filtered through Whatman GF/C filters which permitted the determination of: (i) the quantity of dry mass (DM, g m^{-2}) and ash-free dry mass (AFDM

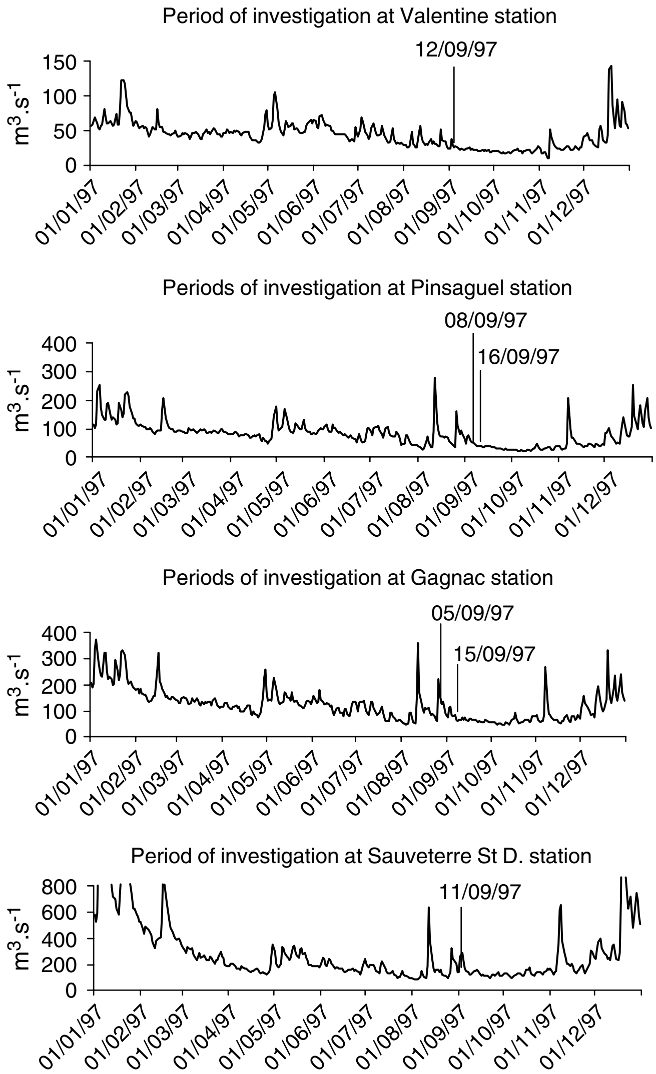


Figure 2. The six periods of investigation in relation to the mean daily discharge (m^3 s^{-1}) at the stations of Valentine, Pinsaguel, Gagnac and Sauveterre

(g m^{-2}), a measure of total organic matter); (ii) the concentration of chlorophyll-*a* (chl*a*, mg m^{-2}) by filtration and extraction with acetone (90%) according to APHA (1992). Analysis of water quality in the river Garonne was carried out by Adour-Garonne Water Agency (1997) (Table II).

APHA (1992) defines the autotrophic index (AI) as a means of determining the trophic nature of the periphyton community. It is calculated as AFDM/chl*a* ratio.

Discharge was estimated by the water management authorities (DIREN Toulouse, DIREN Bordeaux, CACG Tarbes and EDF).

RESULTS

The results of the distribution of epilithic biofilms in the 1997 summer period on the river Garonne at four stations which are 223 km apart (Figure 3) show that the biofilms grow on nearly the whole surface of the

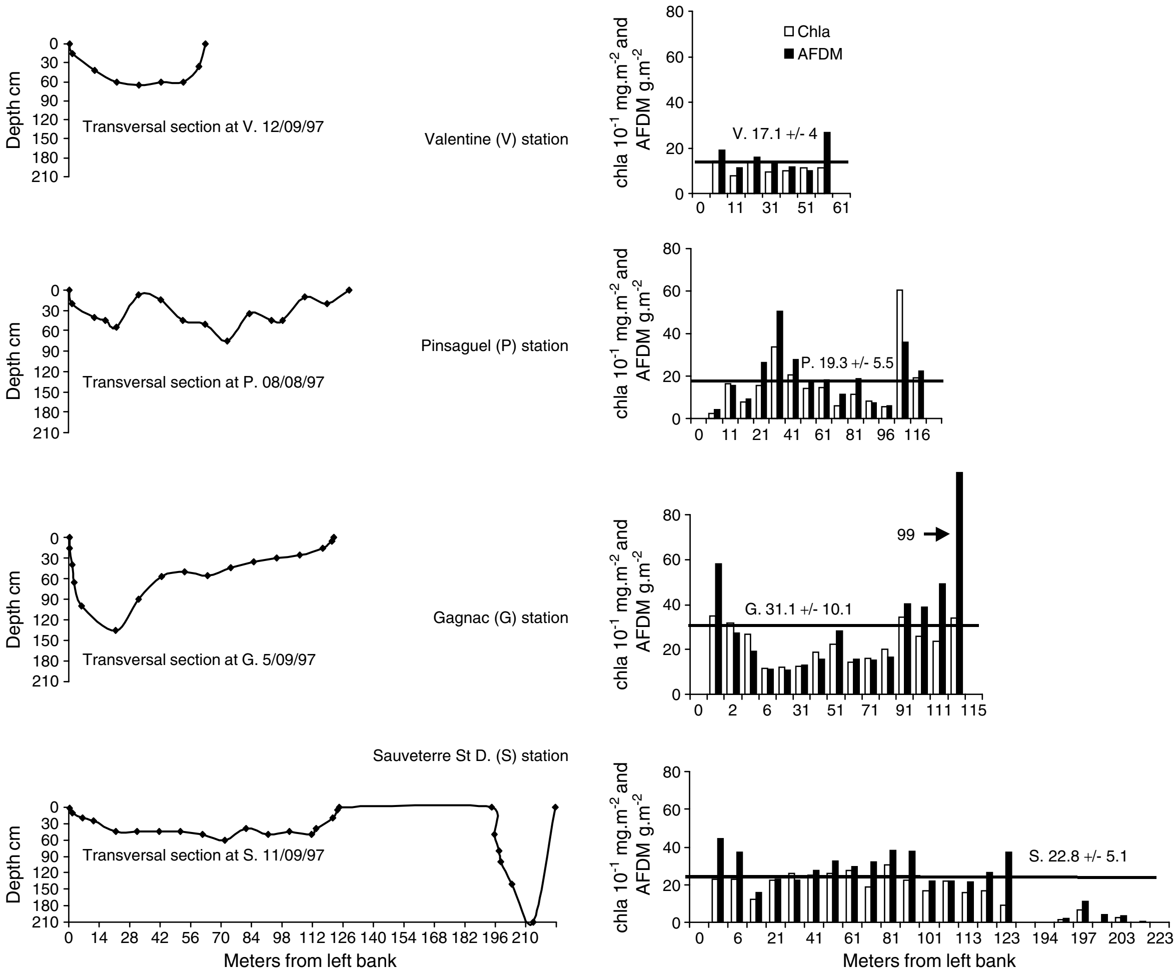


Figure 3. Transverse sections of river at Valentine (V), Pinsaguel (P), Gagnac (G) and Sauveterre St Denis (S) stations: profile of water depth, ash-free dry mass (AFDM) and chlorophyll-*a* (chl*a*) values at different samples and mean of AFDM (±SEM)

gravel bank and with a relevant value even at depth of 1.35 m (G station). The growth is limited at 2 m (S station).

At the V station (with a 'U' transverse profile), chl*a* and AFDM values are relatively homogeneous, with an increase near the edges. The transverse profile of the P station is more heterogeneous as well as biomass values. Maximal values are observed at the lowest water depths, except on the edges that underwent hydraulic fluctuations. At the G station, the curved form of the biomass values tends to a concave curve and to a convex curve at the S station. We observed a heterogeneity of epilithic biomass distribution on transverse profiles often with the maximum at the edges and decreasing with increased depth. However, we do not observe a significant relationship between the biomass values and the water depth. Heterogeneity was also observed on longitudinal sections, more particularly at G and S stations (Figure 4).

The relationship of chl*a* as a function of AFDM gave a significant relationship ($R^2 = 0.6$, $n = 67$, $P < 0.001$) fitting with a log model (Figure 5). Comparisons of individual values of DM, AFDM and chl*a* on each transect for the two fields upstream and downstream of Toulouse (Table III) showed that there was no significant difference between these two stations for AFDM (Mann–Whitney U-test statistic = 286; $p = 0.084$; df = 1). There was a significant difference for DM (Mann–Whitney U-test statistic = 252; $p = 0.022$; df = 1) and chl*a* (Mann–Whitney U-test statistic = 249; $p = 0.013$; df = 1). In the same way, considering the sector of river defined by the four stations, there are no significant differences in DM and

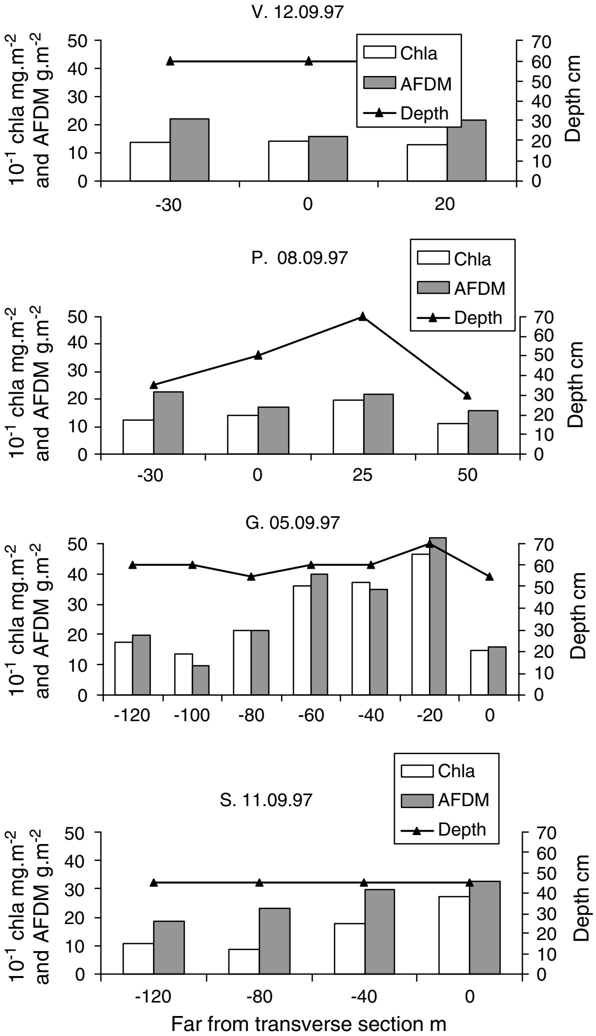


Figure 4. Ash-free dry mass (AFDM) and chlorophyll-*a* (chl*a*) values at different samples on the longitudinal section at Valentine (V), Pinsaguel (P), Gagnac (G) and Sauveterre St Denis (S) stations

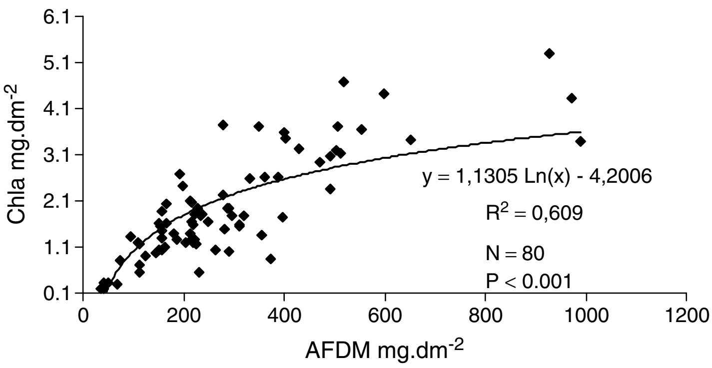


Figure 5. Relationship between chl*a* and AFDM in the river Garonne during low water period 1997

Table III. Comparison of values of dry mass (DM), ash-free dry mass (AFDM) and chlorophyll-*a* (chl*a*) for the two fields (1 and 2) of each two cross-sections upstream (Pinsaguel = P) and downstream (Gagnac = G) of Toulouse by the Mann–Whitney U-test statistic. Bold typeface indicates significant differences ($P > 0.05$)

Biofilm biomasses	Stations	Mann–Whitney U-test statistic	Probability	Degree of freedom	Count
DM	P1 G1	100	0.033	1	17/20
DM	P2 G2	35	0.414	1	10 / 9
DM	(P1+P2)/(G1+G2)	252	0.022	1	27/29
AFDM	P1 G1	113	0.082	1	17/20
AFDM	P2 G2	38	0.568	1	10/9
AFDM	(P1+P2)/(G1+G2)	286	0.084	1	27/29
chl*a*	P1 G1	79	0.003	1	17/20
chl*a*	P2 G2	47	0.870	1	10 / 9
chl*a*	(P1+P2)/(G1+G2)	249	0.013	1	27/29

Table IV. Comparison of values of dry mass (DM), ash-free dry mass (AFDM) and chlorophyll-*a* (chl*a*) for the four cross-sections at the stations of Valentine (V), Pinsaguel (P), Gagnac (G) and Sauveterre Saint-Denis (S) by the Kruskal–Wallis test statistic. Bold typeface indicates significant differences ($P > 0.05$)

Biofilm biomasses	Stations	Kruskal–Wallis U-test statistic	Probability	Degree of freedom	Count
DM	V1 P1 G1 S1	7.289	0.063	3	10/17/20/25
AFDM	V1 P1 G1 S1	4.928	0.177	3	10/17/20/25
chl*a*	V1 P1 G1 S1	17.982	0.001	3	10/17/20/25

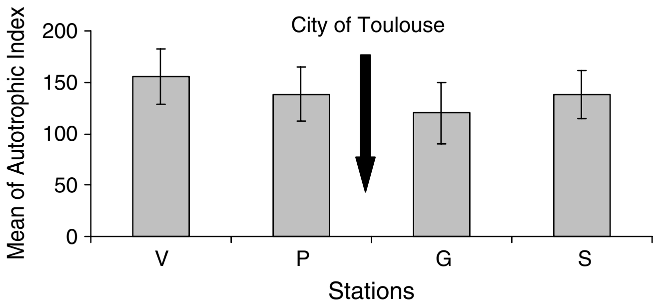


Figure 6. Upstream to downstream variations of mean of autotrophic index in the river Garonne at Valentine (V), Pinsaguel (P), Gagnac (G) and Sauveterre St Denis (S) stations between 5 and 15 September 1997 in low water period without disturbance (values ±SEM)

AFDM (Kruskal–Wallis test statistic = 7.289/4.928, $p = 0.063/0.177$, df = 3/3 respectively) (Table IV). On the other hand, there is a significant difference between the four stations for the chl*a* (Kruskal–Wallis test statistic = 17.982, $p = 0.001$, df = 3).

With regards to the mean values of AI for the four stations studied, these were between 120 and 160 (Figure 6). They did not reveal a difference between upstream and downstream of Toulouse nor along the total sector studied.

DISCUSSION

Epilithic biofilm biomasses

The variation in the distribution of epilithic biomass on transverse profiles confirms the interest of sampling along the cross-section, as done by Cazaubon *et al.* (1995) and Rolland *et al.* (1997) in French Mediterranean rivers (i.e every 1 m on 10–25 m width) and Smolar *et al*. (1998) in streams of Slovenia, in order to have a more representative biomass. However, for a large river such as the Garonne, the samples nearest to the mean of the biomass of the total gravel bed correspond to the samples taken at the mean depth (40–50 cm) (Figure 3), that is to say away from the bank and the maximum depth. In order to continue these investigations we propose to sample six points between 40 and 50 cm of depth over riffles to track these stations. At the four stations, the mean of the AFDM values fluctuated between 17.1 and 31.1 g m^{-2}, and 112 to 254 mg m^{-2} for chl*a* concentration in the 1997 summer low water period. For other studies on artificial substrates in the Garonne at different sites, AFDM varies from 5 to 30 g m^{-2} which corresponds to 50 to 70 mg m^{-2} of chl*a* (Dauta, 1978b; Benmoussa, 1995; Eulin, 1997). Biggs (1995) performed a study at 16 stream sites in New Zealand which were sampled monthly for a year. Means of AFDM varied from 1.8 to 69.5 g m^{-2} and chl*a* from 2.1 to 280.8 mg m^{-2}. Uehlinger *et al*. (1996) measured 0.49 to 247 mg m^{-2} for chl*a* concentration at weekly or two weekly intervals during 17 months in a Swiss pre-alpine gravel-bed river. For a stream linking a chain of lakes in the Laurentian mountains of Quebec (Cattaneo, 1996), biofilm on gravel or boulders is characterized by chl*a* values between 5 and 55 of mg m^{-2}. These values show the variability in biofilm biomass and that most often, AFDM is less than 70 g m^{-2} and chl*a* is less than 300 mg m^{-2}.

Manifestation of eutrophication

The widely quoted OECD guidelines (Anon., 1982) defined eutrophication as water enrichment in nutrients that causes a series of symptomatic changes such as an increase in algal and macrophyte production, damage to water quality, and other changes considered as undesirable and inauspicious to the different uses of water.

In small, non-channelled rivers, eutrophication causes an important growth of fixed vegetation (macrophytes) that obstruct the river bed, while in large rivers, eutrophication is linked to the development of microscopic planktonic algae (Garnier *et al*., 1998). The distribution and abundance of macrophytes change with spatial and temporal variations, mainly dependent on physical factors such as substrate, depth, turbidity and current velocity. Studies performed by IDE Environnement (1994) and Gazagnes (1998) showed that the upper part of the river Garonne (Valentine to Toulouse) is rather more favourable to development of macrophytes than downstream from Toulouse. As regards phytoplankton in the river Garonne, except for some sporadic growth in dams (Manciès; Malause), the suspended algal density is less than 10 μg chl*a* l^{-1}. Microscopic examination showed that this 'phytoplankton' is mainly composed of algal benthic drift (Améziane and Dauta, 1997). So the manifestation of eutrophication in the river Garonne would not appear to be linked to macrophyte or phytoplankton abundance.

Periphytic biomass: is it a manifestation of eutrophication?

Our interest in epilithic biofilm development in the Garonne and the implications of eutrophication on epilithic biofilms in terms of river management have led us to focus on the eutrophication in terms of the origin of epilithic biofilm.

To discuss the epilithic biomass in terms of eutrophication in the river Garonne, we quantified the biofilm by DM, AFDM and chl*a*. As we aim to quantify a complex that is at once autotrophic and heterotrophic (i.e. biofilm), DM, which also takes into account the mineral matter, did not seem to us the relevant biomass evaluation. Moreover, as there is no significant difference in AFDM between upstream and downstream of Toulouse, the significant differences in DM (Table III) are due to an accumulation of mineral matter from the downstream urbanization by sedimentation which maybe increases by the Ariège tributary (David, 1998) and other contributions. As for the chl*a*, used to quantify the biomass of the algae, it represents the most important part of the organic matter in biofilm of rivers. But, as with every cellular component, chlorophyll-*a* varies with the biomass (Capblancq, 1982). This pigment, which is present in every alga, varies in quantity as a function of species, of the cells' nutritive state and of the light environment (Steele and Baird, 1965; Heaney, 1978; Harris, 1980). With the biofilm, there is also a vertical accumulation of cell layers that produce a self-shading effect (Steinman and McIntire, 1987; Hill and Boston, 1991). This is possibly the reason why the relationship of chl*a* as a function of AFDM gave a significant relationship when fitted to a log model (Figure 5), with the quantity of chl*a* levelling off as the AFDM increases. Chl*a* could give more information on biofilm structure (quantity of cell layers) and hence its state of maturity. Consequently, we will keep the interpretation of the AFDM values with regards to our aim. As there were no significant differences in AFDM between upstream and downstream of the urbanization (Mann–Whitney U-test statistic), as for the four stations 223 km apart (Kruskal–Wallis test statistic), this study does not permit us to reveal the biofilm epilithic from the river Garonne as a manifestation of eutrophication.

Autotrophic index

As for the AI, it is a means of determining the trophic nature of the periphyton community. Normal values range from 50 to 200; larger values indicate heterotrophic associations or poor water quality. Non-viable organic material affects this index. Depending on the community, its location and growth habit, and method of sample collection, there may be large amounts of non-living organic material that may inflate the numerator and produce disproportionately high AI values. Nonetheless, the AI is an approximate means of describing changes in periphyton communities between sampling locations (APHA, 1992). Referring to Fayolle (1998), the periphytic community is dominated by algae when AI is lower than 100; for an AI between 100 and 400, the periphytic population is balanced between heterotrophic and autotrophic conditions. Finally, epilithon is dominated by heterotrophs and rubbish when AI is greater than 400. This indicates an imbalance between the communities. Mean values of the AI of the epilithic biofilm at the four stations were measured as between 120 and 160 (Figure 6) and there is no difference between upstream and downstream of the urbanization.

From different studies on natural or artificial substrates and streams (MacIntire, 1966; Cushing, 1967; Bahls, 1971; Dunn, 1976; Elwood *et al.*, 1981 (all in Horner *et al.*, 1983); Capblancq and Cassan, 1979; Dauta, 1978b (river Garonne); Watanabe, 1985; Biggs, 1995; Benmoussa, 1995 (river Garonne); Eulin, 1997 (river Garonne); Smolar *et al.*, 1998; Godillot *et al.* (2001); this study) we examined, by the projection of chl*a* as a function of AFDM values (Figure 7), whether there are different tendencies. There are three groupings of points. The grouping of points near the chl*a* axis (1) characterizes a biofilm with autotrophic dominance (AI $<$ 50). It corresponds to a biofilm grown on gravel introduced in an experimental canal with low nutrients (Godillot *et al.*, 2001). The grouping of points around the AFDM axis (2) characterizes a biofilm grown in a very polluted river with a heterotrophic biofilm (AI = 10 000) (Capblancq and Cassan, 1979). Outside these two cases, the totality of the other points was characterized by an AI between 50 and 200 that corresponds to a balanced biofilm. This is the case for the different values measured in the river Garonne. The AI did not allow the description of changes in periphyton communities between sampling locations in the river Garonne. However, there is a degradation in water quality downstream from Toulouse. The level of the information

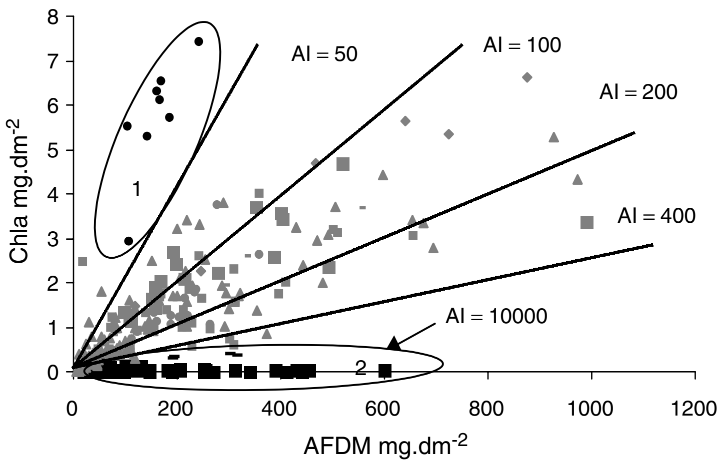


Figure 7. Relationship between chl*a* and AFDM of different studies (272 points). The straight lines represent the different values of AI. The grouping of points 1 corresponds to a biofilm grown in an experimental canal with low nutrients (Godillot *et al.*, 2001). The grouping of points 2 characterizes a biofilm grown in a very polluted river (Capblancq and Cassan, 1979). The other points arises from other studies including those from the river Garonne

given by the AI with regards to the manifestation of eutrophication in moderately polluted rivers, does not permit it to be an indicator of eutrophication.

Nutrient limitation of benthic algae

The notion of nutrient limitation of benthic algae must take into account both concentration of nutrient in the water column and the diffusion in the biofilm. In fact, growth rate saturation always occurred at a phosphate concentration of approximately 0.3–0.6 μg PO_4-P l^{-1} for Bothwell (1988). Because a developing benthic mat will impede molecular and eddy diffusion of nutrients (Borchardt, 1996) a higher P concentration is needed to reach maximum benthic algal biomass: 8 to 25 μg PO_4-P l^{-1} (Horner *et al*., 1983, 1990), 30 to 50 μg PO_4-P l^{-1} (Bothwell, 1989), although 70% of biomass occurred with 1 μg PO_4-P l^{-1}. Nitrogen limitation of benthic algae has been reported in Borchardt (1996) when ambient concentration was 55 to 100 μg NO_3-N l^{-1}.

When nutrient ambient concentrations are near growth-limiting levels, nutrient ratios are useful for assessing limitation. Ambient N : P ratios greater than 20 : 1 are considered P-limited, less than 10 : 1 are N-limited, and between 10 : 1 and 20 : 1 the distinction is equivocal (Schanz and Juon, 1983 in Borchardt, 1996; Vymazal *et al*., 1994).

Insofar as concentrations of nitrogen (1200 μg NO_3-N l^{-1}) and phosphorus (65 μg PO_4-P l^{-1}) in the upper part of the river Garonne are greater than growth-limiting levels, these nutrients do not modify the maximum benthic biomass.

CONCLUSION

Epilithic biofilm and trophic status of the Garonne

The quantitative analysis of the total organic matter in biofilms during the 1997 summer low water period and the autotrophic index measurement did not show any significant difference between the stations upstream and downstream of the urbanization, nor along the total waterway studied. These preliminary results did not confirm the hypothesis that fixed biomass increases with the degradation of the water quality in the river Garonne. As the concentrations of nutrients certainly do not limit the growth of biofilm from the upstream stations, the relative homogeneity in the epilithic biofilm along the Garonne is not surprising. However, nutritive resources constitute only one element (and not necessarily the determinant element) of the principal factors that control the dynamics of vegetal communities: morphometry of the minor bed, hydraulic pattern, hydrodynamics, water transparency, composition of the substrate, and local climate. Because most of the factors are mutually and frequently co-variables, the identification of critical conditions that cause a qualitative and quantitative change in vegetal population is far from evident. Through this study, the epilithic biofilm, evaluated as AFDM and AI, cannot to be used to evaluate the wastewater impact of urbanization nor to define a trophic status along the large gravel-bed river Garonne which is characterized by an important fixed biomass. To understand the functioning of the river Garonne it would be very interesting to take hydrological disturbances into account because it is between floods that the level of nutrient resource supply can influence periphytic growth and community physiology (Biggs *et al*., 1999).

ACKNOWLEDGEMENTS

This work was supported by GIS ECOBAG (Ecology of Adour-Garonne Basin).

REFERENCES

Adour-Garonne Water Agency. 1997. *Atlas et données sur l'eau*. Toulouse.

Améziane T, Dauta A. 1997. Evolution longitudinale du phytoplancton dans la Garonne en période d'étiage estival: contribution des principaux affluents, *CILEF 5 Congress (Belgium)*. Facultes universitaires Notre-Dame de la Paix: Namur.

Anon. 1982. *Eutropication of Waters. Monitoring, Assessment and Control*. Organisation for Economic Co-operation and Development: Paris.

APHA.AWWA.WPCF. 1992. *Standard methods for the examination of water and wastewater* (18th edition); Washington.

Bahls LL. 1971. *Ecology of the diatom community of the upper East Gallatin River, Montana, with in situ experiments on effect of current velocity on features of the Aufwuchs*. PhD dissertation, Montana State University, Bozeman.

Benmoussa M. 1995. *Ecologie des communautés périphytiques (Etude en laboratoire et en milieu naturel des conditions de développement et des caractéristiques de fonctionnement de trois types de biodermes*. PhD thesis. CESAC, University of Paul Sabatier Toulouse III France.

Biggs BJF. 1995. The contribution of flood disturbance, catchment geology and land use to the habitat template of periphyton in stream ecosystems. *Freshwater Biology* **33**: 419–438.

Biggs BJF, Tuchman NC, Lowe RL, Stevenson RJ. 1999. Resource stress alters hydrological disturbance effects in a stream periphyton community. *OIKOS* **85**: 95–108.

Borchardt MA. 1996. Nutrients. In *Algal Ecology, Freshwater Benthic Ecosystems*, Stevenson RJ, Bothwell ML, Lowe RL (eds). Academic Press: New York; 183–227.

Bothwell ML. 1988. Growth rate responses of lotic periphytic Diatoms to experimental phosphorus enrichment: the influence of temperature and light. *Canadian Journal of Fisheries and Aquatic Science* **45**: 261–270.

Bothwell ML. 1989. Phosphorus-limited grwth dynamics of lotic periphytic diatom communities: areal biomass and cellular growth rate responses. *Canadian Journal of Fisheries and Aquatic Science* **46**: 1293–1301.

Capblancq J. 1982. Phytoplancton et production primaire. In *Ecologie du plancton des eaux continentales*, Pourriot R, Capblancq J, Champ P, Meyer J-A (eds). Masson: Paris; 1–48.

Capblancq J, Cassan M. 1979. Etude du periphyton d'une rivière polluée (l'Agout). 1. Structure et développement des communautés sur substrats artificiels. *Annals of Limnology* **15**(2): 193–210.

Cattaneo A. 1996. Algal seston and periphyton distribution along a stream linking a chain of the Canadian Shield. *Hydrobiolagia* **325**: 183–192.

Cazaubon A. 1986. Rôle du courant sur la microdistribution des diatomées épilithiques dans une rivière méditerranéenne, l'Argens (Var, France). *Ninth Diatom Symposium*, Bristol, UK, Koeltz O (ed.). 93–107.

Cazaubon A, Rolland T, Loudiki M. 1995. Heterogeneity of periphyton in French Mediterranean rivers. *Hydrobiologia* **300/301**: 105–114.

Cushing CE. 1967. Periphyton productivity and radionuclide accumulation in the Columbia River, Washington, U.S.A. *Hydrobiologia* **29**: 125–139.

Dauta A. 1978a. Evolution estivale du phytoplancton dans une retenue placée au confluent de deux rivières. *Cahiers Laboratoire Montereau* **7**: 33–40.

Dauta A. 1978b. Colonisation de substrats artificiels dansla retenue de Malause. *Cahiers Laboratoire Montereau* **7**: 41–46.

Dauta A, Améziane T, Garabétian F. 1999. Metric-scale variations of pH and dissolved oxygen around a gravel bar covered by a periphytic biofilm (River Garonne). *Bulletin de la societe linneenne de Bordeaux* **27**: (special volume).

David C. 1998. *Caractérisation des matières en suspension lors d'événements hydrologiques particuliers*. PhD thesis, CESAC, University of Paul Sabatier Toulouse III France.

Dunn RW. 1976. *Seasonal variations in periphyton, chlorophyll a, algal biomass and primary production in a desert stream*. MS thesis, Department of Biology, Idaho State University, Pocatello.

Elwood JW, Newbold JD, Trimble AF, Stark RW. 1981. The limiting role of phosphorus in a woodland stream ecosystem: effects of P enrichment on leaf decomposition and primary producers. *Ecology* **62**: 146–158.

Eulin A. 1997. *Les communautés de diatomées épilithiques de la Garonne*. PhD thesis, CESAC, University of Paul Sabatier Toulouse III France.

Fayolle S. 1998. *Réponses de communautés végétales strictement aquatiques (algues et macrophytes) aux perturbations hydrodynamiques (débits réservés, restitutions) en Durance aménagée (Sud-Est de la France)*. PhD thesis, University of Aix-Marseille III France.

Garnier J, Billen G, Hanset P, Testard P, Coste M. 1998. Développement algal et eutrophisation dans le réseau hydrographique de la seine. In *La seine en son bassin: fonctionnement écologique d'un fleuve antropisé*, Meybeck M, de Marsily G, Fustec E (eds). Elsevier: Amsterdam; 593–626.

Gazagnes G. 1998. *Suivi des manifestations de l'eutrophisation dans la Garonne au cours de l'été 1997*. Agence de l'Eau Adour Garonne Toulouse: France.

Godillot R, Caussade B, Améziane T, Capblancq J. (2001). On periphyton—flow mutual influences. *Journal of Hydrological Research* **39**(3): 227–239.

Harris GP. 1980. Spatial and temporal scales in phytoplankton ecology. Mechanisms, methods, models and management. *Canadian Journal of Fisheries and Aquatic Science* **30**: 1779–1787.

Heaney SI. 1978. Some observations on the use of the in vivo fluorescence technique to determine chlorophyll-a in a natural populations and cultures of freshwater phytoplankton. *Freshwater Biology* **8**: 115–126.

Hill WR, Boston HL. 1991. Community development alters photosynthesis-irradiance relations in stream periphyton. *Limnology and Oceanography* **36**: 1375–1389.

Horner RR, Welch EB, Veenstra RB. 1983. Development of nuisance periphytic algae in laboratory streams in relation to enrichment and velocity. In *Periphyton of Freshwater Ecosystems*, Wetzel RG (ed.). W. Junk: The Hague; 346.

Horner RR, Welch EB, Seeley MR, Jacoby JM. 1990. Responses of periphyton to changes in current velocity, suspended sediment and phosphorus concentration. *Freshwater Biology* **24**: 215–232.
IDE-Evironnement, CESF, LHT. 1994. *Etude des risques d'eutrophisation de la Garonne*; Toulouse.
McIntire CD. 1966. Some effects of current velocity on periphyton communities in laboratory streams. *Hydrobiologia* **27**: 1940–1952.
Petts GE, Bravard JP. 1993. Le réseau hydrographique dans le bassin-versant. In *Hydrosystèmes fluviaux*, Amoros C, Petts GE (eds). Masson: Paris; 21–41.
Reynolds CS, Carling PA, Beven K. 1991. Flow in river channels: new insights into hydraulic retention. *Archives of Hydrobiology* **121**: 171–179.
Rolland T, Fayolle S, Cazaubon A, Pagnetti S. 1997. Methodical approach to distribution of epilithic and drifting algae communities in a French subalpine river: Inferences on water quality assessment. *Aquatic Science* **59**: 57–73.
Schanz F, Juon H. 1983. Two different methods of evaluating nutrient limitations of periphyton bioassays using water from the River Rhine and eight of its tributaries. *Hydrobiologia* **102**: 187–195.
Smolar N, Vrhovsek D, Kosi G. 1998. Effects of low flow on periphyton in three different types of streams in Slovenia. *Advances in River Bottom Ecology*. Backhuys Publishers: Leiden; 107–116.
Steele JH, Baird IE. 1965. The chlorophylla content of particulate organic matter in the northern sea. *Limnology and Oceanography* **10**: 261–267.
Steinman AD, McIntire CD. 1987. Effects of irradiance on the community structure and biomass of algal assemblages in laboratory streams. *Canadian Journal of Fisheries and Aquatic Science* **44**: 1640–1658.
Uehlinger U, Buhrer H, Reichert P. 1996. Periphyton dynamics in a floodprone prealpine river: evaluation of significant processes by modelling. *Freshwater Biology* **36**: 249–263.
Vannote RL, Minshall GW, Cummins KW, Sedell JR, Cushing CE. 1980. The river continuum concept. *Canadian Journal of Fisheries and Aquatic Science* **37**: 130–137.
Vymazal J. 1988. The use of periphyton communities for nutrient removal from polluted streams. *Hydrobiologia* **166**: 225–237.
Vymazal J, Craft CB, Richardson CJ. 1994. Periphyton response to nitrogen and phosphorus additions in Florida Everglades. *Algological Studies* **73**: 75–97.
Watanabe T. 1985. *Etude de la relation entre le périphyton et la qualité chimique de l'eau des rivières: Utilisation de bioessais "in situ" (substrats artificiels) pour caractériser l'état de pollution des eaux*. Thesis, CESAC, University of Paul Sabatier Toulouse III France.