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Abstract. Habitat stability is an important driver of ecological community composition and development. River epilithic biofilms are particularly unstable habitats for the establishment of benthic communities because they are regularly disturbed by floods. Our aim was to determine the influence of habitat instability on meiobenthic organisms. We hypothesized that hydrologic variables are the most important predictors of meiofauna distribution. We monitored epilithic communities (meiofauna and microalgae) with a high sampling frequency during 2 sampling periods with contrasting hydrodynamic patterns in a temperate river (the Garonne, France). Nematodes and rotifers dominated meiofaunal assemblages. The critical flow velocity threshold for their maintenance in the biofilm was ~30 cm/s, a result suggesting that meiofauna can resist higher flow velocity within the biofilm than within sediments. Nematode distribution was primarily influenced by the duration of undisturbed periods, whereas rotifer distribution was also correlated with the thickness of the biofilm. During the periods after floods, rotifers were faster colonizers than nematodes. Collectively, our results show that flow regime was an essential driver for biofilm community development.

Key words: habitat stability, resilience, recolonization, flow velocity, meiobenthos, rotifers, nematodes, periphyton.

Biotope stability is an important driver of community composition and development in both terrestrial and aquatic systems (e.g., Cobb et al. 1992, Death and Winterbourn 1995, Villenave et al. 2001, van der Wurff et al. 2007). Instability in aquatic systems can result from natural variations in flow regime (Death 2002, Lake 2003, Cardinale et al. 2005) or from human-induced perturbations, such as acute pollution, introduced species, and flushing of reservoirs (e.g., Charlebois and Lamberti 1996, Lai and Shen 1996, Carpenter et al. 1998).

River biofilms are a complex assemblage of organisms (bacteria, fungi, algae, heterotrophic protozoans, meiofauna, and macrofauna) embedded in a mucous matrix of exopolymeric substances. These biofilms grow on any hard, submerged substrate (Lock 1993, Costerton 2000) and generally are copiously inhabited by microalgae (Peterson 1996). Consequently, they can constitute the main site of primary production in shallow-water rivers with hard substrates, such as the middle reaches of the Garonne (Ameziane et al. 2003). These biofilms are an important food resource for stream consumers (Fuller et al. 1986, Lawrence et al. 2002, Liess and Hillebrand 2004). Moreover, they play a key role in biogeochemical processes, such as nutrient retention (Battin et al. 2003, Teissier et al. 2007). Nevertheless, these complex habitats are particularly unstable because they can be partially or entirely removed from their substrates—with their inhabitant fauna—on a regular basis by flood events, bacterial degradation processes, grazing, and bioturbation (Biggs and Close

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1989, Lawrence et al. 2002, Boulêtreau et al. 2006, Gaudes et al. 2006, Peters et al. 2007). This instability can shape the biomass, diversity, and viability of algal and bacterial communities inhabiting the mats (e.g., Peterson and Stevenson 1992, Lyautey et al. 2010) and can affect biofilm processes (Cardinale 2011).

Meiobenthic invertebrates (40–500-μm size range; Giere 2009) are particularly abundant within these biofilms (Peters and Traunspurger 2005, Gaudes et al. 2006, Kathol et al. 2011). They are a highly diverse and abundant component of stream communities (e.g., Schmid-Araya 1997, Beier and Traunspurger 2003) and are important foodweb intermediates between micro- and macrofauna (Schmid-Araya and Schmid 2000, Schmid-Araya et al. 2002, Spieth et al. 2011). Moreover, given their short generation times and high fecundity rates (Bergtold and Traunspurger 2005, Stead et al. 2005, Reiss and Schmid-Araya 2010), they are particularly relevant model organisms for testing general ecological theories, especially those relating to population dynamics and community stability (Reiss et al. 2010).

Post-flood periods present an opportunity to study recolonization processes and resilience of lotic meiofauna. In most of the few studies of this topic in rivers, investigators focused on sediment recolonization after catastrophic disturbances (reviewed in Robertson 2000). In a few studies, investigators examined the influence of noncatastrophic flow on the dynamics of sediment-dwelling meiofauna. Palmer (1992) showed that sediment-dwelling meiofauna are frequently found above the sediment–water interface even under low-flow conditions, and Smith and Brown (2006) found that meiofauna can rapidly recolonize sediments in artificial stream channels. However, data are lacking on resistance and resilience of biofilm-dwelling meiofauna to variations of flow in rivers. Sediment-dwelling meiofauna also can respond to environmental constraints, such as temperature, that change seasonally (Stead et al. 2003) and to habitat characteristics, such as sediment grain size distribution or organic matter availability (Swan and Palmer 2000, Beier and Traunspurger 2003, Reiss and Schmid-Araya 2008, Tod and Schmid-Araya 2009). Studies addressing the temporal dynamics of meiofauna in river biofilms are rare. Gaudes et al. (2006) and Caramujo et al. (2008) considered only relatively short time periods, Kathol et al. (2011) highlighted pelagic–benthic coupling via biofilm-dwelling consumers, and Majdi et al. (2011) focused on temporal patterns of nematode assemblages. Therefore, how the complete meiobenthic community responds to the instability of their biofilm habitat is unclear.

Our objective was to determine how biofilm stability influences the composition of biofilm-dwelling meiofauna. We examined the temporal evolution of this relationship and the factors driving its development during 2 periods with contrasting patterns of flood disturbance in a temperate river (the Garonne, France). We hypothesized that hydrologic factors are the most important predictors of meiofauna distribution directly or via modification of biofilm status.

Methods

Study site and epilithic biofilm sampling

The Garonne is the largest river of southwestern France and has a drainage basin of 57,000 km^2 and a length of 647 km. The Garonne is a physically active river (Chauvet and Décamps 1989) with a pluvio-nival flow regime characterized by an intense spring-flood period caused by snowmelt in the Pyrénées Mountains followed by a long low-water period that can continue for the rest of the year. Flash floods caused by heavy rainfall can occur (mostly during autumn and winter) in some years. The river bed consists mainly of cobble and gravel, and large alternating cobble bars are found frequently even in channels up to 7^{th}-order. During low-water periods, a thick biofilm favored by low flow velocities on the river bed and low turbidity typically coats the upper surfaces of cobbles (Boulêtreau et al. 2006, Leflaive et al. 2008).

We sampled the epilithic biofilm at a cobble bar 36 km upstream of the city of Toulouse where the Garonne is 6^{th}-order (lat 01°17′53″E, long 43°23′45″N; elevation: 175 m asl). The epilithic microbial community has been previously described at this site (Lyautey et al. 2005, Leflaive et al. 2008). In this stretch of the Garonne, total P and total N concentrations in the water column vary over a year from 0.01 to 0.05 and 0.4 to 1.4 mg/L, respectively. Dissolved organic C and SiO_4^{4-} vary from 1 to 5 and 2 to 6 mg/L, respectively (Leflaive et al. 2008). The canopy is widely open, but the residence time is too short to allow substantial phytoplankton development, so benthic biofilms are assumed to provide most of the primary production (Ameziane et al. 2002, 2003).

The 1^{st} sampling period (C1) lasted from November 2004 to March 2006 and had 44 sampling occasions. The 2^{nd} sampling period (C2) lasted from September 2008 to March 2010 and had 51 sampling occasions. We sampled weekly when possible, but sample collection was possible only when discharge was <175 m^3/s. On each sampling occasion, we collected 12 randomly selected cobbles (mean diameter = 10 cm) by sliding them into a plastic bag underwater (depth = 30–50 cm) to prevent any detachment of the epilithic biofilm during removal. Within 2 h of

collection, we transported the cobbles to the laboratory in a cool box to reduce pigment degradation. There, we removed the biofilm by scraping the total upper surface of each cobble with a scalpel and a toothbrush. We cut long algal filaments into short segments with scissors and then suspended biofilm samples in ultrapure water (MilliQ filtration; Millipore, Billerica, Massachusetts) to obtain 12 biofilm suspensions (25 mL each). We divided these 12 suspensions into 3 groups of 4 replicates to be used for meiofaunal counts, algal pigment analyses, and estimation of epilithic ash-free dry mass (AFDM). We photographed scraped cobbles and measured the area of the surface from which biofilm had been removed (clearly visible on the cobble) and measured the scraped area (ImageJ software, version 1.38; Abramoff et al. 2004). We expressed meiofauna counts, algal pigments, and AFDM quantitatively per unit area. During C1, AFDM was determined on all 44, algal pigments on 24, and meiofauna on 17 sampling occasions. During C2, AFDM, algal pigments, and meiofauna were determined on all 51 sampling occasions.

Abiotic environmental factors

Mean daily discharge (MDD) was supplied by a gauging station of the French water management authority (DIREN Midi-Pyrénées, Marquefave station) 10 km upstream of the study site. No tributaries or dams occur between the gauging station and the study site. We measured stream flow velocity 5 cm above the streambed (mean of 3 measurements flanking the sampling area) on each sampling occasion with a flow meter (Flo-Mate 2000; Flow-Tronic, Welkenraedt, Belgium). We quantified stability as the number of days between a given sampling occasion and the last critical flood (days after flood [DAF]). Our long-term field observations (including periods during which most of the biofilm had been removed from the cobbles) allowed us to deduce that MDD of critical floods inducing a detachment of the major part of the epilithic biofilm is >300 m^3/s.

During C1, we measured temperature, conductivity, pH, and dissolved O_2 in the water column on each sampling occasion with a LF95 conductivity meter, a pH320 pH meter, and an OXI 320 oximeter, respectively (WTW, Weilheim, Germany). During C2, we measured these variables every 30 min with an automated multiparameter probe (YSI 6000; Yellow Springs Instruments, Yellow Springs, Ohio), which was permanently set 5 cm above the stream bed. We cleaned and calibrated probes monthly to avoid loss of accuracy.

Density, biomass, and resilience of biofilm-dwelling meiofauna

On each sampling occasion, we extracted the organic fraction from the 4 replicate biofilm suspensions with a modified gravity-gradient centrifugation technique (Pfannkuche and Thiel 1988). We used Ludox® HS-40 colloidal silica (Sigma–Aldrich, St. Louis, Missouri) and poured the extract through stacked 500-µm and 40-µm meshes. We preserved the organisms retained on the 40-µm mesh (including meiofauna) in formaldehyde (5% final concentration) with 1% rose Bengal. We counted ≥ 200 meiobenthic organisms per replicate in a Dolfuss cell (Elvetec Services, Clermont-Ferrand, France) under a stereomicroscope (9–90×) to measure their density.

On each sampling occasion, we isolated ≥ 10 meiofaunal chironomid larvae in small Al cups and dried them for 48 h at 50°C to determine their dry mass (DM). We processed meiofaunal oligochaetes and water mites as described for chironomids, but because of their low occurrence in some samples, their DM was not obtained on each sampling occasion. For these organisms, we pooled DM measurements to obtain a mean DM value for each sampling campaign. For nematodes, rotifers, harpacticoid copepods, and tardigrades, we assessed individual DM from biometric conversions of their body dimensions (Giere 2009).

We estimated resilience of nematodes and rotifers (time required for population densities to reach maximum preflood densities; Schmid-Araya 1994) during C2 after 2 critical flash floods caused by rainfall (23 January 2009 and 15 January 2010, both MDD = 462 m^3/s) and after the last critical flood of the spring snowmelt flood period (12 April 2009, MDD = 330 m^3/s). We did not estimate resilience for chironomid larvae because it can be biased by emergence.

Biofilm biomass and extraction of microalgal pigments for high-performance liquid chromatograph and chemotaxonomic analysis

On each sampling occasion, we dried (105°C, 18 h), weighed, and combusted (450°C, 8 h) 4 replicate biofilm suspensions to measure the AFDM content of the biofilm.

We centrifuged the 4 remaining suspensions (3220 × *g*, 20 min) and freeze-dried and thoroughly homogenized the pellets. We removed 250-mg subsamples from each pellet and extracted algal pigments from each subsample 3 times (15 min at −20°C) with a total of 25 mL (10, 10, and 5 mL) 98% cold-buffered methanol (with 2% of 1 M ammonium acetate) by sonication

(Buffan-Dubau and Carman 2000). We filtered 1 mL of the pigment solution through a 0.2-μm polytetrafluoroethylene (PTFE) syringe filter and analyzed the filtrate with a high-performance liquid chromatograph (HPLC) consisting of a 100-μL loop autosampler and a quaternary solvent delivery system coupled to a diode array spectrophotometer (LC1200 series; Agilent Technologies, Santa Clara, California). We prepared and programmed the mobile phase according to the analytical gradient protocol given by Barlow et al. (1997). We identified algal pigments by comparing their retention time and absorption spectra with those of pure standards (DHI LAB products, Hørsholm, Denmark; see Majdi et al. 2011 for further details).

We coupled HPLC-analysis of algal pigments with a chemotaxonomic analysis using CHEMTAX software (version 1.95; Mackey et al. 1996) to estimate the biomass of microphytobenthic groups in the biofilm in terms of contribution to total chlorophyll *a* (Chl *a*) biomass. We used the biomarker pigment ratios of biofilm microalgal groups reported in Majdi et al. (2011) to supply the initial matrix needed to run the chemotaxonomic analysis.

Data analysis

We used Mann–Whitney *U* tests to compare values of abiotic (DAF, conductivity, pH, O_2, temperature, and flow velocity) and biotic (AFDM, Chl *a*, biomass of algal groups, density and individual biomass of meiofaunal groups) variables between the 2 study periods. We used Spearman rank correlation analysis to examine correlations between biofilm AFDM and Chl *a* and between proportions of meiofaunal groups and DAF. We used STATISTICA software (version 8.0; StatSoft, Tulsa, Oklahoma) for these analyses.

We used canonical ordination analyses (CANOCO, version 4.5; Biometris, Wageningen, The Netherlands) to assess the influence of biotic and abiotic factors on the density distribution of main meiofaunal groups (rotifers, nematodes, chironomid larvae, and oligochaetes) in the biofilm. We did not consider tardigrades, harpacticoid copepods, and water mites in this analysis because of their low occurrence in samples. We applied the canonical ordination analyses to $\log(x + 1)$-transformed meiofaunal density data for C1 and C2 separately and for pooled C1 and C2 data. First, we used a detrended correspondence analysis (DCA). The total inertia observed was <2.6, so a predominance of linear group response curves could be expected (ter Braak 1987, 1994). Therefore, we used a redundancy analysis (RDA) in which the ordination axes were constrained to be linear combinations of abiotic and biofilm biotic factors to investigate the relationships between these factors and the distribution of main meiofaunal groups. We chose streambed flow velocity (V) over MDD in the RDA because these factors covaried strongly. We listed factors (conditional effects) according to the variance they explained singly (i.e., without eventual covariability with other factors), given by their eigenvalues (λ). We tested for statistical significance with Monte Carlo permutations (499 unrestricted permutations, $\alpha = 0.05$).

Results

Abiotic background

The 2 study periods contrasted hydrologically. Eight critical floods (MDD >300 m^3/s) occurred during C2, and only 4 occurred during C1 (Fig. 1A, B). Three of the critical floods during C2 and 1 during C1 were flash floods caused by heavy rainfall. The durations of the low-water period were 9 mo (June 2005–February 2006) in C1 and 5 mo (June–October 2009) in C2. DAF of sampling occasions differed significantly between periods (Mann–Whitney *U*, $p < 0.05$). Thus, C1 can be considered less disturbed than C2. Among the other abiotic factors, only conductivity and pH differed significantly between periods (Mann–Whitney *U*, conductivity: $p < 0.001$, pH: $p < 0.01$).

Epilithic biofilm and associated microphytes

AFDM and Chl *a* were strongly correlated (pooled C1 and C2; Spearman rank correlation, $n = 75$, $r = 0.72$, $p < 0.001$). They both decreased drastically after critical floods and tended to increase during low-water periods (Fig. 1A, B). Sudden decreases of AFDM and Chl *a* also were observed in July during C1 and C2, but these decreases were not linked to floods.

Diatoms dominated the algal community of the biofilm (especially during winter) in C1 and C2 (Fig. 1C, D). Their relative biomass was lower during C1 than during C2 (71 vs 82%, respectively; Mann–Whitney *U*, $p < 0.01$). In contrast, the relative biomass of green algae was higher during C1 than during C2 (26 vs 15.5%, respectively; Mann–Whitney *U*, $p < 0.05$). The proportion of green algal biomass was highest during the summer–autumn low-water period. Cyanobacteria generally were minor contributors to total microphytobenthic biomass. However, they peaked up to 14–15% in July during both sampling periods (Fig. 1C, D).

During the recolonization periods after the spring snowmelt floods (June 2005–February 2006 and

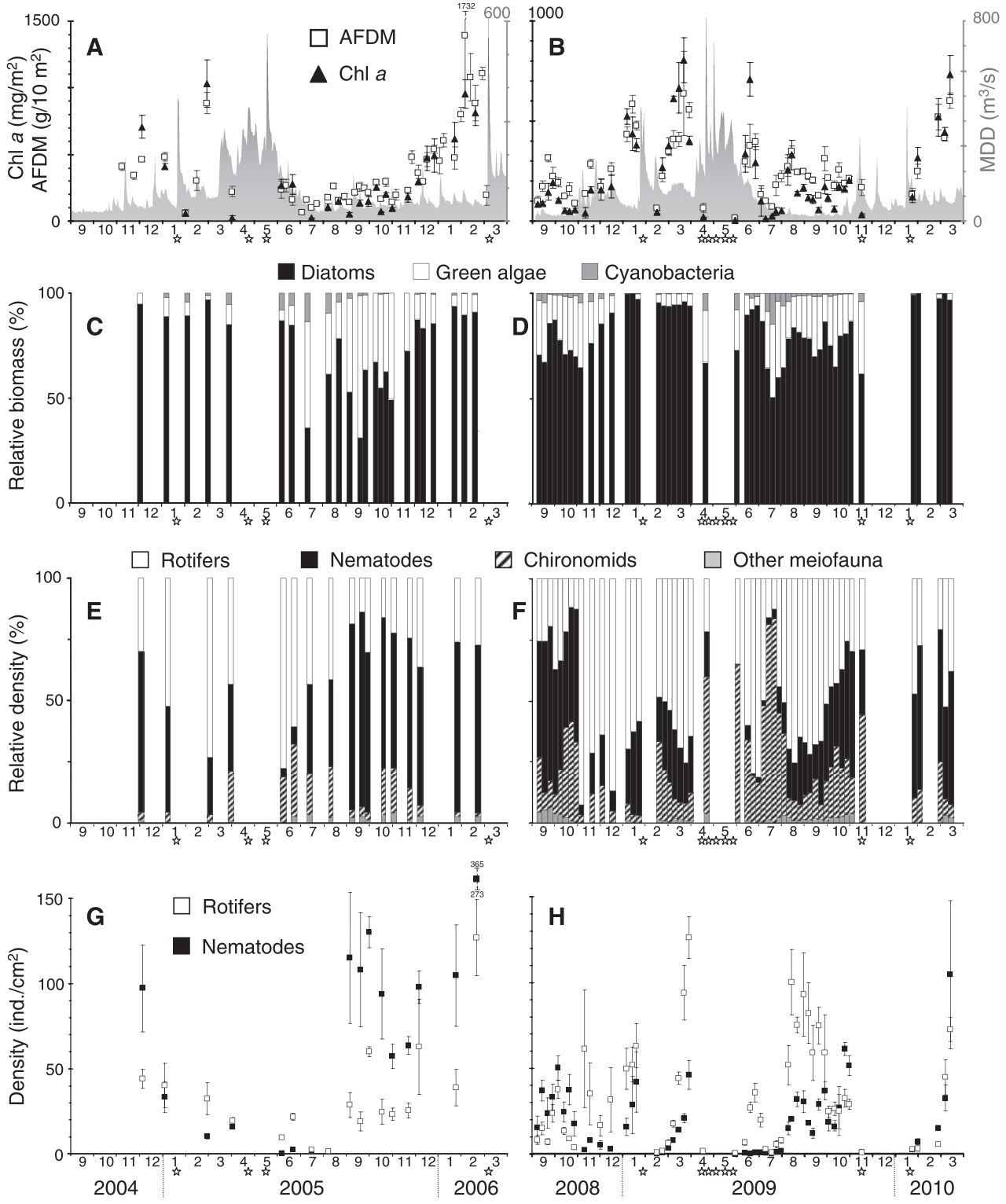

FIG. 1. Mean (±1 SE, $n = 4$) ash-free dry mass (AFDM) of the epilithic biofilm, epilithic chlorophyll *a* concentration (Chl *a*), and daily discharge (MDD) during the 1st (C1) (A) and 2nd (C2) (B) sampling periods; relative biomass of biofilm microalgal groups during C1 (C) and C2 (D); relative density of biofilm-dwelling meiofauna during C1 (E) and C2 (F); and density of biofilm-dwelling nematodes and rotifers during C1 (G) and C2 (H). Critical floods during which mean daily discharge was >300 m^3/s are indicated by stars on *x* axes. Numbers on the *x*-axis represent months of the year (1–12 = January–December, respectively).

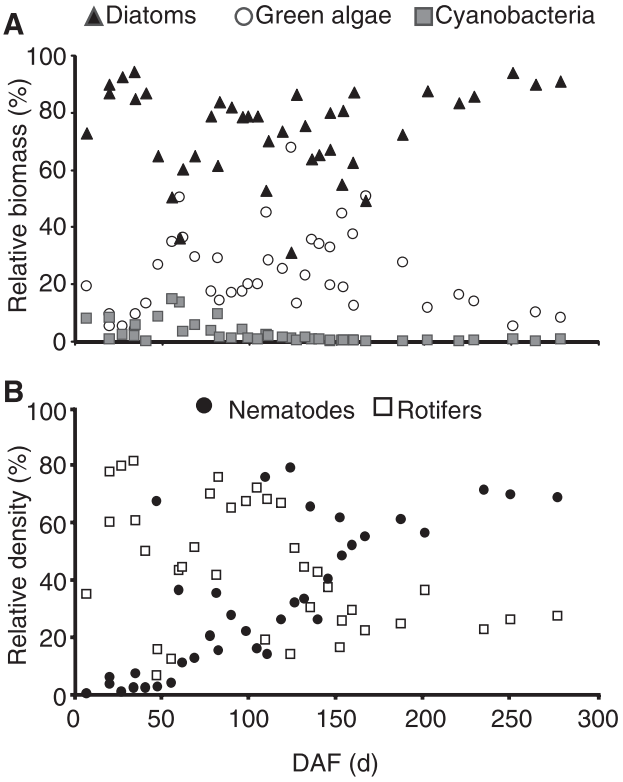

FIG. 2. Relative biomass (n = 41) of diatoms, green microalgae, and cyanobacteria (A), and relative density (n = 37) of nematodes and rotifers (B) in the biofilm relative to the duration of the undisturbed period (days after flood = DAF) after the spring snowmelt floods (pooled data from June 2005–February 2006 and June–November 2009).

June–November 2009), diatom relative biomass was highest during early (10–40 DAF) and late (>200 DAF) successional stages. Relative cyanobacterial biomass peaked at 50–60 DAF, and relative green algal biomass peaked between 50 and 170 DAF, when diatom biomass was relatively low (Fig. 2A).

Composition, density, and biomass of biofilm-dwelling meiofauna

Nematodes and rotifers dominated meiofaunal assemblage density and, on average, accounted for 88% of the total meiofaunal density during C1 and C2 (Fig. 1E, F, Table 1). However, they contributed little to biomass. On average, they accounted for 3.3% of the total meiofaunal biomass, which was dominated by chironomid larvae (66%) and oligochaetes (27%). The means and ranges of density and biomass of meiofaunal-sized chironomid larvae were similar between periods, a result that indicated common patterns of larval development between periods. Chironomid density peaked in October (means ± SE: 28 ± 9, 32 ± 4, 29 ± 9 individuals [ind.]/cm^2 in October 2005, 2008, and 2009, respectively). Chironomid biomass peaked in February 2006 and March 2009 and 2010 because larval DM was high (up to 18 μg/ind.) during these periods (Fig. 3 A, B). Tardigrades, harpacticoid copepods, and water mites were rarely found (Table 1). Nematode and rotifer densities and meiofaunal-sized oligochaete and chironomid biomass decreased drastically after critical floods (Figs 1G, H, 3A, B). However, during July, rotifer density and chironomid biomass decreased suddenly and in the absence of any flood (Figs 1G, H, 3A, B).

Influence of abiotic and biotic factors

The factors (DAF, flow velocity, and conductivity) that significantly influenced the density of the main meiofaunal groups were mainly linked to hydrodynamics (RDA on pooled data from C1 and C2). DAF,

TABLE 1. Mean (n = 17 for C1 and n = 51 for C2) and maximum (Max) density, mean biomass, and relative contribution (%) of each meiofaunal group to the total biofilm-dwelling meiofauna community on cobbles in the Garonne River during 2 study periods (C1 and C2). The resilience times (days to recovery) following winter flash floods (Winter) and spring snowmelt floods (Spring) are for nematodes and rotifers during C2. Ind. = individuals, DM = dry mass.

	C1 period (2004–2006)					C2 period (2008–2010)						
	Density (ind./cm^2)			Biomass (μg DM/ind.)		Density (ind./cm^2)			Biomass (μg DM/ind.)		Resilience (d)	
Meiofauna	Mean	Max	%	Mean	%	Mean	Max	%	Mean	%	Winter	Spring
Nematodes	78	319	65	0.10	8	20	104	32	0.07	2	58–65	148–156
Rotifers	32	127	27	0.02	1	33	126	53	0.03	1	50–58	>340
Chironomids	8	28	6	6.75	56	9	32	14	6.77	82	–	–
Oligochaetes	2	8	2	15.65	32	<1	4	<1	20.16	11	–	–
Harpacticoids	<1	1	<1	0.39	<1	<1	2	<1	0.37	<1	–	–
Tardigrades	<1	4	<1	0.28	<1	<1	1	<1	0.21	<1	–	–
Water mites	<1	<1	<1	81.83	3	<1	1	<1	87.50	4	–	–

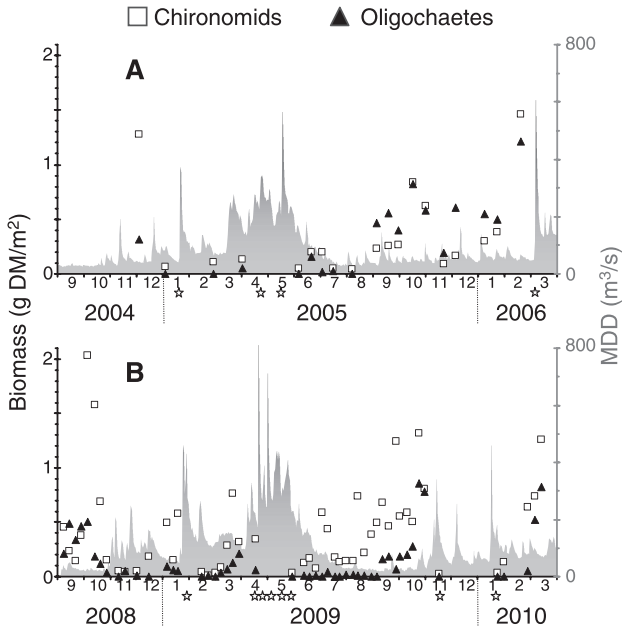

FIG. 3. Dry mass (DM) of meiofaunal chironomid larvae and oligochaetes and mean daily discharge (MDD) during the 1st (C1) (A) and 2nd (C2) (B) sampling periods. Critical floods during which mean daily discharge was >300 m^3/s are indicated by stars on *x* axes.

which can be viewed as an indicator of habitat stability, was the most important predictor of meiofaunal density distribution (Table 2). AFDM and green algal and cyanobacterial biomass, factors related to biofilm status, also significantly influenced meiofaunal density distribution. All meiofaunal groups were on the right side of the biplot (Fig. 4). Axis 1 was correlated mainly with DAF, flow velocity, and cyanobacterial and green algal biomass. Thus, meiofauna were more abundant during stable, undisturbed periods than during disturbed periods. Densities of chironomids, oligochaetes, and particularly nematodes were correlated with DAF (stability), whereas density of rotifers was more strongly correlated with AFDM. RDA analyses done on data C1 and C2 separately gave essentially the same results as the analysis of the total set (not shown).

TABLE 2. Results of the redundancy analysis (RDA) testing the effects of biotic and abiotic factors on the density distribution of biofilm-dwelling meiofauna. Factors are listed by their eigenvalues (λ), i.e., the relative contribution of each factor to the explanation of meiofaunal density variance, without covariability (see Methods). *indicates factors that were statistically significant (Monte Carlo permutation test, $p < 0.05$).

RDA conditional effects		
Factors	λ	*p*-value
Days after flood	0.15	0.002*
Streambed flow velocity	0.07	0.014*
Biofilm ash-free dry mass	0.06	0.014*
Green algae	0.06	0.012*
Cyanobacteria	0.04	0.024*
Conductivity	0.03	0.020*
pH	0.01	0.268
Temperature	0.01	0.420
Diatoms	0.01	0.492
Dissolved O_2	0	0.972
Sum of all λ	0.44	

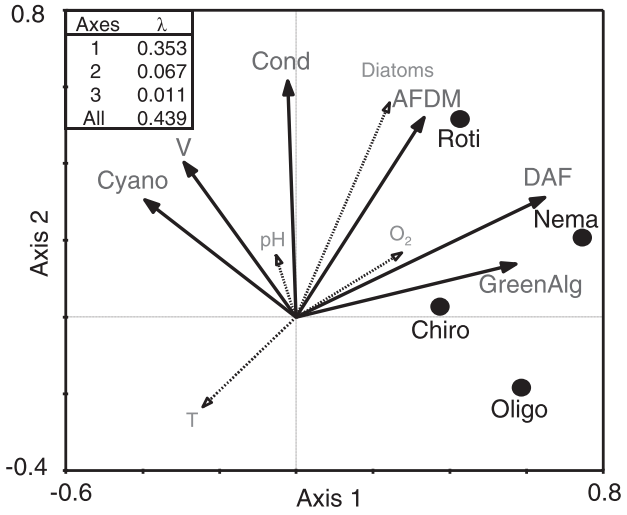

FIG. 4. Redundancy analysis (RDA) biplot showing the density distribution of major meiofaunal taxa under the influence of environmental factors over both sampling periods (C1 and C2). Bold arrows represent statistically significant factors (Monte Carlo permutation test, $p < 0.05$). Slim dotted arrows represent nonsignificant factors. Black points show meiofaunal group positions. The eigenvalues (λ) are indicated for main ordination axes. AFDM = ash-free dry mass of biofilm, GreenAlg = green algae, Cyano = cyanobacteria, DAF = days after flood, V = streambed flow velocity, Cond = conductivity, T = water temperature, Nema = nematodes, Rot = rotifers, Chiro = chironomid larvae, Oligo = oligochaetes.

Response to flood disturbance

Nematodes reached higher average density and biomass during the less-disturbed C1 (Table 1) than during the frequently perturbed C2 (Mann–Whitney *U*, $p < 0.01$, $p < 0.001$, respectively). Mean rotifer density did not differ between C1 and C2, but rotifer biomass was significantly greater during C2 than C1 (Mann–Whitney *U*, $p < 0.05$). These results suggest that nematodes were more negatively affected by the frequency of critical floods than rotifers (Fig. 1E–H).

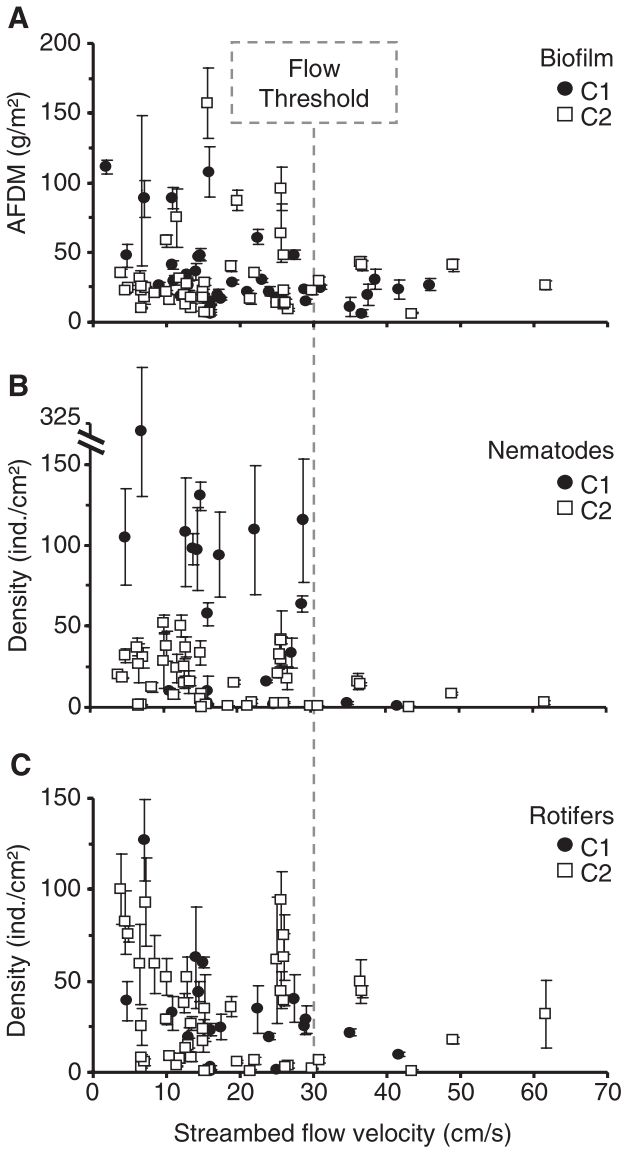

FIG. 5. Mean (±1 SE, $n = 4$) ash-free dry mass (AFDM) of the epilithic biofilm (A), and density of biofilm-dwelling nematodes (B) and rotifers (C) relative to streambed flow velocity during sampling periods (C1 and C2). The vertical dotted line shows the critical flow velocity threshold visually deduced from our data.

Therefore, flood frequency was the main driver of changes in community composition.

Nematodes and rotifers had different resilience times depending on flood type (Table 1). Nematode and rotifer assemblages required more time to recover their preflood densities after snowmelt floods than after flash floods. Mean resilience times after flash floods tended to be lower for rotifers (50–58 d) than for nematodes (58–65 d). During the recolonization periods following spring snowmelt floods (June 2005–February 2006 and June–November 2009), the proportion of nematodes to total meiofauna density was positively correlated with DAF (Spearman rank correlation, $n = 37$, $r = 0.729$, $p < 0.001$), whereas the proportion of rotifers to total density was negatively correlated with DAF ($r = -0.3$, $p < 0.05$). Thus, nematodes and rotifers had different recolonization patterns in the biofilm (Fig. 2B).

Resistance threshold to flow velocity

The method described by Palmer (1992) can be used to deduce critical flow-velocity thresholds from Fig. 5. AFDM reached values >50 g/m^2 only at flow velocities <30 cm/s (Fig. 5A). At higher velocities, AFDM never reached values >43.4 ± 3.8 g/m^2. The mean AFDM reached when velocity was >30 cm/s represented 74% of the mean AFDM reached when velocity was <30 cm/s. A similar resistance threshold of ~30 cm/s was observed for nematodes (Fig. 5B). At velocities >30 cm/s, their density was limited to a maximum of 15 ± 5 ind./cm^2. Rotifer density also tended to be reduced when velocity was >~30 cm/s (Fig. 5C). However, their densities still reached between 7 ± 3 to 50 ± 24 ind./cm^2 at velocities >30 cm/s. Above this flow-velocity threshold, the mean density of nematodes reached only 13.8% of the mean value observed at velocities <30 cm/s (cf. 60.6% for rotifers; Fig. 5B, C). Moreover, nematodes reached their maximum densities only during the less-disturbed C1, but this pattern was not found for rotifers (Figs 5B, 1E–H).

Discussion

We addressed the interaction between hydrological regime and development of biofilm community, considering both its algal and meiofaunal constituents. These factors are clearly linked, but for clarity, we will discuss the aspects essentially related to abiotic factors before discussing the more biotic aspects.

Abiotic factors

Meiofauna were abundant in the epilithic biofilm of the Garonne River, a finding that corroborates the results of the few other studies considering biofilm-dwelling meiofauna in other temperate rivers (Sabater et al. 2003, Gaudes et al. 2006, Kathol et al. 2011). The density distribution of meiofaunal groups depended primarily on biofilm (in)stability imposed by flood disturbance. The frequency of floods and DAF affected the biofilm and its associated meiofauna. Development of biofilm biomass (AFDM), its main microalgae (diatoms and green algae), and its meiofauna are

associated with DAF and flow velocity (Fig. 4). The significance of the conductivity vector seems odd, but can be explained by the covariation of conductivity and temperature. During colder periods, which more or less coincide with flood periods, import of solute ions from the upper drainage basin of the Garonne surpasses the dilution effect of increased runoff and conductivity increases substantially (Probst and Bazerbachi 1986). The hydrological regime in the Garonne River is probably the major determinant of the seasonal distribution of biofilm-dwelling meiofauna because we observed density maxima in winter. In other words, meiofauna peaked when biofilm biomass peaked, and biofilm biomass peaks occurred with increasing periods of stability (DAF). In contrast, density maxima have been reported for spring and summer in most long-term monitoring studies of meiofauna in rivers (Beier and Traunspurger 2003, Stead et al. 2003, Tod and Schmid-Araya 2009). In general, our results agreed with those of other studies pointing out floods as a major shaping force in lotic environments (Lake 2000).

Nematodes were more affected than rotifers by the frequency of critical floods (i.e., habitat instability). Overall, rotifer density was closely coupled to biofilm AFDM, a result suggesting that hydrological scenarios probably influenced rotifer density through biofilm status (e.g., its thickness). Most rotifers consume small algae, protozoans, and bacteria by filtering, scraping, or browsing (Ricci and Balsamo 2000, Kathol et al. 2011). Thus, the abundance of potential food resources within the biofilm might favor rotifer development.

The resilience period of rotifer populations (50–58 d) was slightly shorter than that of nematodes (58–65 d) after flash floods. Gaudes et al. (2010) showed that meiofauna with worm-shaped bodies (e.g., nematodes) have high resilience during recolonization of sediments after a flood. In epilithic biofilms, we showed that rotifers also had high resilience. Rotifers have cilia, short life cycles, parthenogenetic reproduction, and can produce resting eggs or form dormant stages to overcome harsh habitat conditions (Ricci and Balsamo 2000). They can feed on suspended organisms (Kathol et al. 2011) and may be able to take advantage of the increased drifting material shortly after periods of high flow, whereas nematodes would not profit from these circumstances. Moreover, most benthic rotifers found in our study (Bdelloidea and *Proales* spp.) have pedal adhesive glands that secrete a sticky cement used for temporary attachment to the substrate (Ricci and Balsamo 2000). These characteristics apparently render rotifers particularly efficient at recolonizing cobble surfaces cleaned by floods. This ability to colonize early was also observed on submerged wood surfaces (Golladay and Hax 1995). In sediments of artificial stream channels, Smith and Brown (2006) reported very short recolonization periods: i.e., 0.5 and 5 d for rotifers and nematodes, respectively, vs $\geq$50 d in our study. This difference may be caused by differences in flow velocities between the studies. Smith and Brown (2006) used a maximum flow velocity of 12 cm/s, whereas during our study it ranged from 4–62 cm/s, and was >12 cm/s on 62% of the sampling occasions. Palmer et al. (1992) found that meiofaunal density in azoic chambers placed in a 4th-order temperate stream reached values that were 70% of natural stream density within 12 d. The resilience values deduced in our study were within the range of values observed for meiofauna recolonizing sediments of a 3rd-order stream after a flood (42–60 d; Gaudes et al. 2010) and for microcrustaceans recolonizing sediments of a headwater stream after a flood (<54–>243 d; reviewed in Robertson 2000).

Flow velocity also was a significant predictor of the distribution of biofilm-dwelling meiofauna. Palmer (1992) used flume experiments to determine a critical threshold velocity (9–12 cm/s), above which meiofauna (rotifers, oligochaetes, chironomids, and copepods) were removed from the sandy substrate and entered the water column. However, Smith and Brown (2006) reported that a flow velocity = 12 cm/s did not remove meiofauna from gravel substrates in artificial channels. Smith and Brown (2006) suggested that this difference was the result of differences in shear stress needed to displace meiofauna from sand vs from gravel substrates. We found a critical threshold flow velocity ~30 cm/s for nematodes, rotifers, and their biofilm habitat. This result suggests that biofilm-dwelling meiofauna might be more resistant to higher flow velocity than fine-sediment-dwelling meiofauna. However, meiofauna entered the water column when flow velocity was >30 cm/s, probably directly via erosion as particles and indirectly because in detached biofilm fractions. Gaudes et al. (2006) reported that nematodes can be particularly abundant in free-floating biofilm fractions. Streambed flow velocities between 12 and 30 cm/s occurred frequently in the Garonne River (54% of the sampling occasions). Thus, biofilms could serve as refugia for drifting sediment-dwelling meiofauna and could be a source of colonizers for soft-sediment patches in the river bed. Our results also provide support for the idea that epilithic biofilms serve as a refuge for meiofauna (Höckelmann et al. 2004, Mathieu et al. 2007). Interstitial meiofauna can partly resist removal by making small-scale vertical migrations in response to flow variations (Dole-Olivier et al. 1997, Swan and Palmer 2000). Thus, interstitial- and

drifting-meiofauna might be important sources for biofilm recolonization processes after critical floods (i.e., when biofilm is almost totally removed).

Biotic factors

Our results show a close linkage between biofilm biomass (AFDM) and algal biomass (Chl *a*), as is commonly found in the Garonne River (Ameziane et al. 2002, Boulêtreau et al. 2006). Biofilm biomass averaged 34 g AFDM/m^2 and 260 mg Chl *a*/m^2 over the 2 sampling campaigns. These values are high compared to values of 22 g AFDM/m^2 and 77 mg Chl *a*/m^2 from epilithic biofilms of 7 nutrient-rich streams in New Zealand (Biggs 1995). This result strengthens the conclusion that benthic biofilms are important primary producers in the middle reaches of the Garonne and that they probably are an important food source for consumers. However, biofilm biomass suffered when streambed flow velocity exceeded 30 cm/s. Biggs et al. (1998) reported important biofilm biomass losses when flow velocity exceeded 20 cm/s in 5 New Zealand streams, and Poff et al. (1990) reported 30–40× lower biofilm biomass under high (29–41 cm/s) vs low (<1–17 cm/s) flow velocities in troughs connected to the Colorado River.

Flow constraints also determined algal species distribution and succession (Fig. 2A). Diatom relative biomass was highest during early (10–40 DAF) and late succession (>200 DAF). Both green algae and cyanobacteria occurred during middle succession (50–170 DAF), but cyanobacteria were relatively abundant only during a short period (50–60 DAF). Ecological succession of lotic biofilm on artificial substrates occurs after preconditioning by bacteria and organic matter. Early colonists are small diatom species with attachment mechanisms, such as raphes, and succession is completed (>21 d) by filamentous diatoms and green algae (Korte and Blinn 1983, Peterson and Stevenson 1992).

At present, our data do not permit us to determine whether the correlations among microalgal and meiofaunal groups are the result of specific trophic relationships or a common development pattern. Diatoms were the main constituent of the biofilm microalgae during most of both study periods. Diatom abundance was correlated with biofilm biomass (AFDM) and meiofauna (especially rotifer) density. Green algal biomass also was a significant predictor of meiofauna distribution, particularly for nematodes and chironomids. Majdi et al. (2011) examined nematode species distribution in the biofilm during C2 and found correlations between diatom biomass and the distribution of the dominant nematode species *Chromadorina bioculata* (Schultze *in* Carus, 1857) and *Chromadorina viridis* (Linstow, 1876). In our study, the correlations among green algae, nematodes, and chironomids might be explained by the late development of green algae, nematodes, and chironomids after spring snowmelt floods. In contrast, cyanobacteria abundance was negatively correlated with meiofaunal density. Cyanobacteria peaked during July, when rotifer and nematode densities were at their lowest. This negative correlation could be a consequence of a seasonal development cycle of cyanobacteria concomitant with other influences, such as grazing and predation (see below) or temperature-induced self-detachment of the biofilm (Boulêtreau et al. 2006). Moreover, cyanobacteria can produce and release secondary metabolites (Sabater et al. 2003, Leflaive and Ten-Hage 2007) that attract or repel benthic invertebrates, e.g., nematodes (Höckelmann et al. 2004). Thus, we cannot exclude the possibility that cyanobacteria could have a repellent effect on meiofauna. However, considering the relatively minor contribution of cyanobacteria to the biofilm community on most of the sampling occasions, we consider a strong repellent effect unlikely. Both diatoms and green algae are potentially good food sources for meiobenthic organisms (Buffan-Dubau and Carman 2000).

Biofilm and its meiofauna collapsed suddenly in July 2005 and 2009 even though flow was low. Concomitantly, macrofauna crowded the cobbles. In July 2005, mean macroinvertebrate density on cobbles was 12,059 ind./m^2. Large (~5 mm) *Psychomyia pusilla* (Fabricius 1781) (Trichoptera:Psychomyiidae) larvae contributed 71% of the total density (NM, unpublished data). In July 2009, mean density was 11,650 ind./m^2. Psychomyiidae larvae contributed 40% and Ephemeroptera larvae (mainly Baetidae and Ameletidae) contributed 28% of the total density (NM, unpublished data). Psychomyiid larvae construct retreat tubes of small particles held by silk, and they graze the surrounding biofilm by extending their tubes to reach new areas (Wiggins 2004). Their high density and biomass suggests that they could have reduced biofilm biomass directly by grazing or indirectly by destabilizing (bioturbation) deeper biofilm layers. Macrofaunal grazers strongly affect biofilm biomass and community structure (e.g., Feminella and Hawkins 1995, Hillebrand 2009), and meiofauna embedded in biofilm patches can be ingested incidentally by these grazers. However, meiofauna and rotifers can actively migrate from sediment to the water column presumably to avoid predation or habitat disturbances (Palmer et al. 1992, Schmid and Schmid-Araya 1997, Smith and Brown 2006). We speculate that low densities of meiofauna in July could have resulted from indirect predation or from migration of meiofauna subsequent

to depletion of their habitat and resources by macrofaunal competitors.

Overall, the distribution of meiofauna depended primarily on biofilm (in)stability related to flood disturbance. Algal and biofilm biomass were strongly shaped by flow. Densities of nematodes, chironomid larvae, and oligochaetes were related to stability of the biofilm, whereas rotifer density was related to biofilm thickness. These divergences could imply different trophic strategies regarding biofilm resources (e.g., selectivity) that deserve further examination. High grazing activity of macrofaunal insect larvae in early summer could deplete biofilm and weaken meiofauna, but this speculation also needs further examination.

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Literature Cited

Abramoff, M. D., P. J. Magelhaes, and S. J. Ram. 2004. Image processing with ImageJ. Biophotonics International 11: 36–42.

Ameziane, T., A. Dauta, and R. Le Cohu. 2003. Origin and transport of phytoplankton in a large river: the Garonne, France. Archiv für Hydrobiologie 156:385–404.

Ameziane, T., F. Garabetian, D. Dalger, S. Sauvage, A. Dauta, and J. Capblancq. 2002. Epilithic biomass in a large gravel bed river (the Garonne, France): a manifestation of eutrophication? River Research and Applications 18:343–354.

Barlow, R. G., D. G. Cummings, and S. W. Gibb. 1997. Improved resolution of mono- and divinyl chlorophylls a and b and zeaxanthin and lutein in phytoplankton extracts using reverse phase C-8 HPLC. Marine Ecology Progress Series 161:303–307.

Battin, T. J., L. A. Kaplan, J. D. Newbold, and M. E. Hansen. 2003. Contributions of microbial biofilms to ecosystem processes in stream mesocosms. Nature 426:439–441.

Beier, S., and W. Traunspurger. 2003. Temporal dynamics of meiofauna communities in two small submountain carbonate streams with different grain size. Hydrobiologia 498:107–131.

Bergtold, M., and W. Traunspurger. 2005. Benthic production by micro-, meio-, and macrobenthos in the profundal zone of an oligotrophic lake. Journal of the North American Benthological Society 24:321–329.

Biggs, B. J. F. 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, B. J. F., and M. E. Close. 1989. Periphyton biomass dynamics in gravel bed rivers: the relative effects of flows and nutrients. Freshwater Biology 22:209–231.

Biggs, B. J. F., D. G. Goring, and V. I. Nikora. 1998. Subsidy and stress responses of stream periphyton to gradients in water velocity as a function of community growth form. Journal of Phycology 34:598–607.

Boulêtreau, S., F. Garabetian, S. Sauvage, and J. M. Sánchez-Pérez. 2006. Assessing the importance of a self-generated detachment process in river biofilm models. Freshwater Biology 51:901–912.

Buffan-Dubau, E., and K. R. Carman. 2000. Extraction of benthic microalgal pigments for HPLC analyses. Marine Ecology Progress Series 204:293–297.

Caramujo, M. J., C. R. B. Mendes, P. Cartaxana, V. Brotas, and M. J. Boavida. 2008. Influence of drought on algal biofilms and meiofaunal assemblages of temperate reservoirs and rivers. Hydrobiologia 598:77–94.

Cardinale, B. J. 2011. Biodiversity improves water quality through niche partitioning. Nature 472:86–89.

Cardinale, B. J., M. A. Palmer, A. R. Ives, and S. S. Brooks. 2005. Diversity–productivity relationships in streams vary as a function of the natural disturbance regime. Ecology 86:716–726.

Carpenter, S. R., N. F. Caraco, D. L. Correll, R. W. Howarth, A. N. Sharpley, and V. H. Smith. 1998. Nonpoint pollution of surface waters with phosphorus and nitrogen. Ecological Applications 8:559–568.

Charlebois, P. M., and G. A. Lamberti. 1996. Invading crayfish in a Michigan stream: direct and indirect effects on periphyton and macroinvertebrates. Journal of the North American Benthological Society 15:551–563.

Chauvet, E., and H. Décamps. 1989. Lateral interactions in a fluvial landscape: the river Garonne, France. Journal of the North American Benthological Society 8:9–17.

Cobb, D. G., T. D. Galloway, and J. F. Flannagan. 1992. Effects of discharge and substrate stability on density and species composition of stream insects. Canadian Journal of Fisheries and Aquatic Sciences 49:1788–1795.

Costerton, J. W. 2000. Biofilms in the new millennium: musings from a peak in Xanadu. Pages 329–344 *in* D. G. Allison, P. Gilbert, H. M. Lappin-Scott, and M. Wilson (editors). Community structure and co-operation in biofilms. Cambridge University Press, Cambridge, UK.

Death, R. G. 2002. Predicting invertebrate diversity from disturbance regimes in forest streams. Oikos 97:18–30.

Death, R. G., and M. J. Winterbourn. 1995. Diversity patterns in stream benthic invertebrate communities: the influence of habitat stability. Ecology 76:1446–1460.

Dole-Olivier, M. J., P. Marmonier, and J. L. Beffy. 1997. Response of invertebrates to lotic disturbance: is the hyporheic zone a patchy refugium? Freshwater Biology 37:257–276.

Feminella, J. W., and C. P. Hawkins. 1995. Interactions between stream herbivores and periphyton: a quantitative analysis of past experiments. Journal of the North American Benthological Society 14:465–509.

Fuller, R. L., J. L. Roelofs, and T. J. Fry. 1986. The importance of algae to stream invertebrates. Journal of the North American Benthological Society 5:290–296.

Gaudes, A., J. Artigas, and I. Muñoz. 2010. Species traits and resilience of meiofauna to floods and drought in a Mediterranean stream. Marine and Freshwater Research 61:1336–1347.

Gaudes, A., S. Sabater, E. Vilalta, and I. Muñoz. 2006. The nematode community in cyanobacterial biofilms in the river Llobregat, Spain. Nematology 8:909–919.

Giere, O. 2009. Meiobenthology: the microscopic motile fauna of aquatic sediments. Springer-Verlag, Berlin, Germany.

Golladay, S. W., and C. L. Hax. 1995. Effects of an engineered flow disturbance on meiofauna in a north Texas prairie stream. Journal of the North American Benthological Society 14:404–413.

Hillebrand, H. 2009. Meta-analysis of grazer control of periphyton biomass across aquatic ecosystems. Journal of Phycology 45:798–806.

Höckelmann, C., T. Moens, and F. Jüttner. 2004. Odor compounds from cyanobacterial biofilms acting as attractants and repellents for free-living nematodes. Limnology and Oceanography 49:1809–1819.

Kathol, M., H. Fischer, and M. Weitere. 2011. Contribution of biofilm-dwelling consumers to pelagic–benthic coupling in a large river. Freshwater Biology 56:1160–1172.

Korte, V. L., and D. W. Blinn. 1983. Diatom colonization on artificial substrata in pool and riffle zones studied by light and scanning electron microscopy. Journal of Phycology 19:332–341.

Lai, J. S., and H. W. Shen. 1996. Flushing sediment through reservoirs. Journal of Hydraulic Research 34:237–255.

Lake, P. S. 2000. Disturbance, patchiness, and diversity in streams. Journal of the North American Benthological Society 19:573–592.

Lake, P. S. 2003. Ecological effects of perturbation by drought in flowing waters. Freshwater Biology 48: 1161–1172.

Lawrence, J. R., B. Scharf, G. Packroff, and T. R. Neu. 2002. Microscale evaluation of the effects of grazing by invertebrates with contrasting feeding modes on river biofilm architecture and composition. Microbial Ecology 44:199–207.

Leflaive, J., S. Boulêtreau, E. Buffan-Dubau, and L. Ten-Hage. 2008. Temporal patterns in epilithic biofilm-relation with a putative allelopathic activity. Fundamental and Applied Limnology 173:121–134.

Leflaive, J., and L. Ten-Hage. 2007. Algal and cyanobacterial secondary metabolites in freshwaters: a comparison of allelopathic compounds and toxins. Freshwater Biology 52:199–214.

Liess, A., and H. Hillebrand. 2004. Invited review: direct and indirect effects in herbivore-periphyton interactions. Archiv für Hydrobiologie 159:433–453.

Lock, M. A. 1993. Attached microbial communities in rivers. Pages 113–138 *in* T. E. Ford (editor). Aquatic microbiology: an ecological approach. Blackwell Scientific Publications, Boston, Massachusetts.

Lyautey, E., S. Boulêtreau, E. Y. Madigou, and F. Garabetian. 2010. Viability of differentiated epilithic bacterial communities in the River Garonne (SW France). Hydrobiologia 637:207–218.

Lyautey, E., C. R. Jackson, J. Cayrou, J. L. Rols, and F. Garabetian. 2005. Bacterial community succession in natural river biofilm assemblages. Microbial Ecology 50:589–601.

Mackey, M. D., D. J. Mackey, H. W. Higgins, and S. W. Wright. 1996. CHEMTAX - a program for estimating class abundances from chemical markers: application to HPLC measurements of phytoplankton. Marine Ecology Progress Series 144:265–283.

Majdi, N., W. Traunspurger, S. Boyer, B. Mialet, M. Tackx, R. Fernandez, S. Gehner, L. Ten-Hage, and E. Buffan-Dubau. 2011. Response of biofilm-dwelling nematodes to habitat changes in the Garonne River, France: influence of hydrodynamics and microalgal availability. Hydrobiologia 673:229–244.

Mathieu, M., J. Leflaive, L. Ten-Hage, R. de Wit, and E. Buffan-Dubau. 2007. Free-living nematodes affect oxygen turnover of artificial diatom biofilms. Aquatic Microbial Ecology 49:281–291.

Palmer, M. A. 1992. Incorporating lotic meiofauna into our understanding of faunal transport processes. Limnology and Oceanography 37:329–341.

Palmer, M. A., A. E. Bely, and K. E. Berg. 1992. Response of invertebrates to lotic disturbance: a test of the hyporheic refuge hypothesis. Oecologia (Berlin) 89:182–194.

Peters, L., H. Hillebrand, and W. Traunspurger. 2007. Spatial variation of grazer effects on epilithic meiofauna and algae. Journal of the North American Benthological Society 26:78–91.

Peters, L., and W. Traunspurger. 2005. Species distribution of free-living nematodes and other meiofauna in littoral periphyton communities of lakes. Nematology 7: 267–280.

Peterson, C. G. 1996. Response of benthic algal communities to natural physical disturbance. Pages 375–403 *in* R. J. Stevenson, M. L. Bothwell, and R. L. Lowe (editors). Algal ecology: freshwater benthic ecosystems. Academic Press, San Diego, California.

Peterson, C. G., and R. J. Stevenson. 1992. Resistance and resilience of lotic algal communities: importance of disturbance timing and current. Ecology 73:1445–1461.

Pfannkuche, O., and H. Thiel. 1988. Sample processing. Pages 134–145 *in* R. P. Higgins and H. Thiel (editors). Introduction to the study of meiofauna. Smithsonian Institution Press, Washington, DC.

Poff, N. L., N. J. Voelz, J. V. Ward, and R. E. Lee. 1990. Algal colonization under four experimentally controlled current regimes in high mountain stream. Journal of the North American Benthological Society 9:303–318.

Probst, J. L., and A. Bazerbachi. 1986. Transport en solution et en suspension par la Garonne supérieure. Sciences Géologiques Bulletin 39:79–98.

Reiss, J., J. Forster, F. Cassio, C. Pascoal, R. Stewart, and A. G. Hirst. 2010. When microscopic organisms inform general ecological theory. Pages 45–85 *in* G. Woodward

(editor). Integrative biology: from molecules to ecosystems. Academic Press, Oxford, UK.

Reiss, J., and J. M. Schmid-Araya. 2008. Existing in plenty: abundance, biomass and diversity of ciliates and meiofauna in small streams. Freshwater Biology 53:652–668.

Reiss, J., and J. M. Schmid-Araya. 2010. Life history allometries and production of small fauna. Ecology 91: 497–507.

Ricci, C., and M. Balsamo. 2000. The biology and ecology of lotic rotifers and gastrotrichs. Freshwater Biology 44: 15–28.

Robertson, A. L. 2000. Lotic meiofaunal community dynamics: colonisation, resilience and persistence in a spatially and temporally heterogeneous environment. Freshwater Biology 44:135–147.

Sabater, S., E. Vilalta, A. Gaudes, H. Guasch, I. Munoz, and A. Romani. 2003. Ecological implications of mass growth of benthic cyanobacteria in rivers. Aquatic Microbial Ecology 32:175–184.

Schmid, P. E., and J. M. Schmid-Araya. 1997. Predation on meiobenthic assemblages: resource use of a tanypod guild (Chironomidae, Diptera) in a gravel stream. Freshwater Biology 38:67–91.

Schmid-Araya, J. M. 1994. Temporal and spatial distribution of benthic microfauna in sediments of a gravel streambed. Limnology and Oceanography 39:1813–1821.

Schmid-Araya, J. M. 1997. Temporal and spatial dynamics of meiofaunal assemblages in the hyporheic interstitial of a gravel stream. Pages 29–36 *in* J. Gibert, J. Mathieu, and F. Fournier (editors). Groundwater/surface water ecotones: biological and hydrological interactions and management options. Cambridge University Press, Cambridge, UK.

Schmid-Araya, J. M., A. G. Hildrew, A. Robertson, P. E. Schmid, and J. Winterbottom. 2002. The importance of meiofauna in food webs: evidence from an acid stream. Ecology 83:1271–1285.

Schmid-Araya, J. M., and P. E. Schmid. 2000. Trophic relationships: integrating meiofauna into a realistic benthic food web. Freshwater Biology 44:149–163.

Smith, F., and A. V. Brown. 2006. Effects of flow on meiofauna colonization in artificial streams and reference sites within the Illinois river, Arkansas. Hydrobiologia 571:169–180.

Spieth, H. R., T. Möller, C. Ptatschek, A. Kazemi-Dinan, and W. Traunspurger. 2011. Meiobenthos provides a food resource for young cyprinids. Journal of Fish Biology 78: 138–149.

Stead, T. K., J. M. Schmid-Araya, and A. G. Hildrew. 2003. All creatures great and small: patterns in the stream benthos across a wide range of metazoan body size. Freshwater Biology 48:532–547.

Stead, T. K., J. M. Schmid-Araya, and A. G. Hildrew. 2005. Secondary production of a stream metazoan community: does the meiofauna make a difference? Limnology and Oceanography 50:398–403.

Swan, C. M., and M. A. Palmer. 2000. What drives small-scale spatial patterns in lotic meiofauna communities? Freshwater Biology 44:109–121.

Teissier, S., M. Torre, F. Delmas, and F. Garabetian. 2007. Detailing biogeochemical N budgets in riverine epilithic biofilms. Journal of the North American Benthological Society 26:178–190.

ter Braak, C. J. F. 1987. Ordination. Pages 91–173 *in* R. H. G. Jongman, C. J. F. ter Braak, and O. F. R. van Tongeren (editors). Data analysis in community and landscape ecology. Cambridge University Press, Cambridge, UK.

ter Braak, C. J. F. 1994. Canonical community ordination part I: basic theory and linear methods. Ecoscience 1: 127–140.

Tod, S. P., and J. M. Schmid-Araya. 2009. Meiofauna versus macrofauna: secondary production of invertebrates in a lowland chalk stream. Limnology and Oceanography 54:450–456.

van der Wurff, A. W. G., S. A. E. Kools, M. E. Y. Boivin, P. J. Van den Brink, H. H. M. van Megen, J. A. G. Riksen, A. Doroszuk, and J. E. Kammenga. 2007. Type of disturbance and ecological history determine structural stability. Ecological Applications 17:190–202.

Villenave, C., T. Bongers, K. Ekschmitt, D. Djigal, and J. L. Chotte. 2001. Changes in nematode communities following cultivation of soils after fallow periods of different length. Applied Soil Ecology 17:43–52.

Wiggins, G. B. 2004. Caddisflies: the underwater architects. University of Toronto Press, Toronto, Canada.

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