



Role of biodiversity in the biogeochemical processes at the water-sediment interface of macroporous river bed: An experimental approach



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ABSTRACT

This study highlights the effects of interaction between microbial, macro- and meiofauna on NO_3^- -N and DOC reduction in macroporous stream sediment. The tested hypotheses are: the transformation of nutrients and dissolved organic matter (1) is influenced by the presence of invertebrates, (2) is more effective when the diversity of the vertical benthic community increases.

These hypotheses were tested using microcosms reproducing a portion of a river bed water-sediment interface that was colonized with different levels of invertebrate biodiversity. Experimental treatments were abiotic sediment (AS); sediment and biofilm (SB); sediment, biofilm and meiofauna (SBM); and sediment, biofilm, meiofauna and macrofauna community assemblage, which corresponds to the total benthic community of a river bed (SBMM). Reduction rates of nitrates (NO_3^- -N) and dissolved organic carbon (DOC) in the microcosms were measured and considered as a function of the different levels of biodiversity. Nutrient reduction rates were monitored by their decrease from the aqueous phase. Nitrate reduction rates increased significantly with increasing the vertical biodiversity level. After 56 days of biofilm development, NO_3^- -N reduction rates ranged from 3.76 ± 0.35 in SB treatment to 8.92 ± 0.69 $\text{mg N d}^{-1} \text{kg}^{-1}$ sediment Fresh Weight (sed FW) in the treatment with the maximum biodiversity (SBMM). Denitrification rates increased by a factor of 6 in presence of meiofauna and macrofauna compared to that measured in sediment without invertebrates. DOC reduction rates also varied significantly with biodiversity levels but in a lesser extent than nitrate reduction rates (41.89 ± 2.24 $\text{mg C d}^{-1} \text{kg}^{-1}$ sed FW with biofilm alone (SB) to 51.00 ± 1.39 $\text{mg C d}^{-1} \text{kg}^{-1}$ sed FW with the addition of meiofauna community (SBM).

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1. Introduction

Self-purifying capacity or water purification of rivers, as a part of the waste assimilation ecosystem service of regulation, is defined

as their ability to eliminate or breakdown excessive nutrients and compounds that flow in the natural water (Costanza et al., 1997; Haines-Young and Potschin, 2013). In a context of markedly increased nitrogen and carbon loadings in most of the surface water worldwide (Craig et al., 2008; Noe and Hupp, 2008), the study of the river purification capacity associated to nutrient reduction by sediments remains a relevant research domain. A focus on nitrogen and carbon reduction capacities of rivers leads to identification of river compartments including their physical, chemical and biological properties that actively participate to the nutrient transformation pathways.

In rivers, some hydromorphological characteristics tend to facilitate biological and microbiological activities in the free flowing water. For examples, the conditions of (1) low water depth, large proportions of runs and riffles and (2) high granulometry (mainly

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composed of pebbles/gravels) and favour autotrophic biofilm development (Ameziane et al., 2002; Battin, 2000; Sauvage et al., 2003). However, when a hyporheic zone exists, the biofilm biomass may be largely extended with heterotrophic metabolisms in the sediment. This interstitial and attached biomass is composed of bacteria, protozoans and fungi. This biofilm is regarded as an important organic matter storage site and absorption site for dissolved organic matter (DOM) owing to its large internal surface area (Koutny and Rulik, 2007). It is recognized to be the main driver of the carbon and nutrient reduction as required for biomass production and respiration (Baker et al., 2000; Battin et al., 2008). Nitrogen and carbon reduction capacities are now established both for autotrophic biofilm (Majdi et al., 2012b; Mulholland et al., 2004; Ribot et al., 2013; Teissier et al., 2007) and for heterotrophic biofilm in gravel bed sediments (Dahm et al., 1998; Iribar et al., 2015, 2008; Peyrard et al., 2008). The hyporheic zone, a transition zone between groundwater and streams (Orghidan, 1959), is now known as a site of high biological heterotrophic activity that is critical for stream ecosystem functioning (Boulton et al., 2010, 1998; Nogaro et al., 2013). It is an important site for mineralization of organic matter from surface waters. The importance of the hyporheic participation to the global nutrient processing in a river depends, among other factors, on the intensity of ground water/surface water (GW/SW) exchanges linked to the porosity or the clogging of sediment. The permanent water flow through these transition zones explains why hyporheic biogeochemical processes are essential for mediating the chemical quality of adjacent water compartments (Boulton et al., 1998; Janauer, 2000; Sánchez Pérez et al., 2009; Vervier et al., 2009).

One of the major questions concerning the role of hyporheic zones is how and to what extent biodiversity that lives in this habitat is contributing to the riverine ecosystem functioning and resilience. The activities and biodiversity of benthic invertebrates are closely connected to microbial functions and related biogeochemical processes in river beds. The general hypothesis is that biodiversity contributes positively to ecosystem processes and represents an insurance against environmental variations and disturbances (Loreau et al., 2001). Bioturbation, as an inherent benthic activity directly influences the physical structure and consequently the biological and chemical nature of sediments. In fine sediments, the biogeochemical processes dominated by microbial activity are tightly linked to macrofauna and meiofauna. They are (1) particle and solute displacements driven by macrofauna (François et al., 2002; Gerino et al., 2003), (2) agglutination of detritus particles by mucus secretions or proteolytic capacity stimulated by meiofauna (e.g. Nascimento et al., 2012; Riemann and Helmke, 2002). In macro-porous hyporheic sediments, where particle sizes are similar or larger than those of benthic organisms, bioturbation is mainly performed by biofilm consumers and galleries diggers that modify sediment porosity (Mermillod-Blondin et al., 2003; Mermillod-Blondin and Rosenberg, 2006; Nogaro et al., 2007). A change in porosity may thus influence (1) pore water flow and the associated solutes transport, (2) microbial metabolism pathways and intensities, and consequently (3) solutes reduction.

Nutrient cycling and organic matter transformation within the hyporheic zone are mediated mainly by microorganisms which account for over 90% of the community respiration (Pusch, 1996). However, these microorganisms are under a top-down control by organisms of higher trophic level such as scraping or shredding invertebrates (Saleem et al., 2016; Stief, 2013). So interactions between microbial and invertebrate communities could be considered as a controlling factor for biochemical processes (Nogaro et al., 2008). Furthermore, the diversity of invertebrates could also favour these processes (and thus the self-purification capacity of hyporheic zone) (Nogaro et al., 2007). Influences of cross-community interactions (i.e. microorganisms-meiofauna-macrofauna) have been studied in ecosystemic description of

energy fluxes and trophic webs by *in situ* investigations in autotrophic biofilms (Majdi et al., 2012b). Nevertheless, still few well controlled experiments in the literature have explored the effects of this biodiversity on ecosystem function e.g. excessive N load transformation and organic matter degradation (Lillebø et al., 1999; Marshall and Hall, 2004; Webb and Montagna, 1993) in heterotrophic biofilms.

The objective of this paper is to characterize the impact of biodiversity and cross-community efficiency on the ecological processes at the subsurface–surface water. Specifically, this study will consist in characterizing the role of cross-communities (biofilm, meiofauna, macrofauna) diversity on the reduction of nitrates and dissolved organic carbon in hyporheic sediments.

2. Materials and methods

The methodology implemented here relies on laboratory experimentation through the use of microcosms i.e. sediment columns with water circulation to mimic a river hyporheic ecosystem. To test the role of biodiversity on nitrogen and carbon reduction rates, analysis of these elements were performed in water flowing through a series of microcosms reproducing a portion of water-sediment interface. The effects of community combinations in microcosms were tested by comparison of several experimental conditions setting a gradient of increasing communities numbers.

2.1. Microcosm design

The microcosm design was following our previous study as described in Sánchez Pérez et al. (2013). 20 Plexiglas columns (height: 20 cm, internal diameter: 6.8 cm) were independently connected to water tanks to form 20 experimental units or microcosms (Fig. 1a). Abiotic sediment columns were filled with sand and gravel in four successive layers. Their particulate sizes were in ranges of 0.5–1 mm, 1–2 mm, 2–10 mm and 10–20 mm. The thickness of each layer was 2 cm, which was sieved manually with the corresponding mesh before being autoclaved (20 min at 121 °C). The total mass of sediment in each microcosm was 1000 ± 50 g. Mean porosity was $34 \pm 3\%$. A 300 μm filter was placed at the exit of the microcosm to maintain the sediment in the column. Silicone tubes (internal diameter = 3.2 mm) were used for connection to a high-density polyethylene tank with 15 L filtered water (90 μm) from the Garonne River (France). The water was collected before the beginning of the experiment in the Garonne River on April 2008, and conserved in a cold room at 4 °C. Peristaltic pumps (323Du Watson Marlow) were responsible for a downward water circulation in microcosms, realizing a constant infiltration flow rate of 7–8 mL min^{-1} (Darcian velocity = 1.39–1.59 m d^{-1}) similar to the *in situ* range of water flow in hyporheic sediments (Peyrard et al., 2008; Sánchez Pérez et al., 2003; Weng et al., 2003). Supplied water was aerated in tanks to maintain oxygen saturation. All the microcosm-setups ($n=20$) were placed in a dark room to avoid phototrophic biofilm development. Room temperature was fixed at 15 ± 0.5 °C.

2.2. Experimental design

2.2.1. Treatment setup

The experimental design is shown in Fig. 1b. Four different biodiversity levels were set in the microcosms to allow comparison of their functioning: abiotic sediment (AS); sediment and biofilm (SB); sediment, biofilm and meiofauna (SBM); sediment, biofilm, meiofauna and macrofauna community assemblage that corresponds to the total benthic community of a river bed (SBMM). Water circulation was activated in a total of 16 microcosms. After 40 days of incubation, these microcosms were assigned to SB. Another 4

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