



Sedimentary context controls the influence of ecosystem engineering by bioturbators on microbial processes in river sediments

Simon Navel, Florian Mermillod-Blondin, Bernard Montuelle, Eric Chauvet,
Pierre Marmonier

► To cite this version:

Simon Navel, Florian Mermillod-Blondin, Bernard Montuelle, Eric Chauvet, Pierre Marmonier. Sedimentary context controls the influence of ecosystem engineering by bioturbators on microbial processes in river sediments. *Oikos*, 2012, 121 (7), pp.1134-1144. 10.1111/j.1600-0706.2011.19742.x . halsde-00719692

HAL Id: halsde-00719692

<https://hal.science/halsde-00719692>

Submitted on 15 May 2020

HAL is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.

**Sedimentary context controls the influence of ecosystem engineering by bioturbators on
microbial processes in river sediments**

SIMON NAVEL¹, FLORIAN MERMILLOD-BLONDIN¹, BERNARD MONTUELLE^{2,5},
ERIC CHAUVET^{3,4} & PIERRE MARMONIER¹

E-mail addresses: simon.navel@univ-lyon1.fr, mermillo@recherche.univ-lyon1.fr,
bernard.montuelle@thonon.inra.fr, echauvet@cict.fr and pierre.marmonier@univ-lyon1.fr,
respectively.

¹ Université de Lyon, F-69000, Lyon ; Université Lyon 1 ; CNRS, UMR 5023, LEHNA -
Laboratoire d'Ecologie des Hydrosystèmes Naturels et Anthropisés, F-69622, Villeurbanne
Cedex, France.

² Cemagref, CEMAGREF Lyon, 3 bis quai Chauveau, CP 220, 69336 LYON Cedex 09,
France

³ Université de Toulouse; UPS, INP; EcoLab (Laboratoire écologie fonctionnelle et
environnement); 118 route de Narbonne, F-31062 Toulouse cedex 9, France

⁴ CNRS; EcoLab; F-31055 Toulouse cedex 4, France

⁵ present adress: INRA- UMR CARTEL, 75 av. de CORZENT - BP 511, 74203 THONON
Cedex, France

Correspondence: Simon NAVEL, Present adress : UMR - CNRS 5023 Laboratoire
d'Ecologie des Hydrosystèmes Fluviaux - Université Claude Bernard Lyon1 ; 6 rue Dubois,
Campus de la Doua ; 69622 Villeurbanne Cedex, FRANCE.

E-mail : simon.navel@univ-lyon1.fr

1 By modifying the physical environment, ecosystem engineers can have inordinately large
2 effects on surrounding communities and ecosystem functioning. However, the significance of
3 engineering in ecosystems greatly depends on the physical characteristics of the engineered
4 habitats. Mechanisms underlying such context-dependent impact of engineers remain poorly
5 understood whereas they are crucial to establish general predictions concerning the
6 contribution of engineers to ecosystem structure and function.

7 The present study aimed to decrypt such mechanisms by determining how the environmental
8 context modulates the effects of ecosystem engineers (bioturbators) on micro-organisms in
9 river sediments. To test the effects of environmental context on the role of bioturbators in
10 sediments, we used microcosms and recreated two sedimentary contexts at the laboratory by
11 adding a layer of either fine (low permeability) or coarse (high permeability) sand particles at
12 the top of a gravel-sand matrix. For each sediment context, we examined how the sediment
13 reworking activity of a bioturbating tubificid worm (*Tubifex tubifex*) generated changes of the
14 physical (sediment structure and permeability) and abiotic environments (hydraulic discharge,
15 water chemistry) for micro-organisms. The biotic influences of the bioturbation process were
16 measured on microbial characteristics (abundances, activities) and leaf litter decomposition as
17 a major microbe-mediated ecological process.

18 Results showed that *T. tubifex* significantly increased hydraulic discharge (by about 6-fold)
19 and restore aerobic conditions in O₂-limited sedimentary habitats covered by fine sand (low
20 permeability treatment). Consequently, worms stimulated microbial communities developed
21 on buried leaves and leaf litter breakdown increased (+30%). In contrast *T. tubifex* had a low
22 influence on water exchanges and O₂ availability in highly permeable sediments. As
23 bioturbation did not modify the abiotic environment in sedimentary habitats covered by
24 coarse sand, *T. tubifex* did not influence the abundances and activities of microorganisms
25 developed on leaves.

Our study demonstrated that the significance of ecosystem engineering on the functioning of aquatic ecosystems cannot be quantified without assessing the complex interactions between bioturbation activities and sedimentary characteristics. We strongly suggest that context dependency mainly modulates the effects of ecosystem engineers on other biota by controlling the ability of engineers to modulate the availability of limiting factors for other organisms.

Introduction

Habitat modification by engineer organisms has been recognized as a major ecological process with large consequences for biodiversity and ecosystem functions in a broad range of ecosystems (e.g. see Lavelle et al 1997, Crooks 2002, Mermillod-Blondin and Rosenberg 2006, Wright and Jones 2006, Wright et al. 2006, Badano and Marquet 2008, Gutiérrez et al. in press). Ecosystem engineers can have inordinately large effects on communities by modifying their surrounding physical environment (Jones et al. 1994, see Fig.1 for detailed engineering sequence). The beaver (*Castor canadensis*) is a classical example of ecosystem engineer. Its activities (essentially dam-building) can increase (i) the proportion of flooded soils (water and wetlands) in the landscape (Johnston and Naiman 1990) and (2) the retention of sediment, organic material (Naiman et al. 1986) and nutrients (Naiman and Melillo 1984) in the channel by decreasing water velocity, ultimately affecting the structure of animal and plant communities in the landscape (Naiman et al. 1988, Hägglund and Sjöberg 1999, Wright et al. 2002, Anderson and Rosemond 2007). Beside this emblematic example of beavers, it exists a wide diversity of engineer organisms and then engineering mechanisms (Berke 2010) that significantly impact structure and functions. For example, the sediment reworking activities of bioturbators can have marked effects on microbial communities developed on

1 sediments by affecting hydrological fluxes and biogeochemistry at water-sediment interface
2 (Mermillod-Blondin and Rosenberg, 2006; Mermillod-Blondin et al. 2004, Nogaro et al.
3 2009). Whatever the engineering mechanisms (e.g. through bioturbation, physical engineering
4 ...), a change in resource availability relative to the unmodified state may suffice to observe
5 positive or adverse engineering effects on some biotic variables (e.g. absolute and relative
6 abundance and richness of species in the surrounding communities) describing the structure of
7 communities (Bertness 1984a,b, Jones et al. 1997, Menge 2000). The theory of ecosystem
8 engineers also suggests that the highest effects of engineers on physical habitat produced the
9 highest changes of biotic variables (Jones et al. 1994). However, the influence of an
10 ecosystem engineering activity also depends on the environmental context in which it happens
11 (Crain and Bertness 2006, Wright and Jones 2006). Because ecosystem engineers affect
12 community through environmentally mediated interactions, a given engineering process
13 (similar density of a given engineer species) can have contrasted effects on some biotic
14 variables across environmental gradients and thus can appear as idiosyncratic (Jones et al.
15 2004, Moore 2006). However, most of studies dealing with ecosystem engineering only
16 examine the relationships between the engineer (e.g. number of individuals) and (i) the
17 physical characteristics of the environment or (ii) the other species, in a unique environmental
18 context. To establish general principles and predictions (for future changes) about the effects
19 of engineers on communities, there is a need to fully understand mechanisms underlying
20 context dependency and thus to examine the whole ‘cause-effect relationships’ sequence (see
21 Fig. 1) in contrasted environments.

22 In the present study, we aimed to tackle this context dependency in river sediments. The
23 functioning of lotic ecosystems partly depends on the microbially-mediated biogeochemical
24 processes (nutrient cycling, organic matter (hereafter OM) processing) realized in the
25 hyporheic zone (hereafter HZ, sedimentary interface between surface water and groundwater)

(Grimm and Fisher 1984, Findlay 1995, Naegeli and Uehlinger 1997, Boulton et al. 1998, Fellows et al. 2001). By controlling hydrological exchanges between the surface and the sediments, and thus chemical conditions (e.g. availability of dissolved oxygen DO) in the HZ, sediment characteristics such as permeability appears as crucial factors controlling the structure and activities of microbial communities developed on river sediments (e.g. Valett et al. 1990, Brunke and Gonser 1997, Mermillod-Blondin and Rosenberg 2006). In this context, bioturbators can have major influence on the HZ (and *a fortiori* on the whole-stream) ecological functioning through modification of sediment structure and permeability (Nogaro et al. 2009, Nogaro & Mermillod-Blondin 2009). However, the magnitude of bioturbation-driven change in hydraulic conductivity is expected to depend on physical hydraulic conditions which are more or less constraining for micro-organisms living in HZ (Hakenkamp and Palmer 2000, Mermillod-Blondin, in press). As physical hydraulic conditions are essentially linked with sediment characteristics, the effects of bioturbators as ecosystem engineers in river sediment are expected to be sedimentary-context dependent. By applying the conceptual framework of ecosystem engineering in rivers, bioturbators are expected to major influence on hyporheic ecological processes in sedimentary systems where: (1) they are able to drastically modify habitat physical characteristics by actively reworking sediment, (2) these physical changes result in alteration of the hydraulic conductivity in the sedimentary habitat and finally (3) the change in hydraulic conductivity modifies resource availability for interstitial microorganisms involved in studied processes (Fig.1). In riverbeds covered by excessive deposition of fine sedimentary particles, permeability, hydrological exchanges and associated input of resources from the surface for interstitial organisms are reported to be low (Beschta and Jackson 1979, Schächli 1992, Wood and Armitage 1997). In this context, bioturbation would have more influence on water exchanges between surface and hyporheic zone than in highly permeable sediments.

The aim of our study was therefore to examine how the environmental context (sediment characteristics) modulates the effects of an active bioturbator as ecosystem engineer in river sediments. We tested the effects of the bioturbator *Tubifex tubifex* Müller (Oligochaeta, Tubificidae) on the characteristics and activities of microbial communities in two sedimentary habitats with contrasted textures (topped by coarse permeable sand *versus* topped by fine sand with low permeability) by using slow filtration columns. For each habitat, we broke down and examine the ecosystem-engineering process into detailed intermediate steps (see Fig.1) including sediment reworking, physical (hydraulic conductivity) and abiotic (availability of nutrients and electron acceptors used for OM mineralization) changes on the sedimentary habitat. The ecosystem-engineering effects (biotic changes) were measured on the characteristics (bacterial and fungal abundances) and activities (potential aerobic respiration, potential denitrification, hydrolytic exoenzymatic activities) of the microbial community developed on leaves buried in sediments. Decomposition of leaf litter (measurement of breakdown rates) -a crucial microbially-mediated ecological process in river sediment- was used as a final step in the engineering process. Bioturbators were expected to have the most significant influence in sedimentary systems covered by fine particles, where restricted hydrological exchanges (low hydraulic conductivity and associated sharp decrease in O₂ and following electron acceptors) are supposed to constrain microbial communities.

Methods

Experimental design

To address how sediment context modulates the effects of a bioturbator at the water-sediment interface of rivers, we employed a factorial experimental approach in which the occurrence of *T. tubifex* and sediment texture were manipulated. Experiments were carried out

in slow filtration columns (n = 16 experimental units, height = 35 cm and inside diameter = 10 cm; Mermillod-Blondin et al. 2005, Navel et al. 2010) filled with sediment, at constant temperature (15 ± 0.5 °C) under a 12 h light / 12 h dark cycle.

These mesocosms were filled by successively adding gravel (2-4 mm diameter, 300 g) and then sand (100-1000 μm , 170 g), 8 times. We manipulated the surface sediment texture by adding a 2 cm thick layer of either fine sand (90% of particles < 150 μm diameter, low permeability: “fine sand treatment”, n = 8 columns) or coarse sand (90% of particles > 300 μm diameter, n = 8 columns) at the top of the gravel-sand base. The thickness of the top sediment layer was in accordance with observations reported from riverbeds impacted by fine sediment deposits (Wood & Armitage 1997). Analyses performed before the start of the experiment indicated that the specific area and the amounts of total organic carbon (TOC), nitrogen (TN) and phosphorus (TP) in the sediment were higher in the fine than in the coarse sand (Table 1). All the sedimentary material was collected from the Rhône River, elutriated and cleaned with deionised water to eliminate fauna and coarse particulate organic matter (CPOM). Moreover, the whole-sediment layer was kept in the dark to suppress possible photoautotrophic processes.

During sediment installation, a set of 35 leaf discs (diameter: 12 mm) of alder (*Alnus glutinosa* (L.) Gaertner), a common species along rivers characterized by fast leaf degradation (Abelho 2001), was inserted between two circular sieves (3 mm mesh) at a depth of 9 cm below the sediment surface in each column. Discs were cut avoiding central veins of leaves collected from the riparian zone of the Rhône River during abscission (October 2008). Leaves were conditioned in small-mesh bags immersed in a nearby river (located on the campus of the University Claude Bernard Lyon 1, Lyon, France) for 10 days, i.e. a time sufficient to allow microbial colonization (Suberkropp and Chauvet 1995). After installation of sediment and leaf litter, aerated artificial river water ($96 \text{ mg.L}^{-1} \text{ NaHCO}_3$, $39.4 \text{ mg.L}^{-1} \text{ CaSO}_4 \cdot 2\text{H}_2\text{O}$,

60 mg.L⁻¹ MgSO₄ · 7H₂O, 4 mg.L⁻¹ KCl, 19 mg.L⁻¹ Ca(NO₃)₂ · 4H₂O and 1.6 mg.L⁻¹ (CH₃CO₂)₂CaH₂O; pH = 7.5; US EPA 1991) was added at a constant hydraulic head ($\Delta H = 3$ cm), to a depth of 10cm above the sediment surface of each mesocosm. Openings in each mesocosm allowed sampling water at different times during the experiment.

Seven days after sediment installation (T7) (time necessary to obtain a physico-chemical stabilization of the system), we added a set of 100 individuals of *T. tubifex* to half of the experimental units (n = 4 per treatment). The density of tubificid worms in the experimental units (around 12,800 individual.m⁻²) was in accordance with densities reported in field studies (Fruget 1989, Martinet 1993). *T. tubifex* is a common deposit feeder that inhabits sandy and muddy habitats, which can actively rework sedimentary particles (McCall and Fisher 1980) and increase sediment permeability (Nogaro and Mermillod-Blondin 2009, Nogaro et al. 2009). The potential influence of *T. tubifex* on leaf litter degradation was expected to result from the influence of *T. tubifex* as ecosystem engineers rather than a direct feeding on leaf litter. To verify that *T. tubifex* do not feed on leaves, we conducted a preliminary experiment using aerated aquatic microcosms in which 35 alder leaf discs were deposited at the surface of a fine layer of sediment for 59 days. We measured that the occurrence of 100 individuals *T. tubifex* did not significantly influence the leaf litter breakdown rate nor the microbial abundances and activities associated with leaf litter (unpublished data).

During the main experiment, hydraulic discharge rate was measured and water was sampled every 10 days at 4 depths to determine O₂, NH₄⁺, NO₃⁻, NO₂⁻, PO₄³⁻, SO₄²⁻ and dissolved organic carbon (DOC) concentrations, for all columns. At the end of the experiment, columns were dismantled and sediment was cut into slices to quantify sediment reworking and vertical distribution of invertebrates. Fungal biomass, total bacterial abundance, abundance of active eubacteria, potential aerobic and anaerobic activities and

enzymatic activities involved in C and N cycles were determined on leaf discs, as described below. Leaf discs were then dried and weighed to quantify mass loss during the experiment.

Physico-chemical analyses

Every 10 days starting with day 6, a day before fauna addition (T6, T16, T26, T36, T46 and T56), the outlet of each column was closed and water was shunted and sampled at +2 cm above (H1) and -3 cm (H2), -8 cm (H3) and -13 cm (H4) below water-sediment interface under similar hydraulic pressure conditions. An oxygen micro-sensor probe fitted in a glass tube (OX 500, Unisense, Aarhus, Denmark) was used to determine O₂ concentration without contact with atmospheric oxygen during sampling. NH₄⁺, NO₃⁻, NO₂⁻, PO₄³⁻ and SO₄²⁻ concentrations were determined following standard colorimetric methods (Grashoff et al. 1983) after filtration through Whatman GF/F filters (pore size: 0.7 µm; Millipore, Billerica, MA, U.S.A.) using an automatic analyzer (Easychem Plus, Systea, Anagni, Italia). For DOC measurements, water samples were filtered through Whatman HAWP filters (pore size: 0.45 µm; Millipore, Billerica, MA, U.S.A.) and acidified with 3 drops of HCl (35%). The DOC concentration in water samples was measured with a total carbon analyzer (multi N/C 3100, Analytik Jena, Jena, Germany) based on combustion at 900 °C after removal of DOC with HCl and CO₂ stripping under O₂ flow.

Sediment reworking analyses

Particle redistribution induced by worms in the sedimentary matrix was estimated by the luminophore tracer technique (Gérino 1990). In each column, natural sediment particles (150-300 µm) dyed with yellow luminescent paint were deposited uniformly at the top of the sedimentary matrix a few hours after the introduction of *T. tubifex* (at T7). During column

dismantling (T59), the top 4 cm of sediment were cut into 0.5 cm thick slices, dried at 40 °C (48 h) and homogenized before counting luminophores on 500 mg subsamples under U.V. light (3 replicates per sampled slice). Vertical distribution of luminophores in the sediment was obtained by expressing the density of particles (number.g⁻¹ dry sediment) obtained for each slice as percentage of the total amount of luminophores obtained for the whole 4 cm top sediment layer.

Vertical distribution of tubificid worms

After collecting subsamples on the top sediment for luminophore counting, sediment was pooled into 5 cm thick sediment slices that were sieved (using a 500 µm–diameter sieve) to collect living tubificids. Individuals recovered in each slice were preserved in 96% ethanol and counted under a dissecting microscope. For each column, the vertical distribution of tubificid worms in the sediment was determined by reporting the abundance of worms in each slice to the total amount of worms retrieved in the overall sedimentary column (results for each slice were expressed as percent).

Microbial analyses

Fungal biomass

For each column, 5 leaf discs collected at the end of the experiment were stored at -80 °C and freeze-dried for 12 h before analysis. Fungal biomass was estimated with the ergosterol quantification method in which saponified products were obtained by methanol refluxing prior to saponification reaction using KOH/methanol (Gessner et al. 2003), following the protocol detailed in Navel et al. (2010). Ergosterol was isolated from saponified products by

using Oasis HLB 3cc extracting columns (Waters Corporation, Milford, MA, U.S.A) and elution with isopropanol. Mass of ergosterol in the sample was then calculated by using HPLC system (HPLC 360/442, Kontron, Eching, Germany). Fungal biomass was estimated from ergosterol amounts using a 182 conversion factor determined for aquatic hyphomycetes, which are known to dominate fungal assemblages on decomposing litter (Gessner and Chauvet 1993). Results were expressed in mg fungi.g⁻¹ dry mass of leaf litter.

Bacterial abundances

During column dismantling (at T59), 2 leaf discs were immediately collected and fixed in 4% paraformaldehyde in phosphate-buffered saline (PBS; 0.13M NaCl, 7mM NaHPO₄, 3mM NaH₂PO₄; pH=7.2) for 10 h. Fixed samples were subsequently washed twice in PBS and were stored in ethanol and PBS (50:50) at 20 °C. After storage (2 weeks), leaf discs were homogenized in 20 mL of 0.1% pyrophosphate in PBS using a sonicator with a 2 mm-diameter probe at 50 W for two periods of 60 s. All homogenized samples were finally supplemented with the detergent NP-40 (Flucka, Buchs, Switzerland) to a final concentration of 0.01 %. Aliquots (10 µL) of homogenized samples were spotted onto gelatine-coated slides and were hybridized with Cy3-labelled oligonucleotide probe (mix of EUB 338, EUB 338 II and EUB 338 III, eubacteria) and concomitantly stained with the DNA intercalating dye DAPI (200 ng.µL⁻¹, Sigma, Buchs, Switzerland) according to Navel et al. (2010). Numbers of DAPI- and Cy3-bacteria were expressed as numbers of bacteria and numbers of active eubacteria (hybridized with EUB 338, Karner & Fuhrmann 1997) per g dry leaf.

Microbial activities

All microbial activities were measured within the 24 h following columns dismantling, with leaf discs stored at 4 °C before analysis.

Enzymatic activities: β –glucosidase (EC: 3.2.1.21), β –xylosidase (EC: 3.2.1.37) and leucine aminopeptidase (EC: 3.4.11.1) activities were measured on 2 discs (2 times) by fluorimetry using constant volume of substrate analogs: 4-methylumbelliferyl- β -D-glucoside (MUF-glu; 750 μ M, 2 mL), 4-methylumbelliferyl-xylosidase (MUF-xyl; 1000 μ M, 2 mL) and L-Leucine-4-Methyl Coumarinyl-7-amideHCl (MCA-leu; 1000 μ M, 2 mL), respectively. Incubation at 20 °C (40 min) was stopped by transferring into boiling water before centrifugation (5000 G; 4851 rpm, 3 min). Fluorimetry measurements were realised on a mix of supernatant (300 μ L) and buffer (30 μ L, pH 10.4) using a microplate reader (SAFIRE, TECAN Group Ltd, Switzerland) with excitation wavelength of 363 nm and emission wavelengths of 441 nm for MUF-glu and MUF-xyl. Wavelengths were set at 343 nm (excitation) and 436 nm (emission) for MCA-leu. Litter dry mass (drying at 70 °C for 48h) was determined at the end of analyses to express results as nmol of hydrolysed compound.h⁻¹.g⁻¹ dry leaf litter. For each sample, values were corrected by the fluorimetric signal obtained with a formaldehyde-killed control (measurements realised in similar conditions on 2 discs previously treated 30 min with a 39 % formaldehyde solution).

Potential aerobic respiration and anaerobic denitrification activities were measured on leaf discs following the slurry technique (Furutani et al. 1984). Leaf discs (n = 4 for respiration and n = 6 for denitrification) were placed in 150 mL flasks supplemented with feeding solutions to optimize microbial activity. For the measurements of CO₂ production (respiration), the incubation was conducted under aerobic conditions with 5 mL of a feeding solution of glucose (7.5 g.L⁻¹) and glutamic acid (7.3 g.L⁻¹). For the measurements of N₂O production (denitrification), the incubation was under anaerobic conditions with a N₂ atmosphere. The feeding solution was a mixture of 5 mL of a KNO₃ (2.2 g.L⁻¹), glucose (7.5

g.L⁻¹) and glutamic acid (7.3 g.L⁻¹) solution. Acetylene (10% v/v) was introduced in N₂ saturated atmosphere to stop N₂O-reductase activity. CO₂ and N₂O productions were calculated from measurements of concentrations at 2 h and 6 h incubations by using gas chromatography on a microcatharometer (M200 micro gas chromatograph, MTI Analytical Instruments, Richmond, CA, U.S.A.). After the drying of leaf discs (70 °C for 48h), results were expressed in µg of C or N.h⁻¹.g⁻¹ dry leaf litter.

Leaf litter degradation

For each column, the total dry mass of leaf litter after 59 days was calculated as the sum of the dry masses of samples used in microbial analyses and that measured for the remaining leaf material (common drying method: 70 °C for 48 h), with correction for the set of 5 discs that were freeze-dried for fungal biomass assessment. Results were compared to the initial dry mass determined on 5 additional sets of 35 alder discs (228.8 ± 6.25 mg) at the start of the experiment.

Data treatment

Repeated measures of hydraulic conductivity were analyzed using mixed model analysis of variance with “sediment” (“coarse sand” vs. “fine sand”), “worms” (“with” vs. “without”) and “time” as fixed factors, and experimental unit (“column”) as random factor. Repeated measures of vertical profiles in O₂, DOC, NH₄⁺, NO₃⁻, NO₂⁻, SO₄²⁻ and PO₄³⁻ were analysed similarly, with “depth” as additive fixed factor. Vertical distribution of tubificid worms was studied by using two-way analysis of variance (ANOVA) with “sediment”, and “depth” as main factor. Vertical distribution of luminophores was studied by using similar procedure with “sediment”, “depth” and “worms” as main factor. Influences of sediment permeability

and tubificid worms on data obtained on buried leaf litter (daily dry mass loss, fungal biomass, total abundance of bacteria, abundance of active bacteria, % active bacteria, enzymatic activities, potential aerobic respiration and potential denitrification) were examined using two-way ANOVAs with “sediment” and “worms” as main factors. The method of contrast was used to determine significant differences between treatments (Crawley 2002). Hydraulic conductivity and microbial activities on leaves (glucosidase, leucine aminopeptidase activity and potential denitrification activities) were log-transformed before statistical analysis in order to fit the assumption of homoscedasticity. Abundances of luminophores and worms retrieved at the end of experiment for each layer within a same column were expressed as percent of the total abundance for the whole column, and were arcsin-transformed before analyses. Statistical analyses were performed using JMP 8.0 (SAS Institute, Cary, NC, U.S.A.), version 8.0.1. Significance for all statistical tests was accepted at $\alpha < 0.05$.

Results

Influence of sediment physical characteristics on sediment reworking activity.

The physical structure of the habitat influenced the vertical distribution of tubificid worms in sediment (Fig. 2; “sediment-by-depth interaction effect”: $F_{(3,24)} = 83.44$, $P < 10^{-4}$). While the major part of individuals were retrieved in the top sediment layer when covered by fine sand (93% in the first 5 cm), most of worms were retrieved deeper in the sediment of columns covered with coarse sand (around 65% were found after 10 cm depth). The presence of worms in systems increased the transport of luminophores from the sediment surface to the sedimentary column (Fig. 2; “worms-by-depth interaction effect”: $F_{(7,96)} = 29.78$, $P < 10^{-4}$).

This effect of worms on luminophore profiles was strongly influenced by the physical characteristics of the sedimentary habitat ($F_{(7,96)} = 9.61$, $P < 10^{-4}$). The percentage of luminophores buried at depth was less than 5% in the “coarse sand” treatment whereas it was more than 20% in the “fine sand” treatment” (Fig. 2).

Influence of the sediment physical characteristics on hydraulic exchanges and microbial processes involved in CPOM processing

Mean (\pm S.D.) hydraulic conductivity measured for the “fine sand” treatment was about 8-fold lower than for columns topped with “coarse sand” (Fig. 3; 2.02 ± 0.67 and 15.95 ± 4.81 cm.h⁻¹, respectively). The decreases with depth of O₂ and NO₃⁻ concentrations (Fig. 4; $F_{(3,276)} = 933.73$ and 134.90 , respectively, $P < 10^{-4}$ for both) in the interstitial water were higher in “fine sand” than in “coarse sand” treatment (“sediment-by-depth interaction effect”: $F_{(3,276)} = 200.85$ and 159.26 for O₂ and NO₃⁻ respectively, $P < 10^{-4}$ for both). This difference was particularly marked in the top sediment layer (O₂ concentrations reduced by about 87% and 13% in “fine sand” and “coarse sand” treatments, respectively), and led to lower O₂ and NO₃⁻ concentrations in the sedimentary habitat covered by fine sand deposits ($F_{(1,12)} = 631.70$ and 409.18 for O₂ and NO₃⁻ respectively, $P < 10^{-4}$ for both). Peaks of DOC, NH₄⁺, NO₂⁻ and PO₄³⁻ concentrations were only recorded in the “fine sand” treatment, leading to higher concentrations of these solutes in “fine sand” than in “coarse sand” treatment (Fig. 4; $F_{(1,12)} = 93.02$, 665.57 , 193.27 , 16.13 , for DOC, NH₄⁺, NO₂⁻ and PO₄³⁻ respectively, $P < 10^{-4}$ for all).

Determinations of the dry mass of leaf litter retrieved at the end of the experiment (Fig. 5A) showed that the daily mass loss rate was 31% lower in the “fine sand” treatment than in the “coarse sand” treatment ($F_{(1,12)} = 14.39$, $P = 0.043$). In parallel, the total abundance of bacteria (Fig. 5B), the abundance of active bacteria (Fig. 5C), the fungal biomass (Fig. 5D), the

glucosidase activity (Fig. 5H) and the leucine aminopeptidase activity (Fig. 5I) were significantly lower in “fine sand” than in “coarse sand” treatment (contrasts: comparisons without *T. tubifex*: $|t|_{12} = 2.57, 2.87, 2.50, 5.18$ and 7.14 , respectively, $P < 0.028$ for all). Potential aerobic respiration (Fig. 5E), potential denitrification (Fig. 5F) and xylosidase activities measured on leaves were not significantly influenced by sedimentary conditions (contrasts: $|t|_{12} = 1.43, 0.99$ and 0.76 , respectively, $P > 0.176$ for all).

Context-dependent influence of tubificid worms on hydrologic exchanges, biogeochemical processes and CPOM processing

The influence of tubificid worms on hydraulic conductivity (Fig. 3) and water chemistry (Fig. 4) was dependant on the physical characteristics of the top sediment (“sediment-by-worms interaction effect”: $F_{(1,12)} = 24.67, 48.41, 147.73, 13.76, 96.24$ and 4.04 for hydraulic conductivity, O_2 , NO_3^- , SO_4^{2-} , NH_4^+ , and PO_4^{3-} concentrations, respectively, $P < 0.044$ for all). While worms had low influence on hydraulic conductivity and concentrations of solutes in “coarse sand” treatment, they increased by more than 6 fold the hydraulic conductivity in “fine sand” treatment. Consequently, the presence of tubificid worms increased O_2 and NO_3^- concentrations and strongly reduced the peaks of solutes (DOC, NH_4^+ , NO_2^- and PO_4^{3-}) released in the sedimentary columns with “fine sand” treatment.

Similarly, the influence of *T. tubifex* on microbial characteristics and associated processing of buried leaf litter was dependant on the physical structure of the sedimentary habitat (Fig. 4). We did not observe any influence of *T. tubifex* on microbial characteristics measured on leaves buried in “coarse sand” systems (Fig. 5; contrasts: $|t|_{12} = 0.24, 0.83, 0.02, 1.32, 0.97$ and 1.05 for total abundance of bacteria, abundance of active bacteria, fungal biomass, xylosidase, glucosidase and leucine aminopeptidase, respectively, $P > 0.207$ for all) except for potential aerobic respiratory activity ($|t|_{12} = 2.77$, $P < 0.018$). In contrast, *T. tubifex* had a

positive influence on most microbial variables in “fine sand” treatment (contrasts: $|t|_{12} = 4.63$, 4.42, 3.05, 8.04 and 4.46 for total abundance of bacteria, abundance of active bacteria, xylosidase and leucine aminopeptidase respectively, $P < 0.009$ for all; and $|t|_{12} = 1.98$, $P = 0.071$ for glucosidase activity) with the exception of potential denitrification ($|t|_{12} = 0.21$, $P < 0.835$).

In parallel, *T. tubifex* increased the daily loss rate of leaf litter mass in the “fine sand” treatment by about 30% (Fig. 4A; $|t|_{12} = 3.40$, $P < 0.006$) where they counteracted the negative influence of fine sediment deposition on leaf litter degradation ($F_{(1,12)} = 5.61$, $P = 0.036$). Such an effect was not observed in the “coarse sand” treatment (Fig. 5A, $|t|_{12} = 0.17$, $P = 0.867$).

Discussion

Contrasted biogeochemical processes induced by sediment characteristics.

Our study confirmed the expectation that the biogeochemical functioning of the hyporheic zone is strongly influenced by the sediment context. The high hydraulic discharge rates in systems topped by a 2-cm thick layer of coarse sand (highly permeable systems) generated aerobic conditions (O_2 concentration $> 2 \text{ mg.L}^{-1}$) throughout the sedimentary matrix (to a depth of 13 cm). As a consequence of the O_2 availability, NO_3^- and SO_4^{2-} -as less energetically favourable electron acceptors for OM mineralization (Hedin et al. 1998)- were not consumed for OM mineralization. In these conditions, we did not observe any significant production of solutes linked to anaerobic OM degradation in sediments (i.e. NH_4^+ , PO_4^{3-} and DOC) (Nogaro et al. 2007). Hydraulic conductivity and discharge rates were 85% lower in sediment covered by a 2-cm thick layer of fine sand, in comparison with systems topped by coarse sand. As a

consequence of the reduced hydraulic discharge rates, we observed sharp decreases in O_2 and NO_3^- concentrations along depth in these systems. The rapid along-depth succession of metabolic pathways was in accordance with predictable thermodynamic sequences (based on free energy yields): O_2 being consumed first during OM mineralization in the oxic zone, followed by the consumption of NO_3^- (denitrification), manganese and iron oxides, SO_4^{2-} and carbon dioxide (Hedin et al. 1998, Baker et al. 2000, Kristensen 2000). Since O_2 was limiting in the first centimeters of sediments, most of the sedimentary matrix (and thus buried leaf litter) was under anaerobic conditions, leading to the release of NH_4^+ , PO_4^{3-} and DOC. In such O_2 -limited system, microbial abundances (total abundance of bacteria, abundance of active bacteria and fungal biomass) and activities (glucosidase and leucine aminopeptidase activities) were altered, and rates for microbial-mediated ecological processes occurring in the HZ were reduced (leaf litter decomposition was 30% lower than in not-stressed coarse-sand treatment).

The contrasts in biogeochemical conditions between the two sedimentary contexts were consistent with field studies showing that reduced hydrologic exchanges due to clogging favoured the occurrence of anaerobic processes such as denitrification, sulphato-reduction and methanogenesis (Dahm et al. 1987, Brunke and Gonser 1997, Boulton et al. 1998, Lefebvre et al. 2004). Our results are also in accordance with other studies showing that biogeochemical conditions, in particular the availability of electron acceptors (mainly O_2 and NO_3^-), strongly affect (i) the fungal colonization of leaves (Medeiros et al. 2009) and microbial enzymatic activities such as cellulase and peptidase activities (Montuelle and Volat 1997) and (ii) OM degradation rates (Chauvet 1988, Claret et al. 1998, Dahm et al. 1998, Lefebvre et al. 2005). It is therefore clear that sediment contexts that lead to low hydrological exchanges and O_2 concentration in sediments limit the growth and activity of microbial communities and ultimately the rates of microbial-mediated ecological process occurring in the HZ. Finally, we

efficiently recreated the hydrological and biogeochemical functioning of 2 contrasted
sediment contexts and highlighted the key role played by O₂ as resource for microorganisms
developed in sedimentary habitats.

Modulation of bioturbator effects on biogeochemical processes by sediment context.

The present study confirmed our hypothesis that the effects of bioturbators on hydraulic
conductivity and microbial-mediated processes depend on the sediment context.

As upward conveyors (feeding on sediment at depth and ejecting faecal pellets at the
sediment–water interface, Fisher et al. 1980, McCall and Fisher 1980), tubificid worms can
build networks of tubes and burrows that may extend as deep as 20 cm in sediments.
However, our results showed that both the bioturbation activity and the vertical distribution of
tubificid worms were modulated by the sedimentary characteristics. Using luminophore as
particle tracers, we noted that tubificid worms significantly reworked the top of the
sedimentary column with a fine sand layer whereas it was not the case with a coarse sand
layer. This contrast in bioturbation degree was linked to the vertical distribution of worms.
While worms used the whole sediment column in the systems topped with coarse sand, most
of tubificid worms were found in the top 0-5 cm in systems topped by a layer of fine sand.
Fine sand probably acted as preferential feeding zone for *T. tubifex* (Juget 1979, Rodriguez et
al. 2001), which strongly influenced the vertical distribution of worms in the sedimentary
column. The different bioturbation activities exhibited by *T. tubifex* in the two sediment
contexts could explain their contrasting effects on hydraulic conductivity (see Fig.1 for the
successive effects of bioturbators). By producing galleries through the fine sand layer, *T.*
tubifex create water pathways that counteracted the adverse effect of fine sediment deposition
on water exchanges. The 6-fold increase in hydraulic conductivity due to *T. tubifex* in the

fine-sand treatment stimulated the exchanges of water and O₂ from surface to deep sediment layers, restoring aerobic conditions in the sedimentary column (Fig. 1). Modification of aerobic-anaerobic conditions observed through the increase in O₂ and NO₃⁻ concentrations was also associated with a lack of NH₄⁺, PO₄³⁻ and DOC accumulation in the sedimentary habitat bioturbated by tubificid worms. The increase in electron acceptors (O₂ and NO₃⁻) availability for micro-organisms with *T. tubifex* has stimulated microbial communities (abundances and activities) associated to the buried OM, leading to an increase by 30% of OM breakdown rate in sedimentary systems covered by fine sand (Fig. 1). In contrast, tubificid worms did not affect hydraulic conductivity and the subsequent chemical conditions (availability of electron acceptors) in sediments topped by coarse sand. Consequently, bioturbators did not influence microorganisms and microbial-mediated processes in sediments.

Our study clearly demonstrated that the contribution of bioturbating invertebrates on ecosystem processes was negatively correlated with the hydrologic exchanges occurring at the water-sediment interface of the studied system, supporting conclusions from other studies (Hakenkamp and Palmer 2000, Boulton et al. 2002). Bioturbators are able to strongly influence water fluxes (through biological decolmation) in sedimentary habitats characterized by low hydrologic exchanges (affected by the deposition of fine sediment particles), whereas they only slightly modulate existing water fluxes in habitats with high hydrologic exchanges. If the low influence of bioturbators on hydraulic conductivity in systems covered with coarse sand could be linked to their low sediment reworking activity (Fig. 2), it could also resulted from the reduced ability of bioturbation to increase hydrologic exchanges in a system that is already highly permeable.

What underlies context dependency in the impact of engineers on other biota?

Most ecosystem engineering studies quantified the effects of engineers on some biotic variables in a given habitat without taking into account the modulation of organism engineering by environmental conditions (Crain & Bertness 2006). Our study clearly demonstrates that the influences of bioturbators as engineers may vary across environmental contexts. Few studies have already reported similar observations for various types of ecosystems and various types of ecosystem engineers (e.g. Spooner and Vaughn 2006, Nogaro et al 2009, de Moura Quierós A 2011) but they did not finely decrypt the complete mechanisms by which ecosystem engineers and habitat characteristics interacted to shape biological communities and/or ecosystem functions. More precisely, the environmental context can influence the impacts of engineers on communities by modulating (i) the degree with which its activity(ies) (or structure(s)) generates physical changes in the environment and/or (ii) the degree with which physical changes can generate abiotic changes in the environment (Fig. 1, see also Jones et al. 2010). As most of studies dealing with context dependency examined only partially the engineering sequence, they could not provide explanations about the mechanisms underlying context dependency. By examining the whole engineering process as a detailed sequence of successive cause-effect relationships, our study allowed to conclude that the influence of bioturbators on microbial-mediated processes was mainly mediated by the organism ability to change abiotic factors that are limiting for micro-organisms (i.e. O₂). This conclusion supports the general (and not tested before) idea that ecosystem engineering is not an idiosyncratic process and that the magnitude of engineering impacts on a biotic variable controlled by one or more limiting abiotic factors depends on the degree to which the limiting abiotic factor(s) is/are modified relative to the unengineered state (e.g., Gutiérrez et al. 2003, Gutiérrez and Jones 2006, Jones et al. 2010).

Acknowledgments

We thank Bernadette Volat (Cemagref, Lyon), Félix Vallier (LEHF, Villeurbanne) and Didier Lambrigot (EcoLab, Toulouse) for their helpful assistance. This study was funded by the ANR Biodiversity programme (ANR-06-BDIV-007) InBioProcess 2007-2010.

References

- Abelho, M. 2001. From litterfall to breakdown in streams: a review. – *TheScientificWorld* 1: 656-680.
- Aller, R. C. 1988. Benthic fauna and biogeochemical processes in marine sediments: the role of burrow structures. – In: Blackburn T. H. and Sorensen J. (eds), *Nitrogen cycling in coastal marine environments*. John Wiley & Sons Ltd, pp. 301-338.
- Aller, R. C. 1994. Bioturbation and remineralization of sedimentary organic matter: effects of redox oscillation. – *Chem. Geol.* 114: 331-345.
- Baker, M. A. et al. 2000. Organic carbon supply and metabolism in a shallow groundwater ecosystem. – *Ecology* 81: 3133-3148.
- Bertness, M. D. 1984a. Ribbed mussels and *Spartina alterniflora* production in a New England salt marsh. – *Ecology* 65: 1794-1807.
- Bertness, M. D. 1984b. Habitat and community modification by an introduced herbivorous snail. – *Ecology* 65: 370-381.

- 1 Beschta, R. L. and Jackson, W. L. 1979. The intrusion of fine sediments into a stable gravel
2 bed. – *J. Fish. Res. Board Can.* 36: 204-210.
- 3 Boulton, A. J. et al. 2002. Freshwater meiofauna and surface water-sediment linkages: a
4 conceptual framework for cross-system comparisons. – In: Rundle S. D. et al. (eds),
5 *Freshwater meiofauna: biology and ecology*. Backhuys Publishers, pp. 241-259.
- 6 Boulton, A. J. et al. 1998. The functional significance of the hyporheic zone in streams and
7 rivers. – *Annu. Rev. Ecol. Evol. Syst.* 29: 59-81.
- 8 Boulton, A. J. et al. 2010. Ecology and management of the hyporheic zone: stream-
9 groundwater interactions of running waters and their floodplains. – *J. N. Am. Benthol.*
10 *Soc.* 29: 26-40.
- 11 Brunke, M. and Gonser, T. 1997. The ecological significance of exchange processes between
12 rivers and groundwater. – *Freshwater Biol.* 37: 1-33.
- 13 Chauvet, E. 1988. Influence of the environment on willow leaf litter decomposition in the
14 alluvial corridor of the Garonne River. – *Arch. Hydrobiol.* 112: 371–386.
- 15 Claret, C. 1997. Rôle de l'interface rivière-nappe dans la régulation des flux de nutriments.
16 Importance des peuplements microbiens et de la biodiversité des invertébrés interstitiels
17 comme descripteurs de l'effet des aménagements. PhD Thesis, 150pp., Université de
18 Chambéry, Chambéry, France.
- 19 Claret, C. et al. 1998. Seasonal dynamics of nutrient and biofilm in interstitial habitats of two
20 contrasting riffles in a regulated large river. – *Aquat. Sci.* 60: 33-55.
- 21 Cornut, J. et al. 2010. Early leaf decomposition stages are mediated by aquatic hyphomycetes
22 in the hyporheic zone of woodland streams. – *Freshwater Biol.* 55: 2541-2556.

- 1 Crawley, M. J. 2002. Statistical Computing: An introduction to data analysis using S-Plus. –
2 John Wiley & Sons Ltd.
- 3 Dahm, C. N. et al. 1987. Role of anaerobic zones and processes in stream ecosystem
4 productivity. – In: Averett R. C. and McKnight D. M. (eds), Chemical quality of water
5 and the hydrologic cycle. Lewis Publishers, pp. 157–178.
- 6 Dahm, C. N. et al. 1998. Nutrient dynamics at the interface between surface waters and
7 groundwaters. – *Freshwater Biol.* 40: 427-451.
- 8 Daleo, P. et al. 2007. Ecosystem engineers activate mycorrhizal mutualism in salt marshes. –
9 *Ecol. Lett.* 10: 902-908.
- 10 Duffy, J. E. 2002. Biodiversity and ecosystem function: the consumer connection. – *Oikos* 99:
11 201–219.
- 12 Duffy, J. E. et al. 2003. Grazer diversity effects on ecosystem functioning in seagrass beds. –
13 *Ecology Letters* 6: 637–645.
- 14 Fellows, C. S. et al. 2001. Whole-stream metabolism in two montane streams: contribution of
15 the hyporheic zone. – *Limnol. Oceanogr.* 46: 523-531.
- 16 Fisher, J. B. et al. 1980. Vertical mixing of lake sediments by tubificid oligochaetes. – *J.*
17 *Geophys. Res.* 85: 3997-4006.
- 18 Fruget, J. -F. 1989. L'aménagement du bas-Rhône. Evolution du fleuve et influence sur les
19 peuplements de macroinvertébrés benthiques. PhD Thesis, 484pp., Université Claude
20 Bernard Lyon1, Lyon, France
- 21 Furutani, A. et al. 1984. A method for measuring the response of sediment microbial
22 communities to environmental perturbations. – *Can. J. Microbiol.* 30: 1408-1414.

- 1 Gérino, M. 1990. The effects of bioturbation on particle distribution in Mediterranean coastal
2 sediment: Preliminary result. – *Hydrobiologia* 207: 251–258.
- 3 Gessner, M. O. et al. 2003. Qualitative and quantitative analyses of aquatic hyphomycetes in
4 streams. – In: Tsui C. K. M. & Hyde K. D. (eds), *Fungal diversity research series 10 -*
5 *freshwater mycology*. Fungal diversity press, pp. 127-157.
- 6 Gessner, M. O. and Chauvet, E. 1993. Ergosterol-to-biomass conversion factors for aquatic
7 hyphomycetes. – *Appl. Environ. Microbiol.* 59: 502-507.
- 8 Goudard, A. and Loreau, M. 2008. Nontrophic interactions, biodiversity, and ecosystem
9 functioning: an interaction web model. – *Am. Nat.* 171: 91-106.
- 10 Grasshoff, K. et al. 1983. *Methods of seawater analysis*. – Verlag Chemie.
- 11 Grimm, N. B. and Fisher, S. G. 1984. Exchange between interstitial and surface water:
12 implication for stream metabolism and nutrient cycling. – *Hydrobiologia* 111: 219-228.
- 13 Gutiérrez, J. L. et al. 2003. Mollusks as ecosystem engineers: the role of shell production in
14 aquatic habitats. – *Oikos* 101: 79-90.
- 15 Hakenkamp, C. C. and Palmer, M. A. 2000. The ecology of hyporheic meiofauna. *Streams*
16 and ground waters, pp. 307-336. Academic press.
- 17 Hansen, K. and Kristensen, E. 1997. Impact of macrofaunal recolonization on benthic
18 metabolism and nutrient fluxes in a shallow marine sediment previously overgrown
19 with macroalgal mats. – *Estuarine, Coastal Shelf Sci.* 45: 613-628.
- 20 Hedin, L.O. et al. 1998. Thermodynamic constraints on nitrogen transformations and other
21 biogeochemical processes at soil-stream interfaces. – *Ecology* 79: 684-703.

- 1 Holt, R. D. and Loreau, M. 2002. Biodiversity and ecosystem functioning: The role of trophic
2 interactions and the importance of system openness. – In: Kinzig A. P. et al. (eds), The
3 functional consequences of biodiversity: empirical progress and theoretical extensions.
4 Princeton University Press, pp. 246–262.
- 5 Jones, C. G. et al. 1994. Organisms as ecosystem engineers. – *Oikos* 69: 373-386.
- 6 Jones, C.G. et al. 1997. Positive and negative effects of organisms as physical ecosystem
7 engineers. – *Ecology* 78: 1946-1957.
- 8 Karner, M. and Fuhrman, J. A. 1997. Determination of active marine bacterioplankton: A
9 comparison of universal 16S rRNA probes, autoradiography, and nucleoid staining. –
10 *Appl. Environ. Microbiol.* 63: 1208-1213.
- 11 Kristensen, E. 2000. Organic matter diagenesis at the oxic/anoxic interface in coastal marine
12 sediments, with emphasis on the role of burrowing animals. – *Hydrobiologia* 426: 1-24.
- 13 Lawton, J. H. 1994. What do species do in ecosystems? – *Oikos* 71: 367-374.
- 14 Lefebvre, S. et al. 2004. Stream regulation and nitrogen dynamics in sediment interstices:
15 comparison of natural and straightened sectors of a third-order stream. – *River Res.*
16 *Appl.* 20: 499-512.
- 17 Lefebvre, S. et al. 2005. Nutrient dynamics in interstitial habitats of low-order rural streams
18 with different bedrock geology. – *Arch. Hydrobiol.* 164: 169-191.
- 19 Martinet, F. 1989. Le macrobenthos limnivore, descripteur des flux organiques liés aux
20 sédiments : exemple dans diverses annexes fluviales du Rhône. PhD Thesis, 152pp.,
21 Université Claude Bernard Lyon1, Lyon, France.

- 1 McCall, P. L. and Fisher, J. B. 1980. Effects of tubificid oligochaetes on physical and
2 chemical properties of lake Erie sediments. – In Brinkhurst R. O. and Cook D. G. (eds),
3 Aquatic oligochaete biology. Plenum press, pp. 253-317.
- 4 Medeiros, A.O. et al. 2009. Diversity and activity of aquatic fungi under low oxygen
5 conditions. – *Freshwater Biol.* 54: 142-149.
- 6 Menge, B. A. 2000. Testing the relative importance of positive and negative effects on
7 community structure. – *Trends Ecol. Evol.* 15: 46-47.
- 8 Mermillod-Blondin, F. and Lemoine, D. G. 2010. Ecosystem engineering by tubificid worms
9 stimulates macrophyte growth in poorly oxygenated wetland sediments. – *Funct. Ecol.*
10 24: 444-453.
- 11 Mermillod-Blondin, F. et al. 2005. Use of slow filtration columns to assess oxygen
12 respiration, consumption of dissolved organic carbon, nitrogen transformations, and
13 microbial parameters in hyporheic sediments. – *Water Res.* 39: 1687-1698.
- 14 Mermillod-Blondin, F. and Rosenberg, R. 2006. Ecosystem engineering: the impact of
15 bioturbation on biogeochemical processes in marine and freshwater benthic habitats. –
16 *Aquat. Sci.* 68: 434-442.
- 17 Montuelle, B. & Volat, B. 1997. Influence of oxygen and temperature on exoenzyme
18 activities in freshwater sediments. – *Verh. Int. Verein. Limnol.* 26: 373-376.
- 19 Naegeli, M. W. and Uehlinger, U. 1997. Contribution of the hyporheic zone to ecosystem
20 metabolism in a prealpine gravel-bed river. – *J. N. Am. Benthol. Soc.* 16: 794-804.
- 21 Navel, S. et al. 2010. Interactions between fauna and sediment control the breakdown of plant
22 matter in river sediments. – *Freshwater Biol.* 55: 753-766.

- 1 Nogaro, G. and Mermillod-Blondin, F. 2009. Stormwater sediment and bioturbation
2 influences on hydraulic functioning, biogeochemical processes, and pollutant dynamics
3 in laboratory infiltration systems. – Environ. Sci. Technol. 43: 3632-3638.
- 4 Nogaro, G. et al. 2006. Invertebrate bioturbation can reduce the clogging of sediment: an
5 experimental study using filtration sediment columns. – Freshwater Biol. 51: 1458-
6 1473.
- 7 Nogaro, G. et al. 2007. Influence of a stormwater sediment deposit on microbial and
8 biogeochemical processes in infiltration porous media. – Sci. Total Environ. 377: 334-
9 348.
- 10 Nogaro, G. et al. 2009. Ecosystem engineering at the sediment-water interface: bioturbation
11 and consumer-substrate interaction. – Oecologia 161: 125-138.
- 12 Pelegri, S. P. and Blackburn, T. H. 1994. Bioturbation effects of the amphipod *Corophium*
13 *volutator* on microbial nitrogen transformations in marine sediments. – Mar. Biol. 121:
14 253-258.
- 15 Pelegri, S. P. et al. 1994. Denitrification in estuarine sediment stimulated by the irrigation
16 activity of the amphipod *Corophium volutator*. – Mar. Ecol. Progr. Ser. 105: 285-290.
- 17 Pusch, M. and Schwoerbel, J. 1994. Community respiration in hyporheic sediments of a
18 mountain stream (Steina, Black Forest). – Arch. Hydrobiol. 130: 35-52.
- 19 Rodriguez, P. et al. 2001. Selective feeding by the aquatic oligochaete *Tubifex tubifex*
20 (*Tubificidae*, *Clitellata*). – Hydrobiologia 463: 133-140.
- 21 Schälchli, U. 1992. The clogging of coarse gravel river beds by fine sediment. –
22 Hydrobiologia 235/236: 189-197.

- 1 Scheu, S. 2003. Effects of earthworms on plant growth: patterns and perspectives. –
2 *Pedobiologia* 47: 846-856.
- 3 Schmidt, J. et al. 2009. Direct measurement of respiration and decolmation activity of
4 hyporheic meiofauna: first results from a new approach using anesthetizing substances.
5 – *Adv. Limnol.* 61: 185-203.
- 6 Stief, P. and de Beer, D. 2002. Bioturbation effects of *Chironomus riparius* on the benthic N-
7 cycle as measured using microsensors and microbiological assays. – *Aquat. Microb.*
8 *Ecol.* 27: 175-185.
- 9 Suberkropp, K. and Chauvet, E. 1995. Regulation of leaf breakdown by fungi in streams:
10 influences of water chemistry. – *Ecology* 76: 1433-1445.
- 11 Sundbäck, K. et al. 2004. Benthic nitrogen fluxes along a depth gradient in a microtidal fjord:
12 the role of denitrification and microphytobenthos. – *Limnol. Oceanogr.* 49: 1095-1107.
- 13 Svensson, J. M. and Leonardson, L. 1996. Effects of bioturbation by tube-dwelling
14 chironomid larvae on oxygen uptake and denitrification in eutrophic lake sediments. –
15 *Freshwater Biol.* 35: 289-300.
- 16 US EPA 1991. Methods for measuring the acute toxicity of effluents and receiving waters to
17 freshwater and marine organisms, 4th edn. US Environmental Protection Agency.
18 EPA/600/4-90/027, pp 34–35
- 19 Van de Bund, W. J. et al. 1994. Effects of deposit-feeder activity on bacterial production and
20 abundance in profundal lake sediment. – *J. N. Am. Benthol. Soc.* 13: 532-539.
- 21 Wilson, J. B. and Agnew, A. D. Q. 1992. Positive-feedback switches in plantcommunities. –
22 *Adv. Ecol. Res.* 23: 263-336.

Wood, P. J. and Armitage, P. D. 1997. Biological effects of fine sediment in the lotic environment. – Environ. Manage. 21: 203-217.

Wright, J. P. et al. 2002. An ecosystem engineer, the beaver, increases species richness at the landscape scale. – Oecologia 132: 96-101.

Tables

Table 1. Specific area, total organic carbon (TOC), total nitrogen (TN) and total phosphorus (TP) of the sediment used as top-sediment layer (mean \pm SD, n= 4 for specific area, n=5 for TOC, TN and TP).

	Specific area (cm ² g ⁻¹)	TOC (g kg ⁻¹)	TN (g kg ⁻¹)	TP (mg kg ⁻¹)
Coarse sand	59.37 \pm 0.34	0.97 \pm 0.08	0.17 \pm 0.03	3.76 \pm 3.47
Fine sand	1465.00 \pm 33.17	16.6 \pm 1.6	1.35 \pm 0.06	6.62 \pm 1.36

Figure captions

Fig. 1. Successive steps for (i) a general case of ecosystem engineering sequence (in grey) and (ii) the effects of *T. tubifex* on microorganisms in sedimentary habitats (study case, in white). Note that feedbacks to the engineer and other relationships in the ecosystem engineering can exist and are not represented here but can exist (for more information, see Gutiérrez and Jones 2008, Jones et al. 2008).

Fig. 2. Vertical distribution of *T. tubifex* in sediment and their associated effect on vertical profiles of luminophores in sediment columns covered by fine sand or coarse sand.

Fig. 3. Effect of *T. tubifex* on hydraulic conductivity measured every 10 days in sediment columns covered by fine sand or coarse sand.

Fig. 4. Effect of *T. tubifex* on depth profiles for O_2 , $N-NO_3^-$, $N-NH_4^+$, $N-NO_2^-$, and DOC concentrations determined at 4 depths (from H1: 2 cm above sediment interface, to H4: 13 cm below sediment interface) after 36 days in sediment columns covered by fine sand or coarse sand.

Fig. 5. Effect of *T. tubifex* on leaf mass loss rate (A), and microbial characteristics measured at the end of the experiment on leaves buried at 9 cm depth in sediment columns covered by fine sand or coarse sand: total abundance of bacteria (B), abundance of active bacteria (C), fungal biomass (D), potential respiratory activity (E), potential denitrification (F), xylosidase (G), glucosidase (H), and leucine aminopeptidase activities (I).

Figures

Figure 1

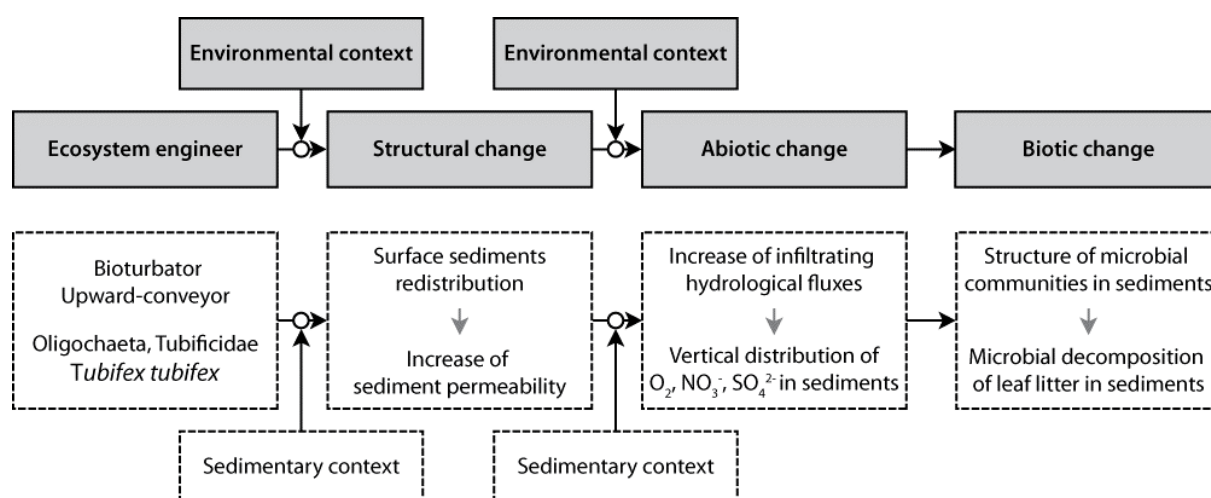
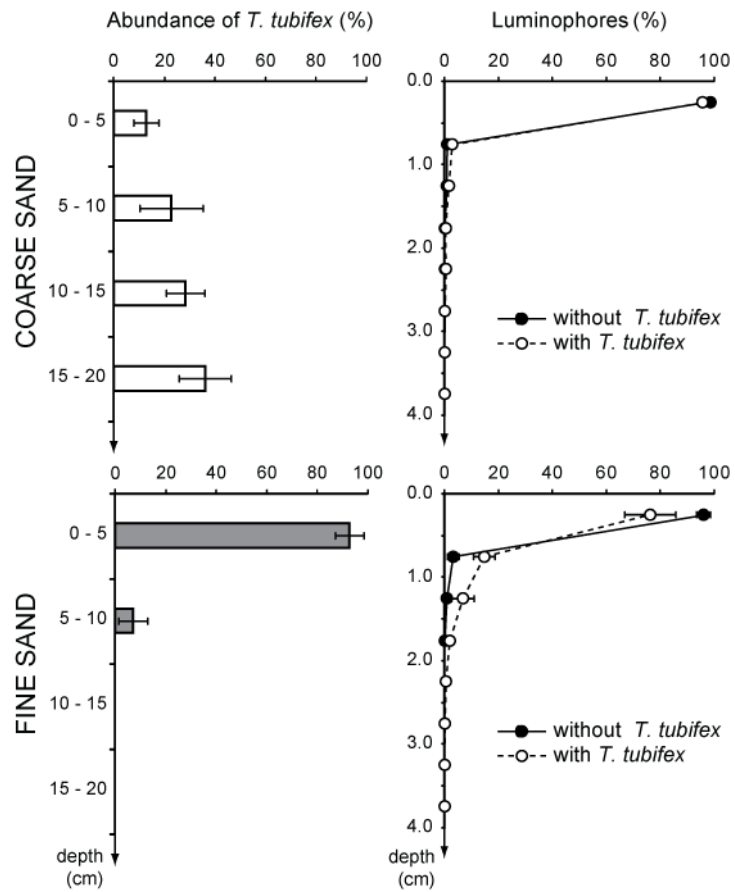
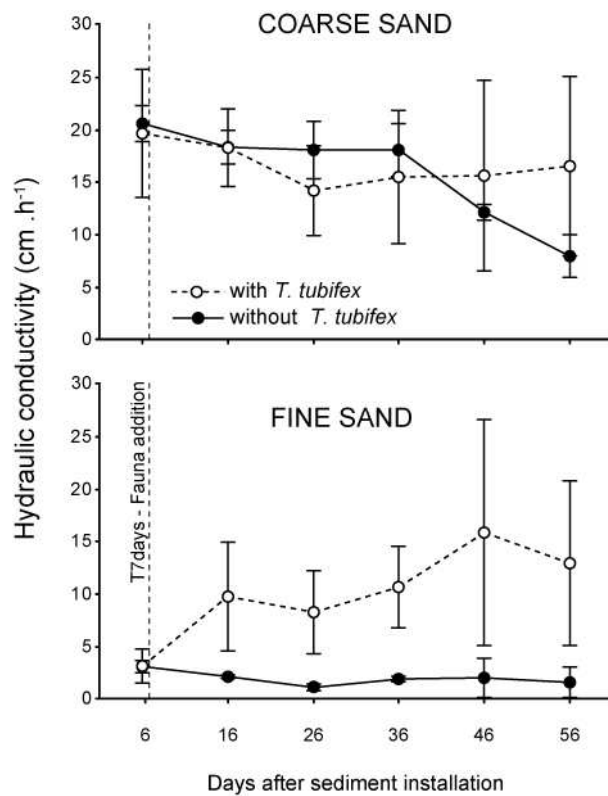


Figure 2

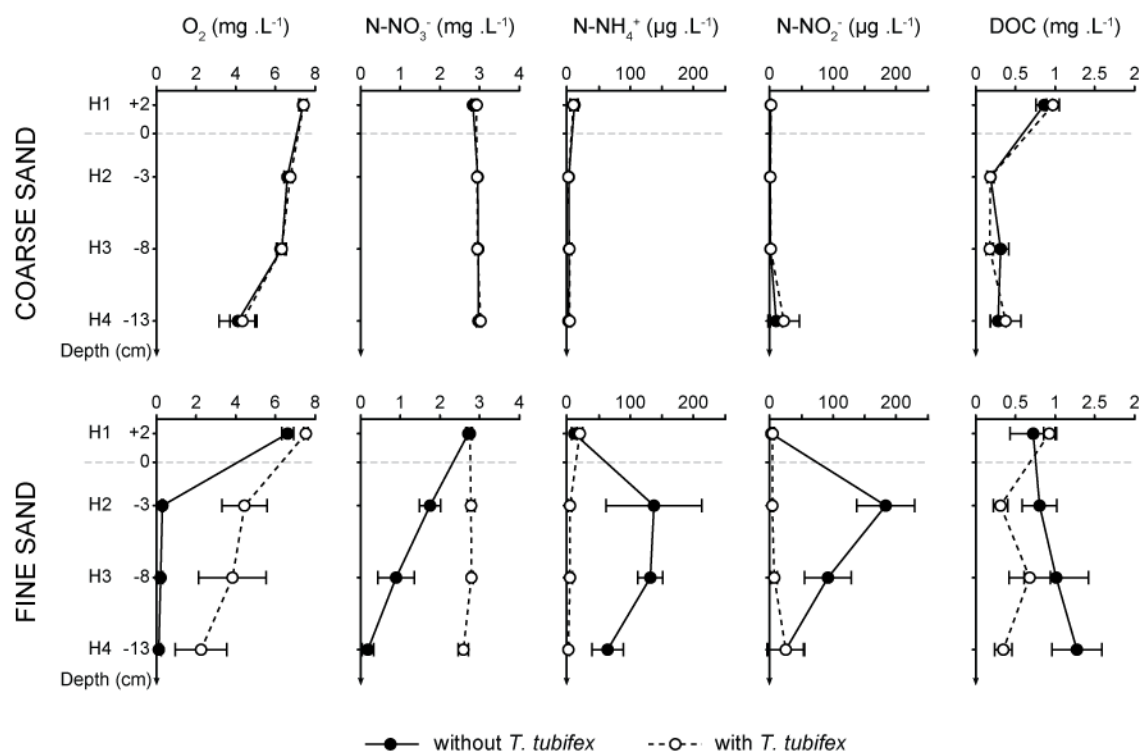


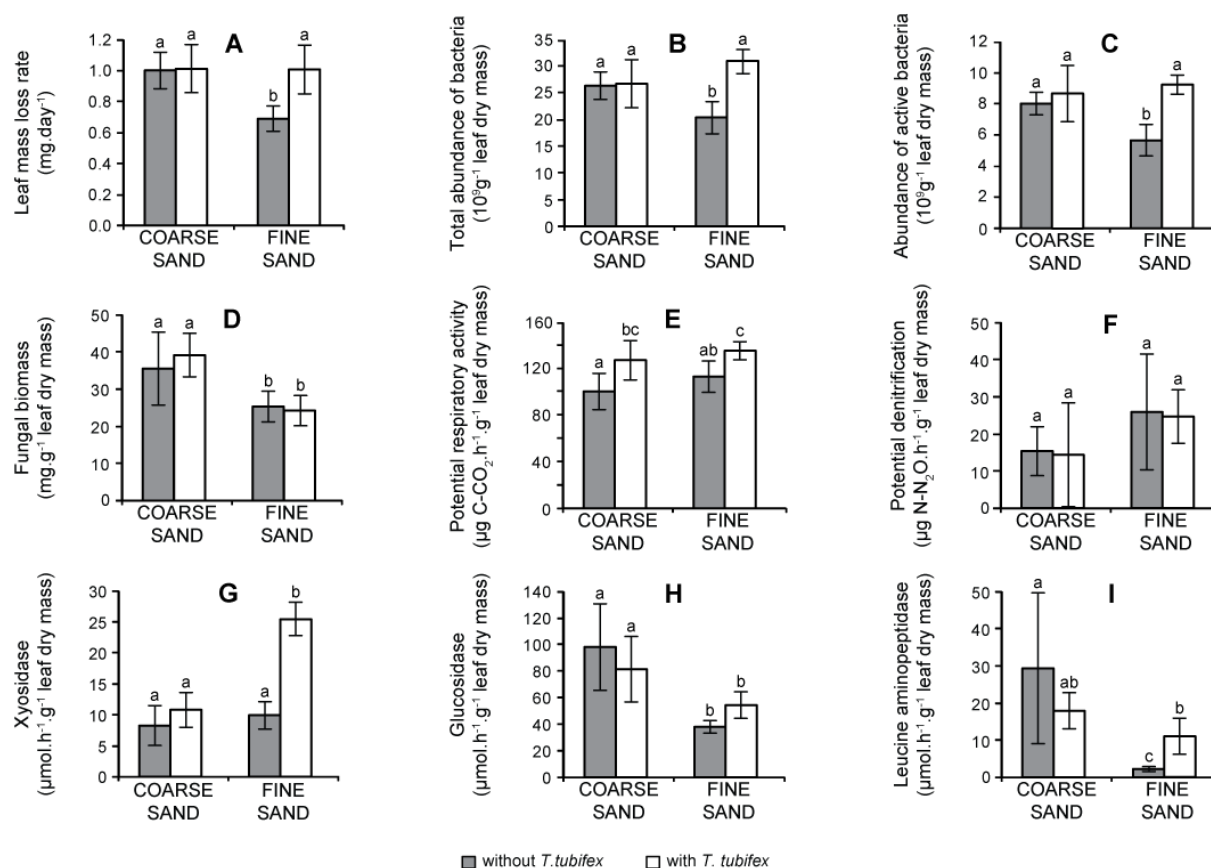
1

2 Figure 3



3





1