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Fine particulate organic matter (FPOM) transport and processing in littoral interstices – use of fluorescent markers

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Abstract

A fluorescent labelling method is presented as a new tool for the investigation of organic particle transport and biogenic carbon cycling processes in sandy littoral interstices at Lake Tegel, Berlin, Germany. Passive particle transport through the pore system was studied by in situ exposition of 2.4 μm monodisperse polymeric resin microparticles stained with 7-amino-4-methylcoumarin (AMC). Uptake of fluorescein-5-isothiocyanate (FITC)-labelled *Chlorella vulgaris* and fine particulate organic matter (FPOM) by the interstitial fauna was investigated in laboratory and field experiments. The major portion (>85%) of the FITC-labelled particles added to sediment cores was recovered from the topmost centimetre of sediment during the study period of 14 days. Uptake of FITC-labelled FPOM was observed in several benthic groups, e.g. chironomids, microcrustaceans, oligochaetes and tardigrads, whereas *C. vulgaris* was ingested by oligochaetes only. There is evidence to suggest that both are suitable materials for investigating the fragmentation and ingestion of organic material by herbivorous and detritivorous fauna. Field experiments with inert microparticles and FITC-labelled FPOM (<1 mm) prepared from dried alder leaves were carried out in plexiglass tubes as in situ whole core technique. Within the investigation period of two weeks, the transport of FPOM was restricted to the topmost 2–3 cm of sediment in conjunction with a distinct fragmentation to finer size classes with respect to increasing sediment depth. Vertical FPOM transport was hindered by a high interstitial concentration of natural POM and an intensive settlement of the interstices by algae (mainly epispammic algae, 65–96% of algae cell number) and extra-cellular polymeric substances (EPS), which formed a dense three-dimensional structure.

Keywords: Fluorescein-5-isothiocyanate (FITC); Fluorescence marker; Littoral sediment; Particulate organic matter (POM); Groundwater recharge; Interstices; Interstitial

Introduction

The transition between streams or lakes and groundwater, the hyporheic biotope or interstitial zone, is a focus of interest in water management, applying the process of water infiltration for groundwater recharge and bank filtration for drinking water supply. Many investigations of the riverine bank filtration zone and slow sand filtration describe the interstitial flora and fauna, and verify self-purification processes during infiltration (Higgins and Thiel, 1988; Graham and

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Collins, 1996; Kühn and Müller, 2000), whereas knowledge of the sandy littoral zone in lenitic ecosystems is scarce and only a little information is available about turnover and retention of particulate organic matter (POM) and dissolved organic matter (DOM; Beulker and Gunkel, 1996; Hakenkamp et al., 2002).

Carbon flux within the interstices during the infiltration process is not only determined by the influx of DOM (among others cyanotoxins), but also by the deposition of POM (detritus, living algae cells), the primary production of epispammic and interstitial algae, the transport of fine POM (FPOM) to deeper sediment layers and the mineralisation of POM and DOM. Mechanical wave effects and topography-related pressure gradients enhance advective forces that favour interstitial transport (Huettel and Rusch, 2000). The infiltration process is also strongly influenced by colonization of sediment surface and interstitial spaces by epispammic and interstitial algae and bacteria. They participate in metabolic processes and have a strong effect to the extent of biological clogging by production of extra-cellular polymeric substances (EPS; Gunkel and Hoffmann, 2006).

The role of bioturbation processes in the upper sediment layer has frequently been studied using different natural and artificial tracer materials, e.g. radionuclides, colour- or fluorochrome-dyed microspheres, laboratory cultivated algae, sediment particles or metal-doped sediment (Gerino et al., 1998; Solan et al., 2003). Studies on fine particle transport in the interstices are mainly reported from rivers using fluorescent-labelled bacteria (Hall et al., 1996), yeast (Paul and Hall, 2002), pollen (Wanner and Pusch, 2000) or radio-labelled seston particles (Georgian et al., 2003). But still the choice of added POM in experiments is insufficient; bacteria, yeast and seston are minor representatives of POM in limnic waters and cannot be used to detect penetration and shredding of POM by interstitial organisms. The use of cultured algae is a better approach (Huettel and Rusch, 2000), but it is not possible to distinguish them from in situ grown algae.

A novel fluorescein-5-isothiocyanate (FITC) fluorescent labelling method allows the study of vertical particle transfer in sediments by the application of FITC-labelled natural organic substrates (e.g. algae, leaves) originating from the sampling site as well as analysis of the biological decay of particulate organic material. With a particle size ranging from 10 to 1000 μm being used, this covers a large and varying size spectrum. The ingestion of FITC-labelled organic matter by the fauna was positively tested in advance by experiments with both cultured organisms and meiofauna inhabiting the studied sediment.

Transport studies were conducted in in situ cores as whole core technique, which were exposed in the shallow (<0.5 m depth) littoral zone of the Lake Tegel, Berlin, a mesotrophic lake. The area studied is characterized by lake shore erosion and reed die-back; about 100 m from the lake shore a well gallery (down to 60 m depth) is used for water abstraction as bank filtrate for drinking water supply.

Focus of the investigations is the analysis of the lake/low land river infiltration zone, the interstitial, as a small boundary layer of about 30 cm depth with high biological activity (Beulker and Gunkel, 1996) and to evaluate the significance of the biotope for the self-purification processes in natural and induced groundwater recharge (Hoffmann and Gunkel, 2006). Clogging phenomena occur, but up to now the significance of the processes such as mechanical (e.g. POM input, gas bubbles), chemical (e.g. precipitation of calcium carbonates) and biological effects (e.g. the formation of extracellular polymeric substances, EPS) are not sufficiently known. The potential for DOC reduction of the infiltration water (in the range 20–40%) and the source of the remaining DOC of about 5 mg L^{-1} DOC in the ground water (Grünheid et al., 2005) makes it necessary to analyse not just the DOC input with the lake's water but also DOC formation and mineralisation within the interstices.

Material and methods

Study area

Lake Tegel is a lowland lake situated in Berlin, a glacial enlargement of the River Havel, with an area of 396 ha and a mean depth of 6.6 m (Table 1). Water balance and water quality is determined by the inflow of two rivulets (Tegeler Fließ, Nordgraben) and by water exchange with the River Havel (Fig. 1). After a severe eutrophication period, a phosphate elimination plant was built and nowadays the lake water quality is mesotrophic, but organic rich anoxic sediments are still typical and periodically cyanobacteria blooms occur (Heinzmann and Chorus, 1994; Schauser et al., 2006a, b).

Lake Tegel has a sedgy littoral zone with forest stands, sporadically interrupted by beach sections caused by erosion, with sparse macrophyte growth (reed *Phragmites australis,* water lily, *Nuphar lutea*). Near the shore of Lake Tegel, bank filtration for Berlin's water supply is done through several galleries with 116 wells, 30–60 m deep, distance to the lake is about 100 m. The pumping rate for each well amounts from 50 to 150 $m^3 h^{-1}$.

Experiments were conducted at a water depth of 30–50 cm, about 3 m away from the splash water zone, and different micro-habitats were studied (sandy erosion shore, *Phragmites* stand, *Nuphar* stand) at the eastern shore of Lake Tegel (N52°34′13.81″ E 13°15′25.52″ to N52°34′10.65″ E13°15′24.42″).

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Table 1. Characteristics of Lake Tegel, Berlin (Jahn and Witt, 2002; Schauser et al., 2006a, b; Senatsverwaltung, 2006).

Lake Tegel	
Length	3.2 km
Width	1.7 km
Area	396 ha
Depth	
Maximum	15.9 m
Mean	6.6 m
Volume	22×10^6 m^3
Water residence time	70 (main basin)–180 d
Inflow:	River Havel, Tegeler Fließ, Nordgraben
Outflow	River Havel
Catchment area	151 km^2
Water quality class	mesotrophic (II)
P_{total} inflow concentration (mean year^{-1}) (Phosphate elimination plant since 1985)	20 µg L^{-1}
P_{total} lake (mean summer 2006)	<30 µg L^{-1}

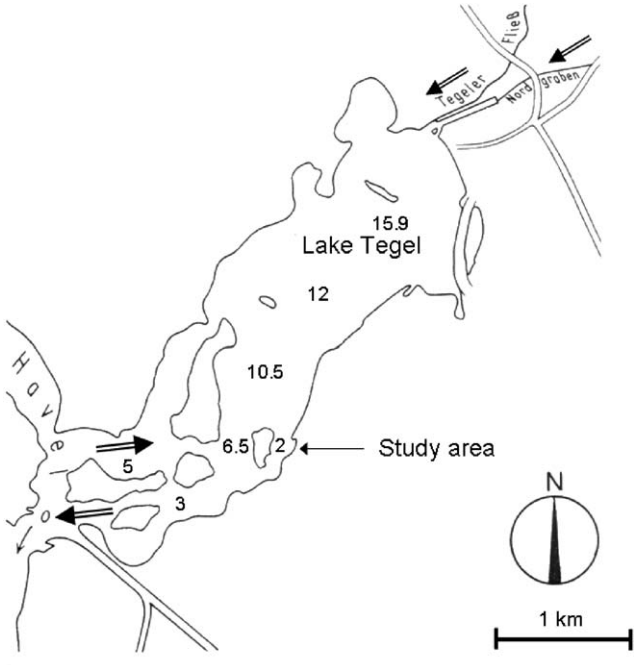

Fig. 1. Lake Tegel and the study area, given are the water depths; the arrows mark water inflow and outflow. The study area includes sandy erosion shore, *Phragmites australis* stands and *Nuphar lutea* stands.

Interstitial characteristics of Lake Tegel shore

Sediment have a medium size distribution range and are composed of medium to fine sand ($D_{50} = 0.38$ mm) with a low content of particulate organic matter (0.7%; Table 2).

Table 2. Granulometric characteristics of the littoral sediment of Lake Tegel (water depth 30 cm) in different sediment depth (each one mean of two 5 cm layers.

	0–10 cm	10–20 cm	20–28 cm
Median particle diameter (mm)	0.14	0.26	0.23
Particle diameter of 25% (D25)	0.08	0.16	0.16
Particle diameter of 75% (D75)	0.19	0.37	0.30
Sorting coefficient (Müller)	1.48	1.54	1.43
Quartile skewness (Müller)	0.91	1.07	1.11
Water content (%)	30.3	13.0	17.2
Organic content (%)	0.77	0.41	0.41
k_f value (Hazen, 10 °C) (m s^{-1})	5.0×10^{-5}	6.5×10^{-5}	9.0×10^{-5}
Porosity	0.40	0.29	0.36

At Lake Tegel the sediment particles were densely colonized mainly by epipsammic algal attached to sediment grains (abundance: 65–95%). Diatoms were the dominant species; the most prevalent diatom species were *Amphora pediculus*, *Cocconeis* spp., *Achnanthes clevei*, *A. minutissima* and *A. lanceolata*. Some interstitial algae such as nanoflagellates (*Trachelomonas*, *Euglena*) were also found.

The littoral sediment is populated by a diverse and abundant benthic biocoenosis with maximum meiofauna abundances of around 20,000 ind. dm^{-2} (Beulker and Gunkel, 1996). The meiofauna consists of nematodes, oligochaetes, harpacticoids, ostracods and chydorids as dominant taxa with a high abundance in 0–5 cm depth, but occurred too in significance abundance in 5–10 cm (Table 3). Studies of depth differentiated profiles in 1991 and 1994 showed that more than two-thirds of the meiofaunal abundance is regularly concentrated in the uppermost sediment layer of 0–10 cm; only nematodes and oligochaetes were found to colonize the sediment much deeper, down to the maximal studied depth of 70 cm (Beulker and Gunkel, 1996).

Sampling and analysis methods

Grain-size distribution of sediment was assessed by wet sieving and subsequent drying and weighing of the screened fractions using the EN ISO 14688 (2002/2004), soil sorting coefficient and the quartile skewness was calculated after Gray (1981); water content was determined at 105 °C, organic content was analysed by loss on ignition (LOI, 550 °C).

Permeability of the sediment was estimated with the Hazen's method (k_f = permeability in m s^{-1}; Beyer,

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Table 3. Colonization of the littoral sediment by frequent meiofaunal taxa; mean and maximum abundance in Lake Tegel, 2004, in 0–5 and 5–10 cm sediment layers ($n = 8$).

Taxon	Abundance (Ind. dm^{-2}) 0-5 cm sediment depth		Abundance (Ind. dm^{-2}) 5-10 cm sediment depth	
	Mean	Maximum	Mean	Maximum
Nematoda	2,671	6,370	1,716	5,366
Tardigrada	231	1,688	35	137
Oligochaeta	111	389	30	130
Ostracoda	163	375	57	230
Harpacticoida	1,210	5,963	237	1,030
Phyllopoda	109	582	122	770
Chironomidae	54	150	19	125

1964) considering particle-size distribution and water temperature.

Pore water velocity ($m\,h^{-1}$) was calculated from infiltration rate and mean porosity ($n = 6$ over a sediment horizon of 0–30 cm), according to the following equation:

$$v_p = \frac{v_f}{n_f} = \frac{Q}{Axn_f}$$

v_p is the pore water velocity ($m\,h^{-1}$); v_f is the filter velocity ($m\,h^{-1}$); n_f is the flow through relevant proportion of the interstitial (= porous volume); Q is the flow through ($m^3\,h^{-1}$); determined with the plexiglass chambers; A is the area (m^2).

Porosity was calculated by

$$\Phi = \frac{(w/\rho_W)}{(w/\rho_W) + (1-w)/\rho_S}$$

Φ is the porosity, w is the water content (%); ρ_W is the density of water at the given temperature ($g\,cm^{-3}$); ρ_S is the density of dried sediment ($g\,cm^{-3}$).

FPOM was measured gravimetrically as LOI after cascade floating separation from sand and silt and elimination of the meiofauna. Depth profiles of meiofauna were studied using the freezing core method with liquid nitrogen, described in detail in Beulker and Gunkel (1996).

The chlorophyll-a content was determined using the photometric method described in DIN 38412 (DEV, 1986). Carbohydrate was analysed after extraction of 1 g sediment d. w. with 1 n EDTA, using the method of Dubois; calibration was done with glucose. Proteins were determined with the Bradford solution, sampling volume was 1 mL, a standard of bovine serum was used (further details see Hoffmann and Gunkel, 2009).

Core experiments were performed as whole core technique in situ in the littoral zone of Lake Tegel in February 2004. Experiments were carried out in plexiglass tubes with a diameter of 6.2 cm and a length of 50 cm, which were pressed into the sediment down to 30 cm. The selected sediment core depth had to be at least 5 cm deeper than the maximal observed transport depth of the fluorescence marker to avoid any environmental contamination. Each core was topped with a PVC cap. FITC-labelled FPOM was added directly to the exposed plexiglass cores as an aqueous suspension to achieve a final load of 5 mg POM (d. w.) cm^{-2}. After injection, the core cap was removed to allow lake water to infiltrate through the sediment core. Exposure times were 4 h (starting after complete FPOM sedimentation), 2 and 14 days.

For sampling, the sediment cores were divided into layers of 0.5 to 1 cm each. Overlying water was removed and added to the surface layer sample. Five to ten parallel samples containing 0.05–0.1 g sediment each were counted for FPOM abundance in Utermöhl chambers. FPOM particles were differentiated into the size classes of $\leqslant 20$, 20–200 μm and > 200 μm. Recovery rate of FPOM particles in the sediment after stocking to the sediment cores amounted to $82 \pm 10\%$ for particles 20–1000 μm. But the smaller fraction of particles $\leqslant 20$ μm had only a recovery rate of $43 \pm 10\%$, due to partial covering by sediment particles in the microscopic analyses.

For scanning electron microscopy with energy dispersive spectrometer (SEM-EDS) wet mini cores (stainless steel tubes, 1 cm height, 0.7 cm ∅) were pressed in the cores sampled at the shore, and superficial sediment as well as sediment from different depth in divided cores were taken; the mini cores were air dried or critical point dried and then sputtered directly with gold and inserted completely into the vacuum chamber of the SEM. A SEM-EDS Hitachi S 2700 electron microscope was used with an acceleration voltage of 20 kV and an IDFix hard and software from SAMx for analysis.

For statistical comparison of distinct layers within different samples of sediment cores and of time intervals one-tailed U-test for two independent parameters were applied to FITC-labelled FPOM data in order to detect significant differences in particle numbers in the upper sediment layer with exposure time. H-test for three independent parameters was used to test for significant alterations in particle number of inert AMC-labelled particles with time: Three sets of samples ($n_i = 9$) from

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corresponding sediment depths at three different times of exposure (3, 7 and 14 d) were tested for significant differences in particle number.

In situ infiltration

In situ infiltration rates at the study site were determined by use of plexiglass chambers (19 cm inner diameter, 20 cm height) pressed 2 cm deep into the sediment and connected pressurelessly via tube fittings to plastic bags (Hollister Inc.), filled with a weighed amount of lake water (about 0.5 L each one). After 24 h the bags were retrieved and reweighed. Daily infiltration rates during a study period of 4 days were assessed monthly from March 2004 to April 2005 at two sampling sites. Simultaneously the hydraulic potential ($n = 3$ per chamber) was measured and k_f values were calculated according to Darcy's Law:

$$k_f = \frac{Q}{A} x \frac{dl}{dh}$$

Q is the amount of infiltrating water per time ($m^3 s^{-1}$); A is the area (m^2); dh is the decrease of the hydraulic potential over the sediment length dl.

Experiments with labelled tracers

The uptake and transport of 7-amino-4-methylcoumarin (AMC)-labelled melamine resin particles and of fluorescein-5-isothiocyanat (FITC)-labelled fine particulate organic matter (FPOM) were studied at the shore of Lake Tegel as well as in laboratory experiments using sediment cores from the same site.

Melamine resin particles of 2.44 ± 0.04 μm diameter (mean ± SD) labelled with 7-amino-4-methylcoumarin (AMC) were used, manufactured by microParticles GmbH, Berlin, Germany; these resin particles are applied as patented particle standards, procedure of labelling resin particles is a trade secret of microParticles GmbH, Berlin. The blue fluorescing MF-AMC particles (λex 360 nm/λem 429 nm) have a density of $1.5\ g\,cm^{-3}$ and a hydrophilic surface charged positively by amino groups. The particles were delivered in aqueous medium. Concentrations of $2.1 \times 10^7 \pm 4.8 \times 10^6$ particles mL^{-1} and $4.8 \times 10^7 \pm 5.6 \times 10^6$ particles mL^{-1} were used for the studies on microfine particle transport in Lake Tegel sediments. Laboratory studies showed a recovery rate of the MF-AMC particles of 64–88% for particles spiked to sediment cores (surface load 7×10^5 particles cm^{-2}), the recovery rate is less than 100% because when using microscopic counting in sedimentation chambers, some particles were covered by sediment particles and thus rendered invisible.

Dried FPOM from alder leaves (*Alnus glutinosa*) and algae (*Chlorella vulgaris;* strain from IGV, Bergholz-Rehbrück, Germany) were labelled with fluorescein-5-isothiocyanate (FITC, λex 506 nm/λem 529 nm), also produced by microParticles GmbH, Berlin. The method of FITC labelling of dried organic material was developed by microParticles GmbH for this study and is a trade secret of the company. Labelled material was delivered in aqueous medium. The fluorescently labelled FPOM was stored at a stock concentration of $67\ g\ L^{-1}$ FPOM (d. w.) at 8 °C. *C. vulgaris* had a mean cell diameter of 13 ± 6 μm and was stored at a concentration of 20 g dried algae L^{-1} at 8 °C. Alder leaves were dried and ground to a particle size of < 1 mm in accordance with the FPOM classification of Wotton (1994).

The particle-size distribution of the FPOM tracers (alder leaf litter) revealed the following size fractions classes: ⩽20 μm: 37.6%; 20–100 μm: 39.8%; 100–200 μm: 18.1%; 200–1000 μm: 4.5% of total number ($n = 2000$). Whereas the smallest particles of ⩽20 μm constitute the most abundant fraction of FPOM, the maximal mass (determined by surface area) is represented by particles of a length of 140 μm.

Microscopic studies for FITC were performed using a light microscope (Zeiss Axioskop) equipped with a UV lamp (Osram HBO 50) and Neofluar objectives of different magnification strengths. Zeiss filter set 09 (BP 450-490, FT 510, LP 515) was used in the FITC studies, and filter set 02 (BP 365, FT 395, LP 420) was used in the AMC fluorescence studies. Filter set 02 was also used to distinguish FITC fluorescence from the green autofluorescence of chitin; chitin fluorescence is bright blue through filter set 02, whereas FITC fluorescence looks green through both filter sets.

Fig. 2 illustrates the microscopic appearance of the AMC and FITC fluorochromes. In general, the microscopic investigation of FITC-labelled FPOM does not show interference with the fluorescence of other particles and organisms in the sediment, the only relevant fluorescence signal from other particles is the red autofluorescence of algae. Background colour and chitinous substances of invertebrates appear dull green, which is normally easily distinguishable from the bright green associated with fluorescein emission. Only the chitinous intestinal tract of some nematode species shows a bright green fluorescence; in cases of doubt, we used filter set 02.

Two in situ core experiments with MF-AMC microfine particles were performed in the littoral zone of Lake Tegel during August 2003 and April 2005 to study the passive vertical transport by water flow. MF-AMC particles were first ultrasonicated to prevent particle agglomeration, then added to the sediment in in situ cores at concentrations of $2.1–4.8 \times 10^7$ particles mL^{-1} ($= 7–16 \times 10^5$ particles cm^{-2}); the particles were exposed to the cores for 3, 7 and 14 days. For sampling, cores were transported to the laboratory and divided into layers (see above). For counting, an aqueous suspension was homogeneously shaken onto a vortex

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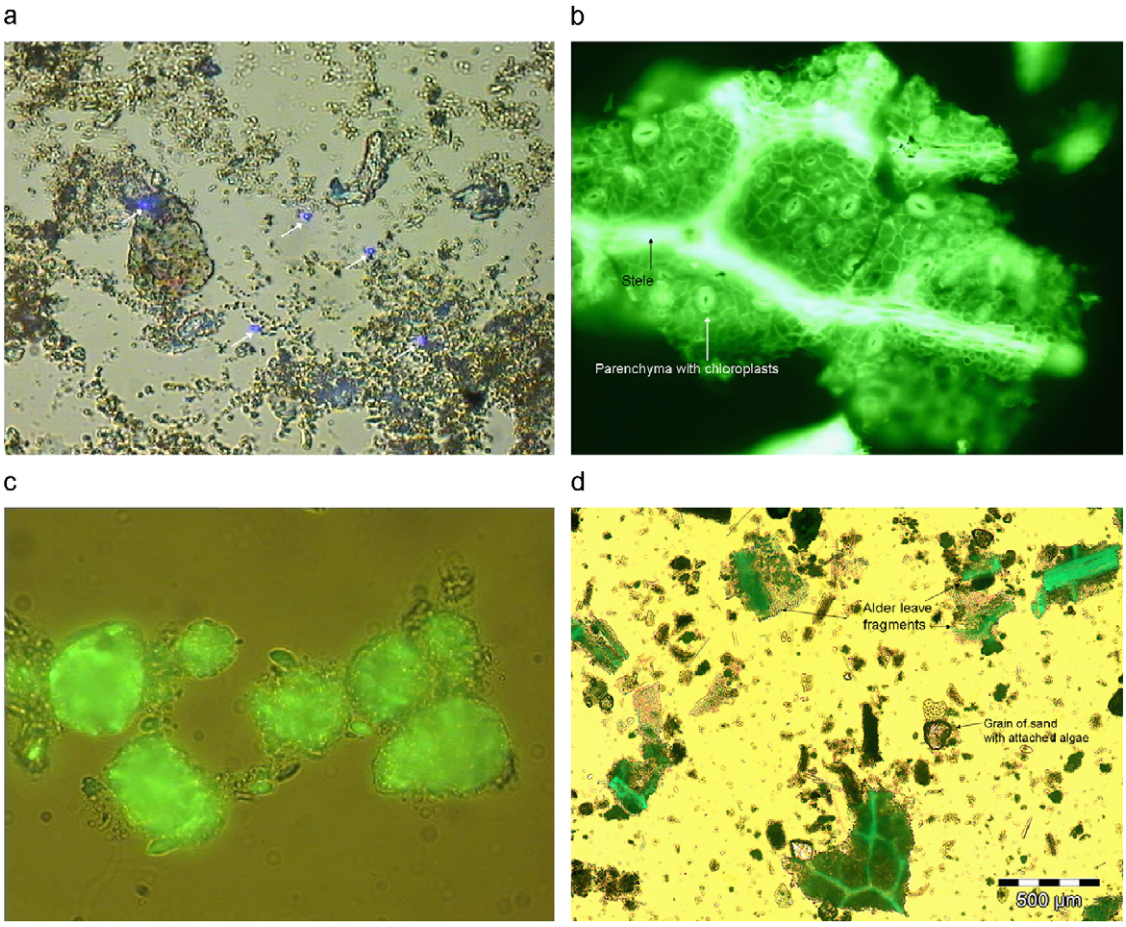

Fig. 2. Microscopic view of the fluorochrome tracers: (a) AMC-labelled resin particles after in situ exposure in the littoral zone of Lake Tegel, arrows mark the AMC resin particles, ∅ 2.44 µm. (b) FITC-labelled FPOM from alder leaves in the stock suspension, (c) FITC-labelled *Chlorella* spec. cells in the stock suspension, (d) FITC-labelled FPOM from alder leaves after in situ exposure in the littoral zone of Lake Tegel. a, c and d by epifluorescence microscopy in combination with transillumination microscopy; b by epifluorescense microscopy.

mixer, and three sub samples of 10 mL were transferred to Petri dishes and dried.

The leaching of the fluorochrome FITC from the labelled algae and leaves into the water was tested with a diluted standard suspension of 0.3 g L^{-1} FITC-labelled FPOM in tap water, which was compared to an unlabelled FPOM suspension of the same concentration. During a period of 40 days, the test suspensions were stored at room temperature in the dark. Subsamples were analysed spectrophotometrically at λ 480 nm after filtration with 2 µm cellulose nitrate filters.

The ingestion of FITC-labelled particles by invertebrates was studied in preliminary tests. Fluorescently labelled *Chlorella* cells were offered in varying concentrations as food for cultured *Daphnia magna* as well as food for algivorous benthic organisms. Sediment samples were spiked with algae to concentrations of 0.05–0.7 mg algae (d. w.) cm^{-3} sediment and incubated for 2–10 days at 6 °C. Fluorescently labelled FPOM from alder leaves was offered to meiofaunal organisms to yield a final concentration of 0.1 g L^{-1} FPOM (d. w.) in tap water. In cases where no fluorescence signal was found in the intestines of the organisms, FITC-labelled FPOM concentration was doubled every day to prevent a possible nutrient limitation.

Results

Leaching and uptake of FITC-labelled fine particulate organic material (FPOM)

Possible leaching of FITC-labelled algae and leaves into water was tested with suspension of FITC-labelled FPOM; the extinction was caused by absorption of FPOM chlorophyll-a as well as by FITC, which both have absorption maxima at a wavelength of 480 nm. In a freshly hydrolysed unlabelled FPOM suspension, a maximum of absorbance was observed after 1 day due

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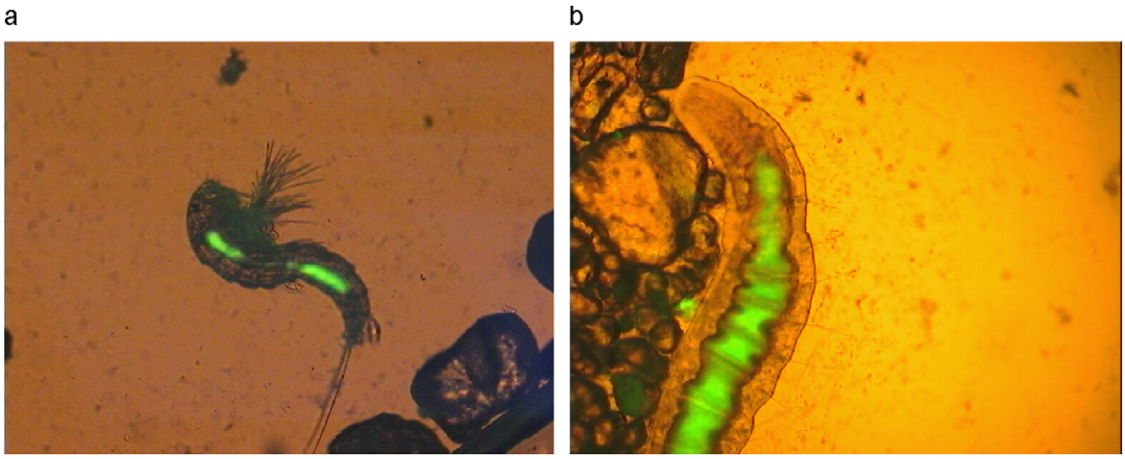

Fig. 3. Fluorescence excitation in the intestines of (a) a harpacticoid and (b) an oligochaete after exposure in in situ cores in the littoral zone of Lake Tegel spiked with (a) 5 mg FITC-labelled FPOM (alder leaves) cm^{-2} sediment for 14 days, and (b) 10 mg FITC-labelled FPOM (alder leaves) cm^{-2} sediment for 3 days. Combined transillumination light and epifluorescence microscopy with Zeiss filter set 09.

to rapid leaching of chlorophyll-a from the leaves. FITC-labelled FPOM did not show this peak, indicating that leaching of chlorophyll-a was already completed in the course of the labelling process and in the aqueous stock suspension. Absorbance in the FITC-labelled FPOM suspension was nearly constant during the 40 days observation period. The higher concentrated stock suspension showed a strong fluorescence signal even after 18 months.

Laboratory tests provided evidence of uptake of FITC-labelled material in form of algae and FPOM from alder leaves by several groups of organisms. After offering FITC-labelled *C. vulgaris* cells to *D. magna*, intestinal fluorescence was observed between 1 h and 8 days, depending on the concentration and prevailing feeding intensity. Offering a mixture of non-labelled and FITC-labelled algae at a ratio of 1:1 did not result in a change of occurrence of intestinal fluorescence. Therefore, ingestion of *C. vulgaris* marked with FITC is not inhibited and no preference for unlabelled algae was observed.

Additionally, FITC-labelled *C. vulgaris* was added to sediment samples containing macroinvertebrates; oligochaetes (*Nais elinguis*), known as typical herbivorous organisms, showed intestinal fluorescence within the observation period of 10 days.

The uptake of FITC-labelled FPOM from alder leaf litter by detritivorous and omnivorous organisms was observed in nearly all of the macroinvertebrates (gastropods, chironomids, cyclopoids, harpacticoids, ostracods, tardigrads, nematodes, oligochaetes; Fig. 3). This material, which was not preconditioned by natural bacterial colonization, was broadly accepted by about 50% of the total population in concentrations to give a clear fluorescence signal. An incubation time of 1–2 days was sufficient to induce the uptake of FITC material. No signs of intestinal fluorescence were found for microfaunal rotifers and ciliates, whose prevailing feeding strategies are bacterivore and saprotrophy, as well as for chydorids, filter feeders, being minor in abundance. Death of the organisms led to rapid leaching of the metabolised dye through the body.

Concentration and vertical distribution of natural particulate organic material

The natural depth distribution of POM and the different size classes of FPOM in the littoral sediments of Lake Tegel between March and October 2004 is given in Fig. 4. Maximal percentages of POM, measured as LOI, were detected in the first 5 cm (1.5% d. w.). Below this sediment horizon, POM declined relatively constantly to values down to $<0.5\%$ in 35 cm. In depths of 0–20 cm, higher proportions of the middle and fine POM size classes indicate a zone where fragmentation takes place due to feeding activities of macroinvertebrates. The occurrence of POM >1 mm in deeper layers (>20 cm) was mainly due to fine rhizomes of the littoral macrophytes.

Besides POM input, primary production is assumed to be another source of organic carbon in the interstices; the vertical distribution of chlorophyll-a (Chl a) confirmed that interstitial algae biomass forms a significant part of the total POM in the upper interstitial zone of 0–6 cm. The Chl a concentration was very high ($21–28\ \mu g\ cm^{-3}$) in the upper 5 cm of sediment and decreased with depth, with only traces of Chl a detected at 9 cm (Fig. 5). The Chl a concentration of about $25{,}000\ \mu g\ dm^{-3}$ must be evaluated as extremely high compared with the lake water, which even under eutrophic conditions contained only about $20\ \mu g\ L^{-1}$.

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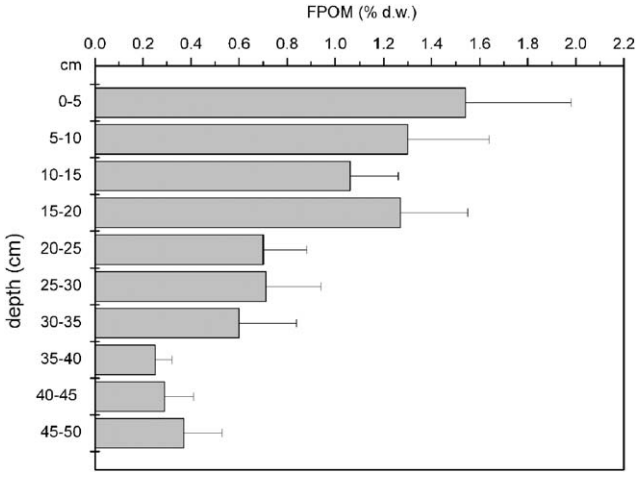

Fig. 4. Depth distribution of different size classes of fine particulate organic matter (determined as loss on ignition) in the littoral zone of Lake Tegel in 2004; data presented are means + SD ($n = 6$).

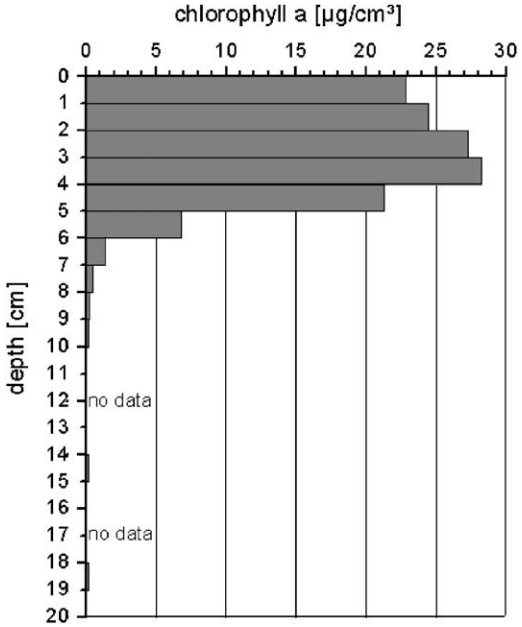

Fig. 5. Vertical chlorophyll-a concentrations within the upper littoral sediment layer of a bank filtration site at Lake Tegel (mean values from March to August 2004, $n = 11$).

The total algal biomass in the upper sandy layer of the interstitial was about 1000 times higher than in the overlying water body of the Lake Tegel.

The origin of these algae were epipsammic species such as *Fragilaria* spp., *Achnanthes* spp., *Cocconeis* spp., *A. pediculus*, *Cymbella* spp., *Gomphonema* spp., *Rhoicosphenia abbreviata* and *Cymatopleura* spp. as well as some interstitial algae (Table 4). Of high interest is the low significance of planktonic algae of the lake water, which reached only three times during the investigation period a significant portion of <24% of the total interstitial algae abundance; in general the relative abundance of the planktonic algae was <3%. Only at the surface of the sand layer a high amount of planktonic algae occurred; this surface serves as a filter with an accumulation of the planktonic algae (Fig. 6a).

Diatoms as well as bacteria are known to produce extra-cellular polymeric substances (EPS) which is of high significance for the three-dimensional structure of the biofilm being spread on the sand grains. The surface of the sand layer is settled by diatoms, which build together with the bacteria and algae a three-dimensional EPS structure at the surface of the sandy sediment (Fig. 6b) as well as in the deeper layers of the interstitial. Different EPS structures like filamentous net or extensive layers were registered which fill up the pore system to a high extent (see Hoffmann and Gunkel, 2009). This EPS contributes to the amount of particulate organic matter and serves as a mechanical filter for the introduction and migration of POM in the interstitial.

It is known that the EPS is built up of proteins and polysaccharides and bacterial DNA; chemical analyses of proteins and carbohydrates done in vertical profiles detected up to 649 $\mu g\,g^{-1}$ carbohydrates and 50.6 $\mu g\,g^{-1}$ proteins in the upper sandy sediment layer, that is 6% carbohydrates and only 0.5% proteins of the total FPOM in the interstices. To the depth of 20 cm the concentrations decreased to 69 $\mu g\,g^{-1}$ carbohydrates, respectively, 18.5 $\mu g\,g^{-1}$ proteins.

Vertical transport of fine particles in the interstices

The k_f values according to Hazen are in the range $10^{-4}\,m\,s^{-1}$ and permit a high potential permeability of the sediment. In contrast to the high k_f values calculated according to Hazen (see Table 2), k_f values based on in situ infiltration rates are significantly lower (5×10^{-5} to $10^{-8}\,m\,s^{-1}$) and indicate severe clogging processes in the interstices. Daily infiltration rates amount to 9.0 $L\,m^{-2}\,h^{-1}$, showing seasonal and local variations (sandy area, *P. australis* and *N. lutea* stands) between 2.2 and 20.9 $L\,m^{-2}\,h^{-1}$ (10 and 90 percentiles, $n = 24$). The infiltration rates correspond to a water flow within the sediment pore system (as median filter velocity) of 54 $cm\,d^{-1}$.

Assuming a monodisperse spherical sediment composition of the littoral sediment in the range of the lake's mean particle size of 380 µm, the packing will create pores of 59 to 154 µm diameter depending on the packing density, due to the rather low sorting coefficient of 1.6 the majority of the pores are probably smaller ones. Therefore, particles of the size class ⩽20 µm should be able to pass the pore system by passive transport with the water flow without hindrance, but only a portion of the particles of size class 20–200 µm are

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Table 4. Ecological typing and abundance of diatom algae in the interstices of the sandy littoral zone of Lake Tegel, + = occurrence with very low abundance.

	1992						1993
	February	April	June	August	October	December	February
Planktonic algae							
Aulacoseira granulata v. *granulata*	+	+	+	0,49	0,48	12,50	0,50
A. granulata Morphotyp curvata							+
Cyclotella cf. *bodanica*		0,99	+			+	+
Cyclotella spp.		2,96				0,50	+
Cyclostephanos dubius	+	+	+			+	+
Stephanodiscus hantzschii	+	+	+	+		1,00	1,50
S. neoastraea	+	+					
Stephanodiscus spp.	0,92	13,79	0,50	0,49		4,00	17,00
Actinocyclus normannii	2,30	6,40	0,50	0,49		2,50	+
Asterionella formosa	+	+				1,00	+
Tabellaria flocculosa	+						
Fragilaria heidenii							
F. ulna sensu lato		+				+	
Sum	3,2	24,1	1,0	1,5	0,5	21,5	19,0
Planktonic/benthic algae							
Diatoma spp.	+	+		+		0,50	+
Fragilaria spp.	+	0,49	1,00	0,49	0,48	0,50	0,50
Pinnularia sp.						+	
Not differentiated algae	9,68	2,96	5,50	1,48	4,83	6,50	10,00
Sum	9,7	3,4	6,5	2,0	5,3	7,5	10,5
Interstitial algae							
Melosira varians							+
Fragilaria ulna v. *acus*	+	0,49					+
Amphora cf. *aequalis*		0,99					
Navicula gregaria	+						+
N. schroeterii	+					+	
N. reinhardtii	+						+
Neidium cf. *apiculatum*		+					+
Pinnularia viridis	0,46						
Nitzschia linearis		+			+	+	+
Nitzschia spp.	+	+				+	1,50
Sum	0,5	1,5	0,0	0,0	0,0	0,0	1,5
Interstitial/epipsammic algae							
Fragilaria neoproducta	0,92		+		+		
Amphora libyca	2,30	4,43	+	+		0,50	1,50
A. ovalis	+	0,49	+				+
Amphora spp.				0,99			1,00
Navicula capitata	0,92	0,49	+				1,00
Sum	4,1	5,4	0,0	1,0	0,0	0,5	3,5

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Table 4. (*continued*)

	1992						1993
	February	April	June	August	October	December	February
Epipsammic algae							
Fragilaria brevistriata	0,92	3,94	0,50	4,93	2,90	+	0,50
F. leptostauron v. *dubia*	+						
F. leptostauron v. *martyi*	1,84	0,99	+	2,46	1,45	0,50	0,50
Achnanthes clevei	4,15	1,48	1,50	0,99	6,76	2,50	5,50
A. delicatula v. *delicatula*	4,15	3,45	5,50	4,43	1,45	3,50	2,00
A. lanceolata ssp. *dubia*	+						+
A. lanceolata ssp. *frequentissima* v. *frequentissima*	2,30	2,96	2,00	2,46	3,86	2,00	2,50
A. lanceolata ssp. *frequentissima* v. *rostrata*	3,23	1,48	6,00	2,96	4,83	1,50	1,50
A. cf. *lauenburgiana*	0,46				0,48	+	
Achnanthes spp.	17,97	5,42	14,50	9,36	18,84	22,00	12,50
Cocconeis neodiminuta[a]	+	+	+	+	+	+	+
C. neothumensis[a]	+	+	+	+	+	+	+
C. pediculus	0,46	+	+		+	+	+
C. placentula[a]	+	+	+	+	+	+	+
C. placentula v. *klinoraphis*[a]	+	+					
Cocconeis spp. *excl. pediculus*[a]	11,98	15,76	35,50	14,78	16,43	19,50	15,50
Amphora pediculus	33,64	28,08	27,00	51,72	37,20	17,00	23,00
Cymbella lanceolata	+						
C. sinuata	0,92	0,99	+	1,48	+	1,00	0,50
Gomphonema olivaceum v. *olivaceum*		0,99				+	+
G. olivaceum v. *minutissimum*	+						
G. angustatum		+					
Gomphonema spp.	0,46		+			1,00	
Rhoicosphenia abbreviata	+					+	0,50
Cymatopleura elliptica v. *elliptica*	+	+					+
C. solea		+					
Sum	82,5	65,5	92,5	95,6	94,2	70,5	64,5

[a]Abundance of *Cocconeis* species, being difficult to determine, was counted as *Cocconeis* spp.

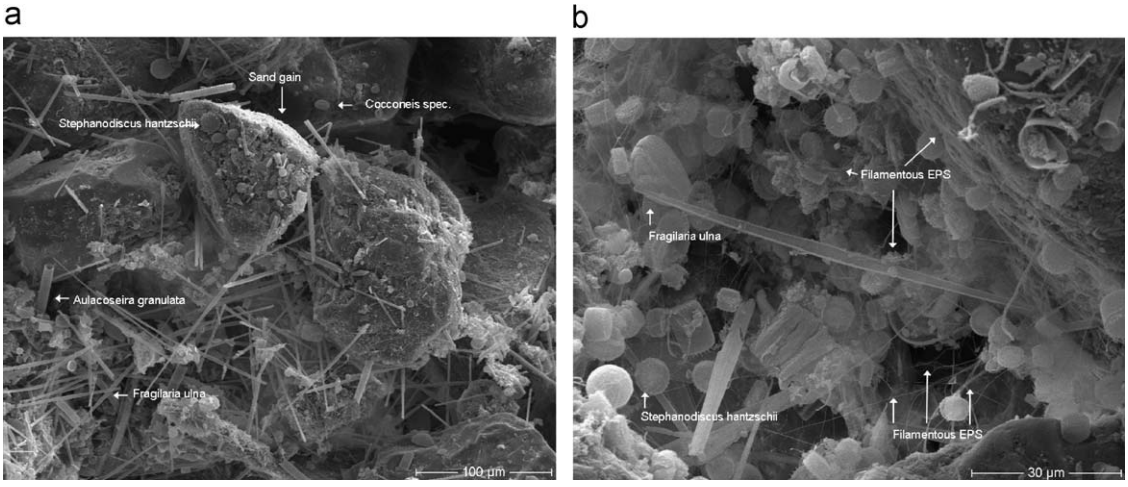

Fig. 6. Surface of the littoral sandy zone of Lake Tegel with an enrichment of diatoms. (a) sand gains with aufwuchs of epipsammic diatoms (*Cocconeis* spp.) and a surficial accumulation of planktonic algae (*Fragilaria ulna*, *Aulacoseira granulata*, *Stephanodiscus hantzschii*); 27.04.2004. (b) Filamentosus EPS with planktonic diatoms (*Fragilaria ulna*, *Aulacoseira granulata*, *Stephanodiscus hantzschii)*; 25.02.2004.

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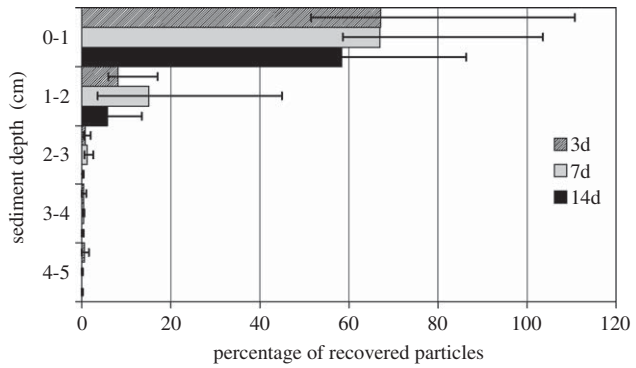

Fig. 7. Percentages of recovered AMC monodispersive resin particles (∅ 2.44 µm) in the upper 5 cm littoral sediment of Lake Tegel in April 2005; no particle occurred deeper than 5 cm. Data presented are means (= bars) and extremes (= lines; $n = 9$).

able to pass the pore system; the largest FPOM fraction studied (200–1000 µm) should not pass the pore system. The mean pore water velocity of 54 cm d^{-1} at the sampling site should transport particles smaller than the sediment pore size at least down to a corresponding sediment depth, if vertical transport is not hindered.

Vertical transport of AMC-labelled microfine particles was studied in littoral sediments over a period of up to 14 days, the experiments showed clearly highest particle retention in the uppermost 5 cm (Fig. 7). The recovery rates of particles varied between 85 ± 22% (after 7 days) and 65 ± 16% (after 14 days) without any significant tendency related to time of exposure. No particle occurred deeper than 5 cm; these data were confirmed by laboratory sediment core experiments with a similar retention rate.

Statistical analysis of the particle distribution could not detect significant differences ($p < 0.05$) for the upper 2 cm of sediment, but the vertical decrease in particle number from the upper to the adjacent layer was significant. Compared with pore water velocities of 54 cm d^{-1}, the observed particle transport with 1–2 cm day^{-1} was significantly slower (about 55 times), but it has to be pointed out that the greatest portion of the fine resin particles were completely hindered in vertical transport. Severe clogging, that means a colmation of the sediment pore system even occurs at the sandy littoral shore of Lake Tegel, formed by erosion processes and exposed to wave action.

Four hours after the beginning of the in situ exposure of fluorescence-labelled FPOM (alder leaf litter) 99% of the FPOM (1.1×10^4 particles cm^{-2}) were found within the topmost sediment layer of 0–0.5 cm (Table 5). After 2 and 14 days, this sediment layer still contained a high percentage of the labelled FPOM (73% and 86%, respectively); nevertheless, slight vertical particle transport was observed. Relevant particle shifting during the 14 day experimental period was observed only down to a depth of 2 cm. Below a depth of 2 cm, FPOM particles only occurred sporadically, representing < 2.5% of the total particle number. Fluorescently labelled FPOM was not registered below a sediment depth of 4 cm.

For the smallest size class, statistical analysis did not indicate significant time-dependent differences between particle abundance in the topmost layer and the total particle number retrieved from the in situ cores, but a significant portion of particles from bigger size classes (20–200 µm and 200–1000 µm) was found to penetrate into deeper layers after 2 and 14 days (Table 5).

Fragmentation of fine particulate organic material (FPOM) in the interstices

The number of FITC-FPOM particles (alder leaf litter, in total 1.1×10^4–1.2×10^4 n cm^{-2}) remained constant within the first 2 days of in situ exposure at Lake Tegel. After 14 days, total particle number had increased by about 110% to 2.5×10^4 particles cm^{-2}; this increase in particle number was mainly due to a statistically significant increase in small particles, that means fragmentation of FPOM to very fine POM ⩽20 µm (*U*-test: $p < 0.05$; Table 5), which increased from 38% in the stock suspension to 61% of the FPOM abundance after two weeks exposure. However no decrease in big and medium-sized particles was observed, because the increase in FPOM < 20 µm is only 1.5% of the total FPOM biomass. The FITC-labelled FPOM undergoes fragmentation in the sediment due to meiofaunal shredding during invertebrate feeding processes, the uptake of the FITC-labelled FPOM was proven microscopically. The process of fragmentation is still in the early stage after a period of two weeks, and only a few bigger FPOM particles shredded into smaller FPOM pieces were registered; it must be assumed that the FPOM is available for a long time, but it must be pointed out, that the FPOM offered was not yet colonized by bacteria.

Discussion

Fluorescein (FITC) tracers

The aim of this study is the development of a novel POM-labelling method and its demonstrative application to in situ experiments for POM transport and fragmentation by abiotic and biotic processes in the sandy littoral sediment of a lowland lake. The use of fluorescently labelled, natural and lake-specific organic matter provides a new potential for studying transport, fragmentation and ingestion of FPOM as well as bioturbation processes by the benthic fauna in detail. Reactive fluorescein (fluorescein-5-isothiocyanate,

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Table 5. Vertical distribution of FITC-labelled FPOM (alder leaf litter) as n $cm^{-3} \pm$ sd after addition of 5 mg FPOM (d. w.) cm^{-2} sediment surface to enclosures and exposure for 4 h (used as control to allow complete deposition), 2 days, and 14 days in the littoral zone of Lake Tegel.

	n $cm^{-3} \pm$ SD ⩽20 μm	n $cm^{-3} \pm$ SD 20–200 μm	n $cm^{-3} \pm$ SD 200–1000 μm	n cm^{-3} All size classes
Exposition time: 4 h (control)				
0.0–0.5 cm	6661 ± 1747	14,490 ± 4778	1300 ± 340	22,451
0.5–1.0 cm	45 ± 22	30 ± 42	0 ± 0	75
1–2 cm	10 ± 15	14 ± 19	0 ± 0	24
2–3 cm	7 ± 0	9 ± 13	0 ± 0	16
3–4 cm	0 ± 0	0 ± 0	0 ± 0	0
Exposition time: 2 d				
0.0–0.5 cm	8563 ± 4575	7939 + 343	793 ± 457	17,295
0.5–1.0 cm	2038 ± 561	1827 ± 197	115 ± 92	3,98
1–2 cm	296 ± 115	225 ± 100	44 ± 33	565
2–3 cm	32 ± 6	0 ± 0	0 ± 0	32
3–4 cm	32 ± 38	4 ± 10	0 ± 0	36
		Significant		
Exposition time: 14 d				
0.0–0.5 cm	30,838 ± 6293	14,361 ± 3912	1638 ± 434	46,837
0.5–1.0 cm	3188 ± 842	3227 ± 498	380 ± 123	6,795
1–2 cm	8 ± 12	64 ± 46	4 ± 8	76
2–3 cm	4 ± 10	22 ± 15	0 ± 0	26
3–4 cm	0 ± 0	0 ± 0	0 ± 0	0
		Significant	*Significant*	

Significant differences (one-tailed *T*-test: $p < 0.05$) between particle number in 0–0.5 cm and total particle number are given.

FITC) can be bound to proteins and other organic components such as dried algae and leaves by conjugation, thus enabling the production of specific fluorochrome compounds. Until now, FITC labelling was mainly restricted to fluorescent dyes used in tracer experiments, enzymatic studies, molecular approaches and biofilm staining (Kemp et al., 1993; Kalmbach et al., 1996; Halpern et al., 2002; Strathmann et al., 2002). Qualitative and quantitative epifluorescence microscope studies of bacteria, protists and tissues are carried out with a variety of fluorochromes binding to nucleic acids (e.g. DAPI, Acridine Orange), cytoplasm (fluorescein-5-isothiocyanate, FITC) or specific cell components. Reactive fluorescein can be bound to proteins and other organic components by conjugation, thus enabling the production of specific fluorochrome-labelled antibodies (Rogers and Keevil, 1992). The development of the fluorescence in situ hybridization (FISH) technique, which allows detection of specific taxonomic units with the help of fluorochrome-labelled oligonucleotide probes, extended the application to ecological studies (Amann et al., 1995). A method using FITC-labelled lectins to visualize glycoconjugates in algal and bacterial exopolymers was introduced by Lawrence et al. (1998) and Böckelmann et al. (2002).

The use of fluorochrome-labelled particles such as yeast, bacteria and pollen as prey for organisms was also developed in planktonic studies, and grazing rates of planktonic protists were assessed by offering fluorochrome-labelled microspheres and bacteria (Hall et al., 1996), yeast (Paul and Hall, 2002) or pollen (Wanner and Pusch, 2000); several authors have extended these methods to the sediment habitat (Starink et al., 1994; Hondeveld, 1998). Difficulties arise due to refusal of microfauna to ingest fluorescence labelled fine resin particles as well as bacteria and yeast (Hammer et al., 1999; Boenigk et al., 2001; Diederichs et al., 2003). The use of natural organic substrates such as algae and leaves originating from the sampling site and labelling of the dried material with FITC will complete the experimental possibilities for POM turnover studies and allow workers to carry out these experiments under in situ conditions. The tests for ingestion of fluorescence-labelled FPOM derived from alder leaves and algae by meiofauna in our study were positive for several taxonomic groups; no rejection of FITC-labelled POM by detritivorous meiofauna was observed.

Vertical transport of particulate organic matter (POM)

The interstices of the sandy littoral zone is partly filled with particulate organic material such as detritus, living

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bacteria and algae cells and EPS, and a high portion of organic matter is found down to 50 cm. Epipsammic algae occur with a high biomass in the upper interstitial zone of about 0–6 cm depth. The interstitial flora is a biocoenosis especially adapted to the sediment pore system, and planktonic algae species from the lake water are only transported into the interstices to a small extent (Beulker and Gunkel, 1996).

Different pathways for the entry of POM into the sediments have to be distinguished, namely passive settling, episodical burial and also physical advection because of infiltrating conditions (Rinck-Pfeiffer et al., 2000). Interfacial currents (infiltration by groundwater recharge) and water exchange due to topography-related pressure gradients (high flood conditions) can also be responsible for vertical transfer of algae (Huettel and Rusch, 2000).

The observed high restriction of small particles to the sediment surface stands in contradiction to the pore size of the sediment and the infiltration velocity of the pore water. This retention effect was proved by artificial monodispersive resin microparticles of only 2.44 μm diameters and FPOM of different size classes. The results indicate that the vertical transport of FPOM is restricted to the upper sediment layer of 1–2 cm; even microfine resin particles undergo much slower migration through the pore system than expected from the calculated pore water velocity (about 55 times slower), and a considerable proportion of particles (about 75%) is retained in the upper sediment layer of 2 cm. Bigger FPOM particles $\geqslant$20 μm are likewise transported to 2–3 cm only, probably induced by bioturbation of the colonizing meiofauna. Passive transport of particles by the infiltrating water does not occur to a noticeable extent in the sandy littoral sediment of Lake Tegel, and the clogging of the sediment surface is of high significance.

In the course of the experiments, the turnover of POM was mainly restricted to the surface layer of the littoral sediment and an accumulation of particles took place in the uppermost centimetre (Gunkel and Hoffmann, 2006; Hoffmann and Gunkel, 2006). This surface layer therefore functions as a mechanical filter for FPOM, which is transported to deeper sediment zones by biological perturbation and mechanical mixing alone. Vertical particle transport obviously is inhibited by intensive clogging processes such as input of seston, fragmentation of POM to FPOM, build-up of gas bubbles, carbonate precipitation, and excretion of EPS (polysaccharides, polypeptides or bacterial DNA; Vandevivere and Baveye, 1992). Recent studies of biofilm formation and structure in littoral sediments prove the significance of clogging processes and provide some insight into the structure of the interstitial (Palmgren and Nielsen, 1996; Rinck-Pfeiffer et al., 2000; Flemming and Wingender, 2001a, b; Langergraber et al., 2003; Gunkel and Hoffmann, 2006). Therefore, the transport processes induced by the water current through interstitial spaces seem to be of low effect for particle transport in the littoral sediments of Lake Tegel.

Fragmentation of particulate organic matter

The FPOM distribution in littoral sediments of Lake Tegel shows an effect of bioturbation and feeding activity, indicated by a higher proportion of smallest size classes with time and a slight particle shift down to 3 cm. The natural depth distribution of POM and especially the presence of middle-sized FPOM-fractions down to 20 cm sediment depth are also an indication of fragmentation by meiofauna and bioturbation. Probably these observations are the consequence of a significantly longer time span for POM than those used in our experiments; in addition, seasonal variations in meiofaunal distribution and activity have to be considered; burial of organic matter through storm events or other disturbances and the input of macrophyte rhizomes also play a role in POM distribution. Nevertheless, little is known about the role of the interstitial fauna in converting FPOM to very fine FPOM by ingestion and the transport of FPOM as faeces by migration of the interstitial fauna (Hakenkamp and Palmer, 2000; Hakenkamp et al., 2002).

The study of FPOM transport is part of the lake/low land river and groundwater interaction research program, concerning natural and artificial ground water recharge and bank filtration processes (Grützmacher et al., 2006; Grünheid et al., 2005; Greskowiak et al., 2005; Gunkel and Hoffmann, 2006; Hoffmann and Gunkel, 2006). Thus, more investigations are needed to follow the fate of the FPOM and the turn over of POM and DOC. The occurrence of cyanobacterial toxins and the possible transport of the cyanotoxins during bank filtration give new perspectives for further studies on FPOM and DOC turn over in the interstices (Sens and Dalsasso, 2007).

Conclusion

The novel method described here is a useful tool for studying the transport and cycling of organic matter in the sediment. The use of the fluorescent labelled, natural, lake-specific organic material provides a new potential for studying not only the transport, but also the fragmentation and the ingestion of FPOM as well as bioturbation processes by the benthic fauna in detail. Due to the in situ sediment core technique and the use of natural organic matter such as algae or alder leaves, artefacts in C turnover studies are nearly excluded.

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The results point out the high significance of the upper sediment layer for the retention of POM and particles in the size range of bacteria, too, due to severe clogging processes, as well as very high biomass of epipsammic algae in the interstices down to a few centimetres. Future application of FITC-labelled FPOM should enable researchers to follow the small scale processes and quantify the transport also in different sediment types, under different infiltration conditions and in regard to seasonal variations.

Acknowledgements

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References

Amann, R.I., Ludwig, W., Schleifer, K.H., 1995. Phylogenetic identification and in situ detection of individual microbial cells without cultivation. Microbiol. Rev. 59, 143–169.

Beulker, C., Gunkel, G., 1996. Studies on the occurrence and ecology of meiofauna and benthic algae in the littoral sediments of lakes. Limnologica 26, 311–326.

Beyer, W., 1964. Zur Bestimmung der Wasserdurchlässigkeit von Kiesen und Sanden aus der Kornverteilungskurve. Wasserwirtsch. Wassertechnik 6, 165–168.

Böckelmann, U., Manz, U., Neu, T.R., Szewzyk, U., 2002. A new combined technique of fluorescent in situ hybridisation and lectin binding analysis (FISH-LBA) for the investigation of lotic microbial aggregates. J. Microbiol. Meth. 49, 75–87.

Boenigk, J., Arndt, H., Cleven, E.J., 2001. The problematic nature of fluorescently labelled bacteria (FLB) in Spumella feeding experiments – an explanation by using video microscopy. Arch. Hydrobiol. 152, 329–338.

DEV, 1986. DIN 38412. Bestimmung des Chlorophyll a Gehaltes von Oberflächenwasser. Deutsche Einheitsverfahren zur Wasser, Abwasser- und Schlammuntersuchung. Loseblattsammlung. L 16. Wiley.

Diederichs, S., Beardsley, C., Cleven, E.J., 2003. Detection of ingested bacteria in benthic ciliates using fluorescence in situ hybridization. System. Appl. Microbiol. 26, 624–630.

Flemming, H.C., Wingender, J., 2001a. Relevance of microbial extracellular polymeric substances (EPSs) – Part I: Structural and ecological aspects. Water Sci. Technol. 43, 1–8.

Flemming, H.C., Wingender, J., 2001b. Relevance of microbial extracellular polymeric substances (EPSs) – Part II: Technical aspects. Water Sci. Technol. 43, 9–16.

Georgian, T., Newbold, J.D., Thomas, S.A., Monaghan, M.T., Minshall, G.W., Cushing, C.E., 2003. Comparison of corn pollen and natural fine particulate matter transport in streams: can pollen be used as seston surrogate? J. N. Am. Benthol. Soc. 22, 2–16.

Gerino, M., Aller, R.C., Lee, C., Cochran, J.K., Aller, J.Y., Green, M.A., Hirschberg, D., 1998. Comparison of different tracers and methods used to quantify bioturbation during a spring bloom: 234-Thorium, Luminophores and Chlorophyll a. Estuar. Coast. Shelf Sci. 46, 531–547.

Graham, N.J.D., Collins, R.M., 1996. Advances in Slow Sand Filters and Alternative Biology Filtration. Wiley, New York.

Gray, J.S., 1981. The Ecology of Marine Sediments. Cambridge University Press, New York.

Greskowiak, J., Prommer, H., Massmann, G., Johnston, C.D., Nützmann, G., Pekdeger, A., 2005. The impact of variably satured conditions on hydrogeochemical changes during artificial recharge of groundwater. Appl. Geochem. 20, 1409–1426.

Grünheid, St., Amy, G., Jekel, M., 2005. Removal of bulk dissolved organic carbon (DOC) and trace organic compounds by bank filtration and artificial recharge. Water Res. 39, 3219–3228.

Grützmacher, G., Wessel, G., Chorus, I., Bartel, H., 2006. Are there limits to cyanobacterial toxin (microcystin) elimination by sand passage? In: UNESCO (Ed.), Recharge systems for protecting and enhancing groundwater resources. In: Proceedings of the Fifth International Symposium on Management of Aquifer Recharge. UNESCO IHP-VI-GW-13, pp. 485–490.

Gunkel, G., Hoffmann, A., 2006. Clogging processes in a bank filtration system in the littoral zone of Lake Tegel (Germany). In: Proceedings of the Fifth International Symposium on Management of Aquifer Recharge. UNESCO IHP-VI-GW-13, pp. 599–604.

Hakenkamp, C.C., Palmer, M.A., 2000. The ecology of hyporheic meiofauna. In: Jones, J.J., Mulholland, P.J. (Eds.), Stream and Groundwaters. Academic Press, New York, pp. 307–336.

Hakenkamp, C.C., Morin, A., Strayer, D.L., 2002. The functional importance of freshwater meiofauna. In: Rundle, S.D., Robertson, A.L., Schmid-Araya, J.M. (Eds.), Freshwater Meiofauna: Biology and Ecology. Backhuys, Leiden, pp. 321–335.

Hall, R.O., Peredney, C.L., Meyer, J.J., 1996. The effect of invertebrate consumption on bacterial transport in a mountain stream. Limnol. Oceanogr. 41, 1180–1187.

Halpern, M., Gasith, A., Broza, M., 2002. Does the tube of benthic chironomid larva play a role in protecting its dweller against chemical toxicants? Hydrobiologia 470, 49–55.

Hammer, A., Grüttner, C., Schumann, R., 1999. The effect of electrostatic charge of food particles on capture efficiency by Oxyrrhis marina Dujardin (Dinoflagellate). Protist 150, 375–382.

Heinzmann, B., Chorus, I., 1994. Restoration concept for Lake Tegel, a major drinking and bathing water resource in a densely populated area. Environ. Sci. Technol. 28, 1410–1416.

Higgins, R.P., Thiel, H., 1988. Introduction to the Study of Meiofauna. Smithsonian Institution Press.

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Hoffmann, A., Gunkel, G., 2006. Physicochemical changes in pore water in the sandy littoral zone of Lake Tegel during bank filtration. In: UNESCO (Ed.), Recharge systems for protecting and enhancing groundwater resources. In: Proceedings of the Fifth International Symposium on Management of Aquifer Recharge. UNESCO IHP-VI-GW-13, pp. 605–610.

Hoffmann, A., Gunkel, G., 2009. Bank filtration in the sandy littoral zone of Lake Tegel (Berlin): Biological-chemical structure of the interstices and clogging processes. Limnologia, submitted.

Hondeveld, B., 1998. Heterotrophic nanoflagellates and their role as bacterivores in marine sediments. Ph.D. thesis. Faculty of Biology, University of Amsterdam, Febodruk B.V. Enschede.

Huettel, M., Rusch, A., 2000. Transport and degradation of phytoplankton in permeable sediment. Limnol. Oceanogr. 45, 534–549.

ISO 14688-1/2, 2002/2004. Geotechnical investigation and testing-Identification and classification of soil. Part 1: Identification and description. Part 2: Principles for a classification.

Jahn, D., Witt, H., 2002. Gewässeratlas von Berlin . Senatsverwaltung für Standentwicklung . UNICOM, Berlin. Download: 〈http://www.berlin.de/sen/umwelt/wasser/wasserrecht/pdf/wasseratlas.pdf〉.

Kalmbach, S., Manz, W., Szewzyk, U., 1996. Untersuchung der Entwicklung natürlicher Biofilme in Trinkwasser-Systemen mit rRNS-gerichteten Gensonden. Schriftenreihe Wasserforschung 1, 107.

Kemp, P.F., Cole, J.J., Sherr, B.F., 1993. Handbook of Methods in Aquatic Microbial Ecology. CRC Press, Boca Raton, FL.

Kühn, W., Müller, U., 2000. Riverbank filtration – an overview. J. AWWA 92, 60–69.

Langergraber, G., Haberli, R., Laber, J., Pressl, A., 2003. Evaluation of stubstrate clogging processes in vertical flow constructed wetlands. Water Sci. Technol. 48, 25–34.

Lawrence, J.R., Neu, T.R., Swerhone, G.D.W., 1998. Application of multiparameter imaging for the quantification of algal, bacterial and exopolymer components of microbial biofilms. J. Microbiol. Meth. 32, 253–261.

Palmgren, R., Nielsen, P.H., 1996. Accumulation of DNA in the exopolymeric matrix of activated sludge and bacterial cultures. Water Sci. Technol. 34, 233–240.

Paul, M.J., Hall Jr., R.O., 2002. Particle transport and transient storage along a stream-size gradient in the hubbard brook experimental forest. J. N. Am. Benthol. Soc. 21, 195–205.

Rinck-Pfeiffer, St., Ragusa, S., Sztajinbik, P., Vandevelde, T., 2000. Interrelationships between biological, chemical, and physical processes as an analog to clogging in aquifer storage and recovery (ASR) wells. Water Res. 34, 2110–2118.

Rogers, J., Keevil, C.W., 1992. Immunogold and fluorescein immunolabelling of Legionella pneumophila within an aquatic biofilm visualized by using episcopic differential interference contrast microscopy. Appl. Environ. Microbiol. 58, 2326–2330.

Schauser, I., Chorus, I., Heinzmann, B., 2006a. Strategy and current status of combating eutrophication in two Berlin lakes for safeguarding drinking water resources. Water Sci. Technol. 54, 93–100.

Schauser, I., Chorus, I., Lewandowski, J., 2006b. Effects of nitrate and phosphorus release: comparison of two Berlin lakes. Acta hydrochim. hydrobiol. 34, 325–332.

Senatsverwaltung, 2006. Bericht über die meteorologische, hydrologische und gewässerökologische Situation im Sommer 2006 in Berlin. Senatsverwaltung für Gesundheit, Umwelt und Verbraucherschutz. Download: 〈http://www.berlin.de/sen/umwelt/wasser/monat/de/pdf/sommer2006.pdf〉.

Sens, M.L., Dalsasso, R.L., 2007. Bank filtration of reservoir as an efficient technique for water supply treatment. In: Gunkel, G., Sobral, M. (Eds.), Reservoirs and River Basins Management: Exchange of Experience from Brazil, Portugal and Germany. Universitätsverlag der Technischen Universität Berlin, pp. 253–264.

Solan, M., Germano, I.D., Rhoads, D.C., Smith, C., Michaud, E., Parry, D., Wenzhöfer, F., Kennedy, B., Henriques, C., Battle, E., Carey, D., Iocco, L., Valente, R., Watson, J., Rosenberg, R., 2003. Towards a greater understanding of pattern, scale and process in marine benthic systems: a picture is worth a thousand worms. J. Exp. Mar. Biol. Ecol. 285/286, 313–338.

Starink, M., Krylova, I.N., Bär-Gilissen, M.-J., Bak, R.P.M., Cappenberg, T.E., 1994. Rates of benthic protozoan grazing on free and attached sediment bacteria measured with fluorescently stained sediment. Appl. Environ. Microbiol. 60, 2259–2264.

Strathmann, M., Wingender, J., Flemming, H.C., 2002. Application of fluorescently labelled lectins for the visualization and biochemical characterization of polysaccharides in biofilms of Pseudomonas aeruginosa. J. Microbiol. Meth. 50, 237–248.

Vandevivere, P., Baveye, P., 1992. Effect of bacterial extracellular polymers on the saturated hydraulic conductivity of sand columns. Appl. Environ. Microbiol. 58, 1690–1698.

Wanner, S.C., Pusch, M., 2000. Use of fluorescently labeled Lycopodium spores as a tracer for suspended particles in a lowland river. J. N. Am. Benthol. Soc. 19, 648–658.

Wotton, R.S., 1994. The Biology of Particles in Aquatic Systems, second ed. CRC Press, Boca Raton, FL.

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