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# An "Integrated Sediment Disturber" (ISD) to study the impact of repeated physical perturbations on sediment geochemistry and the small benthic biota

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### **Abstract**

An Integrated Sediment Disturber (ISD) was developed to study the effects of repeated physical disturbances on sediment geochemistry and associated benthic fauna. The ISD is capable to generate a strong, more or less uniform mixing of surface sediments by three rotating fork-like disturber units of approximately 1 m in diameter. Perturbations can be set at different intensities and frequencies, e.g., on times scales of hours, days, or weeks, depending on the experimental design. The ISD carries an Autonomous Positioning Drive (APD) module, which allows the three-dimensional positioning of sensors by a programmable controller. The three circular disturbed zones and an undisturbed control area in-between can be monitored at specified frequencies using a microprofiler with microelectrodes for measuring various parameters (e.g., oxygen, pH, hydrogen sulphide). All operations are supervised by a digital stills camera. Sediment sampling at the start and end point using coring devices handled by divers (and potentially undertaken using a Remotely Operated Vehicle) provides material to study shifts in biodiversity of the small sediment-inhabiting biota and alterations in ecosystem functioning in response to perturbations and the changing geochemical characteristics of surface sediments.

#### Acknowledgments

This work was part of the Specific Targeted Research Project (STREP) COBO (Coastal Ocean Benthic Observatory) sponsored by the European Union (contract no. 505564), and supported by the Department for Environment, Food and Rural Affairs (contracts A1224, E1409 & E3205). We gratefully acknowledge Malcolm Fulcher, Sergio Vettese, Steve Clarke, and Matthew Brown (Centre for Environment, Fisheries & Aquaculture Science, United Kingdom) for invaluable support in designing and constructing the APD and smart control systems. Lars Damgaard (Unisense A/S, Denmark) is acknowledged for providing new oxygen microsensors, integrating sensing electrodes and micro-electronics in the same housing. We express our thanks to Bruno Bombled (Laboratoire des Sciences du Climat et de L'Environnement, France) for designing a simplified prototype of the ISD and for technical assistance during the deployments of the gear in the Gulf of Fos as well as Philippe Bonte, Irene Lefevre, and Jean-Louis Reyss (Laboratoire des Sciences du Climat et de L'Environnement, France) for γ measurements. We also thank Anja Pappert (Alfred Wegener Institute for Polar and Marine Research, Germany) for biochemical sediment analyses and Melanie Hartwich (Potsdam University, Germany) for assistance with bacterial counts. The constructive comments and criticisms of the Associate Editor David Thistle and the anonymous reviewers are gratefully acknowledged. This is publication "awi-n17148" of the Alfred Wegener Institute for Polar and Marine Research, Bremerhaven, Germany.

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#### Introduction

Disturbances created by bioturbators are an important driving force in sediment community dynamics (Hall 1994). Pickett and White (1985) described a disturbance as a "discrete event in time that interrupts ecosystem, community, or population structure and changes resources, substratum availability, or the physical environment," a definition covering processes acting directly on organisms (e.g., competition, predation) as well as those acting indirectly via changing biogeochemical properties (e.g., changes in sediment granulometry, porewater gradients, food and oxygen availability).

In fact, sediment-dwelling organisms modify sediment characteristics in many ways. They significantly affect sediment stability and frequently alter their nutritional and physiological environment on various spatial scales. Surface deposit feeders remove surficial sediments and might expose deeper sediment layers with often transient chemical signatures (Marinelli 1992). Burrowing organisms alter nutrient and metabolite concentrations by irrigating their tubes and tunnels (Aller 2001). In shallow-water benthic ecosystems in particular, bioturbation can aerate/oxygenate deeper sediment layers, decrease rates of denitrification, and increase the precipitation of phosphate (Widdicombe and Austen 1998). Bioturbation may also lead to the recovery of previously buried organic matter, which could subsequently lead to an increase in the density and biomass of benthic organisms. On the other hand, the release of buried organic matter and nutrients might also result in enhanced bacterial turnover rates, thereby causing hypoxic or even anoxic conditions. This, in turn, might cause mortality in aerobic benthic assemblages. Overall, we can conclude that the relationship between tightly coupled biological and geochemical processes in the marine environment is very complex and still only poorly understood, especially in terms of temporal and spatial variability. Further research and innovative technologies are urgently needed to shed light on the complex interactions between benthic organisms and their sedimentary environment.

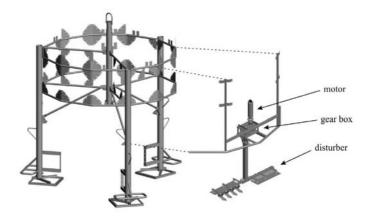
The overall objective of the EU European Strategic Targeted Research Project entitled "Toward a Coastal Ocean Benthic Observatory" (COBO; contract no. 505564) was to integrate emerging and innovative technologies from different disciplines (physics, chemistry, biology, imagery) to provide in-situ monitoring of benthic habitats and to understand complex interactions between the biota (function and diversity) and their chemical environment. One work package of the project was devoted to the development of new technologies in benthic research with which to study the time variable effects of sediment perturbations on the sediment geochemistry and the associated benthic fauna.

The Integrated Sediment Disturber (ISD) developed within COBO is capable of perturbating surface sediments by means of three rotating fork-like disturber units to a fixed depth of a few centimeters. The disturbing elements of the ISD in their present configuration are clearly not able to mimic sediment per-

turbations caused by bioturbation of smaller benthic organisms, which occur in a patchy manner and on spatial scales from millimeters to centimeters. Instead, they create more or less uniform disturbances of approximately 1 m in diameter, and thus mimic perturbations such as the feeding pits of rays. On the other hand, ISD-induced sediment mixing is clearly less intrusive than the radical physical perturbations induced by bottom fishery (causing mass mortality of benthic organisms) or storm events (for shallow water regions), especially because due to the low rotation speed of the disturber units resuspension of fine sediments during ISD disturbances is almost zero. Anyway, ISD disturbing elements cause a strong mixing of surface sediments, and thus open the possibility to study the spatial and temporal development of geochemical and biological characteristics after single and/or repeated sediment disturbances. These investigations will help to understand the time variability within, and the relationship between biological and diagenetic processes in marine sediments.

# Material and procedures

The ISD frame and the disturber units were build by the company K.U.M. Umwelt- und Meerestechnik GmbH in Kiel, Germany. The frame is based on a free-falling device, the GEO-MAR Modular Lander (GML). The tripod stainless steel structure is approximately 2.5 m in height and 2.2 m in diameter (Fig. 1). The frame has a total weight of ca. 150 kg in air (excluding ground weights of 3 × 90 kg). Distances between legs are approximately 1.80 m. The legs provide attachment points for the Autonomous Positioning Drive (APD) (see below). A five mega-pixel high-resolution color digital camera, covering major parts of all three disturbed areas and the control area in-between the perturbed zones, is fixed near the central, vertical axis of the frame at approximately 1.25 m above ground. The ISD frame can be lowered to the seafloor on a wire (shallow-water version). However, there are also attachment points for floats allowing the conversion of the ISD frame into a free-falling device (deep-water version).



**Fig. 1.** GEOMAR Modular Lander frame (left) and a single disturber unit (right).

The ISD frame carries three rotating fork-like disturber units, which allow the surface sediments to be reworked at different frequencies, e.g., on times scales of hours, days, and weeks, depending on the experimental design and subject to limitations of power and data storage requirements. Individual disturber units consist of a motor, a gear box, and disturber arms equipped with fork-like rakes (Fig. 1). The total weight of a single disturber unit (including support frame) is approximately 40 kg in air.

The disturber arms are moved by a 24 V DC motor in a stainless steel pressure housing. The motor has a total length of 35 cm, a diameter of 7 cm, and a weight of 5.4 kg in air. The gear box, inserted between the motor and the disturber arms, determines a constant rotation speed of two rounds per hour. The initiation of the first rotation mechanically lowers the disturber elements from an elevated security position to the sediment surface. Hinges at the central axis (Fig. 2, left) allow a mechanical self-adjustment of the disturber arms to uneven sediment surfaces at max. ± 10°. Disturber arms are 50 cm in length generating a circular reworked area of about 0.8 m<sup>2</sup>. Three interchangeable disturbing elements (i.e., ploughs, harrows, rakes; see Fig. 2, right) are available for use in different sediment types. Maximum penetration depth of the disturbing elements is approximately 10 cm. Spacing between 1 cm thick harrow and rake spikes is 1.25 and 2.5 cm, respectively; spacing between individual ploughs about 6 cm in width is minimum 3 cm. Harrow/rake spikes and single ploughs on opposite disturber arms are orientated along the arms in such a way that, during one turn of the disturber units, they fill the gaps between them; this causes a largely uniform mixing of the sediments. Disturbing elements are mounted in front of plates, which immediately flatten the sediment surface after perturbations, to create even planes for subsequent microprofiling.

A single disturber may perturbate surface sediments at a certain frequency, however, due to the general concept of creating sediment disturbances (i.e., by rotating disturber units), the rate at which a given centimeter of sediment is disrupted increases radially outwards, because the outer elements move faster than the inner elements.

The APD was designed to allow repeated sampling of dis-





**Fig. 2.** Disturber arm with a steel plate to flatten the sediments after perturbations (left), and interchangeable disturbing elements (right), i.e., plough (top), harrow (center), rake (bottom).

turbed and undisturbed regions using in-situ sensors. A number of possible designs were considered before opting for the so-called *r-Theta* alternative. This version consists of a circular stainless steel bar, 1.4 m in diameter, attached to the ISD frame at three points, supporting the ends of a horizontal carriage on which a Unisense® microprofiler is mounted. The carriage can be rotated about a vertical axis by a stepping motor mounted on the ISD frame, and the microprofiler can be moved horizontally by means of another motor and a lead screw (Fig. 3). As far as possible, components were constructed using 316 stainless steel or plastic.

The *r-Theta* design allows unimpeded access to most of the area within the APD ring (apart from the center) and equalarea access to the three disturbed regions. It optimizes the ability to carry out transects through the disturbed regions and give access to regions of undisturbed sediment (Fig. 4). The

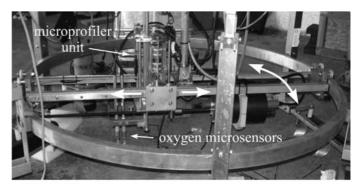
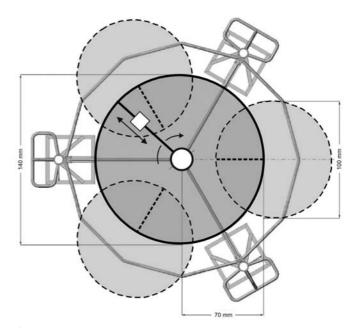


Fig. 3. APD frame carrying a microprofiler along a horizontal carriage.



**Fig. 4.** Top view on the ISD geometry; light gray: areas covered by the disturber units; dark gray: area covered by the APD; white box: microprofiler moving sensors along the APD radius; thick dotted line: optimal sampling line for sensors moved by the APD.

microprofiler has been modified to carry three oxygen microelectrodes in its present configuration; the adaptation of other microsensors (e.g., for pH, hydrogen sulphide) is optional. The time necessary for recording oxygen profiles depends mainly on the desired resolution of the profiles, i.e., the higher the resolution, the longer the time needed. Usually, the time required to get a full set of high-resolution profiles (50  $\mu m$  steps) is around 2 h.

Clark microelectrodes were chosen for their ability to make rapid measurements of oxygen profiles after disturbances. These electrodes are made of glass and are therefore quite fragile when encountering shell debris, but are resistant to breakage in soft sediments eventually containing silt or fine sand. The stability of the microsensors was tested in the laboratory (data not shown) over a period of 2 mo with repeated profile measurements. The reproducibility of the profiles was very good over the entire period with some variation (± 10%) of the signal in free water. The experiment confirmed that oxygen microelectrodes are quite stable over several weeks and therefore are suitable for long-term deployments in the ISD.

During field experiments (see below), the ISD was always equipped with an additional optical oxygen sensor (optode) measuring the dissolved oxygen concentration in the overlaying water. The optode signal was used for the calibration of the oxygen microelectrodes and to compensate for any drift in the microsensors signals. Due to their stability with time, optical microsensors and planar optodes might also be an option for sediment profiling, however, their accuracy is clearly lower than that of microelectrodes (Glud et al. 2005). In their present configuration, planar optodes should be discounted because their measurement mode is intrusive and may modify characteristics of the sedimentary environment.

Control of the various systems and data recording are achieved using a data logger of established design (ESM2, Cefas) and a purpose-built Control Unit (CU). The CU is the interface between the data logger (co-coordinating the high level control of the sampling plan) and the physical mechanical hardware used carry out the measurements and/or actions. This includes control of the APD, the disturber units, the microprofiler, the sampling protocol for the microelectrodes, the z-drive, and the camera system (Fig. 5). The data logger records a log of operations, stores the signals from the microelectrodes, and supports a number of additional environmental sensors.

The CU is based around a Texas Instruments MSP 430 family of FLASH programmable microcontrollers, combined with a series of hardware interfaces: an interface adapter that enables three probe signals to be individually isolated, amplified, filtered, multiplexed and digitized; a digital multiplexer to interface three incremental encoders and eight limit switches (this enables the movements of the probe height, the radius of the probe head from the center of the rig, the azimuth angle, and the movement of the disturbers to be monitored); three DC motor controllers for the oxygen probe

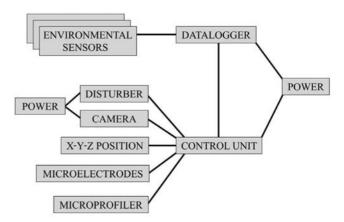


Fig. 5. High-level architecture of the ISD.

height adjustments and two for the radius and azimuth brakes; three solid-state power switches to control the drive to the DC disturber motors; two stepper motor controllers for the radius and azimuth movements (stepper motors were used for their speed, accuracy and repeatability with a relatively short development time - no closed loop feedback system was required); four independent solid-state power switches with shutter release to control the cameras, flash units and shutter release.

Power requirements for the three main elements of the ISD (not including the camera, which has a separate power supply), i.e., the smart data logger, the microprofiler (including movement on three axes), and the disturber units are provided in Table 1. Power is supplied by a battery pack consisting of four Yuasa gel lead-acid batteries, proving 12 volts, and 17 Ah each. The pack is configured as two batteries connected in parallel, in series with the other parallel pair to give a total pack capacity of 24 volts at 34 Ah. The pack is fused and diode protected (to protect against reverse polarity connection).

Firmware has been written to enable the system to issue commands for positioning the sensor head and performing the various measurement functions, according to a configurable timing schedule depending on the duration and purpose of the observational program. To optimize the rate of sampling, preserve the microelectrodes, and conserve battery life, a procedure has been developed that allows the detection of the sediment-water interface (SWI) by the probe. Knowledge of the SWI position permits the probe head to be held clear of the seabed and then lowered relatively rapidly until the sediment-water interface (SWI) is detected.

The algorithm, which has been developed to detect the SWI, is based on continuous monitoring of the oxygen signal as the sensor is lowered through the water column. Each value is compared with a running mean of previous samples (typically eight), and the difference is compared with a threshold criterion that defines the sediment-water interface. The following parameters are configurable: number of samples to take for free water value (same as profile samples per step),

**Table 1**. Approximate power requirements for three exemplary deployment options of the ISD.

Deployment o	pption	Logger (Ah)	Profiler (Ah)	Disturber (Ah)		
Option 1						
Duration (days)	5					
Logger cycle (min)	5					
No. cycles	1.440	4.35	6.13	0.30		
No. profiles	90					
No. disturbances	4					
Option 2						
Duration (days)	30					
Logger cycle (min)	5					
No. cycles	8.640	26.09	36.93	0.60		
No. profiles	545					
No. disturbances	8					
Option 3						
Duration (days)	54					
Logger cycle (min)	5					
No. cycles	15.667	40.34	20.12	2.10		
No. profiles	289					
No. disturbances	28					

SWI detection % of free water level, SWI termination % of free water level, step size, step speed, number of samples in the trend 'window,' value for trend to be seen as negative, retract distance once the SWI is found, and, overall travel limit. All sensors are monitored, and it is the first sensor that detects the SWI that controls the subsequent steps.

Once the SWI is detected, the sensor head is retracted and profiling commences. The sensors are moved down by a step distance, at a set speed, allowing for a settling time, and then taking *y* samples at *z* second intervals. The following profiling parameters are configurable: step size, step speed, probe settling time, number of samples at each step *y*, interval between samples *z*, number of additional profile steps taken after termination level is seen, and overall travel limit. All sensors are

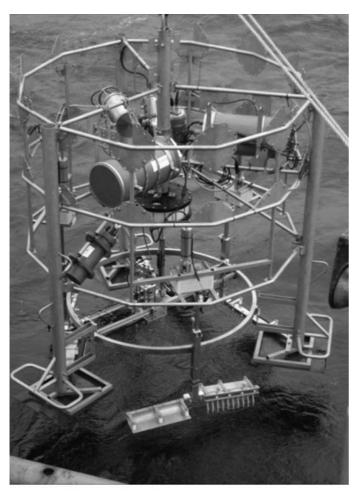
monitored, and at each step, the returned oxygen value is tested against known failure values, both high and low, and the result is logged. The end of the profiling procedure is perceived when the first probe signal reaches a low and constant signal, indicating that it is below the oxic zone. The SWI algorithm reduces the time required to get a full set of high-resolution profiles to approximately 0.5 h.

# **Assessment**

Single components of the ISD were thoroughly tested in the lab, in test tanks, as well as in-situ at approximately 10 m water depth in the Gulf of Fos (Northwestern Mediterranean; Table 2). In May 2006, the complete system (Fig. 6) was deployed in Loch Creran, a shallow-water fjord (~ 25 m water

**Table 2.** Station data as well as abiotic and biotic parameters investigated during ISD experiments in the Gulf of Fos and Loch Creran.

Location	Station	Date	Latitude, °N	Longitude, °W	Depth, [m]	Duration, [h]	Number/frequency [in hours] of disturbances	Luminophores	Natural radionuclides	Oxygen profiles	Chloroplastic pigments	Bacterial activity	Bacterial numbers/biomass	Meiofauna
Gulf of Fos	GF1	10 Mar 2006	43°23.2′	4°54.1′	9.2	~5	1/0	х	х	X	-	-	-	-
Loch Creran	MF1	3 May 2006	56°32.181′	5°19.180′	16.2	~24	4/6	-	-	-	х	х	_	_
Loch Creran	\$5	5 May 2006	56°31.425′	5°21.421′	25.1	~72	11/6	х	-	-	х	X	X	Х
Loch Creran	S11	10 May 2006	56°31.454′	5°21.297′	24.7	~48	11/4	x	_	x	Х	x	x	_



**Fig. 6.** Fully equipped Integrated Sediment Disturber (ISD) prior to deployment in Loch Creran (Scotland) in May 2006.

depth) on the northwestern Scottish coast, to carry out three short-term experiments of 24 to 72 h (Table 2). During these experiments, disturber units were equipped with plough-like disturbing elements and programmed to rework surface sediments every 4 and 6 h, respectively, resulting in a total of eleven disturbances during each experiment (Fig. 7).

The mixing efficiency of the disturbers was evaluated using artificial and natural tracers, i.e., so-called luminophores and different natural radionuclides. Luminophores are fluorescent inert particles of 63-125 µm in diameter. They were spread by divers in the areas covered by the disturber units prior to disturbance. Sediment samples taken from ISD-disturbed areas with diver-operated push-corers (10 cm in diameter, 40 cm in length), at the end of the experiments were then analyzed for vertical gradients of the luminophores by direct counting under UV light using a digital camera (Olympus C-2500L), and image analysis software (Image Pro® Plus). Figure 8 illustrates that due to disturber actions, luminophores were buried to a depth of 4-5 cm in the sediment column. The mean biodiffusive-like coefficient (Db), calculated from luminophore gradients obtained from three replicate sediment cores com-

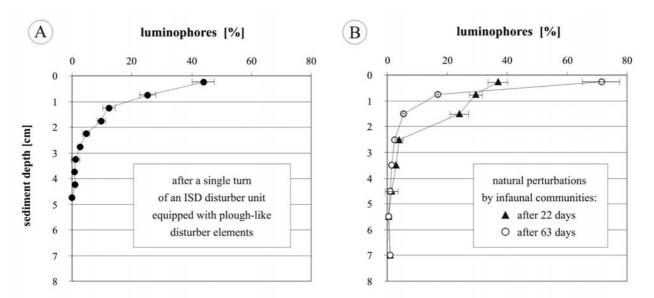


**Fig. 7.** Sediment perturbations after half a turn of the disturber units carrying plough-like disturbing elements (white arrow: direction of circular movement).

ing from the same disturbed area after a single ISD perturbation in the Gulf of Fos, was  $10.6 (\pm 0.3 \text{ SD}) \text{ cm}^2 \text{ y}^{-1}$ . This value is clearly higher than those found for in-situ and ex-situ experiments with natural infaunal communities from the Gulf of Fos after a few weeks (Gilbert et al. 1998: Db =  $3.0 (\pm 0.7 \text{ SD}) \text{ cm}^2 \text{ y}^{-1}$  after 30 d [n = 3]; Grossi et al. 2003: Db<sub>mean</sub> =  $2.2 (\pm 0.4 \text{ SD}) \text{ cm}^2 \text{ y}^{-1}$ , after 22 to 63 days [n = 6]). Gradients of luminophores after ISD perturbations are comparable to those found for natural communities after 1-2 months (Grossi et al. 2003; Fig. 8), as they both show biodiffusive-like distribution patterns with similar relative quantities of buried tracers.

During in-situ tests in the Gulf of Fos, sediment samples from ISD disturbed areas and nearby control sediments were also analyzed for the natural radionuclides beryllium-7 and potassium-40. The cosmogenic radionuclide beryllium-7 is produced by the interaction of cosmic rays with atmospheric gases and has a half-life of about 53 d. Beryllium-7 is delivered to the Earth's surface by dry or wet deposition and can be trapped by particles in the water column, finally reaching bottom sediments. It can be used to determine background steady-state mixing rates, or in certain instances, recent sediment accumulation rates (Cochran et al. 2006). Compared with background samples from the same area, beryllium-7 activities in the uppermost layers of sediments reworked by the disturber units were clearly lower (Fig. 9). This result coincides with data provided by Warnken et al. (2003) and Dellapenna et al. (2006), who measured a lower beryllium-7 inventory after physical sediment perturbations due to bottom fisheries.

Potassium-40 is a tracer of light minerals such as feldspar or muscovite grains. The potassium-40 concentration in marine soft sediments is generally inversely proportional to the grain size (Assinder et al. 1993; Venema and de Meijer 2001). Compared with control sediments, potassium-40 activities in the upper sediment layers of ISD reworked areas were slightly higher (Fig. 9), indicating an uplift of fine materials from



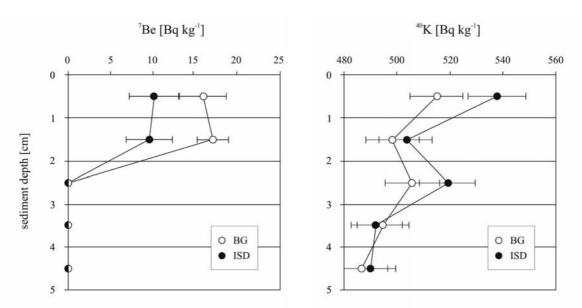
**Fig. 8.** Vertical profiles of luminophores from sediment cores after sediment perturbations created by the ISD during short-term experiments in the Gulf of Fos (A), and gradients produced by natural perturbations (bioturbation) in mesocosm experiments performed by Grossi et al. 2003 (B).

deeper sediment layers. Increased potassium-40 activities associated with decreased beryllium-7 activities in disturbed surface sediments were clearly due to the sediment reworking by the disturber units, demonstrating the ability of the instrument to create the desired sediment perturbations.

The mixing of the upper sediment layers affects geochemical processes in surface sediments, as confirmed by oxygen penetration depths assessed with the ISD-operated microprofiler. Fig. 10 shows oxygen profiles before and approximately 30 min after disturbances (one full turn of the disturber units) from an ISD experiment in the Gulf of Fos (Northwestern

Mediterranean). Oxygen penetration depths of 4 to 6 mm in this eutrophic shallow-water sedimentary environment diminished to 1 to 2 mm following perturbation, most probably due to the uplift of deeper, sub- or anoxic sediments by the disturber units.

Assuming steady state conditions, the diffusive  $O_2$  uptake (DOU) was calculated from the microelectrode profiles using Fick's first law of diffusion:  $DOU = F^{-1}D_0O_2\left[dO_2/dz\right]_{z=0}$ , where is the inverse of the formation factor at the sedimentwater interface (assuming a constant of 0.80 for all profiles),  $D_0O_2$  is the molecular diffusion coefficient of oxygen at the



**Fig. 9.** Beryllium-7 and potassium-40 activities [Bq kg<sup>-1</sup>] in background samples (BG) and ISD reworked sediments (ISD) from the Gulf of Fos (Mediterranean); sediments were perturbated by plough-like disturbing elements.

in-situ temperature, salinity, and hydrostatic pressure, and  $\left[dO_2/dz\right]_{z=0}$  is the oxygen gradient at the sediment-water interface (Rasmussen and Jørgensen 1992).

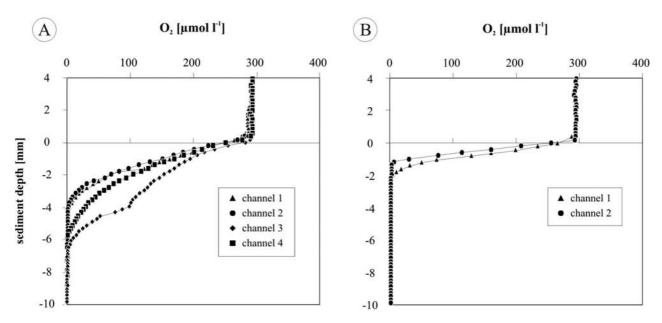
Before the ISD disturbance, DOU was  $9.5 \pm 1.3$  mmol m<sup>-2</sup> d<sup>-1</sup> (n = 5), which is close to the DOU measured in the same site of the Gulf of Fos in October 2002 ( $10.9 \pm 3.3$  mmol m<sup>-2</sup> d<sup>-1</sup>) (Rabouille et al. 2003). After the ISD-induced sediment perturbation, the steady state assumption could not be reasonably put into practice, because transport and reaction processes were clearly not in equilibrium. However, the diffusive oxygen uptake rate calculated from oxygen profiles after sediment perturbation showed an increase by a factor of 2, reaching  $21.9 \pm 2.1$  mmol m<sup>-2</sup> d<sup>-1</sup> (n = 2) 30 min after the ISD-induced sediment mixing.

The uplift of sub- or anoxic sediments will also affect the Redox conditions in the sediments. According to Warnken et al. (2003), dissolved metals in the sediment can be mobilized by the shoaling of the oxygenated layer and fluxes of manganese and cadmium to the water column can significantly increase. Enhanced fluxes of oxygen toward the sediments may deplete oxygen in the sediment overlying waters.

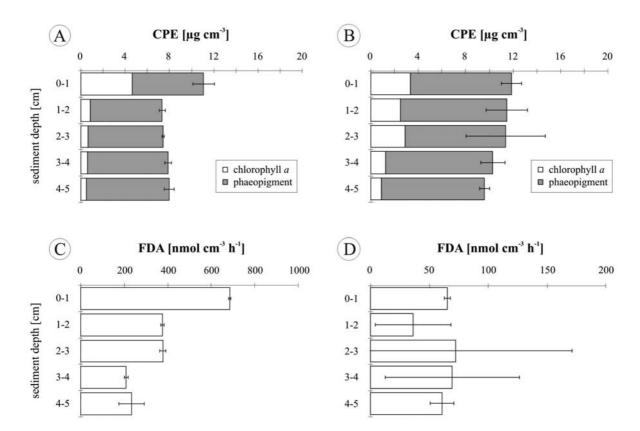
The mixing efficiency of the disturbers and effects of the sediment perturbations performed by the ISD could also be traced by looking at different biogenic sediment compounds: Sediment-bound pigments indicate the input of phytodetrital organic matter to the seafloor, the main food source for benthic organisms. In shallow water systems, when there is sufficient light at the seafloor, concentrations of pigments in sediments are also determined by benthic primary production. Gradients of pigments in the upper sediment layers usually

exhibit their highest values in the uppermost centimeter, with continuously decreasing values at increasing sediment depth. During the experiments in Loch Creran in May 2006, chloroplastic pigments in control sediments taken with a multiple corer (Bowers and Connelly) only a few meters away from the ISD were highest in the uppermost sediment layer (0-1 cm), whereas deeper layers showed lower values at almost constant levels (Fig. 11 A). Chlorophyll a concentrations in the first centimeter were 5 to 6 times higher than in deeper sediment layers. In contrast, sediment samples from ISD disturbed areas (Fig. 11 B) exhibited a smooth gradient in pigment concentrations, with intact Chl a at comparable levels buried down to 3 cm. Generally, higher total pigment concentrations in the disturbed areas might indicate a significant patchiness in the area of investigations. Compared with background samples, ISD disturbed sediments also showed a much higher variability between replicate samples, as indicated by the higher standard deviations, especially at intermediate sediment depths.

Sediment perturbations created by the ISD in the Loch Creran experiments also affected vertical gradients in bacterial activities, estimated as the hydrolytic potential of bacterial exoenzymes in the surface sediments. Gradients with continuously decreasing exo-enzymatic activity toward deeper sediment layers (Fig. 11 C) disappeared and were replaced by comparably heterogeneous patterns of bacterial activity in ISD-perturbed sediments, as confirmed by a much higher variability between replicates from disturbed sediments (Fig. 11 D). Bacterial number days and biomasses generally followed the same trend (data not shown). Much lower bacterial activities in perturbed sediment samples might indicate a comparably high



**Fig. 10.** Oxygen profiles measured at the sediment-water interface before (A) and 15 min after one perturbation by an ISD disturber unit (B) during the field tests in the Gulf of Fos (sediments were perturbated by plough-like disturbing elements; profiling resolution scale was 100 μm, and each data point is an average of five readings; microelectrodes were reinserted at exactly the same location).

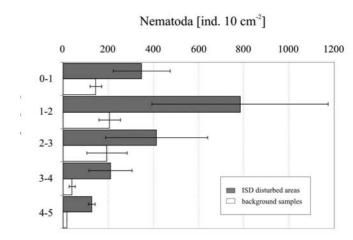


**Fig. 11.** Chloroplastic pigment equivalents (CPE: Chl *α* plus its degradation products, determined with a TURNER fluorometer; Shuman and Lorenzen 1975) and bacterial exo-enzymatic activity (estimated from turnover rates of the fluorogenic substrate fluorescein-di-acetate, FDA; Köster et al. 1991) in control samples (A, C) and in ISD-disturbed areas (B, D) at station S5 in Loch Creran.

patchiness in the study area, however, further investigations are needed to confirm whether or not sediment perturbations also have a direct effect on overall bacterial activity levels.

The redistribution of bacteria in the uppermost sediment layers has various implications for the (re)colonization by higher benthic organisms, e.g., the meio- and macrofauna. On one hand, bacteria are known to be a major food source for nematodes (De Mesel et al. 2003; Moens et al. 2005), representing the dominant taxa of the metazoan meiofauna. On the other hand, microbial processes in the sediments will significantly influence the biochemical milieu for the entire benthic biota. Preliminary results from meiofauna investigations 6.5 weeks after ISD perturbations in Loch Creran confirmed clearly higher nematode abundances in ISD-disturbed areas compared with control sediments sampled at the same time (Fig. 12). However, due to a fairly high variability in nematode numbers in the different sediment horizons, significant differences (P = 0.002; n = 3) were only found for the deepest layer investigated (4-5 cm).

Numerical responses by nematodes have been found in a number of disturbance studies (Warwick et al. 1997; Aarnio et al. 1998; Schratzberger and Warwick 1999), while other studies demonstrated only shifts in community structure (Warwick et al. 1990; Austen and Widdicombe 1998; Austen et al. 1998). Marked reductions in nematode densities due to ongoing biological disturbances were often attributed to predation (Schratzberger and Warwick 1999). Initial biological disturbance, however, might also result in significantly higher



**Fig. 12.** Nematode abundances in ISD disturbed sediments and control samples 6.5 weeks after repeated ISD perturbations in Loch Creran.

nematode abundances, as demonstrated by Schratzberger and Warwick (1999) in mesocosm experiments. Increased nematode densities in ISD-perturbed sediments most likely display a response to the uplift of organic matter by the disturber units.

Images from the ISD still camera provided evidence for an enhanced invasion of epi/megafauna organisms (e.g., crustaceans, echinoderms) within the first hours and days after ISD perturbations (data not shown). These larger benthic organisms are obviously attracted by uplifted organic matter and/or organisms associated with these sediments. Predation of megafauna organisms should have a clear negative impact on the smaller sediment-inhabiting fauna, which makes the enhanced meiofauna densities recognized in ISD disturbed areas even more striking.

Changes in environmental conditions due to ISD perturbations also noticeably affected meiofaunal compositions, at least for the deeper sediment layers. Compared with background samples the relative abundance of nematodes occurring deeper than 1 cm in ISD perturbed sediments increased from approximately 85% to more than 95% (Fig. 13).

#### Discussion

Experimental studies have been an increasing component of recent marine ecology, however, effects of sediment perturbations on the small benthic biota are still only poorly understood (Schratzberger and Warwick 1999; Christoni et al. 2004). Widdicombe and Austen (2001) examined the interaction between physical disturbance as well as organic enrichment on the diversity and community structure of benthic infaunal assemblages in mesocosm experiments. Sediment perturbations in their experiment were caused by a stirring system similar to what is used in the ISD, although in much smaller dimensions.

Such ex-situ experimental approaches are, however, fraught

with limitations (e.g., restrictions in vertical and horizontal movements of the biota that might affect feeding and escape strategies of organisms). When applied to community processes, the inability of some benthic taxa to persist within mesocosms, and particularly the restricted immigration of organisms may lead to artifacts. Therefore, larger-scale field experimentation is urgently required to obtain further evidence demonstrating the effects of different types of physical disturbance on benthic infaunal communities (Austen and Widdicombe 2006)—the development of the ISD represents a major step in this direction.

Initial trials of the ISD confirmed the successful operation of many of the features of this complex system. Results from sediment analyses within our assessments provided clear evidence for the high mixing efficiency of the disturbing elements in physical (e.g., luminophores, radionuclides), geochemical (e.g., oxygen penetration depths), and biological parameters (e.g., various biogenic sediment compounds, bacteria). Comparisons with field data showed that the ISD is capable to rapidly simulate natural disturbances caused by benthic organisms, as confirmed by studies with luminophores in in-situ and ex-situ experiments; few perturbations created with ISD disturbing elements can mimic conditions comparable to those found for natural communities after 1-2 months. Preliminary results from meiofauna investigations confirmed changes in the community structure in reaction to changing environmental conditions due to ISD induced sediment perturbations.

The development of the ISD will allow a large variety of experiments to be performed, including disturbances at different frequencies (even within one experiment), the study of shifts in geochemical parameters at any time during/between sediment perturbations, and sediment sampling and subsequent biogeochemical and faunal analyses at different time intervals after sediment disturbances.

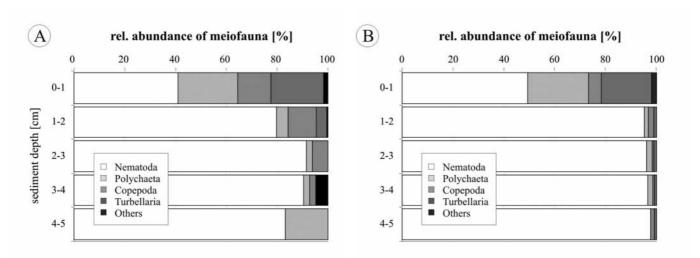


Fig. 13. Meiofauna composition in control samples (A) and ISD disturbed sediments (B) 6.5 weeks after repeated ISD perturbations.

## **Conclusions**

In-situ experiments with the Integrated Sediment Disturber (ISD), developed and built within the EU project COBO, will permit reseachers to study the recovery of geochemical characteristics in perturbed sediments, as well as associated shifts in benthic community structure, in response to changing environmental conditions. This will allow us to assess the vigor and resistance of benthic assemblages to disturbances, and to study the dynamics of recolonization and succession in soft-sediment habitats at different temporal scales. A clear understanding of how episodic and continuous natural disturbances (i.e., bioturbation, "bioengineering") affect the functioning of benthic communities may form the basis for any effective management of coastal ecosystems. In this way, results from ISD experiments will help to establish measures to maintain ecosystem health.

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