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EXtreme ecosystem studies in the deep OCEan :

Technological Developments

Deliverable 7D2

Cruise reports year 2

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Content

<i>WP7- Demonstration Actions</i>	1
<i>3-D reconstruction of small-scale scenes (AWI)</i>	1
<i>WP2 Design of a long-term imaging module (WP2.2)</i>	2
<i>Long-term imaging module, demonstration module (MOB0)s</i>	
<i>WP3, In situ sensors - methane sensor (WP3.3)</i>	4
<i>WP4 INSINC</i>	7



WP7- Demonstration Actions

The objectives of the WP 7 are to organise subsystems testing during the project and to organise the demonstration actions at the end of the project.

The Exocet/d partners participated to several oceanographic cruises during this year: in the Azores on the R/V Archipelago, on the R/V L'Atalante with the ROV Victor 6000 (AWI-ROV 2005 at Håkon Mosby Mud Volcano ; Exomar 05 on hydrothermal vents of the mid-Atlantic ridge), to Mohns Ridge aboard the G.O. Sars in the Norwegian-Greenland Sea, in July-August 2005, to the Costa Rica subduction zone (R/V Meteor cruise M662b with the ROV Quest). Deployments were also done in shallow waters of Wadden Sea near Königshafen on the Sylt Peninsula.

3-D reconstruction of small-scale scenes (AWI)

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Tasks :

The main objective of this WP is to set-up a complete methodology to make projective reconstructions of small-scale scenes from underwater video imagery. 3-D reconstruction through stereo-video technique and stereo-photo methods.

Field tests: R/V L'Atalante

The first field deployments of the stereo camera rig were planned for cruise AWI-ROV 2005 on R/V L'Atalante in September 2005. Only three out of five originally planned dives could be carried out due to bad weather conditions during the better part of this cruise to the Arctic. This restricted the dive time available to all cruise participants and ultimately the time available for field trials of the stereo camera. Our field trial was scheduled for the last dive of the cruise. A test of the manoeuvrability of the stereo camera rig by the Victor 6000 manipulator arm '*maestro*' on deck (Fig. WP2.2a) proved to be successful. Unfortunately, the camera connector turned out not to match the design of the ROV connector. Therefore, an autonomous power supply had to be used. For this purpose, the batteries of the METS (WP3.3), which were already encased in a pressure can were mounted onto the stereo camera rig. Unfortunately, the weather conditions became so bad that the last dive had to be cancelled for an early departure to Tromsø.



Figure ! (a) Stereo camera rig operated by Victor 6000 manipulator arm '*maestro*'; (b) lowering of stereo camera rig to the seabed in Lyngen Fjord.

To test the general functioning and obtain first field data we decided to carry out a deployment in the sheltered Lyngen Fjord. The baited stereo camera rig was lowered from the ship to the sea



floor (20m) by a rope (Fig.1 b). One photograph per minute was taken throughout the 140 min deployment.

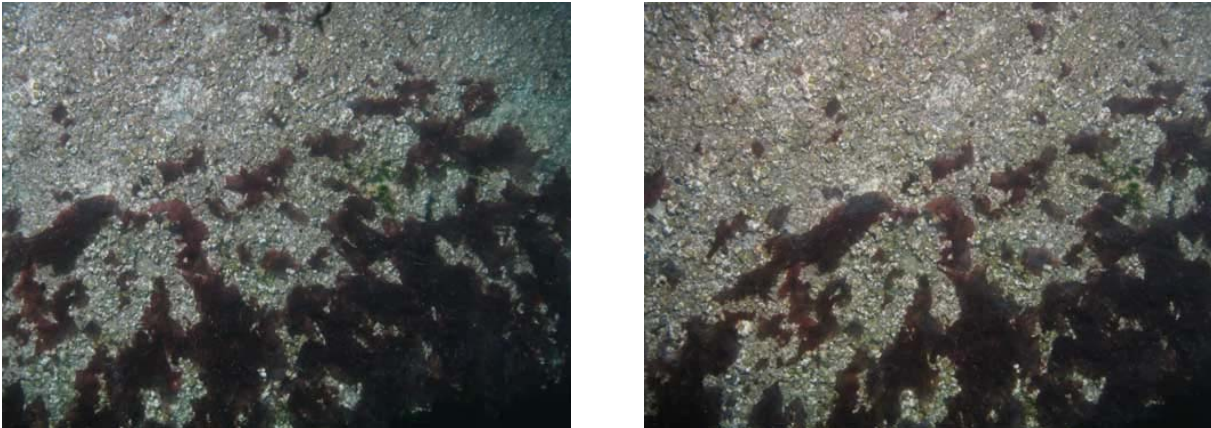


Figure 2 Photographs taken synchronously by the left and right camera during the first field deployment in Lyngen Fjord.

The recovery of the stereo camera rig showed that there were no technical problems. All cameras, cables and the flash returned in good condition. An evaluation of the photographs indicated that the illumination of photographs from both cameras was not the same which may cause problems in the subsequent 3D reconstruction process (Fig.WP2-3). This could have been caused by the angle of the cameras or flash to the seabed. During a ROV dive, the position of the stereo camera rig relative to the seabed could be adjusted, by means of the manipulator arm, which was not possible in this autonomous set-up. By contrast, in this set-up the rig was suspended in the water column and exposed to currents; so there was no control over the camera rig position relative to objects on the seabed. Therefore, some of the photographs turned out dark and blurred.

WP2 Design of a long-term imaging module (WP2.2) ***Long-term imaging module, demonstration module (MOB0)***

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The first leg of the Exomar cruise, held in 2005 on the mid-Atlantic ridge (chief scientist A. Godfroy), allowed the testing of different equipments (MOB0 and protected oxygen optode) developed within the Exocet/d project. MOB0 (a demonstration long-term imaging module) was developed to validate, in the hydrothermal environment, the anti-fouling method developed at ERT/IC on both a camera and lighting system. It was constituted of an aluminum structure (recycled from an old pelagic module), two acoustic beacons (AR391 n° 24 et 25) and an ASSEM energy casing. A Hytech digital still camera, developed for ROV Victor, was installed on the module (Fig. 3). The photo camera porthole was equipped with the antifouling system (ERT/IC, Micrel). Two LED projectors (Dynasub) provided adequate lighting and were controlled by an OSEAN card. One projector was equipped with the anti-fouling system but unfortunately, it did not work in the ship's laboratory and was disconnected before the deployment. The module (Fig. 4) was also equipped with an argos beacon, a gonio and a flash, fixed on the syntactic foam float.



Lentille en mousse syntactique avec flash + gonio
 Bloc de flotteurs cylindriques EUROSORE
 Structure MOB0 avec largueurs et Lest plat intégrés

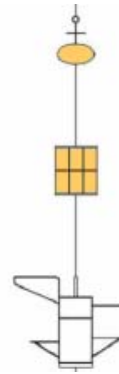


Fig. 3: MOB 0, detail of the digital still camera and of the two LED recover projectors.

Fig. 4: Scheme of the module to

Fig. 5 MOB 0 on the bottom, Exomar cruise, 2005.

The demonstration module (Fig. 5) was recovered after three months on the bottom. The corrosion was important on the structure, particularly near the releasers and on the camera and flash stainless braces. The photo camera porthole was very clean and so was the porthole of N°2 spotlight (control porthole without protection). The porthole of N°1 spotlight (with SnO₂ but without polarisation) was covered by a slight film. The development of biofouling was not important during those three months but the experiment showed that the system worked properly. A SnO₂ coating without polarisation offers a rougher surface than the bare porthole and could favour the formation of a biofilm.

A total of 97 photos were taken. These photos clearly show particle sedimentation or biofouling on the target. The electronics of the MICREL chloration system and of the camera control are not stable when fed by the same energy source. This problem will be addressed further and further testing will be done in deep basin. The reliability of this method over 12 months will be verified next August, during the MoMARETO cruise when the protected Aanderaa oxygen sensor, deployed on a vent mussel community (Fig. 6) will be recovered

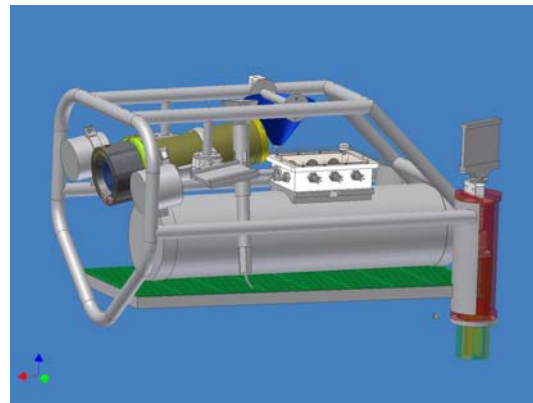
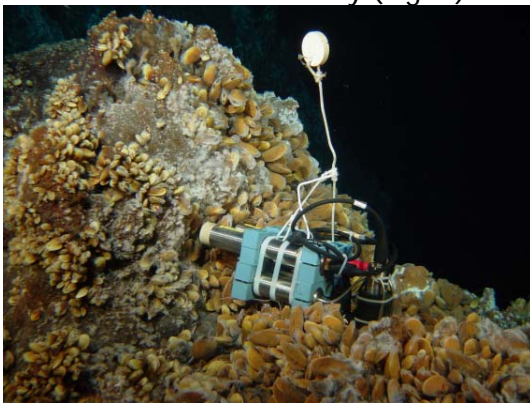


Fig. 6: Mooring with one Aanderaa oxygen sensor, equipped with the antifouling system, and two autonomous temperature probes. Deployed during Exomar cruise in 2005, recovery during MoMARETO cruise in 2006

Fig. 7.: Graphic design of an autonomous video camera module. The EXOCET/D camera will be included in a larger module harbouring different types of sensors and samplers.

The future MOB1 long-term camera module (Fig. 7) will be completed before the MoMARETO cruise, in August 2006. The module will be tested during the first leg of the cruise and if the tests are conclusive, it will be deployed for one year during the second leg of the cruise.

WP3, In situ sensors - methane sensor (WP3.3)

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Field tests

a) Sylt

A METS sensor was deployed several times in the shallow waters of the Wadden Sea near Königshafen on the Sylt Peninsula in co-operation with other AWI projects as well as in the context of the inter-operability of sensor modules (WP5). During one of the trials in August 2005, one of the METS which had been provided and freshly maintained by Capsum was fitted to the moving sea-floor observation system MOVE (Fig. 8), operated by MARUM, University Bremen, Partner #10.

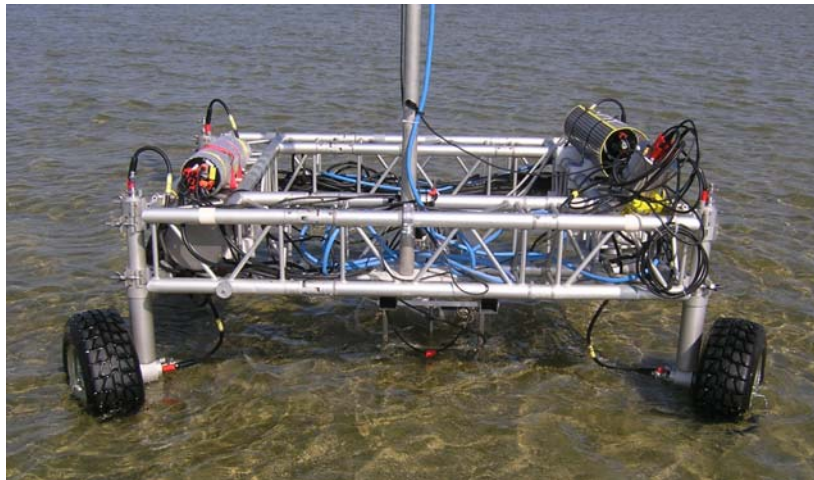


Fig. 8: One of the METS was fitted to the mobile benthic observation system MOVE during a deployment on Sylt Peninsula. The modular system can move in any direction

b) RV Alkor

One of the METS sensors was used during the R/V "Alkor" cruise AL267. The sensor was fitted to both, the tool sled of the ROV (Fig. WP3.13) Cherokee and to a CTD rosette. For these deployments, an oxygen sensor was coupled to the pumping array.



Fig. 9 Deployment of the METS fitted to the ROV Cherokee from R/V Alkor in September 2005.

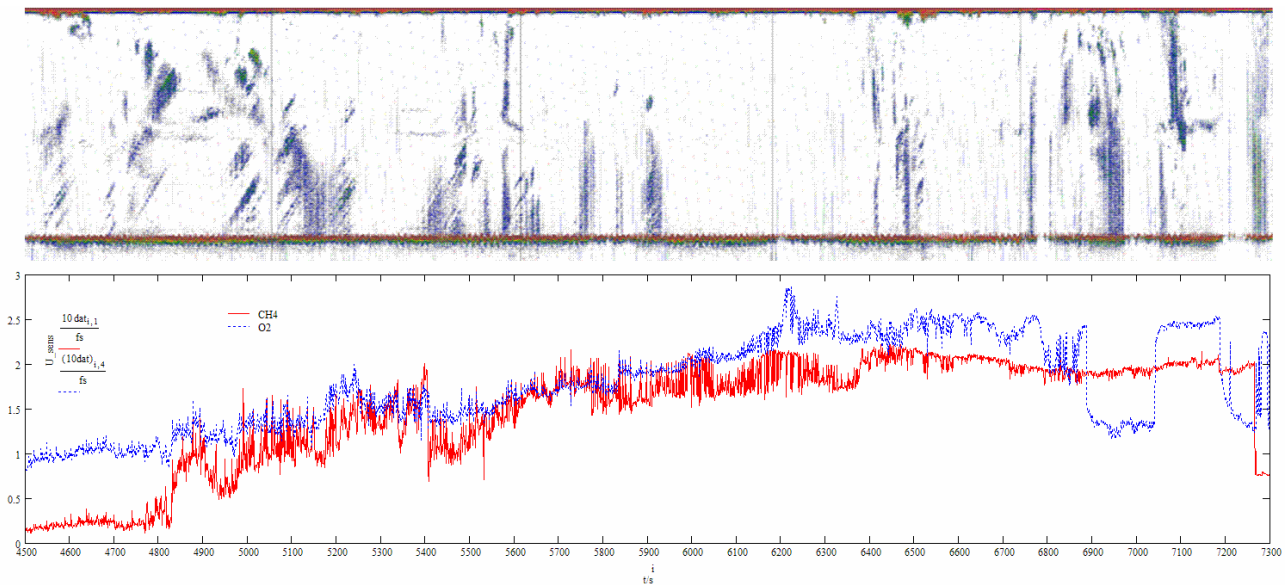


Fig. 10: Methane (blue) and oxygen (red) measurements made above a field of methane plumes. The gas flares appear in the upper part of the graph as blue scatter.

Unfortunately bad weather conditions prevented several of the planned deployments. During a CTD cast, the sensor was moved over a field of methane discharge sites (Fig. 10). At first glance, the sensor's signal is hardly correlated with the acoustic bubble recordings from a fish echosounder. However, the statistical relationship between acoustic plume signals, oxygen and methane values has to be analysed under consideration of the ship's exact course plot and the positions of sensor and echo sounder relative to each other. The methane concentrations may correlate weakly with oxygen concentrations but also show a slow increase over the duration of the entire deployment. This highlights the main problem of METS which lies in the slow response, particularly to decreasing methane concentrations. For more information, please consult the cruise report (Boetius *et al.*, in press).

c) RV L'Atalante

An additional field trial was done during an R/V L'Atalante cruise in September 2005. Here, a METS was integrated into the payload of the AWI AUV planned to record both hydrographic conditions and methane concentrations in the water column above Håkon Mosby Mud Volcano. This volcano discharges large quantities of gaseous methane as gas hydrate coated bubbles. During previous cruises, a methane plume was recorded up to 780 m above the sea floor (Sauter *et al.*, in press). Unfortunately, the weather conditions allowed only for a single deployment in the surface (see cruise report, Sauter *et al.*, submitted). The methane sensor had been configured by Capsum prior to the cruise and was built into the payload section of the AUV. A pumped CTD sensor was connected in series with the METS for simultaneous recordings of temperature, salinity and methane (Fig. 11, left). Unfortunately, several efforts to visit the target area failed due to rough weather and high sea state (Sauter *et al.*, submitted). The only deployment performed further north in surface waters had to be aborted due to rapidly increasing winds (Fig. 11, right). At least, we were lucky to be able to recover the AUV without damage and loss of man. Instead of the planned 3D water column survey above the mud volcano, the trial was performed close to the sea surface, which allowed for an easier communication between the ship and the AUV. The vehicle dived at water depths of up to 30 m which are characterized by very low methane concentrations (background) and air bubbles from enhanced surface mixing due to the stormy weather. This is reflected in the data which also appear to show a drift towards low voltages (increasing methane concentrations) over the entire dive. The final evaluation of the test is not yet available.



Fig. 11: Methane sensor fitted into the AWI AUV. The sensor was connected in series with a pumped conductivity sensor (CTD) leading the ambient water through both sensors (left, middle). Unfortunately, bad weather and sea conditions prohibited the deployment of the AUV at the Håkon Mosby Mud Volcano. The only trial that was possible had to be aborted due to rapidly increasing winds and high swell during the dive of the AUV (right).

EXOCET/D WP4 Report XX

Development and testing of an *In Situ* Incubator (InSinc)

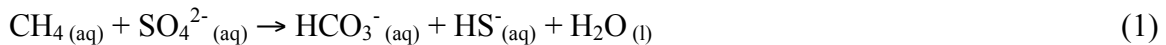
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1. Introduction and Technical Description

The aim of the first year in WP4 was the development of an incubator that allows *in situ* measurements of microbial activity in sediments of marine cold seep systems. Cold seeps are generally characterized by high methane concentrations in the sediment favouring the microbial process of anaerobic oxidation of methane (AOM) according to the following net reaction:



Since the activity of the microbes is determined by the concentrations of methane and sulphate, rate measurements after sample recovery from greater water depths are often biased as a result of depressurisation and methane loss. To overcome this problem it is necessary to conduct such incubations under *in situ* conditions. So far, no such measurements could be accomplished in cold seep ecosystems. Radiotracer measurements are a standard tool to determine microbial turnover rates. For determining AOM activity, ¹⁴C labelled methane and/or ³⁵S labelled sulphate are microbially transformed into ¹⁴CO₂ and H₂³⁵S during incubation. With the respective concentration of total methane and sulphate, the radiotracer turnover rates over time determine the activity of AOM and sulphate reduction (SR). An additional problem is the use of methane tracer (¹⁴C-labeled methane gas dissolved in water), because decompression problems would also affect concentrations of ¹⁴C methane in cores, but not ¹⁴CO₂, the product of the reaction. At cold seeps, methane is commonly the sole electron donor. Hence, AOM and SR occur in a 1:1 relation. Furthermore, a possible discrepancy between AOM and SR can be measured *ex situ* on board. Therefore, the InSinc module is first optimised for sulphate tracer applications, and after successful application, its use for AOM and methanogenesis rate determinations will be tested.

We therefore constructed in collaboration with the German company “Meerestechnik Bremen GmbH” a prototype of an incubator that allows the *in situ* (i.e., at depth) injection and incubation of radiotracer in surface sediments (0-40 cm). The in situ incubator (InSinc, Figs. 1) was designed for applications on remotely operated vehicles (ROVs) and submersibles. This α -prototype contains electrical parts such as pumps and batteries. Hence, the size and weight was unfavourable for deployments by most existing ROV and submersible systems. Furthermore, electrical systems may display several

malfunction under *in situ* conditions. Because of these constraints, we decided to construct a smaller, fully mechanical device leading to an improved β -prototype. The core of the new InSinc module consists of a needle sheath, in which small syringes with a length of 1 cm are packed in a row. The syringes are ejected and loaded by the release of powerful springs in the head of the module. On board, the syringes within the injection needle are filled with $^{35}\text{SO}_4^{2-}$ -tracer. The system, which resembles a standard push core in size and functioning, is then carried by a ROV or submersible to its deployment site. For deployment the module, consisting of head and push core unit, is drawn out of its sheath and a sample is collected by coring a site of choice. In the coring process, the injection needle is plunged into the sediment. The module with the sediment sample is put back into the sheath before the tracer injection is mechanically triggered by releasing the springs in the module head. For an effective *in situ* incubation, InSinc has to stay for at least 4-6 hrs at *in situ* depth and temperature. The incubation is stopped on board, after the recovery of the ROV or the lift (depending which instrument carries the cores). To stop the incubation and fix the sediments, the sediment core is split into defined depth intervals and collected into test tubes prepared with a fixation fluid. The functioning of the tracer injection can be determined immediately by measuring the radioactivity in the supernatant of the sample. The microbial turnover of tracer is then measured in the home laboratory.

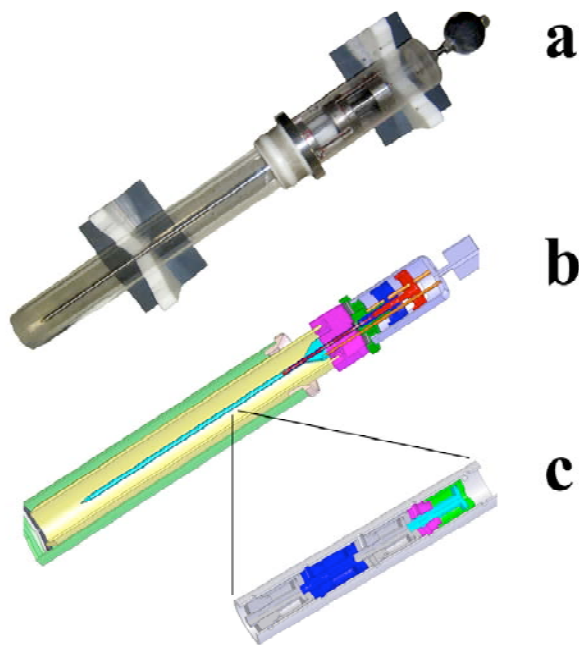


Figure 1. The InSinc module. (a) photograph of the prototype β -version. (b) Schematic drawing of the sheath (green), push core unit (yellow), injection needle (cyan), and the modules head (multi-colour). Panel (c) shows a close up of the injection needle showing the ca. 1 cm long syringes, which are packed in the needle sheath.

2. *In situ* Deployment

After the development and modification phase of InSinc in 2004 and 2005, the next step was a functional test in the field. For this purpose, we joined the R/V Meteor cruise M66-2b with the ROV Quest to the Costa Rica subduction zone (Fig. 2). This area is characterised by a high abundance of seep related structures and was therefore an ideal area for *in situ* measurements on cold seep ecosystems.

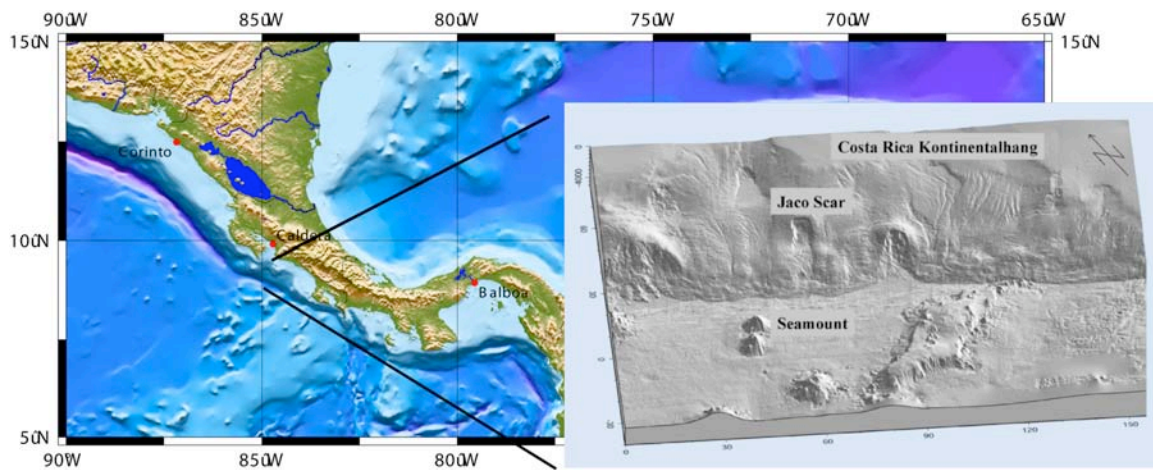


Figure 2. The Costa Rica subduction zone. Seep related features such as slide scars and mounts are indicated. Modified from Brückmann and Niemann (2005) and Rehder (unpubl.)

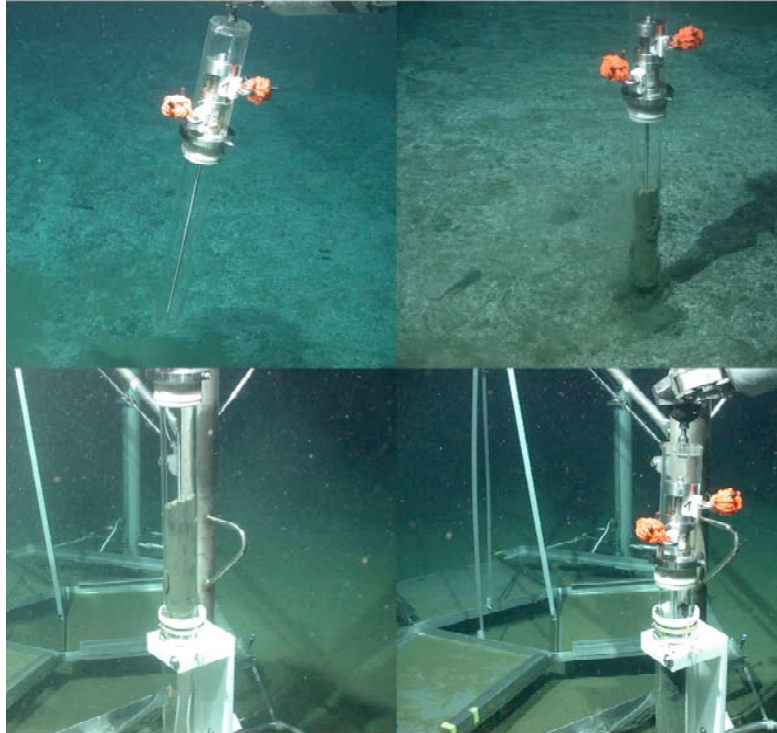
Table 1. Deployments of InSinc

Structure	Lat.N	Long. W	depth
Md. Iguana	11° 12.5'	87° 09.5'	169, 180
Jaco Scar	09° 07.5'	84° 50.4'	ca. 1000 - 2400

In total, we were able to deploy InSinc on two dives, one targeting Mound Iguana and on targeting the Jaco slide scar (Tab. 1). Fig. 3 shows images from the deployment at the Costa Rica stations. Both systems are characterised by elevated fluxes of methane. At both seep structures, the module was placed onto bacterial mats, which indicate elevated sulphide fluxes, possibly derived from anaerobic oxidation of methane. In order to estimate a potential offset between *in situ* and *ex situ* rates push cores were taken in close

proximity to InSinc from which *ex situ* SR rates and diffusive methane fluxes are going to be determined.

Fig 3. InSinc in Action. Upper left: the core is targeted to a bacterial mat. Upper right: a sediment sample is collected by push coring. Lower left: The core is transferred to its container mounted to a lander frame. Lower right: The core is stored safely and the incubation is started by releasing the spring on the core head.



3. Results and Discussion and Outlook

Directly after retrieval, the sediment in the InSinc core was tested for tracer activity. These test showed that the module successfully injected tracer in the sediment (data not shown). However, the variability of tracer injection per volume of sediment is still a bit high, with about 40% variation around the average (average injection is 40000 dpm per ml, i.e. 0.5 kBq g⁻¹). Figure 4 shows the first *in situ* SR rate measurements from cold seep systems.

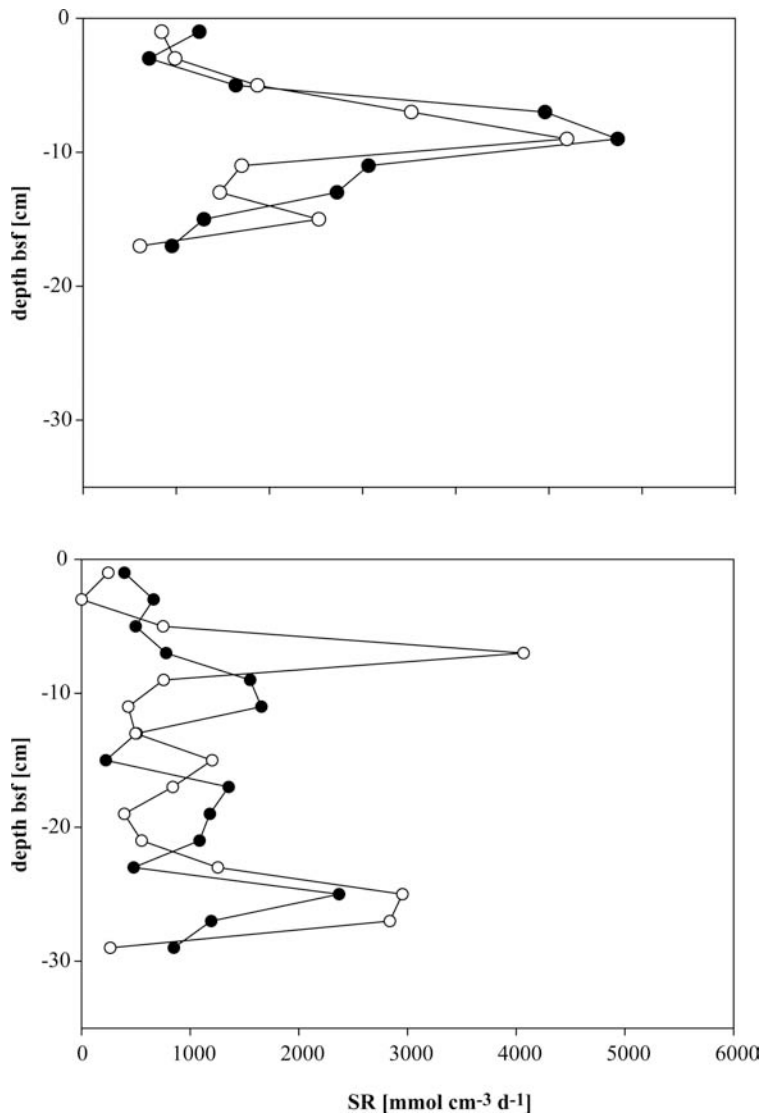


Figure 4. *In situ* SR rates determined from InSinc deployments at Md. Iguana (a) and the Jaco Scar (b). Rate measurements were performed in duplicates (open and closed symbols).

The rate measurements are apparently reproducible as shown by a comparably low data scatter. The somewhat higher scatter encountered at Jaco Scar could be a result of a very small scale environmental heterogeneity as often encountered at cold seeps. The rates are within the top range encountered at cold seeps. Although comparative results are not available yet (see section 2), it may very well be that the *in situ* rates are significantly higher in comparison to *ex situ* rates. The reason may be higher availability of methane *in situ*, before degassing induced by core recovery. However, the availability of electron acceptor may also be lower at *in situ* conditions. It was observed that the ebullition during degassing may introduce bottom water into the core.

As a next step, final improvements on InSink are undertaken. This includes a further reduction of the sizes of module head and core length in order to fit multiple cores into ROV-carried boxes, similar to those, which contain regular push cores. On cruise M66-2b the overall length of a single system (70 cm) required to mount the module on a shuttle frame, as the ROV manipulator was not have been able to operate the module in a vertical position. For 2006, at least two further cruises are planned at which 4 improved InSink modules will be deployed. With this approach, we will be able to statistically determine biases of *ex situ* rate measurements and flux calculations.